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Abstract & Indexing Information

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Conceptual shifts in animal systematics as reflected in the taxonomic history of a common aquatic snail species (*Lymnaea stagnalis*)

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Abstract

Lymnaea stagnalis (L., 1758) is among the most widespread and well-studied species of freshwater Mollusca of the northern hemisphere. It is also notoriously known for its huge conchological variability. The history of scientific exploration of this species may be traced back to the end of the 16th century (Ulisse Aldrovandi in Renaissance Italy) and, thus, *L. stagnalis* has been chosen as a proper model taxon to demonstrate how changes in theoretical foundations and methodology of animal taxonomy have been reflected in the practice of classification of a particular taxon, especially on the intraspecific level. In this paper, I depict the long story of recognition of *L. stagnalis* by naturalists and biologists since the 16th century up to the present day. It is shown that different taxonomic philosophies (essentialism, population thinking, tree thinking) led to different views on the species' internal structure and its systematic position itself. The problem of how to deal with intraspecific variability in the taxonomic arrangement of *L. stagnalis* has been a central problem that made systematists change their opinion following conceptual shifts in taxonomic theory.

Key Words

Introduction

The development of science as a whole, as well as the progress of a particular scientific discipline, is a complicated and diverse process with many separate aspects (Hull 1988) that permit several distinct ways to represent the history of science. In my opinion, at least three approaches are imaginable in this case. Firstly, any history is a *story* telling us about a temporal sequence of events. It means that a narrative approach, which is merely an account of persons and their discoveries, is inevitable. Secondly, one may focus on conceptual shifts in scientists' minds that reflect the theoretical rather than temporal development of science. This approach presupposes a study of continuous changes in scientific concepts and ideas as well as in the methodological foundations of the art of doing science. It may well be a non-linear process since the development of theories does not always run parallel with the progressive sequence of events. Lastly, any description of the practical aftermath of these conceptual shifts may be considered as the third approach to the history of science. It encompasses the "external" manifestations of scientific activity, including modes of representing of knowledge, scientists' social interactions, university curricula, working classifications of studied objects, and so on.

Biological systematics is, probably, the oldest of the branches of life sciences. Its roots may be traced back to the pre-scientific epoch, since so-called "ethnotaxonomy" was just the first attempt to capture biological diversity by using more or less implicit categories and vernacular names (Atran 1990). The picture of the development of biological systematics is usually drawn following either a strictly narrative or conceptual (a history of ideas) approach (Stevens 1994; Wilkins 2009; Pavlinov and Lyubarskiy 2011), but this process has other inter-

esting sides. It includes also the histories of the scientific exploration of particular taxa and their appreciation by practicing systematists. Some of these taxa are so spectacular and valuable practically that the history of their taxonomic treatment is as long as the history of systematics itself. Other species are less important, especially for folktaxonomists, and their taxonomic study started later in the epoch of the early systematics of the sixteenth and seventeenth centuries. To reconstruct the taxonomic history of an individual taxon is an important challenge allowing one to understand deeply the historical development of biological classifications and their practical issues (Schmidtler 2011).

The aim of this paper is to outline the taxonomic history of a widespread and commonly known invertebrate species in Europe and North America – the great pond snail, *Lymnaea stagnalis* (Linnaeus, 1758). The material for the study was obtained during my work with malacological collections of the Zoological Institute, Russian Academy of Sciences (Sankt-Petersburg; ZIN hereafter), Göteborgs Naturhistoriska Muséet (GNM hereafter), Vienna Museum of Natural History, Austria (NHMW hereafter), Naturhistoriska riksmuseet, Stockholm (Sweden), and Zoological Museum of the Copenhagen University, Denmark (ZMUC hereafter). These collections contain a large number of samples of *L. stagnalis* collected and identified by prominent malacologists of the end of the 18th – the first half of the 20th centuries, and examination of these materials helped me to understand how the taxonomists' views changed with time and to trace these changes by analysis of the information available from museum labels. The extensive search through old taxonomic literature has been carried out as well. I used the books kept in ZIN and NHMW libraries and utilized those fantastic facilities provided by electronic archives such as Biodiversity Heritage Library (http://www.biodiversitylibrary.org/).

It is a freshwater pulmonate snail (Fig. 1) using atmospheric air for breathing that allows it to migrate over long distances and to gain a foothold in waterbodies of

Figure 1. A great pond snail in its natural environment. 12.08.2014. Russia, Western Siberia, "Malaya Sos'va" Nature Reserve, Kopanoye Lake (photo: M. Vinarski).

different types. *L. stagnalis* is characterized also by extreme ecological plasticity and enormous variation in its shell size and proportions (Kobelt 1871; Hubendick 1951; Arthur 1982; Vinarski 2014a). Being, very likely, the largest species of freshwater snails in Europe, the great pond snail had attracted the attention of naturalists long before Linnaeus. The first scientific description of this species appeared nearly 400 years ago (see below). The taxonomic history of *L. stagnalis* will serve here as a mirror to reflect shifts in taxonomical practice driven by conceptual changes in animal taxonomy which occurred between the 16th and 21st centuries.

Lymnaea stagnalis in pre-Linnaean zoology

Though the accepted scientific name of this snail should be credited to Linnaeus (1758), it had been repeatedly described under different names by predecessors of the great Swede. It seems a bit strange that such a large and abundant snail was utterly overlooked by the Ancient naturalists. Though, as Eduard von Martens (1860) noted in due time, the Greeks and Romans had very little interest in continental mollusks, both aquatic and terrestrial. The Ancient naturalists generally neglected them, and neither Aristotle nor Pliny the Elder nor any of the secondary Ancient authors described continental mollusks in detail. Aristotle, in his influential *Historia animalium* (HA) mentions only a certain kind of "lake oysters" (*limnostrea*, see HA IV, 40, 67) as well as some obscure "land ostracoderms" not divided into species (? helicid snails; see HA IV, 38).

The only (and rather curious) alleged mention of the pond snail in Antiquity belongs not to a naturalist but to an anonymous poet who was the author of the mock poem *Batrachomyomachia* ("Battle of Frogs and Mice") intended to mimic the Homer's masterpiece *The Iliad*. The poet describes the armour of the Frogs preparing to battle with the Mice:

> "…their bucklers were
> Good thick-leaved cabbage, proof ' gainst any spear;
> Their spears sharp bulrushes, of which were all
> Fitted with long ones; their parts capital
> They hid in subtle cockleshells from blows"
> (Chapman 1888: 10).

The German malacologist Menke (quoted after Jeffreys 1862) tried to determine the identity of these *subtle cockleshells* mentioned by Pseudo-Homer. He supposed it may have belonged to *Lymnaea stagnalis*. This hypothesis was criticized by Jeffreys (1862), who said that it is quite impossible to judge conclusively on this subject. The Greek text is so concise that it gives no chance to choose among large species of aquatic snails inhabiting Greece and to decide which of them provided the Frogs with their helmets. As Jeffreys (1862: 113) stated, "it
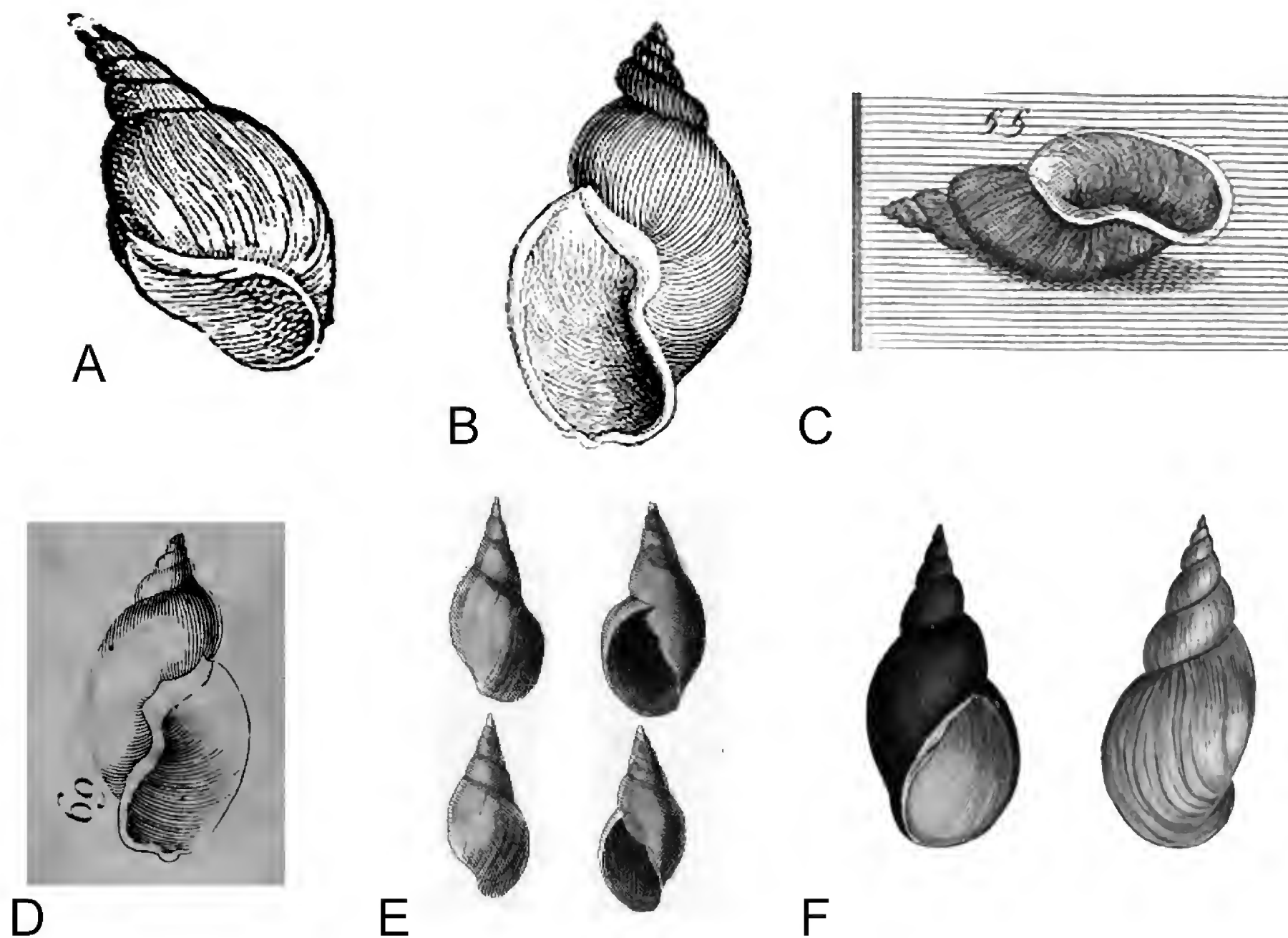
Figure 2. Evolution of accuracy in illustrations of *L. stagnalis* shell through two centuries. Sources of images: **A**. Aldrovandi 1606. **B**. Lister 1678. **C**. Bonanni 1681. **D**. Klein 1753. **E**. Seba 1758. **F**. Schröter 1779.

is not likely that Homer was a conchologist, or distinguished one shell from another for poetical purposes. The kind of shells in question must have made cumbersome helmets for the valiant Frogs".

Medieval descriptive zoology, being deeply dependant on the works of Aristotle and other Ancient naturalists, overlooked *L. stagnalis* as well. One need only envisage the image of this snail in the very crude drawings of aquatic snails which appeared at the end of the fifteenth century (see Allmon 2007, fig. 2). It is impossible, though, to judge on their true taxonomic identity with any certainty.

The first naturalists of early Modern Europe interested in aquatic animals, namely Pierre Belon (1517–1564), Guillaume Rondelet (1507–1566), and Konrad Gessner (1526–1565), did not mention the great pond snail either. Though these authors were not slavish commentators of the Greek and Roman texts and added their own observations on aquatic creatures, they were more interested in marine mollusks than in freshwater ones.

The first record of the great pond snail in the European scientific literature I managed to find is that by Ulisse Aldrovandi (1522–1605), a junior contemporary of Belon and Gessner. His posthumous treatise *De reliquis animalibus* (Aldrovandi 1606) contains a mention of this species that opens a long list of *L. stagnalis* recordings in early animal systematics. Aldrovandi's book also contained the first "scientific" illustration of the *L. stagnalis*

shell (Fig. 2A) that allows us to ascertain its taxonomic identity definitely. It does not matter that the shell in this picture is sinistral (pond snails have normally dextral, or right-coiled, shells). Though sinistral mutant individuals are sometimes found in *L. stagnalis* populations (Vinarski 2007), I believe that Aldrovandi or his engraver had a normal (i.e. dextral) shell of this snail in their hands. The usual technique of engraving in the sixteenth and seventeenth centuries demanded that the plate must be a mirror image of the object to be illustrated. The printers usually were "not preparing a reversed engraving (on wood or copper), but carving the image [of a shell] as it appeared, which would produce a reversed image when printed" (Allmon 2007: 175). The biological mechanism of inversions in shell coiling in snails was not known and thus remained irrelevant for authors which permitted them to present their shells in "wrong" mirror appearance. The picture of *L. stagnalis* shells with right coiling direction did not appear in a printed book until 1681 (see Fig. 2C).

Aldrovandi (1606) gave no formal description of the great pond snail. More precisely, the Latin name of the snail ("Turbo laevis item in stagnis degens"; Aldrovandi 1606: 358) served as its proper description at this time. The early taxonomists were far from using binomial nomenclature consistently, and the species' names produced by them were *polynomials*. Each polynomial should contain several words whose quantity corresponded to the number of es-

Table 1. Polynomial taxonomic names proposed for the great pond snail in the 17th and 18th centuries.

Author	Latin name	English translation
Aldrovandi 1606: 358–359	Turbo levis item in stagnis degens	*Turbo* with smooth shell, living in stagnant waters
Lister 1678: 137	Buccinum longum 6 spirarum, omnium & maximum & productius, subflavum, pellucidum, in tenue acumen ex amplissima basi mucronatum	*Buccinum* with long [shell] having six whorls, whole, large, oblong, yellowish coloured, transparent, [apex] sharp and narrow, [shell] basis very ample
Bonanni 1709: 453	Longior antecedenti Turbo, levissimus, colore atro cum nitore	*Turbo* longer than the antecedent [species], smoothest, [shell] dark colored, glossy
Gualtieri 1742: [34]	Buccinum fluviatile, testa tenuissima, & fragillissima, prima spira notabiliter ventricosa, & elongata, in mucronem aculeatum statim definens, subflavum, pellucidum	*Buccinum* riverine, shell very narrow and fragile, the first whorl notably inflated and oblong, [shell] ends with sharp apex, yellowish, transparent
Linnaeus 1746: 374	Cochlea testa producta cuminata opaca, anfractibus senis subangularis, apertura ovata	*Cochlea* with elongated dark coloured shell having six subangulate whorls and ovale aperture
Seba 1758: 119	Cochlea fluviatilis, indigena, ex oblongo acuminata, lineolis veluti taeniata	*Cochlea* riverine, native [= European], with oblong and pointed shell, covered by thin lines
Klein 1755: 54–55	Auricula stagnorum – subflava, pellucida, in tenue acumen ex amplissima basi mucronata	*Auricula stagnorum* – [shell] yellowish and transparent, with pointed apex and very wide [shell] basis
Schlotterbeccius 1762: 283	Turbo fluviatilis major, corpore oblongo ampullaceo definente in mucronem acutissimum & limacem continente fuscum	*Turbo* riverine, large, body [= shell] oblong, inflated [in its base] and ending by a sharpest apex; it contains soft body of contunuosly dark coloration.
Geoffroy 1767: 72	Buccinum testa oblonga, fusca, anfractibus senis	*Buccinum* with oblong shell of brownish black colour, having six whorls
Favart d'Herbigny 1775: 139	Buccinum fluviatile, testa tenui et fragili, forma oblonga, ventricosa; sex spiris exertis parum convexis in apice acuto definentibus compositum; colore corneo, pellucido, apertura spatiosa, elongata, integra, et labio expanso distinctum	*Buccinum* riverine, its shell is narrow and fragile, oblong and inflated; it consists of six slightly convex whorls ending with a sharp apex; [shell] horny-coloured, transparent, aperture ample, elongated, whole, differs by a wide lip
Chemnitz 1786: 166	Helix <...> testa albida, pellucida, superne turrita, inferne ventricosa, apertura effusa seu ampliata, columella sinuosa	*Helix* <...> [with] whitish pellucid shell, in its upper part it is turreted; the lower part is inflated. Aperture ample or wide; columella folded

sential characters needed to be revealed in order to express what the species *is* and how to distinguish it from its congeners. The more species there were in a genus, the longer the species' names had to be produced (Pavlinov 2013). A polynomial is a name bearing the diagnosis of the taxon itself and it is very far from the Linnaean binomen, which is merely a useful verbal label serving to be remembered quickly (Vinarski 2013). The Linnaean name of the species under discussion, *Helix stagnalis*, contains no information about the *essence* of this species since it highlights the ecological preference of the great pond snail to live in stagnant waters. Certainly, this characteristic may be applied to numerous other species of European aquatic snails.

The essence of a taxon was an Aristotelian category not seen directly by the eyes, but being a mental construction based on the subjective weighting of animal characters aimed to distinguish between essential and secondary (accidental) ones. A modern scientist would say the essential diagnosis is a hypothesis since different authors may come to different views on which characters are essential and which are accidental. Therefore there was no commonly accepted scientific name for the great pond snail before Linnaeus' (1758) work. I collected a series of different polynomial names proposed from 1606 to 1786 for the designation of this species (it is by no means complete), and it is easy to see that no two authors had the same definition of its "essence" (Table 1).

The next step in the study of the great pond snail was undertaken almost 70 years later, in England. Martin List-

er (1639–1712) was an English physician and naturalist, and vice-president of the Royal Society. He contributed extensively to many branches of science, including arachnology (Roos 2011) and chemistry (Roos 2008). Lister was a devoted conchologist and became the author of the first European treatises on mollusks (*Historiae Conchyliorum*, 1685; *Conchyliorum Bivalvium*, 1696). O.F. Müller (1774: xiii) called him "Conchyliologorum princeps" (head of conchologists) proposing a clear analogy with Linnaeus' informal title "Princeps botanicorum". Lister made numerous observations of mollusks' morphology (both external and internal), feeding, ecology, and distribution. His conchological works also included a description of the fossil species of shells.

In Lister's *Historiae animalium Angliae tres tractatus* (Lister 1678), one may find a detailed account of *L. stagnalis* that follows much higher standards of zoological descriptions compared with Aldrovandi's. This text contains not only the polynomial name (= short diagnosis) for this species but also a relatively long two-page sketch of the great pond snail's bionomics. Lister provides a lengthy general description of the animal's external morphology (including the pattern of mantle pigmentation), the shape of its excrements, the mode of copulation, the structure of egg-masses alongside a list of aquatic plants being its food. Some localities of *L. stagnalis* in England were also mentioned. Lister's species' account was almost 100 years ahead of its time. This high standard of publication of malacological data was not established un-
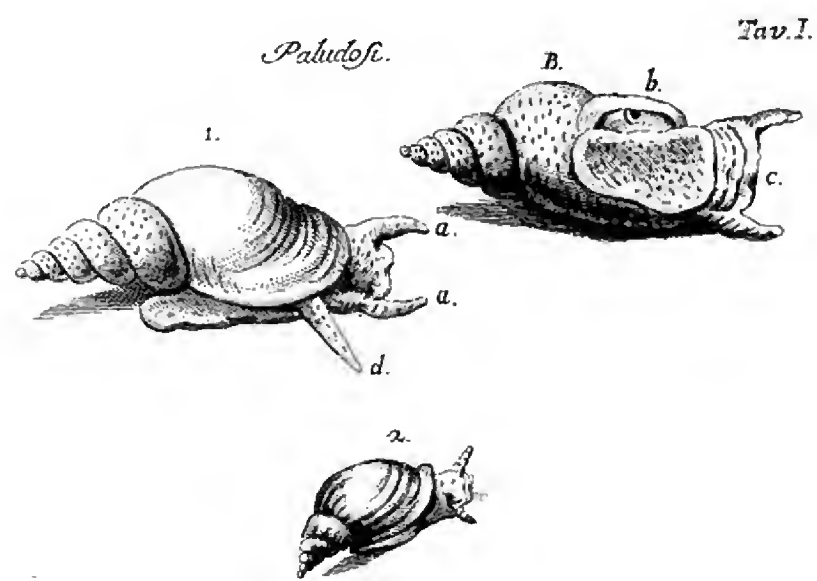
Figure 3. Illustrations of living *L. stagnalis* by Ginanni (1757).

til the end of the 18th / early 19th centuries when comparable works of European naturalists appeared (Müller 1774; Draparnaud 1805).

The picture of the great pond snail shell given by Lister (1678) is more informative than that by Aldrovandi though it is still rather crude and depicts a sinistral shell (see Fig. 2B).

Later on, Lister (1695) published a very detailed account of the *L. stagnalis* internal structure accompanied by engravings. Trained as a *medicus*, Lister was a brilliant anatomist aiming to dissect mollusks belonging to different taxa, both terrestrial and aquatic. He even estimated his own purely medical works as being of lower importance than his studies of molluscan anatomy (Heppel 1995). Another perfect anatomist of the age, Jan Swammerdam (1637–1680), was also interested in freshwater mollusks, and his study of the *L. stagnalis* anatomy was published posthumously in the author's prominent book *Bybel der natuure* (Swammerdam 1738). However, in both cases the advance of anatomical research did not enhance the progress in taxonomy. Systematists of the 17th and 18th centuries typically did not use anatomical information in their works, and the classification of mollusks long remained purely conchological (Vinarski 2014b). The use of anatomical data in lymnaeid taxonomy did not start until the first half of the 20th century (Baker 1911; Roszkowski 1914).

Most subsequent authors, whose works were published between the works of Lister and Linnaeus, were mere collectors of deposited shells of *L. stagnalis* and other freshwater species in their private museums and shell "cabinets". This form of hobby was extremely popular among European noblemen and the educated part of the middle class in the 17th and 18th centuries (Dance 1966). Some of these proud collectors published voluminous books targeted to present their treasures to a wide audience. Sometimes such books contained high quality hand-colored illustrations and therefore were extremely expensive (Dance 1966). Typically, their authors did not give lengthy accounts on species' morphology and bionomics and restricted themselves to the simplest scheme of exposition: a short diagnosis (i.e. polynomial name) of a species plus a picture of its shell. This scheme was used,

among others, by Bonanni (1709), Gualtieri (1742), and Seba (1758). The quality and accuracy of shell images greatly increased through the 17th and 18th centuries, with the most accurate illustrations appearing in the 1770s (Fig. 2). Sometimes, rather realistic portraits of living snails appeared; for example, those of crawling pond snails in Ginanni's (1757) posthumous book. Ginanni's image is morphologically correct and depicts some important details such as the shape of tentacles and the respiratory opening of the animal (Fig. 3 compare with Fig. 1).

Klein (1753) was the first author to separate lymnaeid snails into a taxon of their own – the genus *Auricula* with three species included. Before Linnaeus' seminal work (Linnaeus 1758), Klein already used binomial nomenclature and introduced the first two-part name for the great pond snail – *Auricula stagnorum*. This name has a formal priority before the Linnaeus' *Helix stagnalis* but, being published before 1758, it was not available for taxonomical and nomenclatorial purposes.

Post-Linnaeus taxonomy: discovery of an intraspecific variation

Carolus Linnaeus, the Swede, was a great botanist and reformist of biological taxonomy, but his malacological (or, more correctly, conchological) works received rather low esteem among next authors. For example, Maton and Rackett (1804: 175) stated that "there has been a very general belief that less attention was devoted by Linnaeus to the history and arrangement of the Testacea than to any other order of the animal kingdom, and that he even thought their external coverings, or shells, scarcely worthy of becoming subjects of scientific distribution". Donovan (1807) expressed his disgust with the Linnaean conchological works in stronger phrases: "an opinion is pretty generally prevalent that less attention was devoted by Linnaeus to the history and arrangement of the testacea, than any other order of nature; and that he even thought them unworthy of becoming objects of scientific arrangement. These points have been contested. The truth however dill appears to be, that Linnaeus had not really bestowed much critical attention on this subject <...> When therefore the completion of the Systema required that some attention should be paid to testaceology, he was unprepared, and referring to the authorities of others, comprised this department in the smallest compass possible, more with the view of filling up a chasm, which the omission of a tribe so generally admired would occasion, than from any idea of elucidating the subject <...> It is time we should lay aside the trammels of servile adherence, and speak decidedly: – those early attempts of this celebrated writer, we do not scruple to say, if examined with candour, will be found only *a slight and ill conceived compendium* of what has been handed down to us by antecedent writers" (italics added by me).

The examination of several descriptions of *L. stagnalis* found in Linnaeus works (Linnaeus 1746, 1758, 1761,

1767) allows one to agree with the abovequoted words. Linnaeus did not move forward as compared with his predecessors (except for Lister). In 1746 he still used polynomials as the means of species designation (see Table 1). Since 1758, his species descriptions became two-part: the short binomial name proposed for the sake of utility accompanied by more detailed diagnosis being, in essence, nothing other than a traditional polynomial name (Pavlinov 2013). Thus, the great pond snail was christened in 1758 as *Helix stagnalis* and provided with a short diagnosis "*H*[*elix*] testa imperforata ovato-subulata subangulata, apertura ovata" (Linnaeus 1758: 774). Subsequent editions of "Systema Naturae" brought no new information on the great pond snail (Linnaeus 1767; Gmelin 1791).

Linnaeus and his immediate follower Johann Friedrich Gmelin (1791) did not recognize any variation within the species *Helix stagnalis*. The diagnosis of this taxon was presented as a list of essential conchological traits as if all snails were completely identical in their shell appearance. Though Linnaeus himself paid much attention to the problem of intraspecific variation in his theoretical works (i.e. in *Philosophy of Botany*; Linnaeus 1751), he was interested mainly in variations in plants. In the zoological part of *Systema Naturae* only a few animal species were mentioned as having varieties in their structure (notably, *Homo sapiens* Linnaeus, 1758 was among these species).

As a rule, animal taxonomists of the second half of the seventeenth century were still not aware of the existence of intraspecific variation (Vinarski 2013), though the most gifted observers among them knew well that there is some degree of phenotypical heterogeneity within common and widespread species. Possibly, the Danish naturalist Otto Frederick Müller (1730–1784) was the first student of non-marine mollusks to make an attempt to reflect this heterogeneity in a taxonomic work. Müller's opus magnum *Vermium terrestrium et fluviatilium* (Müller, 1774) became the most influential monograph on terrestrial and freshwater mollusks published between *Systema Naturae* and the dawn of the nineteenth century. Müller was a really great naturalist with a special interest in aquatic creatures, including infusorians, rotifers, crustaceans, and mollusks. He was the first naturalist to use a dredge for sampling benthic animals of inland waterbodies (Anderson and Rice 2006). Müller's species descriptions were rather lengthy and informative though *Vermium terrestrium et fluviatilium* did not contain illustrations of any described objects. The Dane established a new standard of arrangement of intraspecific variation in malacological treatises. His approach presupposed the enumeration of as many intraspecific varieties as was possible, each given a polynomial name serving as the diagnosis. The Greek alphabet letters were used for formal designation of these varieties. In some species of mollusks, especially of terrestrial ones, a number of such varieties could be rather high. For instance, Müller (1774) could distinguish 27 varieties of the common European garden snail, *Cepaea nemoralis* (L., 1758), designated by combinations of Greek letters, α to δδ. In aquatic snails, Müller (1774) identified far fewer varieties, and no intraspecific group was described

within *L. stagnalis* at all. Nevertheless, this approach was accepted by subsequent students of freshwater mollusks (Draparnaud 1805; Pfeiffer 1821; Nilsson 1822).

At the end of the 19th century, the practice of recognition of varieties in *L. stagnalis* reached its summit in the works of S. Clessin (1884, 1887) and C. A. Westerlund (1885, 1897). Westerlund was, perhaps, the most prolific maker of varieties. He was able to determine as many as 19 varieties of the great pond snail in the Scandinavian region alone (Westerlund 1897), though he did not give a key for their exact categorisation. The users of his taxonomic catalogues were provided with only German or Latin diagnoses of the varieties with no chance to know the characters to distinguish among them precisely. The readers of Clessin's books (1884, 1887) were in a slightly more favourable position since the author illustrated shells of most varieties. This simple scheme of „species and its varieties" was applied in several influential malacological works of the 20th century; for instance, in those by Ehrmann (1933) and Zhadin (1952).

Having searched through the old malacological literature, I compiled a synonymy of *L. stagnalis* s. lato that contains nearly 80 varieties of this species described between the 1820s and 1920s (Electronic Appendix 1) A closer examination of this „zoo" reveals a plethora of causes serving as grounds for establishing new varieties. I had an opportunity to examine many of these varieties using samples of *L. stagnalis* identified by malacologists of the 19th century (Fig. 4) and now kept in the Europe's scientific institutions. It gave me a possibility to outline a rough classification of the varieties depending on the basis of their recognition. At least seven large groups of varieties may be separated:

1. Those reflecting variation in shell size. Example: *L. stagnalis* var. *major* Moquin-Tandon, 1855. Sometimes, shells of knowingly juvenile individuals were described as a distinct variety (*L. stagnalis* var. *junior* Nilsson, 1822).
2. Sinistral mutants: *L. stagnalis* var. *sinistrorsa* Jeffreys, 1862.
3. Variants of the shell surface colouration: *L. stagnalis* var. *bicolor* Hartmann, 1840; *L. stagnalis* var. *roseolabiata* Beck, 1837; *L. stagnalis* var. *fasciata* Merkel, 1908.
4. Varieties based on shell proportions: *L. stagnalis* var. *ampliata* Clessin, 1876; *L. stagnalis* var. *producta* Colbeau, 1859.
5. Varieties based on peculiarities of shell structure, including structure of aperture and umbilicus, whorls' shape: *L. stagnalis* var. *umbilicata* Hutton, 1905.
6. Varieties based on ecological preferences: *L. stagnalis* var. *lacustris* Studer, 1820; *L. stagnalis* var. *alpicola* Gredler, 1859
7. Geographical races: *L. stagnalis* var. *bottnica* Westerlund, 1884; *L. stagnalis* var. *gallica* Bourguignat, 1864.
8. Varieties based on conchological similarity with other lymnaeid taxa: *L. stagnalis* var. *palustriformis* Kobelt, 1870.
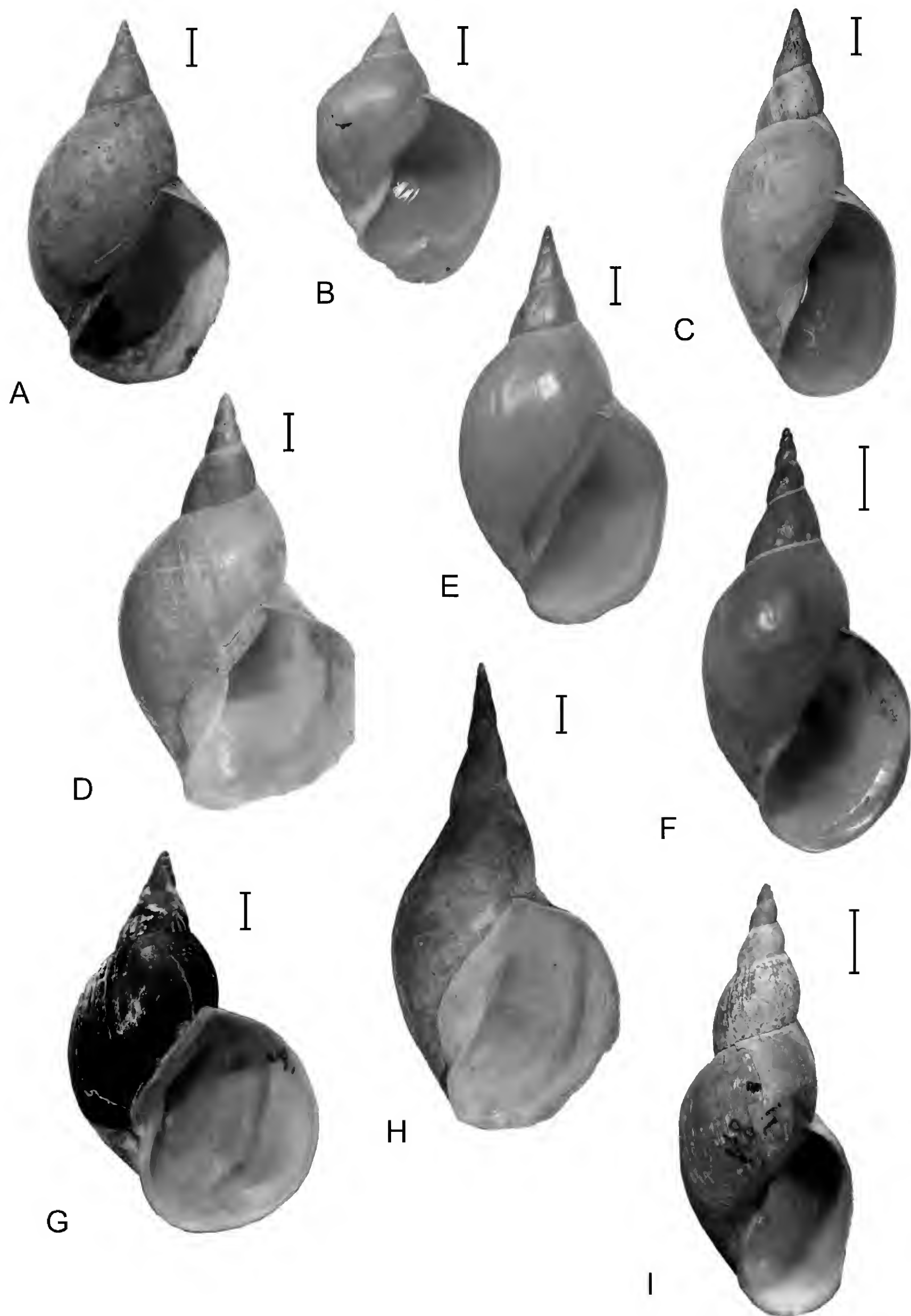
Figure 4. Varieties of *L. stagnalis* as they were identified by malacologists of the 19th century. **A**. *L. stagnalis* var. *typica* (det. S. Clessin; ZIN). **B**. *L. stagnalis* var. *media* (det. C.A. Westerlund; GNM). **C**. *L. stagnalis* var. *producta* (det. C.A. Westerlund; GNM). **D**. *L. stagnalis* var. *rosea* (identified by a unknown person; ZMUC). **E**. *L. stagnalis* var. *colpodia* (det. C.A. Westerlund; GNM). **F**. *L. stagnalis* var. *variegata* (det. C.A. Westerlund; GNM). **G**. *L. stagnalis* var. *turgida* (det. C.A. Westerlund; GNM). **H**. *L. stagnalis* var. *raphidia* (det. C.A. Westerlund; GNM). **I**. *L. stagnalis* var. *palustriformis* (det. A. Fuchs, NHMW). Scale bars 5 mm.

This short review shows how vague and indeterminate was this common practice of the discrimination of varieties. Zhadin (1952; 166) rightly advised the readers / users of his key that „we do not recommend to fit each collected shell to some variety; in most cases it is enough to give the shell measurements and only very divergent shells should be illustrated by drawings or photographs".

Some authors attempted to reduce this vagueness by making demand that only those varieties that are found to have a hereditary basis are real. For instance, Chaster (1907: 28) proposed a new definition of variety in malacology: „a variety is a group of individuals that differs from the typical or normal form in more or less distinct characters which are transmissible to the offspring".

Nevertheless, the most peculiar approach to the systematization of pond snails was realized by a group of French malacologists of the second half of the nineteenth century known under the name "Nouvelle École" (Dance 1970). The followers of it believed that a new species should be established if an individual is found to differ from all others by three characters or more (Davies 2004). Such a method led them to accept a huge number of nominal taxa of species' rank on the basis of very slight differences, usually in shell shape and proportions. Thus Locard (1893) was able to recognize no less than 22 "species" corresponding to *L. stagnalis* s. lato in the fauna of France alone (see Electronic Appendix 2). Most of these taxa were plain varieties raised to the rank of species. The unreliability of this approach was obvious, and no malacologist outside France could accept it.

A seemingly more productive approach to the *L. stagnalis* group taxonomy was proposed in Germany by Wilhelm Kobelt (1840–1916), a prominent zoologist who was also a strong critic of the "Nouvelle École" methods. Kobelt (1871) supposed that the shell variation in great pond snails has essentially ecological character, and their shell shape and proportions are moulded environmentally. Kobelt considered *L. stagnalis* as a Formenkreis ("circle of forms"), i.e. as a polymorphic species consisting of a series of ecological races that, in turn, embrace a plethora of varieties described prior to 1871. He distinguished at least four ecological forms (races) denoted by vernacular German names:

1. "Normalform" (a typical morph, see Fig. 4A).
2. "Hungerform" (a starvation morph) – dwarf phenotype of *L. stagnalis* arising allegedly as a result of food shortage.
3. "Seeform" – a phenotype of large lakes.
4. "Kanalform" – a phenotype produced in canals.

Kobelt's idea helped to reduce the mammoth number of varieties to a few comprehensible entities with relatively clear content. Its influence may be traced in the works of David Geyer (1927) in Germany and Vladimir Zhadin (1933, 1952) in USSR, who used the Formenkreis concept to outline a circle of varieties within widespread species of freshwater snails, including *L. stagnalis*, *Radix*

auricularia (L., 1758), *Planorbarius corneus* (L., 1758) and others. In this context, Hans Modell's (1922) attempt to build a taxonomic framework for freshwater unionid mussels (very variable like lymnaeids) on the basis of the recognition of intraspecific ecological morphs is of high interest as another realization of Kobelt's idea.

Population thinking and its consequences: twentieth century systematics

The advent of population genetics in the 1910–1920s brought to malacologists a new way of thinking capable of explaining the great pond snail variation. The futility of establishing an endless number of obscure varieties was demonstrated by Mozley (1937), who insisted that it is almost senseless to use standard shell measurements and their ratios for the exact determination of *L. stagnalis* varieties. The cause was that the overall conchological variation in a particular waterbody is sometimes so wide that "it may approach the range of variation which is to be found over the whole of the geographical range of the species" (Mozley 1937: 185). Instead, Mozley proposed to use the "local race" concept, according to which each more or less isolated habitat, pond or lake, has its own recognizable race of *L. stagnalis*. Mozley himself observed such a situation in Finland, where these races are common and, more importantly, "do *not* appear to be specially adapted to the local conditions under which they live" (Mozley 1937: 186). Thus the local races are not identical to ecological races according to Kobelt (1871). The main factor of their origin is the spatial separateness of waterbodies preventing the gene flow among populations.

This idea was a consequence of a quite novel form of biological thought known under the label "population thinking" (Mayr 1982). This population thinking shifted biologists from the study of single (or few) individuals to the examination of representative samples of animals by means of the rigorous statistical methods developed by biometrics. Most students agreed that character variation in natural populations is a norm rather than an annoying exception (Vinarski 2013), and the biometric studies revealed the continuous characters of shell variation not comparable with the concept of many distinct varieties within a species.

Further progress in biometric studies and conchometry based on measurements of large samples for drawing statistical inferences led to the complete renunciation of the local race concept in "freshwater" malacology. Eventually it became a trivial fact that any well isolated population of a given species should differ phenotypically from other populations of the same species. A total number of local races of *L. stagnalis* would be positively immense, so that makes the concept itself impracticable. Another cause of neglect of local races and similar intraspecific entities were anatomical studies focused mainly on the reproductive morphology of lymnaeids. Though the first information on *L. stagnalis* anatomy was obtained as ear-

Table 2. Taxonomy of the *Lymnaea stagnalis* species complex according to Kruglov and Starobogatov (1993).

Subgenus	Section	Species

Lymnaea	*Lymnaea* s.str.	1. *L. doriana* (Bourguignat, 1862)
		2. *L. fragilis* (L., 1758)
		  – *L. fragilis fragilis*
		  – *L. f. producta* (Colbeau, 1859)
		3. *L. stagnalis* (L., 1758)
		  – *L. stagnalis stagnalis*
		  – *L. s. turgida* (Hartmann, 1840)

	Kobeltilymnaea Kruglov & Starobogatov, 1993	4. *L. araratensis* Kruglov & Starobogatov, 1985
	Stagnaliana Servain, 1881	5. *L. media* (Kobelt, 1877) 6. *L. bodamica* (Miller, 1873)

ly as the 17th century (M. Lister, J. Swammerdam), its taxonomic relevance was not acknowledged until the 1910s (Baker 1911; Roszkowski 1914). Pioneering work with broad taxonomic use of data on lymnaeid anatomy was published by Frank C. Baker (1911), a prominent American malacologists, who tried to construct a system of Lymnaeidae on the joint conchological-anatomical basis. Bengt Hubendick, the Swedish malacologist, put anatomy as the only cornerstone of his worldwide system of the family (Hubendick 1951). He failed to find any qualitative anatomical differences among conchologically distinct morphs and varieties of *L. stagnalis*. Hubendick (1951) identified all these intraspecific entities as mere conchological variants of the same biological species, having no real taxonomic significance. Anatomical features of lymnaeid snails were thought to be of much higher taxonomic value compared to the conchological ones. After Hubendick, most authors viewed *L. stagnalis* as a conchologically heterogenous but anatomically uniform species with no intraspecific taxa in its structure (Piechocki 1979; Jackiewicz 1998; Glöer 2002).

In the twenthienth century, the Synthetic Theory of Evolution (STE) absolutely dominated over biologists' minds. One of the most influential STE constituents was the so called 'biological species concept' (BSC) that is universally known and needs no detailed exposition here. I wish only to remind that BSC sees species as isolated gene pools able to keep their integrity due to complete or almost complete absence of gene flow among them (Mayr 1982; Wilkins 2009). Hubendick (1951) acknowledged the theoretical significance of BSC but refuted its practical application to the systematization of lymnaeids. He stated that a realization of BSC guidelines "offers considerable difficulties. A strictly practical application involves the conducting of crossing experiments between individuals from different populations. To carry out such a noteworthy degree with the Lymnaeids <…> is impossible in practice" (Hubendick 1951: 35).

The first attempt to apply BSC in lymnaeid systematics was undertaken in the USSR in the 1970s and 1980s by N.D. Kruglov and Ya.I. Starobogatov. They adopted two main methods of systematization: artificial crossing experiments (Kruglov 1975; Kruglov and Starobogatov 1985) and the so called "comparatorial (or comparatory) method" invented by Logvinenko and Starobogatov (1971). The latter is based on an analysis of the subtle differences in shell growth patterns of closely allied species of snails and bivalves (Kruglov and Starobogatov 1985; Shikov and Zatravkin 1991; Graf 2007). Starobogatov and his numerous disciples applied this method in order to revise all families of freshwater mollusks of the former USSR. Though Graf (2007) considers the comparatorial method as a peculiar "species concept", his statement is not fully correct since Starobogatov himself was a follower of BSC and published several papers on its application to the systematics of freshwater mollusks (Starobogatov 1968, 1977).

As a consequence, Kruglov and Starobogatov (1985, 1993) proposed a new taxonomic structure of the *L. stagnalis* complex drastically dissimilar to the Hubendick (1951) system. The great pond snail was thought to represent at least six independent species distributed among three sections of the genus *Lymnaea* (Table 2). I have to note, however, that only two species out of the six (*L. stagnalis* s.str., *L. fragilis* L., 1758) were really involved in the artificial breeding experiments conducted by the authors. The species status of the remaining taxa was proposed for the reason of lack of morphological intermediates between them under condition of syntopy. According to Starobogatov (1968, 1977), the absence of such intermediate specimens could serve as an indirect proof of the reproductive isolation between studied taxa and might be taken into account by taxonomists even if this alleged isolation was not tested experimentally. In particular, differences in geometric patterns of shell coiling between two forms revealed by means of the comparatorial method was regarded by some Russian workers as a solid proof of their belonging to different species (Shikov and Zatravkin 1991).

The fate of Kruglov and Starobogatov's (1985) crossing experiments is remarkable. Though their results appeared in an international malacological journal and the language of the publication was English, not Russian, a serious discussion on the subject did not started. Only Meier-Brook (1993) briefly discussed these experiments in his article devoted to the species problem in "freshwater" malacology: "If the criteria for the assessment of the descendants are convincingly as described in their paper, we have to admit logically that Kruglov & Starobogatov are right to consider distinct species in this case" ("Wenn die Kriterien für die Beurteilung der Nachkommenherkunft so überzeugend sind, wie in ihrer Publikation beschrieben, wird man konsequenterweise zugestehen müssen, daß Kruglov & Starobogatov Recht haben, hier von

getrennten Arten auszugehen"" Meier-Brook 1993: 136). Other malacologists were not so loyal and either avoided any discussion or restricted themselves to general phrases with no sympathy for the "Russian" system ("opinions of Russian malacologists on the lymnaeid taxonomy <...> raised great doubts and <...> have not been taken into consideration"; Jackiewicz 1998: 3).

I think this question could not be resolved in principle before the modern "revolution" in taxonomy that was triggered by introduction of the molecular genetic techniques in the 1980s.

The great pond snail in the brave new world

The 'brave new world' of animal taxonomy relies heavily on inferences based on molecular studies and the quasi-cladistic way of bringing up and testing phylogenetic hypotheses (Mooi and Gill 2010). Though molecules are not the panacea for all puzzles systematists seek to solve, genetic methods provide us with an independent set of characters giving an opportunity to test hypotheses based on classical morphological studies. This has brought new possibilities and launched a new cycle of taxonomic studies on biological species, including the most well-studied ones such as *L. stagnalis*.

Already the first attempts to examine the internal diversity of *L. stagnalis* by molecular methods (Remigio and Blair 1997; Remigio 2002) revealed that its populations from different countries (Italy, Germany) and continents (Europe, North America) are separated by rather great genetic distances comparable with those separating distinct species of Lymnaeidae or even slightly exceeding them. Remigio (2002: 691) suggested that these populations "probably deserve at least subspecies status". The number of studied specimens/populations of *L. stagnalis* and the geographical scope in these early works were, however, not enough for making sound conclusions.

In 2008, a group of Ukrainian malacologists (Mezhzherin et al. 2008) submitted the two alleged species of the Kruglov and Starobogatov (1985) system, *L. stagnalis* s.str. and *L. fragilis*, to allozyme electrophoresis. They demonstrated that there are no significant genetic differences between these two taxa and they should be synonymized. Their reproductive isolation has also been questioned by Mezhzherin et al. (2008). An interesting pattern of congruence between genetic diversity and geography in the great pond snail was found: in the Ukraine, populations of *L. stagnalis* s. l. form two genetically distinct groups, "western" and "eastern", with the boundary between them lying somewhere in the central part of the country. Mezhzherin et al. (2008) proposed to treat the two groups as allospecies (sensu Amadon 1966) within the superspecies *L. stagnalis* s. l.

Having used the DNA sequencing technique and four gene markers (two nuclear and two mitochondrial), Vinarski et al. (2012) obtained results similar to those of Mezhzherin et al. (2008). In their study, two large genetically distinct groups whose distribution is clearly correlated with geography (geogroups) were found to exist within Palearctic *L. stagnalis* s. l. One of them is of mainly western distribution being found throughout Europe (except for the eastern parts of Ukraine and European Russia), and the second is widely distributed in Asia, from Transcaucasia eastwards to Mongolia, Tajikistan, and Lake Baikal. Most probably, these geogroups correspond to the two allospecies discovered by Mezhzherin et al. (2008) and therefore the use of the superspecies concept (Amadon 1966) is justified in this case. The phylogeographic pattern of the great pond snail, with separation of the species into two large divisions with eastern and western Palearctic distribution, resembles a number of similar examples found in different animal taxa, including freshwater amphipods (Vainio and Väinölä 2003), amphibians (Borkin et al. 2004), fish (Makhrov and Bolotov 2006) and mammals (Marmi et al. 2006). The usual explanation for such phylogeographic patterns is that of invoking Pleistocene glacial events, including the long isolation of groups of populations in refugia and the following recovery of the former range with the formation of zones of secondary intergradation.

However, a thorough analysis of morphological variation in *L. stagnalis* s. l. shows that the internal structure of the species cannot be restricted to molecularly defined groups. Though numerous morphs and varieties of the great pond snail lack the genetic support, the reality of some morphologically distinct entities within it has been proved by statistical methods (Vinarski 2014a). Four or five conchologically defined "morphotypes" can be delineated within *L. stagnalis* in Palearctic, with two of them, *L. stagnalis* (f. typica) and *L. fragilis* sensu Kruglov and Starobogatov being the most widely distributed (Vinarski 2014a). From the phylogenetic point of view, however, these morphotypes do not constitute separate clades and may arise in both geogroups in parallel. Their spatial distribution is apparently not governed by ecological or physical geographical factors (Vinarski et al. 2012; Vinarski 2014a).

These results represent a clear example of drastic incongruence between molecular and morphological data. Though the validity of "minor" species of the great pond snails accepted by Kruglov and Starobogatov (1985, 1993) was not corroborated genetically, the question of a possible existence of "cryptic" species (or other taxonomically significant entities of lower rank) within *L. stagnalis* s. lato remains opened. In my opinion, both "dimensions" of the species, genetic and morphological, are worthy of study since provide us with additional evidence and as such may be used in an integrative taxonomic approach. The geogroups teach us something of the history of the species' range and alleged Plestocene refugia. The morphotypes reflect another, functional aspect of biodiversity additional to the taxonomic one that may be measured at the infraspecific level (Albert et al. 2012). It has been assumed that conchological differences between the morphotypes may be of some adaptive value (Vinarski 2014a).

Nevertheless, this brave new view on *L. stagnalis* will, possibly, change in the nearest future since new, more

powerful methods of molecular study (next generation sequencing, transcriptomics) are coming. The application of these methods may bring essentially new results concerning the great pond snail – this long studied but still not completely understood species.

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Leaping towards a saltatorial lifestyle? An unusually long-legged new species of *Rhombophryne* (Anura, Microhylidae) from the Sorata massif in northern Madagascar

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Abstract

The Madagascar-endemic microhylid genus *Rhombophryne* consists of a range of partly or completely fossorial frog species. They lead a poorly known, secretive lifestyle, and may be more diverse than previously thought. We describe a new species from the high altitude forests of the Sorata massif in north Madagascar with unusual characteristics for this genus; *R. longicrus* **sp. n.** has long, slender legs, unlike most of its fossorial or semi-fossorial congeners. The new species is closely related to *R. minuta*, a much smaller frog from the Marojejy massif to the southeast of Sorata with similarly long legs. We discuss the morphology of these species relative to the rest of the genus, and argue that it suggests adaptation away from burrowing and toward a more saltatorial locomotion and an accordingly more terrestrial lifestyle. If this is the case, then these frogs represent yet more ecological diversity within the already diverse Cophylinae. We recommend an IUCN Red List status of Endangered B1ab(iii) for *R. longicrus* **sp. n.**, because it is known only from a single site in a forested area of roughly 250 km², which is not yet incorporated into any protected area.

Introduction

The microhylid frog genus *Rhombophryne* (Anura, Microhylidae, Cophylinae) is receiving renewed taxonomic attention in the wake of recent genetic barcoding efforts on the frogs of Madagascar. These DNA barcode studies revealed that less than 60% of the island's amphibian diversity might so far have been formally described (Vieites et al. 2009, Perl et al. 2014). The taxonomic gap between what has been described and the total diversity that exists is extreme in the cophyline microhylids, a Madagascar-endemic radiation of narrow-mouthed frogs. The cophyline genus *Rhombophryne* typifies the taxonomic gap: In 2007, it contained eight described species (Glaw and Vences 2007). Thirteen candidate species were identified by integrative inventories taking into account morphology and mtDNA and nuclear gene sequences (Wollenberg et al. 2008, Vieites et al. 2009, Perl et al. 2014). Still more have been discovered by additional fieldwork. Five of these have been described recently (D'Cruze et al. 2010, Glaw et al. 2010, Scherz et al. 2014, 2015), facilitated and accelerated by integrative approaches. These approaches are the key to closing the taxonomic gap in Madagascar's amphibians, which is in turn key to understanding and protecting them.

Repeated swapping of ecological niches (i.e. transitions between gross ways of life, such as between terrestrial and arboreal lifestyles) has led to high ecological diversity in the Cophylinae (Andreone et al. 2005, Wollenberg et al. 2008). The genus *Rhombophryne* is genetically most closely related to the dwarf frogs of the genus *Stumpffia* but

is morphologically very similar to *Plethodontohyla* (Andreone et al. 2005). *Rhombophryne* contains obligate and facultative burrowing frogs (Andreone et al. 2005, Wollenberg et al. 2008), and the enigmatic *R. minuta*, the ecology of which we discuss below. *Stumpffia*, a genus in need of revision, consists of mostly terrestrial dwarf frogs, some of which number among the smallest frogs in the world (Glaw and Vences 2007). Recently, Peloso et al. (2015) proposed the synonymy of *Stumpffia* and *Rhombophryne*. However, we here refrain from adopting their changes pending further investigation, as these genera are morphologically distinct. *Plethodontohyla* species resemble *Rhombophryne* more closely in size, but consist of a mixture of probably facultative burrowers and arboreal or semi-arboreal climbers. Reasons for this ecological flexibility, and the ancestral states of the genera, have so far been poorly explored (Andreone et al. 2005).

Here, we describe a new *Rhombophryne* clearly distinct from all other known species, although phylogenetically nested. We describe its skeleton by means of micro-computed tomography (micro-CT), revealing additional differences to the other *Rhombophryne* species for which skeletal data are available (Scherz et al. 2014, 2015, unpublished results). We discuss the implications of this potentially novel morphology for the evolution of the genus *Rhombophryne*, and highlight the need for herpetological survey work in less accessible areas of Madagascar.

Materials and methods

Specimens were collected, euthanized, fixed in 90% ethanol and transferred to 70% ethanol for long-term storage. One specimen was deposited in the Université d'Antananarivo Département de Biologie Animale (UADBA), and the other in the Zoologische Staatssammlung München (ZSM).

In this work we refer to the species *R. ornata* and *R. tany*. Their descriptions are in press (Scherz et al. 2015), and we therefore give the disclaimer that the use of these two names here should be considered conditional, and these two names herein are not made nomenclaturally available, in accordance with Articles 8.3 and 15.1 of the International Code of Zoological Nomenclature (ICZN 1999).

Morphological measurements were taken with a digital calliper to 0.01 mm and rounded to the nearest 0.1 mm for presentation here. Fig. 1 shows our measurement scheme except cumulative measures (HIL, FORL, and FOTL). Abbreviations are used as follows: SVL (snout–vent length), HW (maximum head width), HL (head length, from the maxillary commissure to the anterior-most point of the mouth), ED (horizontal eye diameter), END (eye–nostril distance), NSD (nostril–snout tip distance), NND (internarial distance), TDH (horizontal tympanum diameter), TDV (vertical tympanum diameter), HAL (hand length, from the metacarpal–radioulnar articulation to the tip of the longest finger), LAL (lower arm length, from the carpal–radioulnar articulation to the centre of the radioulna–humeral articulation), UAL (upper arm length, from the centre
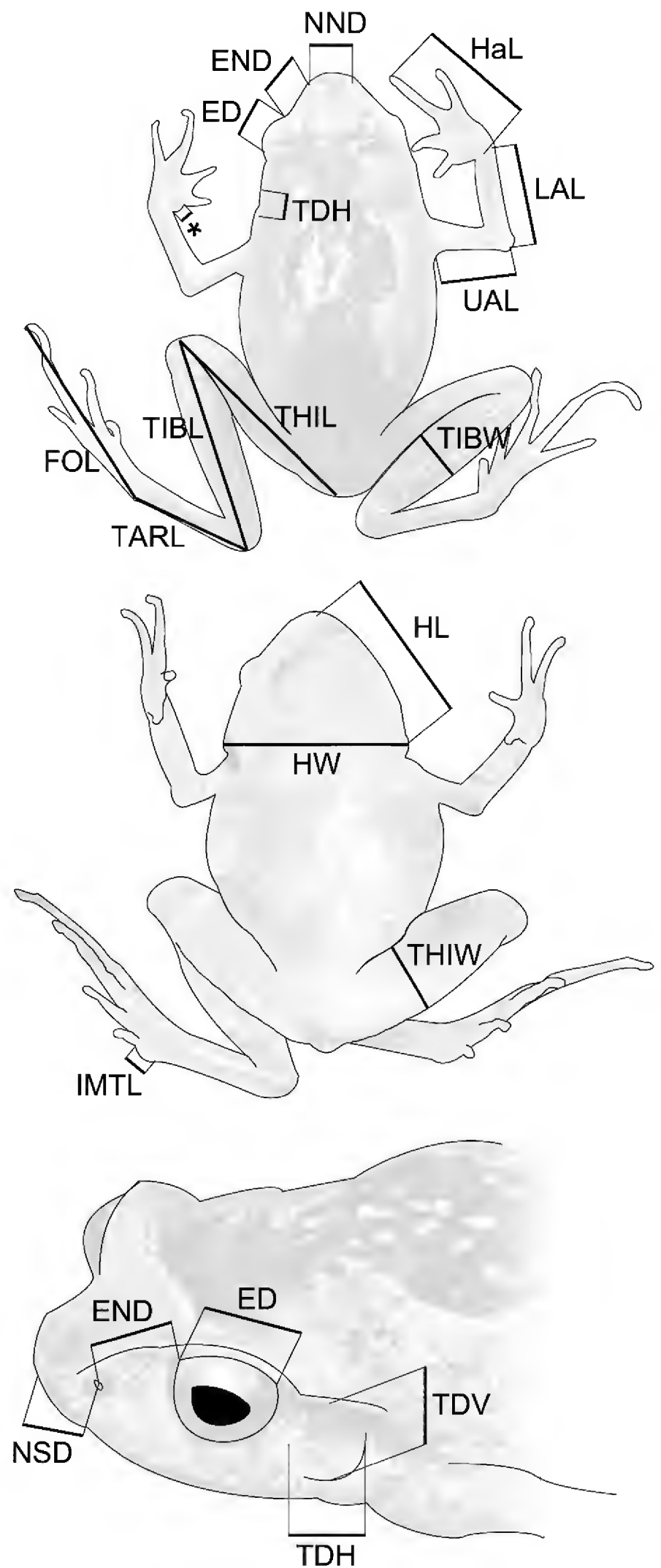
Figure 1. Measurement scheme used to measure *Rhombophryne longicrus* sp. n. and congeners for this study. Abbreviations are explained in Materials and Methods, as are cumulative measures such as forelimb and hindlimb length. * indicates IMCL.

of the radioulna–humeral articulation to the trunk, measured along the posterior aspect of the arm), FORL (forelimb length, given by the sum of HAL, LAL, and UAL), FOL (foot length, from the tarsal–metatarsal articulation to the tip of the longest toe), TARL (tarsal length, from the tarsal–metatarsal articulation to the tarsal–tibiofibular articulation), FOTL (foot length including tarsus, from the

tibiotarsal articulation to the tip of the longest toe, given by the sum of FOL and TARL), TIBL (tibiofibula length), TIBW (tibiofibula width at thickest point, measured in dorsal aspect), THIL (thigh length, from the vent to the femoral–tibiofibular articulation), THIW (thigh width at thickest point, measured in supine position), HIL (hindlimb length, given by the sum FOL, TARL, TIBL, and THIL), IMCL (maximum length of inner metacarpal tubercle), IMTL (maximum length of the inner metatarsal tubercle).

Micro-CT scanning was carried out on a nanotom m (GE Measurement & Control, Wunstorf, Germany). The holotype of the new species, ZSM 1630/2012, was mounted on a polystyrene board inside a sealed polyethylene vessel, and secured in place using small wooden struts and additional polystyrene. A small volume of 80% ethanol was added to achieve air saturation, preventing desiccation of the specimen. The vessel was mounted on a polyvinylchloride tube, and placed inside the micro-CT scanner. Scanning was conducted at a voltage of 140 kV and a current of 80 mA, with a timing of 500 ms for 20 minutes (2440 projections). Scan data were assembled in phoenix DATOS|X 2 RECONSTRUCTION CT software (GE Measurement & Control, Wunstorf, Germany) and visualised in VG STUDIO MAX 2.2 (Volume Graphics GmbH, Heidelberg, Germany) and subsequently processed into a 3D surface render in AMIRA 5.4.5 (FEI Visualization Sciences Group, Burlington MA, USA). Skeletal description is based on both surface and volume renderings of micro-CT data, due to artefacts produced in surface rendering. Readers are advised that micro-CT scanning does not render poorly calcified structures, especially cartilage. Cartilages are therefore omitted from the osteological description below and the respective figures; additional specimens will need to be cleared and stained in order to assess cartilaginous characters of these frogs. We provide a PDF-embedded interactive 3D model of the skeleton of the holotype as Suppl. material 1. Osteological terminology follows Trueb (1968, 1993). Skull ratio measurements were calculated from high resolution TIFF images of prepared models in ImageJ 1.48v (Schneider et al. 2012).

We extracted total genomic DNA from ethanol-preserved tissue samples using proteinase K digestion (final concentration 1 mg/mL) and a standard salt extraction protocol (Bruford et al. 1992). We amplified a fragment of the mitochondrial large ribosomal subunit or 16S rRNA (16S) via polymerase chain reaction (PCR) using the primers 16Sar-L and 16Sbr-H (Palumbi et al. 1991). Sequences were resolved on an ABI 3130xl automated DNA sequencer (Applied Biosystems) and newly determined sequences were deposited in GenBank (accession numbers KR025897 and KR025898).

We calculated a phylogenetic tree from the 16S sequences by Bayesian inference (BI) with MRBAYES 3.2 (Ronquist et al. 2012) after determining a GTR+I+G substitution model as best fitting the data using JMODELTEST (Darriba et al. 2012). We considered all sections of the 16S gene with more than two consecutive gaps in one or more sequences as ambiguous and excluded these sections from the alignment (total alignment length after exclusion: 397 nucleotides). Explorative analyses including these stretches resulted in an identical topology and similar support values for all nodes supported with PP > 0.9. We ran two independent analyses for 20 million generations, each comprising four Markov Chains (three heated and one cold), and sampled every 10,000 generations. Chain mixing and stationarity was assessed by examining the standard deviation of split frequencies and plotting the –lnL per generation using TRACER 1.6 software (Rambaut et al. 2014). Results were combined to obtain a 50%-majority rule consensus tree and the respective posterior probabilities of nodes, after discarding 25% of the generations as burn-in (all compatible nodes with probabilities <0.5 kept).

Results

The new species described below is phylogenetically nested in the genus *Rhombophryne* (Fig. 2) and placed as sister to *R. minuta* with strong support. The relatively
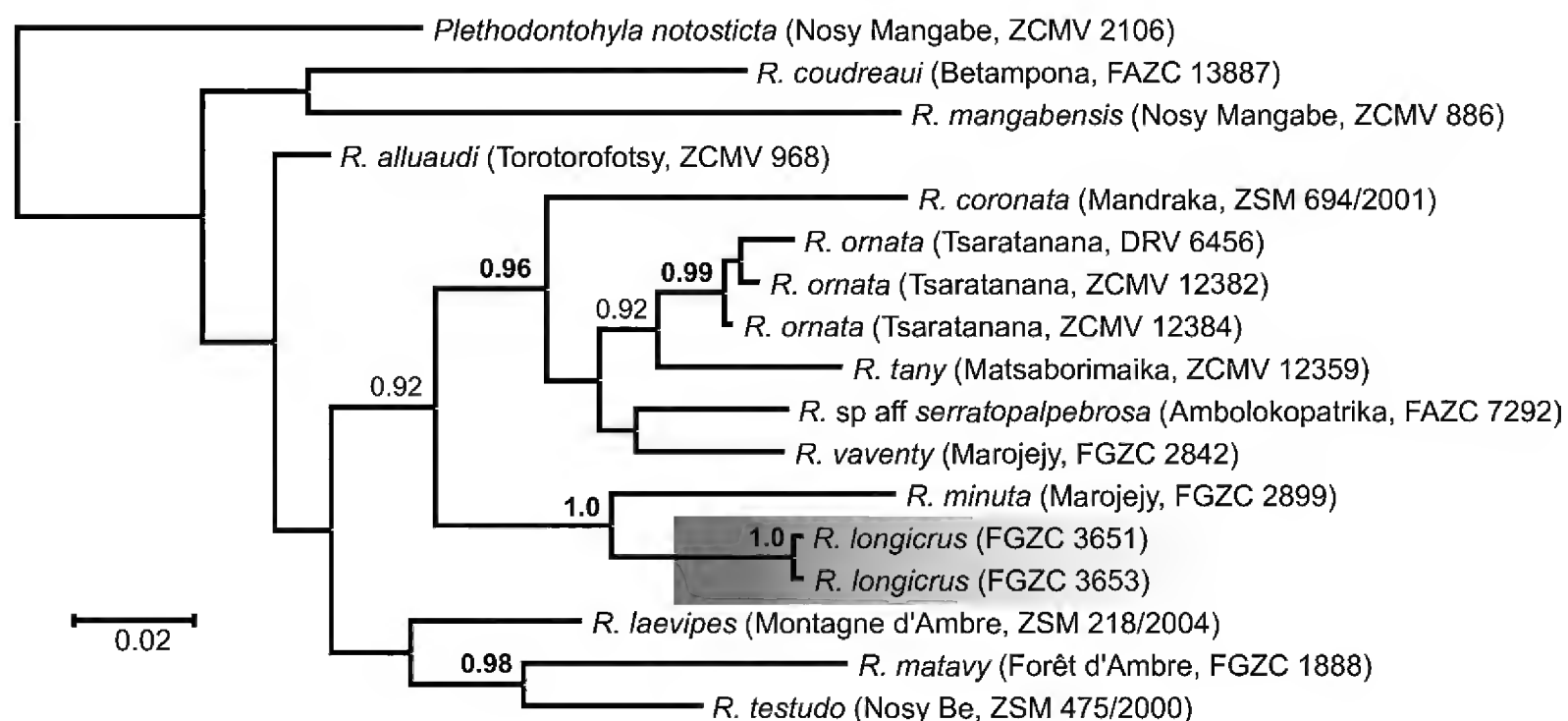

Figure 2. Majority-rule consensus tree derived from Bayesian inference analysis of the genus *Rhombophryne* based on the mitochondrial 16S rRNA gene. Numbers at nodes represent posterior probability (PP). PP values greater than 0.95 are bolded. Values lower than 0.8 are not shown.

long branch length indicates, however, a strong genetic differentiation from its probable sister species. Uncorrected pairwise differences (*p*-distances) using the entire amplified 16S fragment (including hypervariable regions; alignment length 536 bp) was 6.8% to *R. minuta* and >8.6% to all other species of *Rhombophryne* (including *R. guentherpetersi*; M. Vences, analyses in progress). The concordance of such a high genetic divergence (>3% in the 16S fragment; see Vieites et al. 2009) with clear morphological differentiation is strong evidence for status as an independent evolutionary lineage, warranting its recognition as a distinct species. We therefore formally describe this new species here as:

Rhombophryne longicrus sp. n.

http://zoobank.org/35E10C49-8211-4E30-BDB0-48E753348738
Figs 3, 4

Holotype. ZSM 1630/2012 (FGZC 3653), an adult female with immature oocytes, collected in the montane forest of the Sorata Massif, north Madagascar (ca. 13.675°S, ca. 49.4392°E, ca. 1580 m; datum = WGS84) on 28 November 2012 by A. Rakotoarison, A. Razafimanantsoa, T. Rajoafiarison, F. M. Ratsoavina, O. Hawlitschek and F. Glaw.

Paratype. UADBA-A 60271 (FGZC 3651), an adult male with the same collection data as the holotype.

Diagnosis. A microhylid assigned to the genus *Rhombophryne* on the basis of overall morphology, including the possession of maxillary and vomerine teeth, absence of expanded toe pads, and absence of an enlarged prepollex. Confirmed as a member of the genus *Rhombophryne* on the basis of its phylogenetic relationships as assessed by mitochondrial DNA, as there are no known morphological characters by which *Rhombophryne* may be distinguished from *Plethodontohyla*.

Rhombophryne longicrus sp. n. is distinguished from all other Madagascan frog species by the following set of characters: SVL 23.8–27.9 mm, head wider than long, horizontal tympanum diameter 47% of eye diameter, absence of superciliary spines, weak supratympanic fold, dark supratympanic region and nostril, tibiotarsal articulation reaching the nostril, total hindlimb length 183–185% of SVL, second finger shorter than fourth, and fifth toe shorter than third. It is also separated by a pairwise genetic distance of at least 6.8% in the 16S mitochondrial gene from all other known species of the genus *Rhombophryne*.

Within the genus, *R. longicrus* sp. n. may be distinguished from all *Rhombophryne* species, except *R. laevipes*, *R. minuta*, and *R. vaventy,* by tibiotarsal articulation reaching the nostril (versus not exceeding the eye); from *R. coronata, R. ornata, R. serratopalpebrosa, R. tany,* and *R. vaventy* by the absence of superciliary spines (versus presence); from *R. alluaudi, R. laevipes, R. mat-*

avy, R. testudo, and *R. vaventy* by its smaller size (SVL 23.8–27.9 mm versus 32–53 mm); from *R. minuta* by its larger size (SVL 23.8–27.9 mm vs. up to 17.1 mm); from *R. testudo* by the absence of barbels on the throat and tympanum smaller than eye; from *R. alluaudi, R. coronata, R. serratopalpebrosa, R. tany,* and *R. vaventy* by its weak, almost absent supratympanic fold; from *R. coudreaui* and *R. vaventy* by smooth dorsal skin (versus granular/tubercular); from *R. mangabensis* by lack of paired dark dorsal tubercles; from *R. laevipes, R. mangabensis, R. ornata, R. testudo,* and *R. vaventy* by absence of dark crossbands on hindlimbs; and from *R. coronata, R. minuta, R. testudo,* and *R. vaventy* by dark supratympanic region.

Osteologically, a micro-CT scanned specimen of *R. longicrus* sp. n. tentatively differs from *R. ornata* (3 specimens: ZSM 1815/2010, 1816/2010, and 2859/2010), *R. serratopalpebrosa* (1 specimen: MNHN 1975.24), *R. tany* (1 specimen: ZSM 1814/2010), and *R. vaventy* (1 specimen: ZSM 375/2005) (as described in Scherz et al. 2014, 2015) by its relatively larger nasals (nasal length at longest point 18.5% of skull length versus 11.1–16.4%), which are in contact with the sphenethmoid (versus not contacting any other bones), its relatively longer and less broad skull (skull length 81.6% of skull width versus 66.5–79.4%), and its relatively longer brain case (frontoparietals+sphenethmoid length 74.0% of skull length versus 63.3–71.5%; length of frontoparietals+sphenethmoid 197.7% of width of frontoparietals anterior to prootic versus 173.4–185.6%). A thorough osteological treatment of this genus is needed to confirm further differences and their values.

Rhombophryne species can be confused with *Plethodontohyla* species. *Rhombophryne longicrus* sp. n. differs from them in the following ways: absence of a sharp dorsolateral colour border and expanded finger and toe pads (versus presence in *P. notosticta, P. guentheri, P. mihanika,* and *P. inguinalis*), absence of inguinal spots (versus presence in *P. mihanika, P. inguinalis, P. ocellata,* and *P. bipunctata*), tibiotarsal articulation reaching the nostril (maximally reaching to the mid-eye in all *Plethodontohyla* except *P. mihanika*), absence of crossbands on legs (versus presence in *P. fonetana, P. inguinalis, P. notosticta, P. guentheri,* and *P. mihanika*), and smooth skin (versus granular to rough in *P. tuberata*).

Description of the holotype. Adult female in an excellent state of preservation. A ventral incision was made in order to check the sex and access the stomach contents. The incision runs laterally and posteroventrally anterior to the pubis and up the middle of the venter.

Body gracile; dorsal and ventral skin smooth. Head wider than long (HW 122.5% of HL), snout rounded in dorsal view, squarish in lateral view; nostrils weakly protuberant, directed laterally, equidistant between eye and snout; canthus rostralis concave; loreal region concave; tympanum indistinct, oval, horizontally 47% of eye diameter; pupil round; supratympanic fold weak, almost
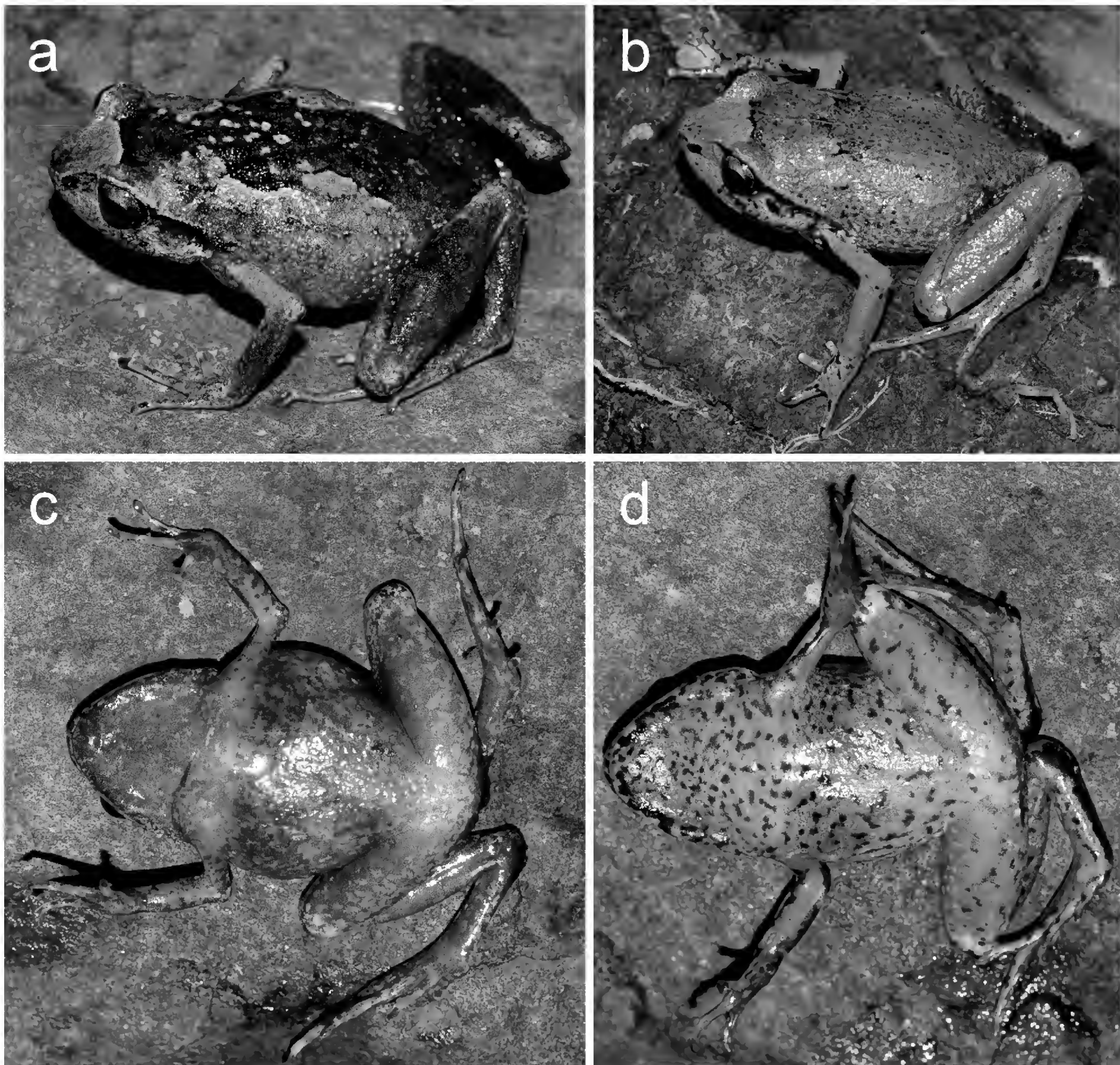
Figure 3. *Rhombophryne longicrus* sp. n. in life. Holotype ZSM 1630/2012 in (**a**) dorsolateral and (**c**) ventral view. Paratype UADBA-A 60271 in (**b**) dorsolateral and (**d**) ventral view.

absent; tongue unlobed, posteriorly free; vomerine teeth present in a straight row with a small medial gap (<1 mm; see Osteology below); choanae small, oval.

Arms slender and long; fingers without webbing, long, without distinct subarticular tubercles, relative lengths 1<2<4<3, second finger much shorter than fourth, without enlarged terminal discs; inner metacarpal tubercle present; nuptial pads absent. Legs exceptionally long and slender (HIL 185% of SVL), tibiotarsal articulation reaching the nostril when hindlimb is adpressed along body; toes long, unwebbed, with indistinct subarticular tubercles, relative toe lengths 1<2<5<3<4, third toe much longer than fifth; inner metatarsal tubercle present and indistinct.

Colouration of the holotype: (Fig. 3a, c). In life, snout anterior to eyes, above eyes, side of head, and upper arms bronze to tan in colour; tip of snout darker, lightening

posteriorly; area around nostril black; supratympanic region dark brown, fading below to the tan of the lateral side of the head. Body laterally light brown, becoming increasingly yellowish brown dorsolaterally until tan border with dark dorsal marking; this marking is flecked with additional tan spots, and extends from a black horizontal bar between the eyes to the legs, where dark ashy grey dominates; border between dorsal and dorsolateral colouration almost symmetrically emarginated. Hands and feet tan with dark flecks. Ventral skin pinkish and slightly translucent; chin dark relative to rest of venter, posteriorly lightening with few darker patches interspersed with translucent areas lacking pigment. Anteroventral surface of legs with dark pigment, becoming less pigmented more ventrally; ventral surface mottled pinkish and light brown.

After three years in 70% ethanol, all browns have faded to shades of grey. Dorsal areas that were lightest in shade
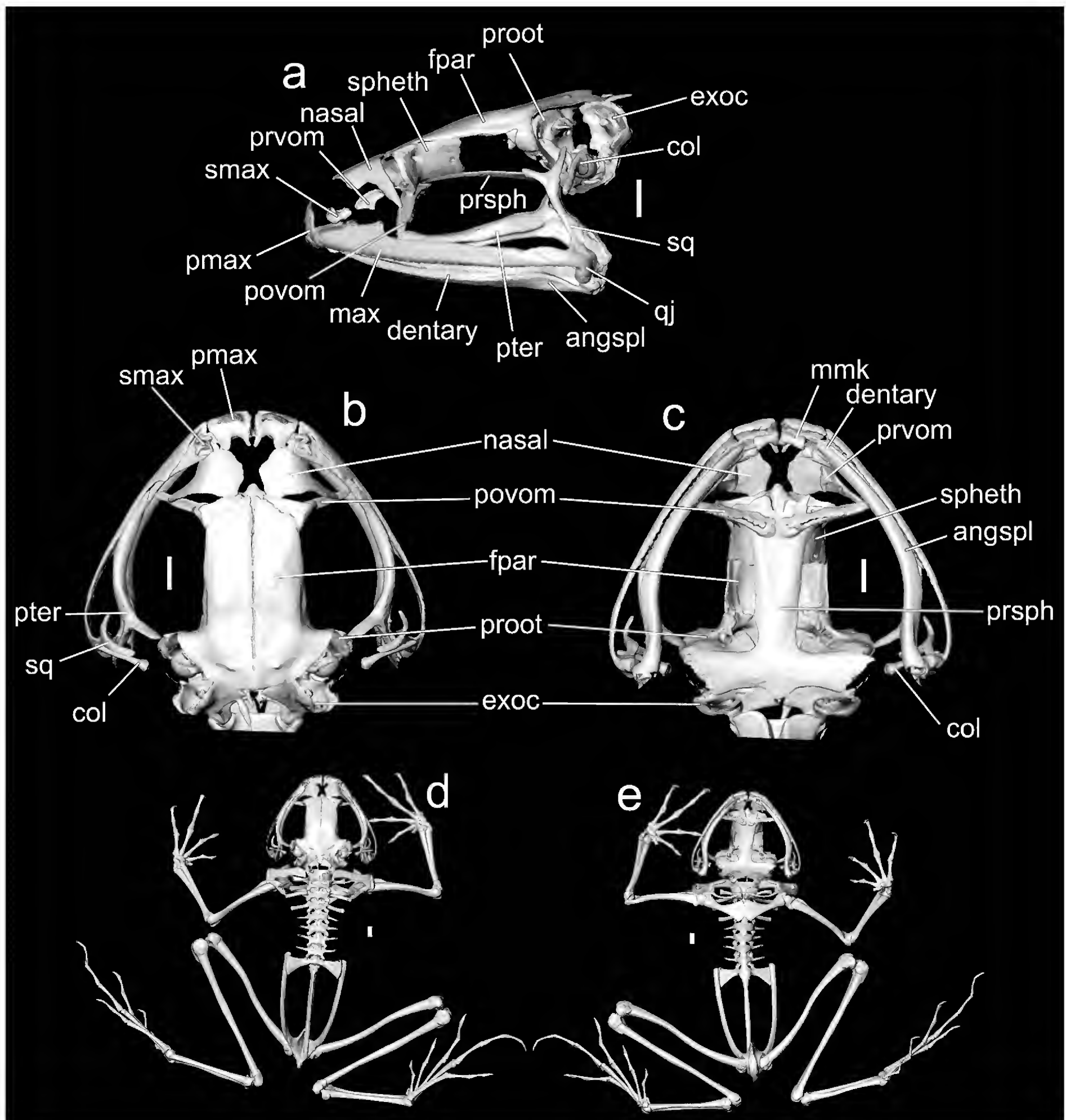
Figure 4. Osteology of the holotype of *Rhombophryne longicrus*, ZSM 1630/2012. Skull in (**a**) lateral, (**b**) dorsal, and (**c**) ventral view. Skeleton in (**d**) dorsal and (**e**) ventral view. Note: figures display only calcified structures; cartilages are omitted due to limitations of micro-CT scanning. Abbreviations: angspl = angulosplenial, col = columella, exoc = exoccipital, fpar = frontoparietal, max = maxillary, mmk = mentomeckelian, pmax = premaxilla, povom = postchoanal vomer+neopalatine, proot = prootic, prsph = parasphenoid, prvom = prechoanal vomer, pter = pterygoid, qj = quadratojugal, smax = septomaxilla, spheth = sphenethmoid, sq = squamosal.

are whitish, particularly between the eyes anterior to the dark inter-ocular bar. Ventrally, all areas that lack pigmentation and were pink in life are cream in preservation. Chin the same colour as the snout.

Osteology of the holotype: (Fig. 4, Suppl. material 1). All bones of the skull paired except the parasphenoid and sphenethmoid. Vomer divided into pre- and postchoanal portions; prechoanal part small, longer than broad, subtri-angular; postchoanal part overlapping neopalatine, bearing ventral serrations (vomerine teeth), separated medially from its counterpart by a gap of 0.7 mm. Postchoanal vomer+neopalatine in dorsal contact with anterior end of cultriform process of parasphenoid, and through it with the sphenethmoid; laterally not in contact with maxilla. Teeth present on maxilla and premaxilla. Premaxilla medially not fused to counterpart, anterodorsal alary processes

rising dorsolaterally, pars palatina with two well-defined processes, the medial (palatine) process thin, lateral process broad; pars dentalis bearing small teeth. Septomaxilla roughly spiralling upward from posterior ramus to lateral ramus to anterior ramus to medial ramus. Nasal medially in contact with the sphenethmoid posteriorly, possessing a pointed maxillary process extending ventrolaterally toward the maxilla, lacking an anterolateral ramus. Maxilla long, bearing small, poorly resolved teeth, possessing a horizontal pars palatina along its lingual margin; in broad lateral contact with anterior ramus of pterygoid; posteriorly without clear distinction from quadratojugal. Pterygoid broad and triradiate, with anterior, medial, and posterior rami; the ventrolateral edge of its anterior ramus, posterior margin of its medial ramus, and lateral face of its posterior ramus sculpted inward; medial ramus much shorter than posterior ramus; posterior ramus not in contact with quadratojugal. Quadratojugal L-shaped, anterior process without clear distinction from posterior of maxilla; posteriorly possessing a ventral bulbous process with a concave posterior face; dorsally without clear distinction from squamosal. Squamosal thin and distally bifurcated, extending anterodorsomedially from quadratojugal to level of otic capsule passing anterior to columella; otic ramus longer than zygomatic ramus. Columella with a long shaft that exceeds the level of the squamosal; dorsal edge of columella straight even to end of footplate; columellar footplate broad and concave. Frontoparietal medial and lateral edges straight and parallel, lateral edge curved ventrally to form dorsolateral border of brain case; possessing paired bumps at the transverse level of the columellae; posterolateral sutures with prootics and exoccipitals not clear from micro-CT scans; anterior process contacting sphenethmoid. Parasphenoid T-shaped; cultriform process broadening anteriorly, contacting sphenethmoid at its anterior end; broad posterior alary processes perpendicular to cultriform process, in dorsal contact with prootics anteriorly and exoccipitals posteriorly.

Mandible slim, edentate. Mentomeckelian small, in narrow medial contact with counterpart (possibly artefactual), and in dorsolateral contact with dentary. Dentary long and thin with a sculpted outer face and smooth inner face, overlapping angulosplenial for much of its length. Angulosplenial broadening posteriorly, with a posterior dorsomedial crista; possessing a lateral channel running from the posterior into the sculpted outer edge of the dentary.

Posterolateral processes of hyoid shovel-like, a medial crista running along posteromedial process, the base of which is broad and flat with a rounded anteromedial edge and sharp anterolateral and posteromedial corners; parahyoid absent.

Humerus long, slim and straight; crista lateralis weak, crista ventralis short (~30% of humerus length), crista medialis absent. Radioulna broadening distally. Finger phalangeal formula 2-2-3-3. Terminal phalanges of fingers 2, 3, and 4 with distal knobs. Prepollex 31% of first finger.

Pectoral girdle composed of paired coracoids, clavicles, scapulae, cleithra and suprascapulae. Sternal characters not visible in CT render. Coracoids in medial contact; medially dorsoventrally flattened, laterally rounded, posterior surface straight, anterior surface strongly concave. Clavicle thin and curved approximately parallel to the anterior edge of the coracoid, its lateral end broadened, posteriorly in contact with ventral edge of scapular pars acromialis. Scapula thick, hourglass shaped, its posterior edge less strongly curved than its anterior edge, medioventrally bifurcated; pars acromialis distally rounded, in contact with the lateral end of the clavicle, its anterior surface concave; pars glenoidalis curved ventrally, in contact with lateral face of coracoid, posterior face concave; dorsal edge of scapula approaching cleithrum. Cleithrum thin and long, not possessing any cristae, anteriorly thicker than posteriorly. Suprascapula with highest X-ray absorption ventrally and posteriorly suggesting possible ossification in these areas.

Toe phalangeal formula 2-2-3-4-3; terminal phalanges without distinct distal knobs. Leg bones long and thin. Femur without any crests. Tibiofibula slightly longer than femur. Tibiale and fibulare proximally and distally fused, articulating distally with metatarsals V and IV, tarsals 1–3, and the centrale. Prehallux present, short.

Ilium, ischium, and pubis forming ossified acetabulum, each composed of paired, medially fused elements. Iliac shafts oval in cross-section, dorsal-ventral diameter larger, possessing a weak dorsal tubercle posterior to shallow oblique groove. Iliosacral articulation type IIA sensu Emerson (1979).

Eight presacrals present; no vertebrae fused. Posterior articular processes round. Transverse processes of presacrals II–IV broader than those of V–VIII. Neural spines decreasing in size from presacral II to absent by V. Sacrum wide, with broad diapophyses articulating with the ilia; anterior edge of each diapophysis roughly perpendicular to body axis, posterior edge oblique. Urostyle long and thin, with a dorsal ridge along a third of its length, beginning at its anterior end; articulation with sacrum bicondylar.

Measurements. Holotype (paratype in brackets), measurements in mm: SVL 28.0 (23.8), HW 9.9 (9.7), HL 8.0 (7.0), ED 3.3 (2.8), END 2.0 (2.0), NSD 1.9 (1.7), NND 3.0 (2.2), TDH 1.5 (1.3), TDV 1.8 (1.4), HAL 8.4 (7.0), UAL 5.7 (4.7), LAL 6.8 (5.7), FORL 20.9 (17.4), THIL 13.2 (11.7), THIW 3.9 (3.4), TIBL 14.6 (11.7), TIBW 2.97 (2.64) TARL 8.6 (7.3), FOL 14.8 (12.5), FOTL 23.4 (19.8), HIL 51.2 (43.2), IMCL 1.0 (0.9), IMTL 1.3 (1.0).

Variation. Only two specimens are known. The paratype is male, and smaller than the holotype (SVL 23.8 mm). It agrees in all aspects of its morphology with the holotype, but differs strongly in colouration (see Fig. 3b, d). In life, the dorsum has a yellow-brown base colour, with distributed black or dark brown flecks. A black inter-ocular bar is present, behind which the skin fades from brown to the base colour; the back does not possess the dark marking of the holotype, but instead two darker areas with a few black

flecks lie in the suprascapular region. The lateral skin fades to grey ventrally, also speckled with black. A dark line runs from the preocular region to the axial pit through the supraocular and supratympanic regions. The nostril is surrounded by black, and the tympanum has a dark fleck on it. The legs are grey at the hip, but this lightens to the yellowish brown of the dorsum further away; the arms are dorsally yellow, the hands possessing a few black spots. Ventrally, the pink areas of the holotype are orange in life in the paratype, particularly over the pectoral girdle and beneath the chin. The venter is marked with many irregular black flecks. The arms are ventrally orange, bordered posteriorly in black.

Etymology. The species epithet is an invariable noun in apposition to the genus name, derived from the Latin words *longus* (meaning long), and *crus* (meaning leg), and refers to the unusually long legs of this species.

Distribution. This species has only been found at high altitude in the montane forests of the Sorata massif in north Madagascar. Its distinctiveness leads us to hypothesize that it has never been found elsewhere and misidentified, so it may be microendemic to this small area. Additional surveys are required in areas in and around Sorata to identify its full distribution.

Ecology. Both specimens described here were captured in the early evening on the ground along a path through primary montane forest. The stomachs of both specimens contained remains of several small insects (mostly Coleoptera) and a spider (possibly belonging to the family Salticidae), mixed with moss, suggesting an opportunistic diet of arthropods. Calls of this species are unknown. The female holotype had more than twenty immature oocytes with the largest having diameters ranging from 1.3 to 1.6 mm. As a member of the Cophylinae, it is likely that *R. longicrus* lays its eggs away from running water or large water bodies, and has endotrophic tadpoles.

Conservation status. The forests of Sorata are currently unprotected. All locally endemic species are threatened by uninhibited deforestation and forest degradation. The greatest pressure on forests is at their edges. High altitude species like *R. longicrus* may therefore be the least threatened by this. However, a sustained rate of deforestation will increase the threat level to species at ever-higher altitudes. It is conceivable that a restriction of this species to high altitudes may mean that it is susceptible to climate change (Raxworthy 2008, Raxworthy et al. 2008). We consider this threat far less serious than that of deforestation. *Batrachochytrium dendrobatidis* has now been confirmed from numerous localities in Madagascar (Bletz et al. 2015). So far no negative impacts on native frogs have been observed. The water-independent lifestyle of *Rhombophryne* species suggests that they are probably at relatively low threat from chytridiomycosis.

While this species is, at present, known from just two specimens collected on one expedition, the fact that it has not been collected by previous expeditions suggests it may be scarce, seasonal, or have a scattered distribution. Even if it were distributed throughout the forests of the Sorata massif, its distribution would still only constitute an area of ~250 km² (as calculated in Google Earth® Pro 6.1.0.500, Google Inc., Mountain View, CA). Thus, because of its potentially limited range inside an unprotected forest, the on-going and intensifying threat of deforestation, potential threat by climate change, and potential scarcity or seasonality, it qualifies as Endangered B1ab(iii) under the IUCN Red List Criteria (2012).

Discussion

Hindlimb length is significantly associated with habitat and mode of life in frogs (Gomes et al. 2009). Longer legs relative to body length result in greater relative leaping performance (Zug 1978, Choi et al. 2003, Gomes et al. 2009). In general, fossorial frogs have the poorest jumping performance, while arboreal and semi-aquatic frogs are the strongest jumpers (Zug 1978). Terrestrial frogs are intermediate, but generally poor leapers, preferring to hop rather than leap. Emerson (1976) noted that adaptations involved in hopping in terrestrial frogs are similar to those required for burrowing. This can lead to difficulties disentangling the morphology involved in these two habits. We may also expect this to lead to frequent evolutionary transitions in preference between hopping and digging.

Robust and at least partly burrowing frogs typify the genus *Rhombophryne*. Some species are specialised burrowers (*R. matavy* and *R. testudo*), while others are probably facultative burrowers (*R. serratopalpebrosa* group, and probably *R. alluaudi*, *R. laevipes*, and *R. mangabensis*). Two species however seem to have at least partly abandoned burrowing: *R. minuta* and *R. longicrus*. *Rhombophryne minuta* lives at high altitude (close to and above the tree line) on the Marojejy massif and calls from low bushes that make up the complex matrix of ground in its unusual habitat (Glaw and Vences 2007). *Rhombophryne longicrus* lives at high altitude on the Sorata Massif to the north of Marojejy; its ecology is more or less unknown. These species form a strongly supported sister group in our phylogeny (Fig. 2), and resemble each other in morphology. Most notably, both have exceptionally long, slender legs relative to those of their congeners: HILs of both species are between 178.5% and 183.8% of their SVLs, considerably above the genus mean of 158.0%; and relative to their lengths, the thighs of *R. longicrus* are thinner than any other *Rhombophryne* species (THIW 29.1–29.2% of THIL) except *R. minuta* (whose THIW/THIL ranges from 24.4–38.0%; Scherz et al. unpublished data). We therefore expect these frogs to be capable of leaps of relatively greater distance than their congeners.

In addition to leg length, several other characters are also associated with more saltatorial locomotion. Emer-

son (1979) characterised three types of iliosacral articulation correlated with locomotion patterns: Type I, expanded sacral diapophyses without ligament attachment, allowing great anteroposterior freedom of movement, most common in aquatic frogs, but also found in burrowers and climbers (Reilly and Jorgensen 2011); Type IIA, broad sacral diapophyses and proximal attachment of a broad ligament, the most adaptable and widespread of the articulation types, typical of walking and hopping locomotion, common in burrowers; and Type IIB, distal attachment of a narrow ligament to thin and posteriorly pointed sacral diapophyses, typical of long-distance leapers (although frogs with this type of articulation are not necessarily better leapers; see Reilly and Jorgensen 2011). Reilly and Jorgensen (2011) expanded this classification into seven types, by incorporating also dorsal ridges of the iliac shafts, and the nature of the urostyle. The iliosacral articulation of *R. longicrus* is Type IIA sensu Emerson (1979), and the ilia possess no ridges, while the urostyle is bicondylar and bears a ridge on its anterior third (as in all other *Rhombophryne* spp. so far investigated).

The iliosacral articulation of *Rhombophryne longicrus* is almost the same as members of the *R. serratopalpebrosa* group (Scherz et al. 2014, 2015), but is slightly modified such that its iliac shafts are closer together. This may produce faster launch speeds and thus greater leaping distances (Choi et al. 2003). This, coupled with its long, slim legs and lack of burrowing specialisations, such as enlarged internal metatarsal tubercles, suggests adaptation to saltatorial locomotion. In external morphology, and based on preliminary osteological data, its sister species, *R. minuta*, appears to share most of these characteristics. Therefore, we hypothesise that these two species constitute a divergent, ancestrally saltatorial lineage that diverged from possibly semi-fossorial ancestors. A thorough treatment of the osteology of *Rhombophryne* will shed light on this question. It is clear already, however, that this genus constitutes an osteologically and ecologically diverse group of frogs, rivalling the diversity seen in other cophylines.

The discovery of such a distinctive new species highlights the incompleteness and patchiness of herpetological survey work in Madagascar. Whilst some forests, particularly accessible, protected ones, are receiving a lot of research attention (e.g. Betampona: Andreone et al. 2010; Rosa et al. 2012, 2014), others, like the forests of the Sorata massif, are receiving little study. Sorata is part of a constellation of high-altitude massifs, linked to the massifs Tsaratanana to the west, and Marojejy to the east by narrow stretches of remaining forest. Further survey work will be needed to understand its diversity and role in this network of massifs. At the same time, however, its forests are unprotected and highly threatened by anthropogenic habitat destruction and modification. Protected status must therefore be pursued together with an enhanced knowledge of this area's flora and fauna.

Acknowledgements

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Supplementary material 1

PDF-embedded 3D Skeletal Model

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Data type: Adobe PDF file

Explanation note: This file contains a PDF-embedded interactive 3D model of the skeleton of the holotype of *Rhombophryne longicrus* sp. n., ZSM 1630/2012, generated via X-ray micro-Computed Tomography. The model can be opened in Adobe® Acrobat Pro or Reader, versions IX and above

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Life in the spray zone – overlooked diversity in West African torrent-frogs (Anura, Odontobatrachidae, *Odontobatrachus*)

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Abstract

Key Words

West African torrent-frogs of the genus *Odontobatrachus* currently belong to a single species: *Odontobatrachus natator* (Boulenger, 1905). Recently, molecular results and biogeographic separation led to the recognition of five Operational Taxonomic Units (OTUs) thus identifying a species-complex. Based on these insights, morphological analyses on more than 150 adult specimens, covering the entire distribution of the family and all OTUs, were carried out. Despite strong morphological congruence, combinations of morphological characters made the differentiation of OTUs successful and allowed the recognition of five distinct species: *Odontobatrachus natator*, and four species new to science: *Odontobatrachus arndti* **sp. n.**, *O. fouta* **sp. n.**, *O. smithi* **sp. n.** and *O. ziama* **sp. n.** All species occur in parapatry: *Odontobatrachus natator* is known from western Guinea to eastern Liberia, *O. ziama* **sp. n.** from eastern Guinea, *O. smithi* **sp. n.** and *O. fouta* **sp. n.** from western Guinea, *O. arndti* **sp. n.** from the border triangle Guinea-Liberia-Côte d'Ivoire. In addition, for the first time the advertisement call of a West African torrent-frog (*O. arndti* **sp. n.**) is described.

Introduction

For a long time all West African torrent-frogs have been assigned to the genus *Petropedetes* Reichenow, 1874, until their generic distinctiveness from Central African species was revealed by Barej et al. (2014a). Based on molecular and osteological characters Barej et al. (2014b) even placed them in their own family, the Odontobatrachidae, which is endemic to West Africa and the Upper Guinea region.

West African torrent-frogs are nocturnal, inhabit lotic waters and usually occur close to streams with strong currents, cascades or rapids in forested areas. However, Rödel (2003) also collected specimens in gallery forests surrounded by savannah in Mont Sangbé National Park, Côte d'Ivoire. While females are usually found in close proximity to rapids and waterfalls, males may sit on rocky surfaces further away (Rödel 2003). Tadpoles are well adapted to life in torrents. With a dorso-ventrally depressed body and sucker-like mouthparts with enlarged labials, they live attached to rocks in strongest currents or adhere to rock-surfaces in the spray zone. These special mouthparts are only reduced at the very last stages of metamorphosis, which is typical for rheophilous larvae (Lamotte and Zuber-Vogeli 1954; Guibé and Lamotte 1958; Channing et al. 2012). The adult frogs are characterised by a medium to large body length (females reaching > 60 mm snout-urostyle length), the possession of dilated, heart-shaped toe tips, a rough dorsal skin texture with glandular ridges, mandibular fangs in both sexes and femoral glands in males (Boulenger 1905; Barej et al. 2014a). These frogs have a patchy distribution within the Upper Guinea forest region, roughly ranging from western Guinea through Sierra Leone and Liberia to western Côte d'Ivoire (Boulenger 1905; Guibé and Lamotte 1958; Böhme 1994b; Rödel 2003; Rödel et al. 2004a; Hillers and Rödel 2007; Hillers et al. 2008a).

Since the first description, West African torrent-frogs have been regarded as a single species: *Odontobatrachus natator* (Boulenger, 1905). Although inter-population differences in colouration and shape of dorsal glands have been reported (Rödel and Bangoura 2004; Rödel et al. 2004a), this has not resulted in taxonomic actions. Based on molecular data, five distinct lineages (therein treated as Operational Taxonomic Units, OTUs) were recognised in this supposedly monospecific family, indicating hitherto overlooked cryptic species (Barej et al. 2015). All samples from Sierra Leone, the type locality of *O. natator*, are grouped in a single clade which has consequently been assigned to the nominate taxon. Two OTUs occur in the westernmost and two more in the easternmost distribution of the family Odontobatrachidae. While the largest area is occupied by the nominate taxon *O. natator*, OTUs show a tendency to parapatric distribution with little overlap in their potential distribution areas; exceptionally the two western OTUs possess a similar range according to modelled distribution. The recognition of a potential species-complex in the presumably monospecific frog family Odontobatrachidae demonstrated that the current threat classification as "Near Threatened (NT)" (IUCN 2011) is insufficient as recognised OTUs possess very small distribution ranges, demanding a higher threat classification. However, a reassessment of threat categories and subsequent conservation actions require formal description of new species.

We herein present morphological results gathered from more than 150 specimens, covering the entire geographic distribution of the family Odontobatrachidae. Morphological characteristics were analysed and interpreted in combination with the published molecular data and biogeographic insights after Barej et al. (2015). Consequently, we re-describe *Odontobatrachus natator* (Boulenger, 1905), describe four new species, and provide the first call analysis for *Odontobatrachus*.

Material and methods

Species concept and species delimitation

We herein follow the General Lineage Concept of species (de Queiroz 1998, 1999) and accept distinctiveness on species level based on both morphological and genetic data. The genetic data have already been presented by Barej et al. (2015), indicating four undescribed candidate species and their relationships (compare Vieites et al. 2009). OTUs defined by Barej et al. (2015) were the basis of our morphological analyses. Consequently these molecular clades were taken as *a priori* group assignments to ensure understanding of the overall morphological character diversity within and between OTUs. Herein, we accept all five OTUs sensu Barej et al. (2015) and consequently four new species are described in the following. We re-describe the nominate species *Odontobatrachus natator* (blue colour code) and describe OTU1 as *O. ziama* sp. n. (red), OTU2 as *O. smithi* sp. n. (yellow), OTU3 as *O. fouta* sp. n. (green) and OTU4 as *O. arndti* sp. n. (orange). For convenience we will use these names without the suffix sp. n. throughout the manuscript, anticipating their formal description below. Environmental Niche Models (see Barej et al. 2015) confirm the overall distribution pattern of the family. No major range extensions are expected and modelled niches of the individual species are very similar. Interestingly niche similarity identified two groups: *O. smithi* and *O. fouta* in the first and the remaining three taxa in the second. The distribution of all five OTUs and herein recognised species is illustrated in Figure 1.

Morphology

Collected frogs were anesthetized either with chlorobuthanol or benzocaine solutions and thereafter fixed in 4% formalin or 70% ethanol. All voucher specimens have been transferred to 75% ethanol for long-term storage. Abbreviations of museum collections hosting the investigated vouchers are as follows: The Natural History Museum (BMNH), London, United Kingdom; Natural History Museum of Geneva (MHNG), Geneva, Switzerland; Zoologisches Forschungsmuseum Alexander Koenig (ZFMK), Bonn and Museum für Naturkunde Berlin (ZMB), Berlin, both Germany.

Measurements follow standard procedures and were taken on preserved material with an electronic dial calliper (± 0.1 mm) by one person (MFB). Webbing formulae are composed as follows: dividing different toes by a '-'

and differentiating inner and outer side of toe by a '/', thus the example 0-0.5/0-1/0-1/1-0 translates to: the webbing reaches the disc at toe I, webbing extends halfway between the most proximate tarsal tubercle to the disc at the external side of toe II and the disc at the internal side of this toe, etc. Tarsal tubercles are counted from tip of the toe to toe base. Additional qualitative characters such as skin granulation were recorded, but not always ascertainable in all vouchers, probably due to different preservation procedures.

Recorded measures comprise: snout–urostyle length (SUL); head width at level of jaw articulation (HW); horizontal orbita diameter (O); interorbital distance (ID); horizontal tympanum diameter (TD); eye–nostril distance (EN); eye–snout distance (ES); length of femur (FM); femoral gland length (GL); femoral gland width (GW); tibiofibula length (TI); foot length without tarsus (FL); inner tarsal tubercle (IT). Additionally, the following ratios have been calculated and analysed: TI/SUL, FM/TI, FL/SUL, GL/FM, GL/GW, HW/SUL, TD/O, FM/SUL, IT/FL, O/EN, ES/O, TD/SUL. Measurements are summarised separately for males (Table 1) and females (Table 2).

Statistical analyses

Potential statistical discrimination of OTUs by morphological data was tested in SPSS 20 and R 3.1.0 (default packages; R Core Team 2013). We only included complete data sets in statistical analyses; damaged specimens or specimens with preservation artefacts were excluded. Furthermore, only measurements of adult frogs were taken into consideration. In order to consider sex-specific characters, e.g. femoral glands (present in males only) or size dimorphism (females growing larger than males), sexes were analysed separately.

Natural Log (ln) transformations were applied on measurements before analysis to obtain a homogeneous data distribution. Principal component analyses (PCA) were performed to explore the overall morphological variation between the putative taxa. Subsequently, we tested for significance of differences between OTUs with non-parametric tests (Kruskal-Wallis H test) since morphological datasets often violated the assumptions of standard parametric statistics and non-parametric tests are generally considered to be more conservative, not relying on assumptions such as random sampling, normality and homogeneity of variance (Anderson 2001). We finally tested for sex-based morphological characteristics within each OTU (Mann-Whitney U test). A Type I error of p < 5% was chosen to reject the null hypothesis. Sampling of included vouchers per OTU/species was as follows (N_{male}/N_{female}): *Odontobatrachus natator* (22/29), *O. ziama* (11/30), *O. smithi* (3/6), *O. fouta* (3/4) and *O. arndti* (26/24).

Finally, canonical discriminant function analyses (CDA) were performed on ln-transformed mensural variables to test whether our *a priori* groupings could be confirmed. These analyses maximised separations between groups based on within-group variance and correlation. CDA were again implemented on female and male datasets independently. Both the PCA analyses and the CDA were performed separately on absolute values and morphometric ratios.

Advertisement calls

Odontobatrachus call recordings were collected from specimens in terraria (vouchers collected on Nimba Mts., Guinea). Oscillograms (waveforms) and audiospectrograms (sonograms) as results of the Fast Fourier Transformation (FFT; frequency spectrum) were examined for spectral and temporal characters (analysis settings: 44.1 kHz sample ratio, 16 bits resolution, FFT length = 256). Call recordings were analysed with the software package Soundruler v0.9.6 (Gridi-Papp 2007), spectrograms and oscillograms were prepared with the software package Seewave for R (Sueur et al. 2008). Values of call duration, dominant frequency, fundamental frequency and number of notes are presented as minimum and maximum only, because of low numbers of recorded calls.

Genetics

Phylogeographic analyses included samples from the entire family range and were based on mitochondrial (12S, 16S, CYTB) and nuclear (BDNF, SIA, RAG1) genes (Barej et al. 2015). Herein, we present uncorrected 16S p-distances between species from Barej et al. (2015); a table providing inter and intra-species distances is provided in the Appendix 1: Table A. A list of samples gathered in addition to Barej et al. (2015) and respective GenBank numbers are given in Appendix 2: Table B. For laboratory procedures see Barej et al. (2014a).

Conservation status

Following IUCN Red List criteria, Barej et al. (2015) calculated the Extent of Occurrence (EOO) and the Area of Occupancy (AOO) using GeoCat (2011) for *Odontobatrachus natator* and four additional OTUs, herein described as new species. While EOO, often measured by a minimum convex polygon, corresponds to the contained area of a species, AOO refers to the area within the species EOO, excluding cases of unoccupied or unsuitable habitat. According to IUCN regulation, the higher of the two classifications is crucial for assessing global extinction risk.

Results and discussion

All specimens have been assigned to five OTUs *a priori* (OTUs at the putative species level following molecular results in Barej et al. 2015). Phenetic differences of all OTUs were assessed by carrying out a principal component analysis (PCA) on the respective datasets of a total of 65 males and 93 females. Due to the low number of available data points for *O. smithi* and *O. fouta*, placement of their centroids has to be regarded with caution.
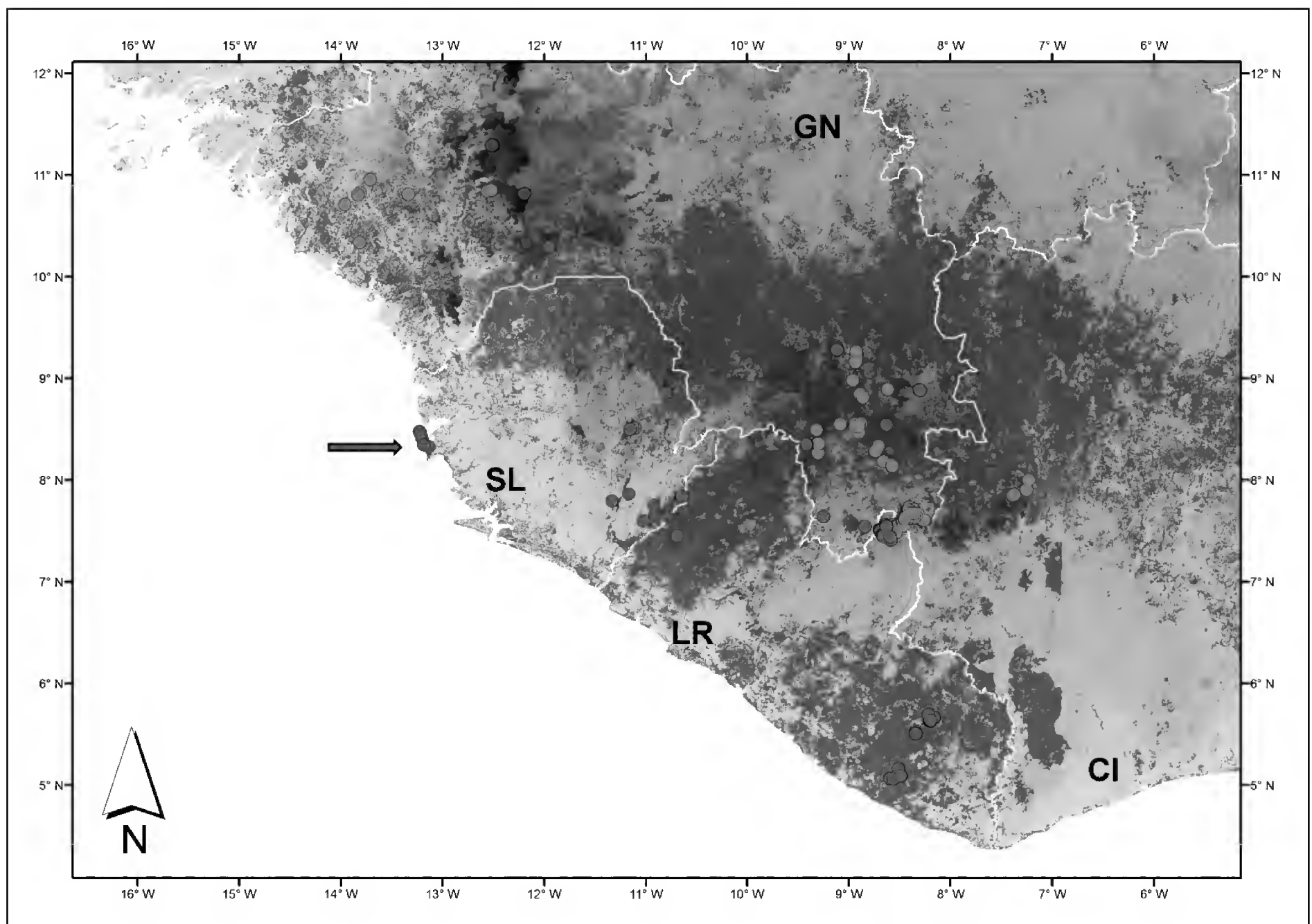
Figure 1. Distribution of *Odontobatrachus* spp. in the western Upper Guinea forest zone (country code: GN = Guinea, SL = Sierra Leone, LR = Liberia, CI = Côte d'Ivoire). The map shows forest cover (Arino et al. 2012; green shading) and elevation (increasing from light to dark grey). The arrow indicates the restricted type locality of *O. natator*. Colour code reads as follows: blue = *O. natator*, red = *O. ziama* sp. n., yellow = *O. smithi* sp. n., green = *O. fouta* sp. n., orange = *O. arndti* sp. n.

PCA results of absolute values in male *Odontobatrachus* (Fig. 2a) revealed that GL and GW contributed most to axis 1 and axis 1 accounted for 52.84% of the total variance. Axis 2 contributed an additional 23.23%, summing up to a total of more than 75% of the variance explained (Table 3). Axis 2 consisted mostly of contributions by EN, TD and GW. All contributors to axes 1 and 2 in the analysis of absolute values in males are given in Table 3. Axis 3 increased the total explained variance by only 6.60% (contributors not shown). The centroid of *O. ziama* was separated from those of *O. natator* and *O. smithi* on the second axis and from that of *O. arndti* by the first axis. Centroids of *O. ziama* and *O. fouta* were separated on both axes. The centroid of *O. smithi* was separated from that of *O. fouta* on the second axis and from that of *O. arndti* on both axes. The centroid of *O. fouta* was separated from those of *O. natator* and *O. smithi* on the second axis and from that of *O. arndti* on the first axis. Centroids of *O. fouta* and *O. arndti* were separated on the second axis. Centroids of *O. arndti* and *O. natator* were separated on both axes (Fig. 2a).

PCA results of morphometric ratios in male *Odontobatrachus* (Fig. 2b) revealed that main contributors to axis 1 were GL/FM, OD/EN, TD/OD and TD/SUL and axis 1 accounted for 41.28% of the total variance. Axis 2 contributed an additional 17.06%, summing up to a total of 58.35% of the variance explained (Table 4). The loading of this axis was mostly made up of contributions by GL/FM and TD/SUL. All contributors to axes 1 and 2 in the analysis of morphometric ratios in males are given in Table 4. Axis 3 explained an additional 13.44% of the variance (contributors not shown). The centroid of *O. ziama* from was separated from those of *O. natator*, *O. smithi* and *O. fouta* on the first axis and from that of *O. arndti* on the second axis. The centroid of *O. arndti* was separated from those of *O. natator*, *O. smithi* and *O. fouta* on both axes. Individuals of *O. natator*, *O. smithi* and *O. fouta* strongly overlapped in this plot (Fig. 2b).

PCA results of absolute values in female *Odontobatrachus* (Fig. 2c) revealed that IT and TD contributed mostly to axis 1 and axis 1 accounted for 59.02% of the total variance. Axis 2 contributed an additional

Table 1. Summary of morphological measures (in mm) of adult male *Odontobatrachus* species. Minimum (min), maximum (max), mean values (mean), standard deviation (SD) and number of included vouchers (N) are given. Measurements of holotypes are provided separately. For abbreviations see material and methods section.

Taxon		SUL	HW	FM	GL	GW	TI	FL	IT	TD	O	ID	EN	ES	TI/SUL	FM/TI	FL/SUL	GL/FM	GL/GW	HW/SUL	TD/O	FM/SUL	IT/FL	O/EN	ES/O	TD/SUL
O. natator	min	42.6	16.1	21.9	8.7	4.0	23.3	19.6	2.8	2.4	6.1	4.6	3.2	5.9	0.49	0.91	0.42	0.36	1.71	0.34	0.33	0.47	0.14	1.55	0.84	0.05
	max	52.5	19.0	25.5	13.7	7.4	26.8	23.1	4.3	3.1	7.5	5.9	4.1	6.8	0.57	1.02	0.51	0.56	2.32	0.39	0.47	0.54	0.20	2.06	1.03	0.06
	mean	48.0	17.3	23.8	10.9	5.6	24.8	21.4	3.6	2.7	6.8	5.2	3.7	6.2	0.52	0.96	0.45	0.46	1.96	0.36	0.40	0.50	0.17	1.82	0.92	0.06
	SD	2.2	0.8	1.0	1.3	0.8	1.1	0.9	0.4	0.2	0.4	0.3	0.2	0.2	0.02	0.03	0.02	0.06	0.16	0.01	0.04	0.02	0.02	0.11	0.05	0.00
	N	23	23	23	23	23	23	23	23	23	23	23	23	23	23	23	23	23	23	23	23	23	23	23	23	23
O. ziama sp. n.	ZMB 78300 (holotype)	46.1	15.5	24.1	12.8	7.6	24.4	21.4	3.7	2.4	6.5	5.2	3.3	6.4	0.53	0.99	0.46	0.53	1.69	0.34	0.37	0.52	0.17	2.00	0.98	0.05
	min	43.0	15.0	19.6	7.5	4.7	22.4	19.0	2.4	1.8	6.3	4.0	2.7	5.3	0.49	0.86	0.42	0.38	1.60	0.34	0.27	0.45	0.12	1.95	0.84	0.04
	max	50.3	17.5	24.9	13.7	7.7	24.8	23.2	3.9	2.7	7.1	5.2	3.4	6.7	0.54	1.00	0.53	0.59	2.13	0.37	0.40	0.52	0.18	2.51	1.00	0.06
	mean	45.10	15.86	22.24	11.54	6.31	23.52	20.83	3.29	2.25	6.54	4.75	3.05	5.95	0.52	0.95	0.46	0.51	1.83	0.35	0.34	0.49	0.16	2.16	0.91	0.05
	SD	1.99	0.70	1.46	1.77	0.83	0.65	1.17	0.43	0.26	0.25	0.37	0.25	0.44	0.01	0.05	0.02	0.07	0.17	0.01	0.04	0.02	0.02	0.17	0.05	0.01
	N	12	12	12	11	11	12	12	12	12	12	12	12	12	12	12	12	11	11	12	12	12	12	12	12	12
O. smithi sp. n.	ZMB 78310 (holotype)	60.4	21.9	29.9	15.6	7.2	30.4	25.7	4.3	3.3	7.9	5.7	4.7	7.7	0.50	0.98	0.42	0.52	2.17	0.36	0.42	0.49	0.17	1.69	0.98	0.05
	min	40.1	15.0	19.4	7.4	3.8	19.8	17.4	3.1	2.3	5.6	4.5	3.5	5.8	0.49	0.92	0.42	0.38	1.95	0.36	0.41	0.48	0.15	1.51	0.98	0.05
	max	60.4	21.9	29.9	15.6	7.2	30.4	25.7	4.3	3.3	7.9	5.7	4.7	7.7	0.53	0.98	0.45	0.52	2.24	0.39	0.49	0.49	0.19	1.69	1.07	0.06
	mean	48.9	18.3	23.9	11.3	5.3	24.8	21.3	3.6	2.8	6.4	4.9	4.0	6.6	0.51	0.96	0.44	0.46	2.12	0.38	0.44	0.49	0.17	1.60	1.03	0.06
	SD	10.4	3.5	5.4	4.1	1.7	5.3	4.2	0.6	0.5	1.2	0.7	0.6	1.0	0.02	0.03	0.01	0.07	0.16	0.01	0.04	0.01	0.02	0.09	0.05	0.00
	N	3	3	3	3	3	3	3	3	3	3	3	3	3	3	3	3	3	3	3	3	3	3	3	3	3
O. fouta sp. n.	ZMB 78314 (holotype)	55.6	21.6	27.8	14.3	8.4	28.9	25.8	4.1	3.1	7.7	5.9	3.7	7.1	0.52	0.96	0.46	0.51	1.70	0.39	0.41	0.50	0.16	2.07	0.93	0.06
	min	47.8	17.3	23.1	8.9	5.3	25.6	22.1	3.2	2.8	6.3	5.3	3.7	5.8	0.52	0.90	0.44	0.35	1.69	0.36	0.40	0.48	0.14	1.49	0.92	0.05
	max	57.0	21.6	27.9	14.4	8.4	29.8	25.8	4.2	3.1	7.8	5.9	4.3	7.4	0.53	0.97	0.46	0.52	1.79	0.39	0.46	0.51	0.18	2.07	1.06	0.06
	mean	52.5	19.6	26.0	12.5	7.2	27.6	23.8	3.9	3.0	7.0	5.6	4.0	6.8	0.53	0.94	0.45	0.46	1.72	0.37	0.43	0.50	0.16	1.78	0.96	0.06
	SD	4.5	2.3	2.3	3.2	1.7	2.0	1.9	0.5	0.2	0.8	0.3	0.2	0.7	0.01	0.03	0.01	0.10	0.06	0.01	0.03	0.01	0.02	0.26	0.06	0.00
	N	4	4	4	3	3	4	4	4	4	4	4	4	4	4	4	4	3	3	4	4	4	4	4	4	4
O. arndti sp. n.	ZMB 78355 (holotype)	48.8	17.1	24.6	13.0	7.9	26.3	24.1	3.7	2.7	7.6	5.2	3.4	6.0	0.54	0.93	0.49	0.53	1.65	0.35	0.36	0.50	0.15	2.24	0.78	0.06
	min	43.5	15.2	22.5	10.4	5.1	23.4	20.0	2.9	2.2	6.0	4.2	3.0	5.2	0.50	0.80	0.44	0.40	1.50	0.33	0.32	0.45	0.13	1.81	0.76	0.04
	max	53.6	19.3	27.2	15.1	8.7	28.6	24.8	4.4	3.0	7.6	5.7	3.8	7.0	0.59	1.04	0.50	0.59	2.14	0.36	0.44	0.56	0.19	2.47	1.05	0.07
	mean	49.1	17.3	24.9	12.8	6.9	26.2	23.0	3.5	2.6	7.0	5.0	3.4	6.1	0.5	0.95	0.47	0.51	1.86	0.35	0.37	0.51	0.15	2.05	0.89	0.05
	SD	2.3	1.0	1.3	1.3	1.1	1.2	1.2	0.3	0.2	0.4	0.4	0.2	0.4	0.0	0.04	0.02	0.05	0.17	0.01	0.03	0.02	0.01	0.17	0.07	0.01
	N	28	28	28	26	26	28	28	28	28	28	28	28	28	28	28	28	26	26	28	28	28	28	28	28	28

Table 2. Summary of morphological measures (in mm) of adult female *Odontobatrachus* species. Minimum (min), maximum (max), mean values (mean), standard deviation (SD) and number of included vouchers (N) are given. For abbreviations see material and methods section.

| Taxon		SUL	HW	FM	TI	FL	IT	TD	O	ID	EN	ES	TI/SUL	FM/TI	FL/SUL	HW/SUL	TD/O	FM/SUL	IT/FL	O/EN	ES/O	TD/SUL
O. natator	min	44.6	15.9	22.0	23.7	20.2	2.5	2.4	6.2	4.6	3.4	5.4	0.45	0.87	0.39	0.32	0.34	0.43	0.12	1.60	0.82	0.04
	max	61.1	21.2	28.1	29.8	26.1	4.9	3.4	7.9	6.4	4.7	7.5	0.58	1.01	0.51	0.38	0.46	0.55	0.20	2.18	1.11	0.06
	mean	53.6	18.6	26.0	27.2	23.7	3.8	2.9	7.1	5.4	3.9	6.6	0.51	0.96	0.44	0.35	0.40	0.49	0.16	1.80	0.93	0.05
	SD	5.0	1.4	1.6	1.5	1.6	0.5	0.3	0.5	0.5	0.3	0.5	0.03	0.03	0.03	0.02	0.03	0.03	0.02	0.13	0.07	0.00
	N	31	31	31	31	31	29	31	31	31	31	31	31	31	31	31	31	31	29	31	31	31
O. ziama sp. n.	min	44.3	15.1	21.9	23.9	20.7	2.7	1.9	6.0	4.3	2.9	5.8	0.45	0.88	0.38	0.30	0.29	0.41	0.12	1.69	0.84	0.04
	max	60.3	19.8	27.7	29.7	25.4	4.2	2.7	7.7	5.8	3.8	7.4	0.56	1.03	0.49	0.40	0.40	0.53	0.18	2.38	1.10	0.06
	mean	52.1	17.5	24.8	26.3	22.8	3.6	2.4	6.9	5.0	3.4	6.5	0.51	0.94	0.44	0.34	0.34	0.48	0.16	2.03	0.94	0.05
	SD	4.3	1.2	1.6	1.4	1.2	0.3	0.2	0.4	0.4	0.2	0.5	0.03	0.03	0.03	0.02	0.02	0.03	0.01	0.17	0.08	0.00
	N	30	30	30	30	30	30	30	30	30	30	30	30	30	30	30	30	30	30	30	30	30
O. smithi sp. n.	min	48.7	18.1	22.7	24.7	21.8	3.0	2.7	6.7	4.2	3.8	5.9	0.49	0.91	0.42	0.35	0.39	0.45	0.14	1.61	0.87	0.05
	max	61.9	22.4	28.6	31.2	27.9	5.1	3.7	7.8	6.3	4.8	8.1	0.57	0.96	0.51	0.39	0.48	0.52	0.19	1.79	1.12	0.07
	mean	54.1	20.1	25.9	27.9	24.7	4.0	3.2	7.1	5.6	4.2	7.0	0.52	0.93	0.46	0.37	0.44	0.48	0.16	1.69	0.98	0.06
	SD	4.4	1.7	2.4	2.6	2.5	0.8	0.4	0.5	0.7	0.3	0.8	0.03	0.02	0.03	0.01	0.03	0.03	0.02	0.07	0.10	0.01
	N	6	6	6	6	6	6	6	6	6	6	6	6	6	6	6	6	6	6	6	6	6
O. fouta sp. n.	min	44.1	16.8	22.5	23.7	20.9	2.4	2.8	6.0	4.7	3.6	5.9	0.48	0.95	0.41	0.34	0.41	0.47	0.11	1.57	0.97	0.05
	max	62.5	22.2	29.4	30.2	26.3	4.8	3.9	7.7	6.4	4.5	8.5	0.55	0.98	0.49	0.40	0.50	0.52	0.18	1.88	1.10	0.07
	mean	51.1	18.6	24.8	25.8	22.5	3.6	3.1	6.6	5.3	3.9	6.8	0.51	0.96	0.44	0.37	0.47	0.49	0.16	1.71	1.02	0.06
	SD	8.2	2.4	3.2	3.0	2.6	1.0	0.5	0.8	0.8	0.4	1.2	0.03	0.01	0.03	0.03	0.04	0.02	0.03	0.13	0.06	0.01
	N	4	4	4	4	4	4	4	4	4	4	4	4	4	4	4	4	4	4	4	4	4
O. arndti sp. n.	min	45.9	16.2	22.3	24.7	20.6	3.1	2.4	6.1	4.5	3.0	5.7	0.47	0.88	0.42	0.31	0.31	0.44	0.13	1.65	0.75	0.04
	max	64.0	20.9	29.8	31.3	27.7	4.7	3.4	8.3	5.9	4.3	7.3	0.56	0.97	0.51	0.37	0.45	0.53	0.18	2.59	1.00	0.06
	mean	56.1	18.9	27.1	29.2	25.7	3.8	2.8	7.4	5.3	3.6	6.5	0.52	0.93	0.46	0.34	0.38	0.48	0.15	2.04	0.89	0.05
	SD	4.5	1.2	2.0	1.9	1.8	0.4	0.2	0.6	0.4	0.3	0.4	0.02	0.03	0.02	0.01	0.04	0.02	0.01	0.19	0.06	0.00
	N	25	25	25	25	25	24	25	25	25	25	25	25	25	25	25	25	25	24	25	25	25
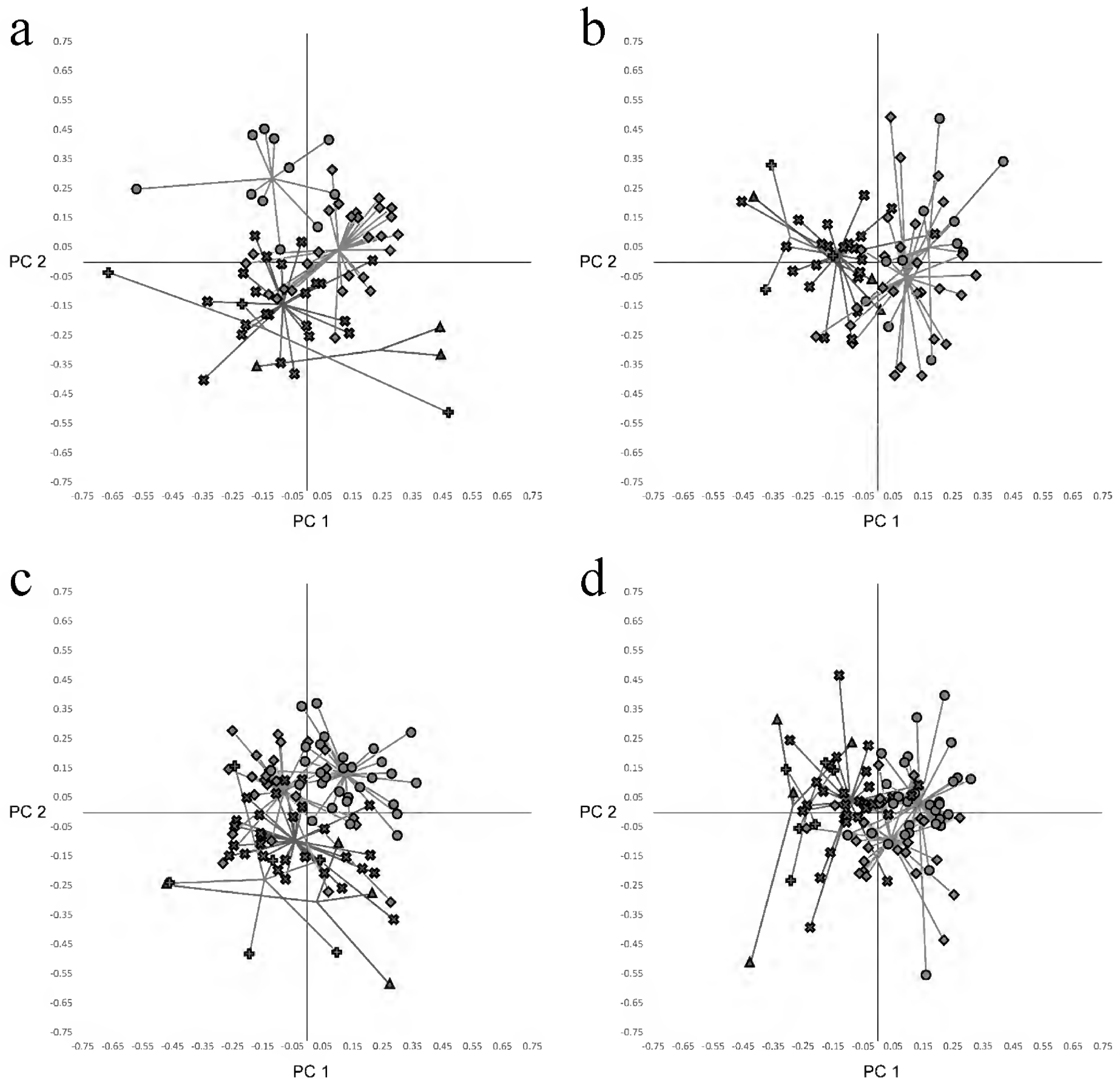
Figure 2. Scatter plot of the first and second axis of the principal component analyses for absolute values of morphological measurements in males (**a**) and females (**c**), and respective ratios in males (**b**) and females (**d**). For PCA loadings see Tables 3 and 4. Lines are drawn to indicate the centroid of points for each species. For colour code see Fig. 1.

14.27%, summing up to a total of more than 73% variance explained (Table 3). Axis 2 was mostly made up of contributions by TD and EN. All contributors to axes 1 and 2 in the analysis of ratios in males are given in Table 3. Axis 3 increased the total explained variance by only 8.79% (contributors not shown). The centroid of *O. ziama* was separated from those of *O. natator* and *O. smithi* on both axes, from that of *O. fouta* on the second axis and from that of *O. arndti* on the first axis. Centroids of *O. smithi* and *O. natator* were separated from that of *O. fouta* on the first axis and from that of *O. arndti* on the second axis. Centroids of *O. arndti* and *O. fouta* were separated on both axes from each other (Fig. 2c).

PCA results of morphometric ratios in female *Odontobatrachus* (Fig. 2d) revealed that main contributors to axis 1 were TD/O and TD/SUL and axis 1 accounted for 48.38% of the total variance. Axis 2 contributed additional 17.84%, summing up to a total of 66.22% of the variance explained (Table 4). Axis 2 was mostly made up of IT/FL. All contributors to axes 1 and 2 in the analysis of ratios in females are given in Table 4. Axis 3 explained an additional 12.49% (contributors not shown). The centroid of *O. ziama* was separated from those of *O. natator*, *O. smithi* and *O. fouta* on the first axis and from that of *O. arndti* on the second axis. The centroid of *O. arndti* was separated from those of *O. natator*, *O. smithi* and *O. fouta* on both axes (Fig. 2d).

Table 3. Principle component loadings for male (left) and female (right) absolute values. Eigenvalues, percent of explained variance for the first two axis and the cumulative variance are given.

	male		female	
	PC1	PC2	PC1	PC2
Eigenvalue	0.0766	0.0337	0.0581	0.0141
Percent variance	52.84	23.23	59.02	14.27
Cumulative variance	52.84	76.07	59.02	73.29
Loadings (absolute values)				
Snout-urostyle length (SUL)	0.2444	-0.1534	-0.4375	0.1178
Head width (HW)	0.2224	-0.2185	-0.4029	-0.0173
Femur length (FM)	0.2774	-0.1283	-0.3644	0.1217
Femoral gland length (GL)	0.6581	0.1982	---	---
Femoral gland width (GW)	0.7881	0.3470	---	---
Tibiofibula length (TI)	0.2547	-0.1241	-0.3523	0.1340
Foot length (FL)	0.2796	-0.0701	-0.3901	0.1429
Inner metatarsal tubercle length (IT)	0.2712	-0.2628	-0.5340	0.2196
Tympanum diameter (TD)	0.1145	-0.3954	-0.4940	-0.4614
Orbita diameter (OD)	0.2316	-0.0549	-0.3427	0.0844
Interorbital distance (ID)	0.2106	-0.1749	-0.3683	-0.0191
Distance eye to naris (EN)	0.0921	-0.3951	-0.3608	-0.3051
Distance eye to snout (ES)	0.1818	-0.1573	-0.3180	0.0216

Table 4. Principle component loadings for male (left) and female (right) ratios. Eigenvalues, percent of variance for the first two axes (PC1 and PC2) and the cumulative variance are given.

	male		female	
	PC1	PC2	PC1	PC2
Eigenvalue	0.0362	0.0150	0.0338	0.0124
Percent variance	41.28	17.06	48.38	17.84
Cumulative variance	41.28	58.35	48.38	66.22
Loadings (ratios)				
Tibiofibula length to snout-urostyle length (TI/SUL)	0.0345	-0.0091	-0.0750	-0.1917
Femur length to tibiofibula length (FM/TI)	-0.0043	0.0246	0.0024	-0.0115
Foot length to snout-urostyle length (FL/SUL)	0.1005	-0.0628	-0.0746	-0.2220
Femoral gland length to femur length (GL/FM)	0.4487	-0.3126	---	---
Femoral gland length to femoral gland width (GL/GW)	-0.1595	0.0494	---	---
Head width to snout-urostyle length (HW/SUL)	-0.0784	0.0093	-0.1872	-0.1029
Tympanum diameter to orbita diameter (TD/OD)	-0.5397	-0.2602	-0.6797	0.0372
Femur length to snout-urostyle length (FM/SUL)	0.0302	0.0155	-0.0726	-0.2032
Inner tarsal tubercle length to foot length (IT/FL)	-0.1794	0.0933	-0.0964	0.5165
Orbita diameter to distance eye naris (OD/EN)	0.4411	-0.2198	0.4463	-0.0762
Distance eye snout to orbita diameter (ES/OD)	-0.1440	0.2105	-0.1403	0.1742
Tympanum diameter to snout urostyle length (TD/SUL)	-0.4402	-0.3605	-0.6957	-0.0986

Generally, PCA results on morphology supported the separation of the five molecular OTUs sensu Barej et al. (2015; *O. natator*, *O. ziama*, *O. smithi*, *O. fouta*, *O. arndti*), although males and females often showed overlap in the variance in both analyses (absolute values and ratios) between OTUs. Two major morphological groupings *O. natator*, *O. smithi*, *O. fouta* vs. *O. ziama*, *O. arndti* were uncovered in all analyses, with the groups being separated from each other on at least one of the two major axes (Fig. 2).

We tested for statistical differences in particular morphological characters and ratios between species (Kruskal-Wallis H test), considering potential sex-dependant characters (Appendix 3: Table C). These non-parametric tests revealed significant differences in males and females of the five species highlighting their morphological distinctness (Table 5).

Due to the overlap in morphological variation of species (see above), the correct assignment of single individuals would be difficult if their geographic origin is unknown. We therefore applied Detrended Correspondence Analyses (DCAs) to assess how reliably individuals can be assigned to a particular species. DCA results showed high levels of correctly assigned males and females, based on absolute values and ratios of mensural data sets, respectively (Table 6). Combined correct assignments of all five species for absolute values in male *Odontobatrachus* summed up to 89.2% and combined values of ratios referred to 87.7%. DCA results in female *Odontobatrachus* were 82.9% for absolute values and 74.2% in ratios. The lowest percentages of correctly assigned individuals were recovered in the species with the lowest voucher numbers (Table 6). Despite the high percentage of correct assignments, the persisting mismatches reflected the PCA results and highlight the morphological overlap between some species.

Based on the combination of the molecular data recognising five OTUs and their respective distribution patterns (indicating spatial partitioning) presented in Barej et al. (2015), as well as morphological distinction presented herein, we consider the five *Odontobatrachus* OTUs as distinct species. Until now, all Upper Guinean populations were assigned to *Odontobatrachus natator* (Boulenger, 1905) and thus no synonyms are available. We provide diagnostic characters (Table 7) and formally describe four new species.

Systematics of the *Odontobatrachus natator*-complex

Frogs belonging to the genus *Odontobatrachus* are all characterised by the following external morphological characters: tusk-like odontoids on the lower mandible in both sexes; posteriorly curved teeth on premaxillaries and anterior maxillaries; presence of vomerine teeth; eye diameter distinctly larger than tympanum diameter; pupil horizontally elliptical; tympanum rather indistinct; skin

Table 5. Morphological comparison of absolute measurements and ratios in males (lower left corner) and females (upper right corner). Number of samples per sex in each species and level of significance are given; lack of significant differences in absolute values or ratios is marked with "X". See material and methods section for abbreviations.

Taxon	O. natator (N_male,female = 22/29)	O. ziama sp. n. (N_male,female = 11/30)	O. smithi sp. n. (N_male,female = 3/6)	O. fouta sp. n. (N_male,female = 3/4)	O. arndti sp. n. (N_male,female = 26/24)
O. natator		♀♀ O. natator < O. ziama: O/EN (p< 0.001) / ♀♀ O. natator > O. ziama: HW (p< 0.05), TD (p< 0.001), ID (p< 0.05), EN (p< 0.001), HW/SUL (p< 0.01), TD/O (p< 0.001), TD/SUL (p< 0.001)	X	X	♀♀ O. natator < O. arndti: TI (p< 0.05), FL (p< 0.01), O/EN (p< 0.001) / ♀♀ O. natator > O. arndti: EN (p= 0.06), FM/TI (p< 0.05), HW/SUL (p< 0.05)
O. ziama sp. n.	♂♂ O. ziama < O. natator: HW (p<0.01), TD (p< 0.01), EN (p< 0.001), HW/SUL (p< 0.01), TD/O (p< 0.01), TD/SUL (p< 0.05) / ♂♂ O. ziama > O. natator: O/EN (p< 0.01)		♀♀ O. ziama < O. smithi: HW (p< 0.01), TD (p< 0.001), ID (p< 0.05), EN (p< 0.001), HW/SUL (p< 0.001), TD/O (p< 0.001), O/EN (p< 0.001), TD/SUL (p< 0.001)	♀♀ O. ziama < O. fouta: TD (p< 0.001), HW/SUL (p= 0.07), TD/O (p< 0.001), TD/SUL (p< 0.001) / ♀♀ O. ziama > O. fouta: O/EN (p< 0.05)	♀♀ O. ziama < O. arndti: HW (p< 0.01), FM (p< 0.001), TI (p< 0.001), FL (p< 0.001), TD (p< 0.001), O (p< 0.05), ID (p= 0.09), TD/O (p< 0.05)
O. smithi sp. n.	X	♂♂ O. smithi < O. ziama: O/EN (p< 0.01) / ♂♂ O. smithi > O. ziama: EN (p< 0.01), GL/GW (p< 0.01), HW/SUL (p< 0.05), TD/O (p< 0.05)		X	♀♀ O. smithi < O. arndti: O/EN (p< 0.01) / ♀♀ O. smithi > O. arndti: EN (p< 0.05), HW/SUL (p< 0.01), TD/O (p< 0.05), TD/SUL (p< 0.05)
O. fouta sp. n.	♂♂ O. fouta > O. natator: FL (p= 0.08)	♂♂ O. fouta < O. ziama: GL/GW (p= 0.05) / ♂♂ O. fouta > O. ziama: SUL (p< 0.001), HW (p< 0.01), TI (p< 0.001), FL (p< 0.05), TD (p< 0.01), ID (p< 0.05), EN (p< 0.01), HW/SUL (p= 0.07)	♂♂ O. fouta > O. smithi: GL/FM (p< 0.05)		♀♀ O. fouta < O. arndti: FL (p< 0.05), O/EN (p< 0.05) / ♀♀ O. fouta > O. arndti: TI (p< 0.05), TD/O (p= 0.06), ES/O (p< 0.05), TD/SUL (p= 0.06)
O. arndti sp. n.	♂♂ O. arndti < O. natator: EN (p< 0.01), HW/SUL (p= 0.06), TD/SUL (p= 0.05), TD/O (p= 0.06), IT/FL (p< 0.01) / ♂♂ O. arndti > O. natator: GL (p< 0.01); GW (p< 0.01), TI (p<0.01), FL (p< 0.001), TI/SUL (p<0.01), FL/SUL (p< 0.01), O/EN (p< 0.001)	♂♂ O. arndti > O. ziama: SUL (p< 0.01), HW (p< 0.01), FM (p< 0.01), TI (p= 0.08), TD (p= 0.08), O (p<0.01)	♂♂ O. arndti > O. smithi: FL/SUL (p= 0.06), GL/GW (p< 0.05), O/EN (p< 0.01), ES/O (p< 0.05)	♂♂ O. arndti < O. fouta: TD (p= 0.06), EN (p= 0.08) / ♂♂ O. arndti > O. fouta: GL/GW smaller (p= 0.05)	

Table 6. Results from statistical discrimination of species (DCA) using morphological data and pooling individuals according to sex. Percentage of correct assignments, number of cases (N) and overall correct classification rate for absolute values and ratios in males and females are given.

male absolute values	*O. ziama* sp. n.	*O. smithi* sp. n.	*O. fouta* sp. n.	*O. arndti* sp. n.	*O. natator*	N
O. ziama sp. n.	100.0	0.0	0.0	0.0	0.0	11
O. smithi sp. n.	0.0	66.7	33.3	0.0	0.0	3
O. fouta sp. n.	0.0	0.0	100.0	0.0	0.0	3
O. arndti sp. n.	3.8	0.0	0.0	84.6	11.5	26
O. natator	0.0	4.5	0.0	4.5	90.9	22
all taxa (combined)	89.2%					
male ratios						
O. ziama sp. n.	81.8	0.0	0.0	18.2	0.0	11
O. smithi sp. n.	0.0	100.0	0.0	0.0	0.0	3
O. fouta sp. n.	0.0	0.0	100.0	0.0	0.0	3
O. arndti sp. n.	7.7	0.0	3.8	80.8	7.7	26
O. natator	0.0	0.0	0.0	4.5	95.5	22
all taxa (combined)	87.7%					
female absolute values						
O. ziama sp. n.	93.3	3.3	0.0	3.3	0.0	30
O. smithi sp. n.	0.0	83.3	16.7	0.0	0.0	6
O. fouta sp. n.	0.0	0.0	75.0	0.0	25.0	4
O. arndti sp. n.	0.0	0.0	0.0	83.3	16.7	24
O. natator	10.3	10.3	3.4	3.4	72.4	29
all taxa (combined)	82.8%					
female ratios						
O. ziama sp. n.	83.3	3.3	0.0	10.0	3.3	30
O. smithi sp. n.	0.0	83.3	16.7	0.0	0.0	6
O. fouta sp. n.	0.0	25.0	50.0	0.0	25.0	4
O. arndti sp. n.	12.5	0.0	0.0	70.8	16.7	24
O. natator	10.3	6.9	3.4	10.3	69.0	29
all taxa (combined)	74.2%					

texture granular and heterogeneous; males with femoral glands, external vocal sacs, velvety nuptial excrescences on finger I. These characters apply to all species treated herein and are not repeated in the specific diagnoses below. For further osteological characters see Barej et al. (2014a, b).

Odontobatrachus natator (Boulenger, 1905)

OTU *natator* sensu Barej et al. (2015)

Syntypes. BMNH 1947.2.30.65-69 (syntypes: 1 male, 3 females, subadult), Sierra Leone, no more details available, coll. Major F. Smith.

Examined material. Sierra Leone: BMNH 1961.1248-54 (5 juveniles), Western Area; BMNH 1963.1047 (female), Southern Province; BMNH 1964.178 (female), Western Area; ZMB 78196 (juvenile), Western Area Peninsula Forest (Latitude: 8.35; Longitude: -13.18), 178 m a.s.l.; ZMB 78197 (female), Western Area Peninsula Forest

(8.47; -13.22), 367 m a.s.l.; ZMB 78198 (female), Northern Province (9.21; -11.14), 1325 m a.s.l.; ZMB 78199 (female), Eastern Province (8.86; -10.79), 748 m a.s.l.; ZMB 78200 (male), Northern Province (9.21; -11.14), 1345 m a.s.l.; ZMB 78202, ZFMK 95469 (2 females), ZMB 78203, MHNG 2731.51, ZFMK 95470 (3 males), Eastern Province (7.66; -10.90), 334 m a.s.l. Guinea: ZMB 78207 (juvenile), ZMB 78208 (female), N'Zérékoré Region (8.89; -8.31), 1019 m a.s.l.; ZMB 78209 (female), Kankan Region (9.28; -9.11), 637 m a.s.l.; ZMB 78210 (juvenile), ZMB 78211 (female), N'Zérékoré Region (7.54; -8.84), 403 m a.s.l.; ZMB 78212 (female), ZMB 78213 (male), N'Zérékoré Region (8.88; -8.29), 939 m a.s.l.; ZMB 78214 (male), ZMB 78215-6 (2 females), N'Zérékoré Region (7.64; -9.25), 533 m a.s.l.; ZMB 78217-19 (3 males) Mamou Region (10.30; -11.94), 527 m a.s.l.; ZMB 78303 (female), N'Zérékoré Region (8.35; -9.42), 487 m a.s.l. Liberia: BMNH 1982.631 (male), Iti Valley; ZMB 78220 (female), Grand Cape Mount County (7.45; -10.69), 299 m a.s.l.; ZMB 78221 (female), ZMB 78222 (male), Nimba County (7.54; -8.63), 595 m a.s.l.;

ZMB 78223-24, ZMB 78232, ZMB 78234, ZMB 78236-7, ZMB 78239 (7 females), ZMB 78227, ZMB 78229-31, ZMB 78233, ZMB 78235, ZMB 78238, ZMB 78240-42 (10 males), ZMB 78228 (juvenile), Nimba County (7.44; -8.66), 634 m a.s.l.; ZMB 78225 (female), Nimba County (7.44; -8.59); ZMB 78226 (female), Nimba County (7.46; -8.67), 591 m a.s.l.; ZMB 78244 (female), ZMB 78245 (male), Grand Gedeh County (5.66; -8.16), 316 m a.s.l.; ZMB 78246 (juvenile) Grand Gedeh County (5.69; -8.21), 247 m a.s.l.; ZMB 78247 (male), Grand Gedeh County (5.64; -8.19), 367 m a.s.l.; ZMB 78248 (juvenile), Grand Gedeh County (5.64; -8.19), 345 m a.s.l.; ZMB 78249 (female), ZMB 78250 (female, juvenile), Grand Gedeh County (5.63; -8.19), 388 m a.s.l.; ZMB 80504 (male), Nimba County (7.51; -8.70), 429 m a.s.l.; ZMB 80505 (female), Nimba County (6.44; -9.06), 533 m a.s.l.

Boulenger's (1905) species description is based on a series of five specimens in the BMNH collection (1947.2.30.65-69, formerly: 1905.1.27.4-5 and 1905.2.2.15-17). The type series collected by Major F. Smith Royal Army Medical Corps (R.A.M.C.) contains one male, three females and a subadult female. The type locality is given as "Sierra Leone".

During his service in western Africa, Captain (Local Major) F. Smith researched tropical diseases, prepared species lists of pests and elaborated respective preventive measures (Smith 1902, 1905). A part of his contribution contains the local fauna around barracks (Smith 1905) and Major F. Smith mentioned "a local frog (a new species named *Petropedetes natator*)…". He was based in Freetown predominantly surveying the area of Mt. Aureol, Tower Hill and Kortright but likewise carried out short travels to Port Lokkoh (today: Port Loko) and Rotifunk in the close hinterland (Smith 1902). However, Smith (1902) searched in the latter region for swampy areas as potential breeding habitats of the mosquito genus *Anopheles*, a habitat type inappropriate for torrent-frogs. Consequently, we herein restrict the type locality of *Petropedetes natator* Boulenger, 1905 to the Freetown area, Sierra Leone. A more detailed restriction appears unreasonable.

We refrain from designating a single lectotype as subsequent species descriptions are possible with comparison to the whole syntype series.

Genetics. *Odontobatrachus natator* is genetically well differentiated from all congeners and known populations form a well-supported and monophyletic clade (Barej et al. 2015). Uncorrected 16S p-distances between *O. natator* and other *Odontobatrachus* species range from 3.40–5.40% (Appendix 1: Table A), while maximum intrataxon differences of *O. natator* reach 1.98% (one-to-one pairwise comparisons N = 703), maximum intra-subclade difference values for the two subclades of *O. natator* are 0% (N = 1) and 0.72% (N = 630) respectively (Appendix 1: Table A). These two subclades correspond to the disjunct distribution of I) the Freetown area and II) all remaining localities further inland; divided by unsuitable

habitat in-between (Barej et al. 2015). In case taxonomic changes are made in the future, the Freetown clade should retain the nominate form following the restriction of the type locality.

Description of male syntype. The male syntype (BMNH 1947.2.30.68) has been assigned to this taxon in both DCA analyses (absolute values and ratios). The male syntype has a robust body shape: snout-urostyle length of 46.1 mm; head width 17.0 mm; head slightly longer than broad; snout in lateral view short, slightly rounded at the snout tip (Fig. 3); snout in dorsal view fairly rounded; lower jaw with sharp tusk-like prolongations and single small knob at lower jaw symphysis with corresponding socket in-between premaxillae; upper premaxillae and maxillae with numerous teeth, posteriorly curved; vomerine teeth present, arranged in two small odontophores, closer to each other than to choanae; tongue broadly heart shaped; horizontal eye diameter 6.4 mm; interorbital distance 5.3 mm; pupil horizontally elliptical; eye diameter distinctly larger than tympanum diameter (Fig. 3); tympanum indistinct (horizontal diameter 2.7 mm); nares closer to snout than to eye; snout as long as eye diameter; *canthus rostralis* rounded; loreal region concave; paired lateral vocal sacs (Fig. 3); forelimbs moderately slender, forearms slightly hypertrophied, fingers slender; prepollex absent; relative finger lengths III>IV≥II>I (Fig. 3); velvety nuptial excrescences on finger I weakly developed; subarticular tubercles large, subconical; supernumerary tubercles absent; fingertips dilated, triangular, notched in the middle; femur length 23.2 mm; tibia length 23.8 mm; femoral glands large (length × width: left: 10.3 × 5.4 mm, right: 9.5 × 5.5 mm); femoral glands positioned on the posterior part of the ventral side of femur (Fig. 3); relation femoral gland length to femur length: 0.43; minuscule circular glands running along upper side of tibia; foot length (incl. longest toe) 29.9 mm; relative toe lengths IV>III≥V>II>I; inner metatarsal tubercle elliptical; toe tips broadened forming triangular dilated discs; inner metatarsal tubercle prominent (2.8 mm); number of subconical subarticular tubercles on toes I-V: 1, 1, 2, 3, 2; supernumerary tubercles absent (Fig. 3); prominent skin fold on posterior side of feet; dorsal skin texture heterogeneous; dorsum and flanks covered with slender dorsal ridges of app. 3.0 mm length (partially flattened on the dorsum due to preservation); venter smooth; flank texture as on dorsum; webbing fully developed (0-0.5/0-1/0-1/1-0), running as a skin fold along toes III and IV to the disc, webbing between toes hardly concave, almost straight.

Colouration in preservation. Specimen overall brownish in colour (Fig. 3); dorsum darker than ventrum; throat darker than belly, ventrum lacking any marbling or patterns. Damage of the male syntype: third toe of left foot (in dorsal view) cut off (Fig. 3); left side (in dorsal view)
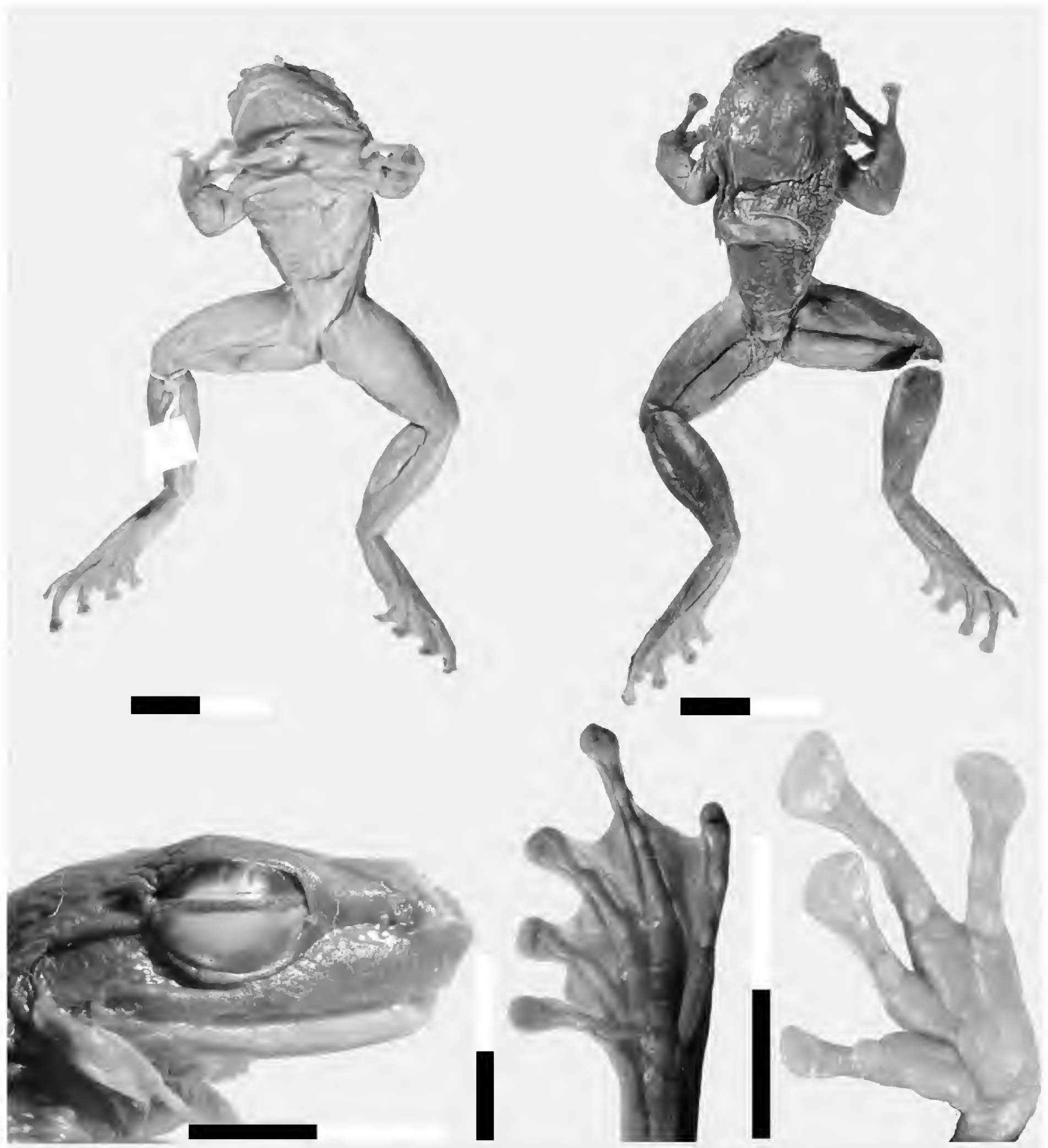
Figure 3. Male syntype of *Odontobatrachus natator* (BMNH 1947.2.30.68) in ventral and dorsal view (top from left to right, scale bar: 10 mm); head in lateral view, details of pedal webbing, details of hand (bottom left to right, scale bar: 5 mm).

with cut along flank; transverse cut on throat; discs on toes and fingers partially shrivelled due to drying-out.

Variation. Females are significantly larger than males (SUL: $Z = -3.814$, $p < 0.001$, $N_{males} = 22$, $N_{females} = 29$), mean SUL in females 53.6 mm and 48.0 mm in males, and consequently possess longer extremities (FM: $Z = -4.395$, $p < 0.001$; TI: $Z = -4.746$, $p < 0.001$; FL: $Z = -4.623$, $p < 0.001$), broader heads (HW: $Z = -3.570$, $p < 0.001$) and longer snouts (EN: $Z = -2.533$, $p < 0.01$; ES:

$Z = -3.285$, $p < 0.05$) in absolute measurements (Tables 1 and 2). However, ratios are predominantly similar between the two sexes, with males only showing higher values in HW/SUL ($Z = -2.796$, $p < 0.01$), IT/FL ($Z = -1.978$, $p < 0.05$) and TD/SUL ($Z = -2.701$, $p < 0.01$); for details see Tables 1 and 2. Both sexes possess enlarged tusk-like prolongations in the lower jaw as well as the name-bearing 'teeth' on the upper jaw. Male secondary sexual characters are femoral glands, velvety nuptial excrescences on finger I and presence of vocal sacs.
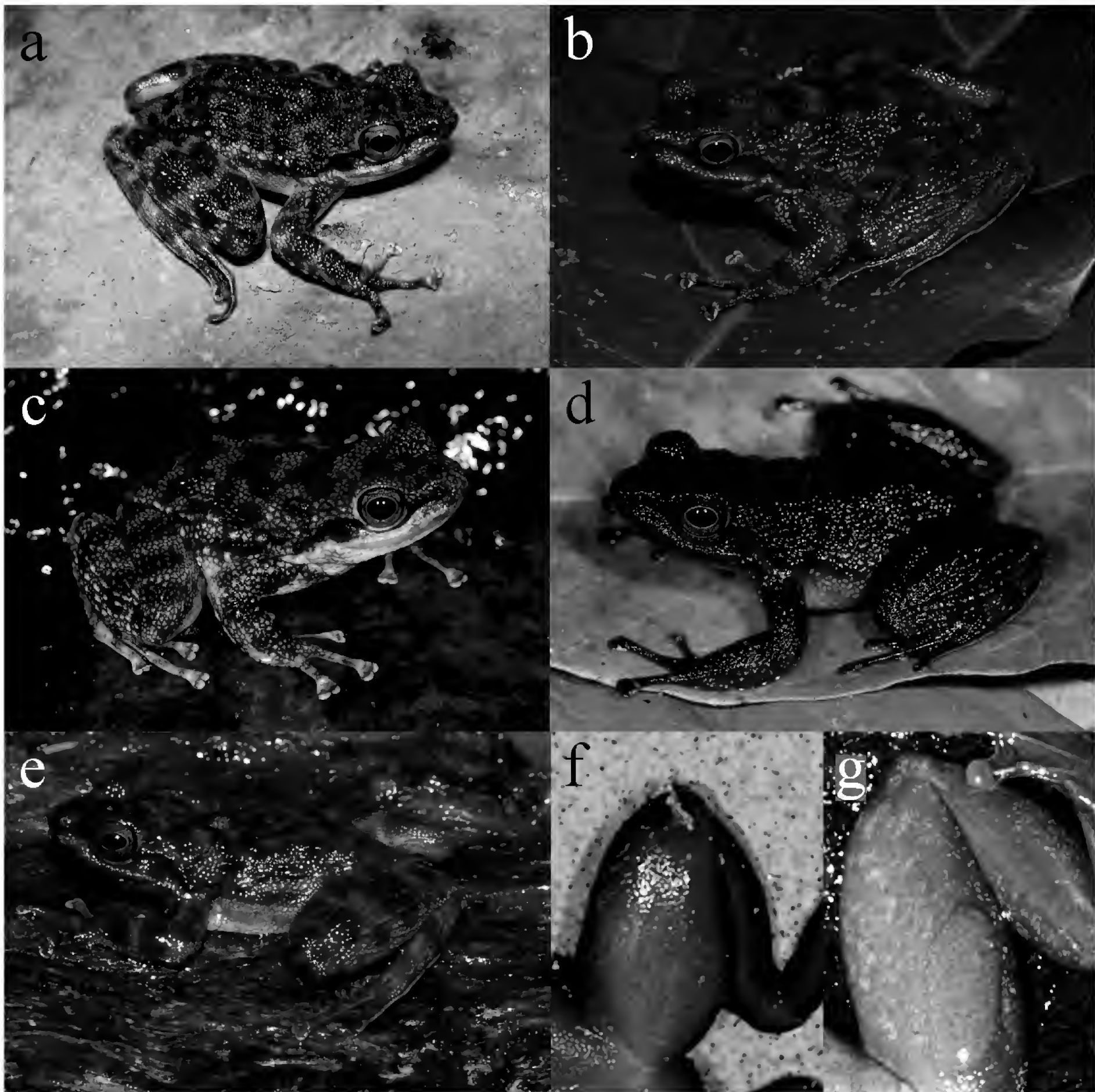
Figure 4. *Odontobatrachus natator* in life: **a)** female ZMB 78303, Ziama Forest, Guinea; **b)** male ZMB 78214, N'Zérékoré Region, Guinea; **c)** Gola Rainforest National Park, Sierra Leone; **d)** Freetown Area (type locality of *Petropedetes natator* Boulenger, 1905), Sierra Leone; **e)** ZMB 80504, Nimba County, Liberia; **f)** colouration of male femoral glands hardly visible (male shown in **d**); **g)** colouration distinctly contrasted against the femur (male shown in **e**).

Variation in webbing formulae of examined specimens in the covered distribution range corresponds to the extent in the type series (Table 7); although skin folds running along toes in the male syntype are more distinct than in many other specimens, showing an almost fully extended webbing state. Dorsal ridges form either slender lines as in the male syntype (Fig. 3, see also Fig. 4b, c) or are short and knob like (Fig. 4a). Number of distinct dorsal ridges (counted from spine to flank) range between two and six, usually three to five ridges per body site. However, both characters were not recognisable due to preservation artefacts in many specimens. Glandular ridges on tibia are usually built of small to large conic glands and form more or less interrupted lines (Fig. 4a–e). Dorsal colouration (in life) varies from uniform brownish, to mottled patterns with greenish or light brownish background and darker spots, usually arranged along dorsum. Male femoral glands are rose-coloured but colouration may be attenuated by the ventral colouration (Fig. 4e). Belly colouration (in alcohol) ranges from completely pale, dirty whitish, dark throat and pale belly, dark with few pale markings, to entirely dark colouration, showing no sex-dependant colour differentiation.

Distribution. *Odontobatrachus natator* has the widest distribution of all congeners (Fig. 1). The species is known

from Sierra Leone, Liberia and Guinea. While the species distribution overlaps with *O. ziama* and *O. arndti* in eastern Guinea, westernmost localities reach extensions of the Fouta Djallon area, close to the range of *O. fouta*. Two distinct molecular clades have been uncovered in *O. natator* (Barej et al. 2015), one of them being restricted to the Freetown Peninsula in coastal Sierra Leone (FP sensu Barej et al. 2015) and the other covering all remaining localities (IL sensu Barej et al. 2015) of this taxon.

Conservation status. The EOO, combining both subclades of *O. natator* (Barej et al. 2015; therein *natator*) sums up to 180,231 km², resulting in the IUCN Red List category "Least Concern (LC)". However, due to the habitat requirements of this family the AOO is restricted to 224 km² and thus classifies the species as "Endangered (EN)". When considering the genetic subdivision of *O. natator* (see Barej et al. 2015), the distribution areas further diminish dramatically, especially for the Freetown Peninsula subclade. While IUCN categories remain constant for the widely distributed subclade, the Freetown Peninsula subclade possesses an AOO of only 20 km² classifying it as EN and an EOO of 34 km² placing it as "Critically Endangered (CR)" if treated as its own taxonomic unit.

***Odontobatrachus ziama* Barej, Schmitz, Penner, Doumbia, Hirschfeld, Brede, Bangoura & Rödel, sp. n.**

http://zoobank.org/89BB73CC-EC8E-42E9-A075-990A52E711C5
OTU1 sensu Barej et al. (2015)

Holotype. ZMB 78300 (male), Republic of Guinea, Ziama Classified Forest (Latitude: 8.35790; Longitude: -9.29993), 668 m a.s.l., 22 November 2008, coll. C. Brede, M.A. Bangoura and J. Doumbia.

Paratypes. Guinea: ZMB 78298 (female), N'Zérékoré Region (8.36; -9.31), 878 m a.s.l., 11 July 2011; ZMB 78299 (female), same data as holotype; ZMB 78301, ZFMK 95464-65, MHNG 2731.46 (4 females), N'Zérékoré Region (8.36; -9.29), 558 m a.s.l., 30 July 2010; ZMB 78302, MHNG 2731.45 (2 males), N'Zérékoré Region (8.49; -9.31), 960 m a.s.l., 5 August 2010.

Additional material. Guinea: ZMB 78251 (male), ZMB 78252 (female), Kankan Region (9.21; -8.93), 1119 m a.s.l.; ZMB 78253-58 (5 females), N'Zérékoré Region (7.98; -9.12), 472 m a.s.l.; ZMB 78259 (female), Kankan Region (8.982; -8.96), 606 m a.s.l.; ZMB 78260, ZMB 78263, ZMB 78264 (juvenile), ZMB 78265-7 (5 females), ZMB 78261-2, ZMB 78268-9 (4 males), Kankan Region (9.26; -8.93), 754 m a.s.l.; ZMB 78271 (juvenile), N'Zérékoré Region (8.55; -9.08), 529 m a.s.l.; ZMB 78272 (male), Kankan Region (9.16; -8.93), 999 m a.s.l.; ZMB 78273 (male), ZMB 78274-5 (2 females), N'Zérékoré Region (8.89; -8.62), 646 m a.s.l.; ZMB 78276-7 (2 females), ZMB 78278 (juvenile), N'Zérékoré Region (8.55; -8.90), 1201 m a.s.l.; ZMB 78279-80 (2 females), N'Zérékoré Region (8.85; -8.89), 937 m a.s.l.; ZMB 78281 (female), ZMB 78282 (male), N'Zérékoré Region (8.82; -8.86), 726 m a.s.l.; ZMB 78283 (juvenile), N'Zérékoré Region (8.52; -8.94), 600 m a.s.l.; ZMB 78284 (male), ZMB 78285-6, ZMB 78288 (3 females), ZMB 78287 (juvenile), N'Zérékoré Region (8.53; -8.91), 1310 m a.s.l.; ZMB 78289-91 (3 males) ZMB 78292 (female), N'Zérékoré Region (8.14; -8.57), 622 m a.s.l.; ZMB 78295 (female), N'Zérékoré Region (8.28; -8.74), 908 m a.s.l.; ZMB 78296 (male), ZMB 78297 (female), N'Zérékoré Region (8.33; -8.71), 701 m a.s.l.

Diagnosis. Medium sized frogs, robust body shape; head narrow, smallest tympanum diameter/eye diameter ratio in the family, webbing fully developed, leaving up to 0.5 of the distal phalange free at the inner side of toe II, leaving up to 0.5-0.75 of the distal phalange free at toe IV; male femoral glands dark orange; glandular lines on tibia contain minuscule to small conic glands forming a pretty continuous line, belly pattern highly variable. Genetically *O. ziama* differs by a minimum of 2.89% in the mitochondrial 16S gene from its congeners.

Differential diagnosis. *Odontobatrachus ziama* can be distinguished from its congeners by a combination of characters (for all significant differences see Table 5): SUL in *O. ziama* is smaller than in *O. smithi* and *O. fouta* (Tables 1 and 2); male *O. ziama* differ from their congeners in the following ratios (Table 1): HW/SUL smaller than in *O. natator*, *O. smithi* and *O. fouta*; TD/O smaller than in *O. natator* and *O. smithi*; O/EN larger than in *O. natator* and *O. smithi*; TD/SUL smaller than in *O. natator*; GL/GW smaller than in *O. smithi* but larger than in *O. fouta*; female *O. ziama* differ from their congeners by the following ratios (Table 2): HW/SUL smaller than in *O. natator*, *O. smithi* and *O. fouta*; TD/O smaller than in *O. natator*, *O. smithi*, *O. fouta* and *O. arndti*; O/EN larger than in *O. natator*, *O. smithi* and *O. fouta*; TD/SUL smaller than in *O. natator*, *O. smithi* and *O. fouta*. Webbing of *O. ziama* is more more extensive than in *O. natator*, less extensive than in *O. smithi* and *O. fouta* and possesses a similar extent to *O. arndti* (Table 7). Femoral glands are dark orange in *O. ziama* but rose-coloured in *O. natator*, pale orange in *O. smithi* and bright orange in *O. fouta* (Figs 4, 6, 8, 10). Glandular lines on tibia contain minuscule to small conic glands forming almost continuous lines (Fig. 6a–d), while small to large glands form more or less interrupted lines in *O. natator* (Fig. 4a–e), small to mean conic glands form predominantly interrupted lines in *O. smithi* (Fig. 8a, b), small to large glandular conic glands, rather interrupted lines in *O. fouta* (Fig. 10b, c). and similar to *O. ziama* small to mean glandular conic glands form hardly interrupted lines in *O. arndti* (Fig. 12b, c). Morphologically the species is most similar in size and colour pattern to *O. arndti* (Table 7); however, they differ in several mensural characters: male *O. ziama* have larger SUL, but smaller HW, TD, O and extremities (FM, TI,
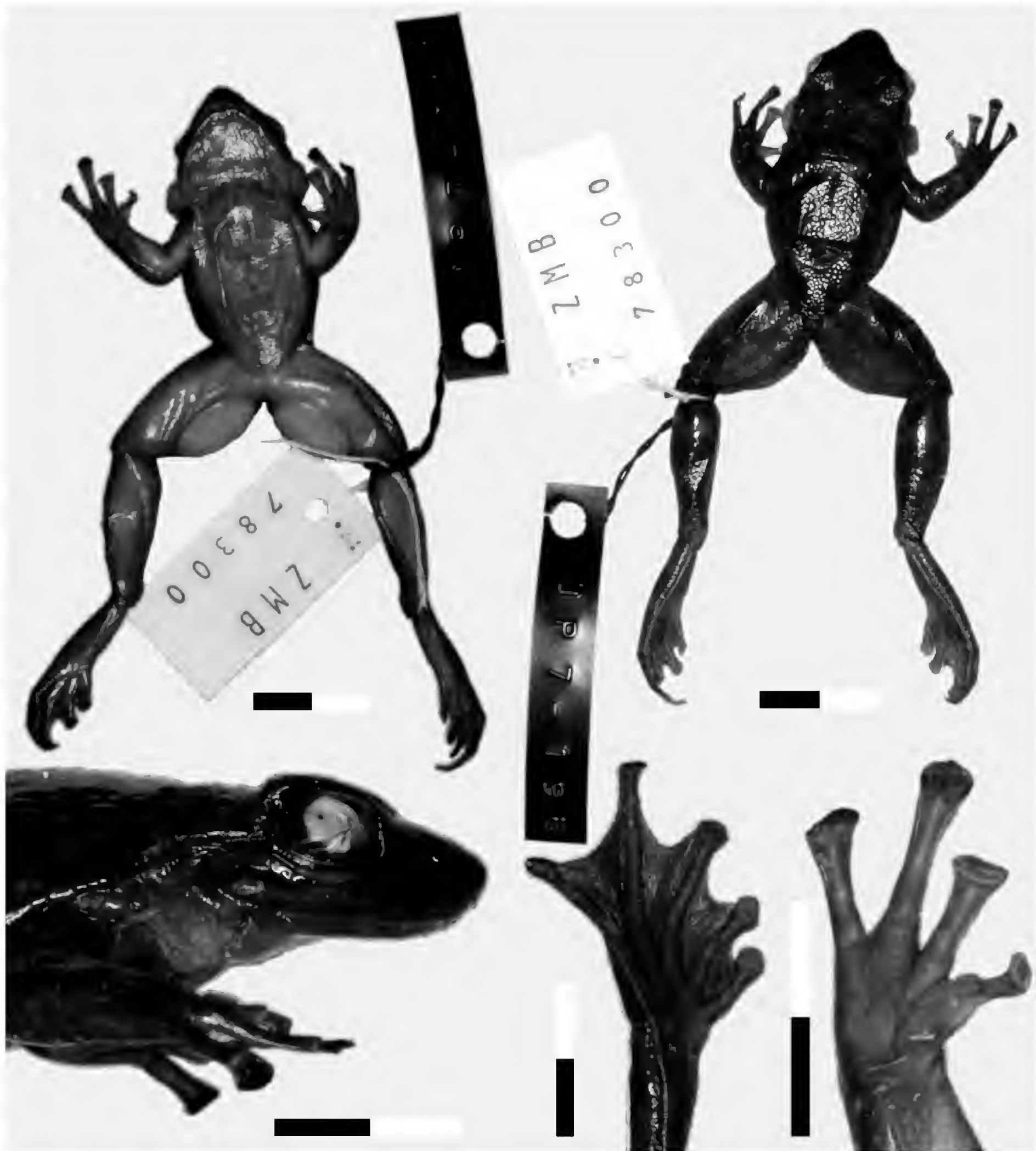
Figure 5. Male holotype of *Odontobatrachus ziama* sp. n. (ZMB 78300) in ventral and dorsal view (top from left to right, scale bar: 10 mm); head in lateral view, details of pedal webbing, details of hand (bottom left to right, scale bar: 5 mm).

FL); female *O. ziama* have smaller HW, O, ID and extremities (FM, TI, FL).

Genetics. The species is genetically well differentiated from all congeners and known populations form a well-supported and monophyletic clade (Barej et al. 2015). Uncorrected 16S p-distances between *O. ziama* and other *Odontobatrachus* species range from 2.89–5.41%, while maximum intrataxon differences of *O. ziama* add up to 0.38% (mean value 0.18%; N = 496; Appendix 1: Table A).

Holotype description. The male holotype has been correctly assigned to this taxon in both DCA analyses (absolute values and ratios). The holotype is an adult male with a moderately robust body shape (Fig. 5): snout-urostyle length of 46.1 mm; head width 15.5 mm; head slightly longer than broad; snout in lateral view short, flattened and slightly pointed at the snout tip; snout in dorsal view pointed; lower jaw with sharp tusk-like prolongations and single small knob at lower jaw symphysis with corresponding socket in between

premaxillae; upper premaxillae and maxillae with numerous teeth, posteriorly curved; vomerine teeth present, arranged in two small odontophores, closer to each other than to choanae; tongue broadly heart shaped; horizontal eye diameter 6.5 mm; interorbital distance 4.8 mm; pupil horizontally elliptical; eye diameter distinctly larger than tympanum diameter; tympanum indistinct (horizontal diameter 3.5 mm); nares closer to snout than to eye; snout as long as eye diameter; *canthus rostralis* rounded; loreal region concave; paired lateral vocal sacs; forelimbs moderately slender, forearms slightly hypertrophied, fingers slender; prepollex absent; relative finger lengths III>IV>II>I; velvety nuptial excrescences on finger I weakly developed; subarticular tubercles large, subconical; supernumerary tubercles absent; fingertips dilated, triangular, notched in the middle; femur length 24.1 mm; tibia length 24.4 mm; femoral glands large (length × width: left: 12.7 × 7.6 mm, right: 12.8 × 7.5 mm); femoral glands positioned on the posterior part of the ventral side of femur; relation femoral gland length to femur length: 0.53; minuscule circular glands running along upper side of tibia; foot length (incl. longest toe) 32.0 mm; relative toe lengths IV>III≥V>II>I; shortest toe 5.5 mm; inner metatarsal tubercle elliptical; toe tips broadened forming triangular dilated discs; inner metatarsal tubercle prominent (3.7 mm); number of subconical subarticular tubercles on toes I-V: 1, 1, 2, 3, 2; supernumerary tubercles absent; prominent skin fold on posterior side of feet; dorsal skin texture heterogeneous; dorsum and flanks covered with slender dorsal ridges of app. 2.6 mm length (partially flattened, but recognisable as darker spots); venter smooth; flank texture as on dorsum; webbing fully developed (0-0.25/0-0.75/0-1/1.25-0); webbing between toes hardly concave. Damage to the male holotype: left femur (in dorsal view) with short cut; third toe of left foot (in dorsal view) clipped for tissue sample; glandular dorsal ridges partially not recognisable due to preservation.

Colouration of holotype in alcohol (Fig. 5). Dorsum brownish, marbled with small dark spots (partially indicating presence of former dorsal gland ridges), a pale marking between shoulders; hind limbs on upper side with large dark blotches, surrounded with blurred pale lines; throat dark showing pale markings of scratches (scars); venter dark; femoral glands pale, clearly contrasted against femora, with few minuscule dark dots; femora and tibia dark as belly.

Variation. Females are significantly larger than males (SUL: Z = -4.164, p < 0.001, N_{males} = 11, $N_{females}$ = 30), mean SUL in females 52.1 mm and 45.1 mm in males, and consequently possess longer extremities (FM: Z = -3.649, p < 0.001; TI: Z = -4.665, p < 0.001; FL: Z = -3.694, p < 0.001), broader heads (HW: Z = -3.638, p < 0.001), longer snouts (EN: Z = -3.261, p < 0.01; ES: Z = -2.402, p < 0.05) and larger eyes (O: Z = -2.431,

p < 0.05) in absolute measurements (Tables 1 and 2). However, ratios are predominantly similar between the two sexes, although males show higher values in FL/SUL (Z = -2.119, p < 0.05), FM/SUL (Z = -1.883, p = 0.06), HW/SUL (Z = -3.119, p < 0.01) and TD/SUL (Z = -1.942, p = 0.52). Both sexes possess enlarged tusk-like prolongations in the lower jaw as well as the name-bearing 'teeth' on the upper jaw. Male secondary sexual characters are femoral glands, velvety nuptial excrescences on finger I and presence of vocal sacs. Male femoral glands are dark orange (Fig. 6f). Several specimens marked as males in the field lacked obvious secondary sexual characters (femoral glands) and flanks were opened to assess primary sexual characters. Probably due to preservation, femoral glands were not contrasted against the femora, and skin of vocal sacs shrivelled and retracted; however, one male showed no trace of skin modification on femora even if the typical gland position was cut open (ZMB 78262). Webbing formulae show very extensive webbing (Table 7). However, few specimens show a little reduced webbing on toe IV leaving almost the whole distal phalange free (1/1). Dorsal ridges are either long and slender (Fig. 6a–c) or are roundish and knob-like (Fig. 6d). Number of distinct dorsal ridges (counted from spine to flank) ranges between three and seven, usually four to five ridges per body site. However, both characters were not recognisable due to preservation artefacts in many specimens. Glandular ridges on tibia are usually built of small to large conic glands and form more or less interrupted lines (Fig. 6a–d). Dorsal colouration (in life) varies from uniform dark brown or olive, to dark brownish with pale irregular markings, to ochre with brownish spots and dorsal ridges are set off in terms of colour by usually being darker than the remaining dorsum (Fig. 6; Rödel and Bangoura 2004). Male femoral glands are dark orange (Fig. 6f). Belly colouration (in alcohol) is very variable, ranging from completely pale, to dirty smeared pale-dark, to pale reticulation on dark belly, to dark throat and pale belly, to dark throat and belly with pale longitudinal lines to dark with few pale markings, to completely dark, showing no sex-dependant differentiation.

Distribution. Distribution of *Odontobatrachus ziama* is restricted to isolated mountains north of the Nimba Mts. in south-eastern Guinea (Fig. 1). Its range apparently overlaps with *O. natator* as the latter is found in proximity to the Simandou Mountain Range, Massif du Ziama or Mt. Going. However, no syntopic populations are known so far. At present no differing habitat requirements or ecological adaptations are known (Barej et al. 2015), which could explain their spatial separation. Presence of *O. natator* in lower altitudes (e.g. Liberia, Grand Gedeh 250-500 m a.s.l.) could be a factor but both species co-occur in altitudes of app. 500–1300 m a.s.l. in the distribution range of *O. ziama*.
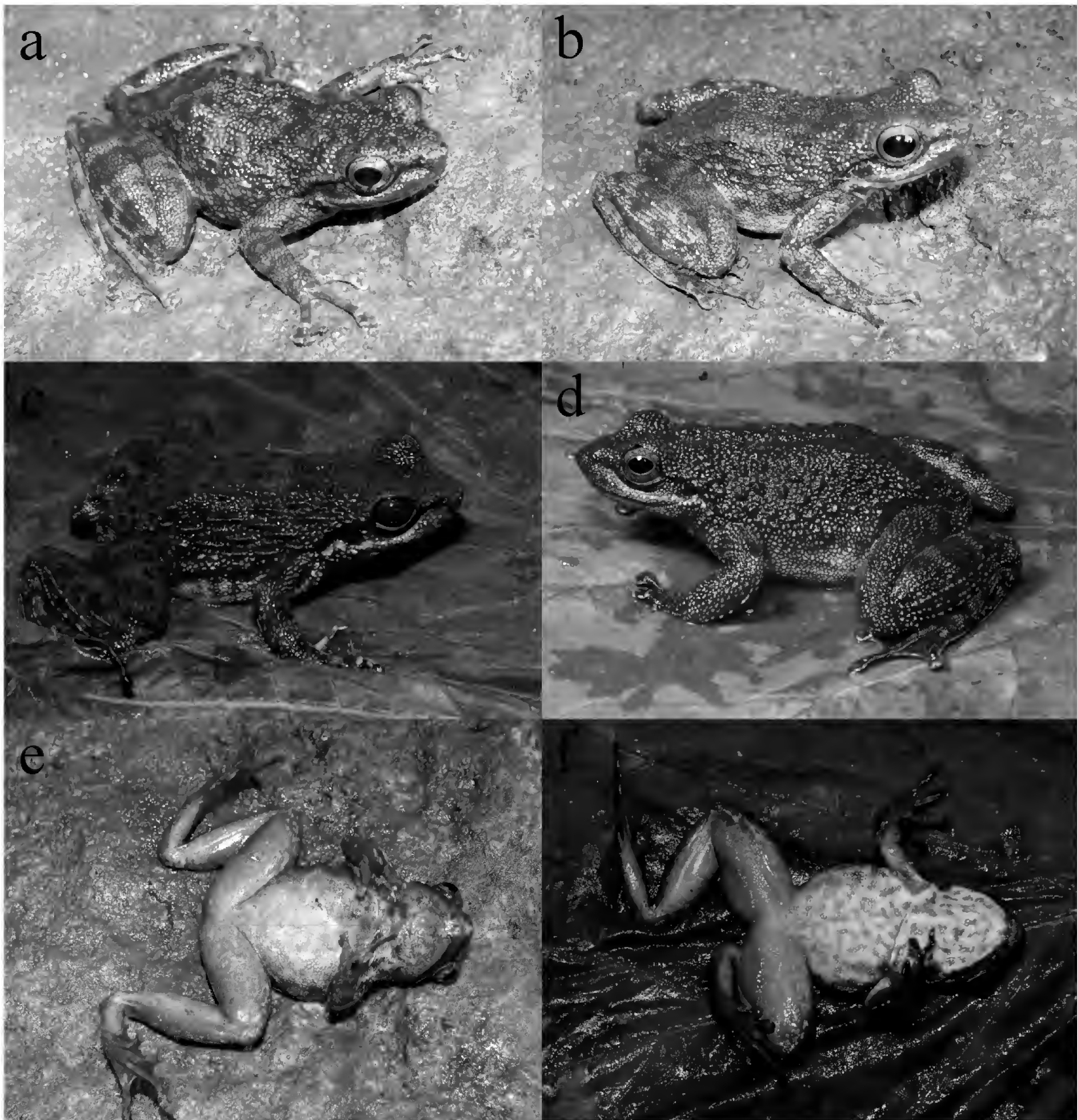
Figure 6. *Odontobatrachus ziama* sp. n. in life: **a**) female paratype ZFMK 95465 Ziama Forest, Guinea; **b**) female paratype MHNG 2731.46, from Ziama Forest, Guinea; **c**) female ZMB 78267, Kankan Region, Guinea; **d**) female ZMB 78263, Kankan Region, Guinea; **e**) ventral view of ZFMK 95465; **f**) colouration of femoral glands in male ZMB 78269. Mind the variation in shape of snout in lateral view from rounded (**b**) to pointed (**d**) and the variation in shape of dorsal ridges ranging from sub-elliptical (**a, b**), elongated (**c**) to conic (**d**).

Natural history remark. *Odontobatrachus ziama* is known as a host of the endoparasitic mite *Endotrombicula pillersi*, otherwise known from members of the family Phrynobatrachidae (Wohltmann et al. 2007).

Etymology. The species epithet *ziama* is a noun in apposition, therefore invariable, referring to the species' type locality, the Ziama Forest, in eastern Guinea.

Common name. We advise to use the term "Ziama torrent-frog" in English and "grenouilles des torrents de Ziama" in French.

Conservation status. The EOO of *O. ziama* is 7797 km^2, placing the species in the category "Vulnerable (VU)" while the AOO of 104 km^2 classifies the species as "Endangered (EN)" (Barej et al. 2015).

***Odontobatrachus smithi* Barej, Schmitz, Penner, Doumbia, Sandberger-Loua, Hirschfeld, Brede, Emmrich, Kouamé, Hillers, Gonwouo, Nopper, Adeba, Bangoura, Gage, Anderson & Rödel, sp. n.**

http://zoobank.org/94C996AB-8A52-4439-8F75-13938558A3EB
OTU2 sensu Barej et al. (2015)

Holotype. ZMB 78310 (male), Republic of Guinea, Fouta Djallon, Pita, Hörè Binti (Latitude: 10.83964; Longitude: -12.55572), 510 m a.s.l., 23 July 2010, coll. C. Brede and J. Doumbia.

Paratypes. Guinea: MHNG 2731.47 (female), Mamou Region (10.85; -12.52), 664 m a.s.l., 22 July 2010; ZFMK 95466, ZMB 78306 (2 females), Kindia Region (10.81; -13.34), 314 m a.s.l., 3 October 2010; ZMB 78311 (female), same data as holotype.

Additional material. Guinea: ZMB 78304-05 (2 juveniles), Kindia Region (10.83; -13.81), 253 m a.s.l.; ZMB 78307 (male), Kindia Region (10.81; -13.34), 314 m a.s.l.; ZMB 78308 (female), Kindia Region (10.96; -13.71), 312 m a.s.l.; ZMB 78309 (male), Kindia Region (10.00; -12.34), 92 m a.s.l.; ZMB 78312 (female), ZMB 78313 (juvenile), Mamou Region (10.85; -12.52), 664 m a.s.l.

Diagnosis. Medium to large sized frogs, robust body shape; head narrow, smallest tympanum-eye ratio in the family, highest eye diameter/eye-naris-distance ratio in the family, webbing fully developed, leaving up to 0.5 of the distal phalange free at the inner side of toe II, leaving 0.5-0.75 of the distal phalange free at toe IV; belly pattern very variable, male femoral glands pale orange; glandular lines on tibia contain mean conic glands forming frequently interrupted lines. Genetically *O. smithi* differs by a minimum of 3.79% in the mitochondrial 16S gene from its congeners.

Differential diagnosis. *O. smithi* can be distinguished from its congeners by a combination of characters (characters distinguishing *O. smithi* vs. *O. ziama* see above; for all significant differences see Table 5): growing larger than *O. natator* and *O. arndti* (Tables 1 and 2); male *O. smithi* differ from their congeners by the following ratios (Table 1): GL/GW larger than in *O. natator* and *O. fouta* but smaller than in *O. arndti*; TD/O larger than in *O. natator*; O/EN smaller than in *O. natator* and *O. arndti*; ES/O smaller than in *O. arndti*; female *O. ziama* differ from their congeners by the following ratios (Table 2): HW/SUL larger than in *O. natator* and *O. arndti*; larger TD/O than in *O. natator* and *O. arndti*; O/EN smaller than in *O. natator* and *O. arndti*; TD/SUL larger than in *O. arndti*. Webbing formulae of *O. smithi* are similar to *O. fouta* and *O. arndti*; however, *O. smithi* possesses less webbing on the inner side of toe II, and webbing in *O. natator* is more extensive (Table 7). Femoral glands are pale orange in *O. smithi* but rose-coloured in *O. natator*, dark orange

in *O. ziama* and bright orange in *O. fouta* (Figs 4, 6, 8, 10). Glandular lines on tibia contain small to mean sized conic glands forming predominantly interrupted lines (Fig. 8a, b), while small to large glands form more or less interrupted lines in *O. natator* (Fig. 4a–e), small to large glandular conic glands, rather interrupted lines in *O. fouta* (Fig. 10b, c) and small to mean glandular conic glands form hardly interrupted lines in *O. arndti* (Fig. 12b, c). Morphologically, the species is most similar in size and colour pattern to *O. fouta*. However, they differ in a few characters, namely colouration in male femoral glands and belly pattern with both taxa possessing a dark belly colouration, but only *O. fouta* specimens show a smeared pattern.

Genetics. The species is genetically well differentiated from all congeners and known populations form a well-supported and monophyletic clade (Barej et al. 2015). Uncorrected 16S p-distances between *O. smithi* and other *Odontobatrachus* species range from 3.79–5.55%, while maximum intrataxon differences of *O. smithi* add up to 0.54% (mean value 0.20%; N = 45; Appendix 1: Table A).

Holotype description. The male holotype has been assigned to this taxon in the DCA analysis of ratios. The holotype is an adult male with a robust body (Fig. 7): snout-urostyle length of 60.4 mm; head width 21.9 mm; head slightly longer than broad; snout in lateral view short, flattened and slightly pointed at the snout tip; snout in dorsal view triangular, pointed; lower jaw with sharp tusk-like prolongations protruding the skin and single triangular knob at lower jaw symphysis, corresponding socket in between premaxillae weakly developed; upper premaxillae and maxillae with numerous teeth, posteriorly curved; vomerine teeth present, single prolongations; odontophores arranged in short lines, closer to each other than to choanae; tongue broadly heart shaped; horizontal eye diameter 7.9 mm; interorbital distance 5.6 mm; pupil horizontally elliptical; eye diameter distinctly larger than tympanum diameter; tympanum distinct (horizontal diameter 4.0 mm); nares closer to snout than to eye; snout as long as eye diameter; *canthus rostralis* rounded; loreal region concave; paired lateral vocal sacs; forelimbs robust, forearms hypertrophied, fingers slender; prepollex absent; relative finger lengths III>IV>II>I; velvety nuptial excrescences covering finger I; subarticular tubercles large, subconical; supernumerary tubercles absent; fingertips dilated, triangular, notched in the middle; femur length 29.9 mm; tibia length 30.4 mm; femoral glands large (length × width: left: 15.7 × 7.0 mm, right: 15.4 × 7.3 mm); femoral glands positioned on the posterior part of the ventral side of femur; relation femoral gland length to femur length: 0.52; minuscule circular glands running along upper side of tibia; foot length (incl. longest toe) 40.0 mm; relative toe lengths IV>III≥V>II>I; shortest toe (7.1 mm); inner
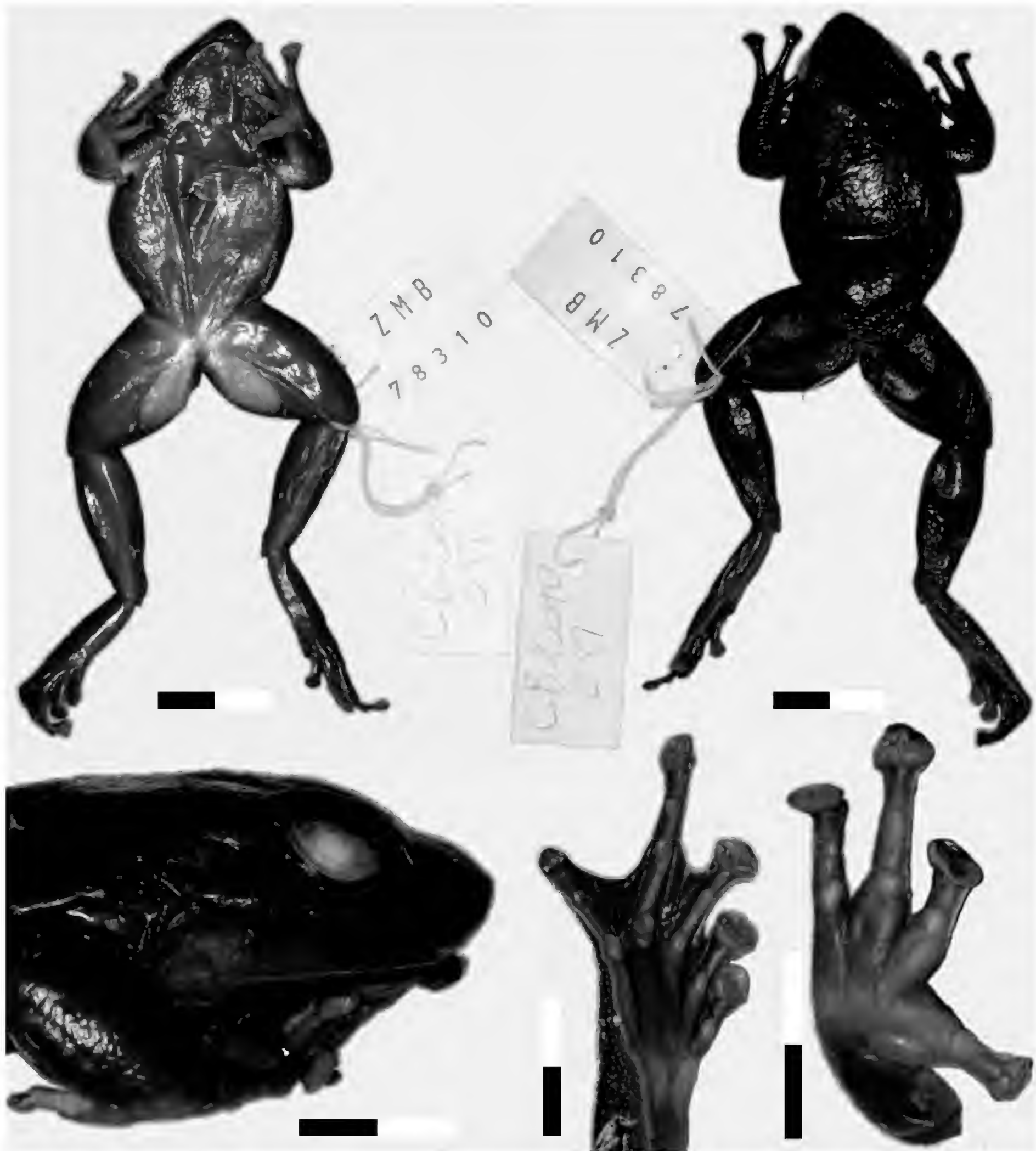
Figure 7. Male holotype of *Odontobatrachus smithi* sp. n. (ZMB 78310) in ventral and dorsal view (top from left to right, scale bar: 10 mm); head in lateral view, details of pedal webbing, details of hand (bottom left to right, scale bar: 5 mm).

metatarsal tubercle elliptical; toe tips broadened forming triangular dilated discs; inner metatarsal tubercle prominent (4.7 mm); number of subconical subarticular tubercles on toes I-V: 1, 1, 2, 3, 2; supernumerary tubercles absent; prominent skin fold on posterior side of feet; dorsal skin texture rough; dorsum and flanks covered with broad dorsal ridges of app. 3.0–4.0 mm (partially flattened, but recognisable as darker spots); venter somewhat rough and slightly granular; flank texture rough and granular as dorsum; webbing ful-

ly developed (0-1/0-1/0-1/1-0)), skin fringe running along toe III, webbing between toes hardly concave. Damage to the male holotype: transverse cut at pectoral region (liver tissue sampled) and skin cut on throat; glandular dorsal ridges partially not recognisable due to preservation.

Colouration of holotype in alcohol (Fig. 7). Dorsum dark brownish, few pale marblings recognisable; hind limbs with dark blotches on upper side; entire throat dark
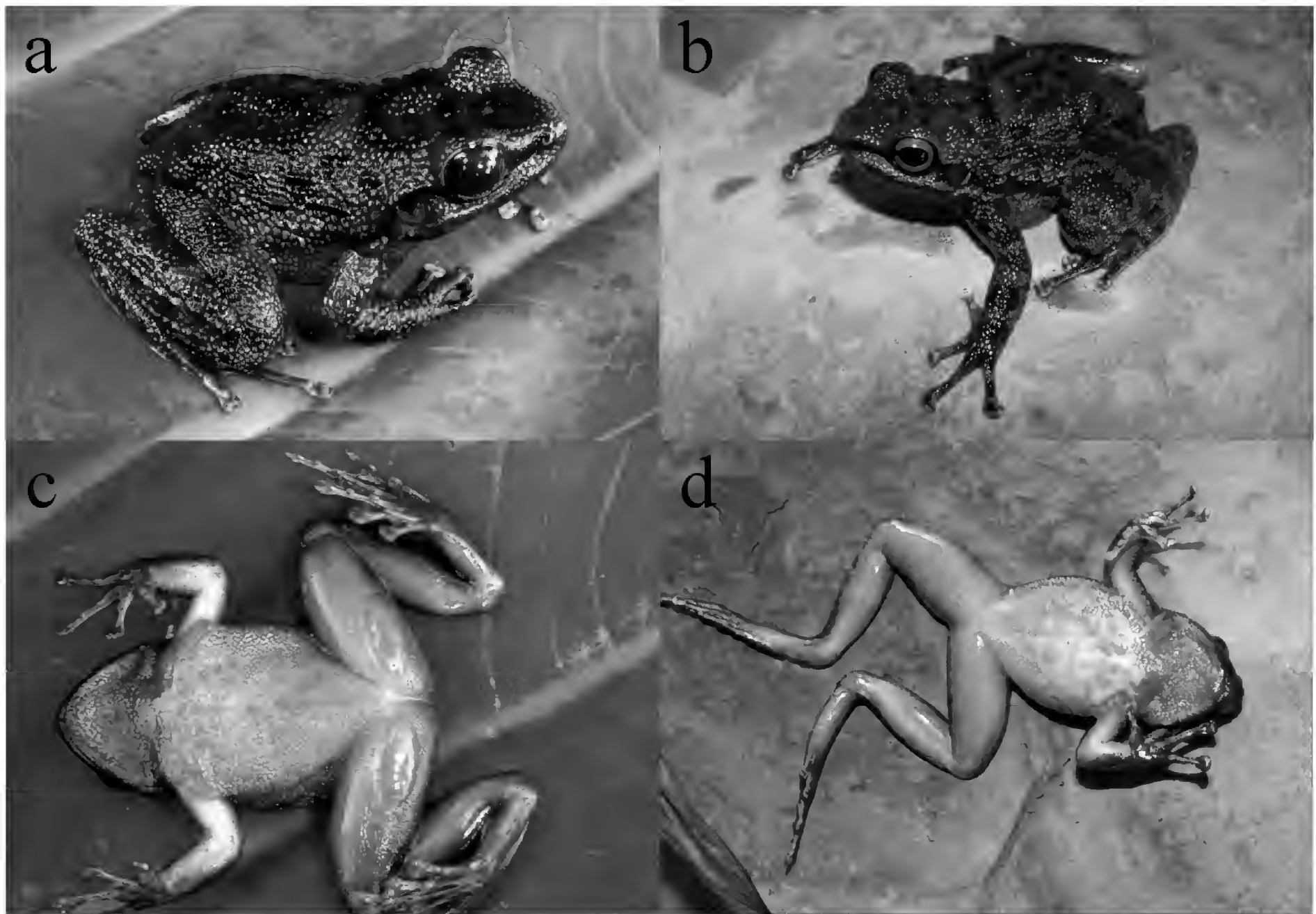
Figure 8. *Odontobatrachus smithi* sp. n. in life: **a**) male ZMB 78307, Kindia Region, Guinea; **b**) female paratype MHNG 2731.47, Fouta Djallon: Pita, Hörè Binti, Guinea); **c**) colouration of femoral glands in ZMB 78307; **d**) ventral view of female paratype MHNG 2731.47.

showing minuscule pale dots; venter dark; pale colouration between axillaries and elbows, femoral glands pale, clearly silhouetted from femora, with blurred minuscule dark dots; femora and tibia dark as belly.

Variation. Females ($N_{females}$ = 6) grow larger than males (N_{males} = 3), mean SUL in females 54.1 mm and 48.9 mm in males and accordingly absolute values for extremities are larger too. However, ratios between the two sexes overlap in their range, and are very similar in their mean values showing only minor differences (Tables 1 and 2). Both sexes possess enlarged tusk-like prolongations in the lower jaw as well as the name-bearing 'teeth' on the upper jaw. Male secondary sexual characters are femoral glands, velvety nuptial excrescences on finger I and presence of vocal sacs. Male femoral glands are pale orange (Fig. 8d). Webbing formulae showed little variance in this character (Table 7). Dorsal ridges are usually elongated and slender (Fig. 8a, b). Number of distinct dorsal ridges (counted from spine to flank) ranges between four and seven, usually five to six ridges per body site. However, this character was not recognisable in all vouchers due to preservation artefacts. Glandular ridges on tibia usually are built of small to large conic

glands and form more or less interrupted lines (Fig. 8a, b). Dorsal colouration (in life) ochre coloured with dark brown markings along dorsal glandular ridges or greenish with dark grey markings. Male femoral glands are pale orange (Fig. 8c). Belly colouration (in alcohol) is mainly uniform dark, only few specimens possess paler markings or show a dirty smeared colouration, showing no sex-dependant differentiation.

Distribution. Distribution of *Odontobatrachus smithi* is restricted to localities in western Guinea on the western and southern edge of the Fouta Djallon Highlands and its western extensions to the Kindia region (Fig. 1). Its easternmost localities are in proximity of *O. fouta*. However, *O. smithi* seems to occupy lowland to mid-altitudes (app. 100–650 m a.s.l.) while *O. fouta* occurs in mid-altitudes (app. 650–900 m a.s.l.).

Etymology. The species epithet *smithi* refers to Major F. Smith of the Royal Army Medical Corps (R.A.M.C.). In addition to his studies on blackwater fever he contributed to our knowledge on West African amphibians and collected the first specimens of *Petropedetes natator* Boulenger, 1905 in Sierra Leone during his military service in West Africa.

Common name. We advise to use the term "Smith's torrent-frog" in English and "grenouilles des torrents de Smith" in French.

Conservation status. The EOO of *Odontobatrachus smithi* is 12673 km², placing the species in the category "Vulnerable (VU)" while the AOO of 40 km² even classifies the species as "Endangered (EN)" (Barej et al. 2015).

Odontobatrachus fouta Barej, Schmitz, Penner, Doumbia, Brede, Hillers & Rödel, sp. n.

http://zoobank.org/D7A22E4A-430A-45E6-81DC-8E0792B442A2
OTU3 sensu Barej et al. (2015)

Holotype. ZMB 78314 (adult male), Republic of Guinea, Fouta Djallon, Labé, Sala (Latitude: 11.29389; Longitude: -12.50178), 916 m a.s.l., 18 July 2010, coll. C. Brede and J. Doumbia.

Paratypes. Guinea: ZMB 78314, MHNG 2731.48 (2 females), same data as holotype.

Additional material. Guinea: ZMB 78316 (female), same data as holotype; ZMB 78317-18 (2 males), Mamou Region (10.82; -12.19), 760 m a.s.l.; ZMB 78319 (juvenile), Labé Region (11.29; -12.51), 882 m a.s.l.; ZMB 78320, ZMB 78323 (2 females), ZMB 78322 (male), ZMB 78321, ZMB 78324-5 (3 juveniles), Mamou Region (10.34; -12.17), 652 m a.s.l.

Diagnosis. Medium to large sized frogs, robust body shape; head narrow, low mean eye diameter/eye-naris distance ratio, highest tympanum diameter orbita diameter ratio in the family, webbing fully developed, leaving 0.75 of the distal phalange free at the inner side of toe II, leaving the distal phalange at toe IV free; belly colouration typically dark, male femoral glands bright orange; glandular lines on tibia contain mean conic glands forming frequently interrupted lines. Genetically *O. fouta* differs by a minimum of 3.79% in the mitochondrial 16S gene from its congeners.

Differential diagnosis. *O. fouta* can be distinguished from its congeners by a combination of characters (characters distinguishing *O. smithi* vs. *O. ziama* and *O. fouta* see above; for all significant differences see Table 5): SUL in *O. fouta* is larger than in *O. natator* and *O. arndti* (Tables 1 and 2); male *O. fouta* differ from their congeners by the following ratios (Table 1): GL/GW smaller than in *O. natator* and *O. arndti*; TD/O and O/EN smaller than in *O. natator*; female *O. fouta* differ from their congeners by the following ratios (Table 2): TD/O larger than in *O. natator* and *O. arndti*; O/EN smaller than in *O. natator* and *O. arndti*; ES/O and TD/SUL larger than in *O. arndti*. Webbing in *O. fouta* is generally less extensive than in *O. natator* and shows less webbing on the inner side of toe II than in *O. arndti* (Table 7). Femoral glands are bright orange in *O. fouta* but

rose-coloured in *O. natator*, pale orange in *O. smithi* and dark orange in *O. ziama* (Figs 4, 6, 8, 10). Glandular lines on tibia contain small to large glandular conic glands, rather interrupted lines (Fig. 10b, c), while similar to *O. fouta* small to large glands form more or less interrupted lines in *O. natator* (Fig. 4a–e), and small to mean glandular conic glands form hardly interrupted lines in *O. arndti* (Fig. 12b, c).

Genetics. The species is genetically well differentiated from all congeners and known populations form a well-supported and monophyletic clade (Barej et al. 2015). Uncorrected 16S p-distances between *O. fouta* and other *Odontobatrachus* species range from 3.79–4.98%, while maximum intrataxon differences of *O. fouta* reach 0.36% (mean value 0.15%; N = 55; Appendix 1: Table A).

Holotype description. The male holotype has been assigned to this taxon in both DCA analyses (absolute values and ratios). The holotype is an adult male with a robust body (Fig. 9): snout-urostyle length of 55.6 mm; head width 21.6 mm; head slightly longer than broad; snout in lateral view short, flattened and slightly rounded; snout in dorsal view triangular, tip fairly rounded; lower jaw with sharp tusk-like prolongations protruding the skin and single triangular knob at lower jaw symphysis, corresponding socket in between premaxillae weakly developed; upper premaxillae and maxillae with numerous teeth, posteriorly curved; vomerine teeth present, single prolongations; odontophores arranged in short lines, closer to each other than to choanae, skin around vomerine teeth dark; tongue broadly heart shaped; horizontal eye diameter 7.7 mm; interorbital distance 5.9 mm; pupil horizontally elliptical; eye diameter distinctly larger than tympanum diameter; tympanum distinct (horizontal diameter 3.1 mm); nares closer to snout than to eye; snout as long as eye diameter; *canthus rostralis* rounded; loreal region concave; paired lateral vocal sacs; forelimbs robust, forearms hypertrophied, fingers slender; prepollex absent; relative finger lengths III>IV>II>I (Fig. 9); velvety nuptial excrescences covering finger I; subarticular tubercles large, subconical; supernumerary tubercles absent; fingertips dilated, slightly triangular; femur length 27.8 mm; tibia length 28.9 mm; femoral glands large (length × width: left: 14.2 × 8.0 mm, right 14.3 × 8.7 mm); femoral glands positioned on the posterior part of the ventral side of femur; relation femoral gland length to femur length: 0.51; minuscule circular glands running along upper side of tibia; foot length (incl. longest toe) 38.0 mm; relative toe lengths IV>III≥V>II>I (Fig. 9); shortest toe 7.2 mm; inner metatarsal tubercle elliptical; toe tips broadened forming triangular dilated discs; inner metatarsal tubercle prominent (4.5 mm); number of subconical subarticular tubercles on toes I-V: 1, 1, 2, 3, 2; supernumerary tubercles absent; prominent skin fold on posterior side of feet; dorsal skin texture rough; dorsum and flanks covered with slender dorsal ridges of app. 2.0–5.0 mm, mainly positioned dorsolaterally (partially
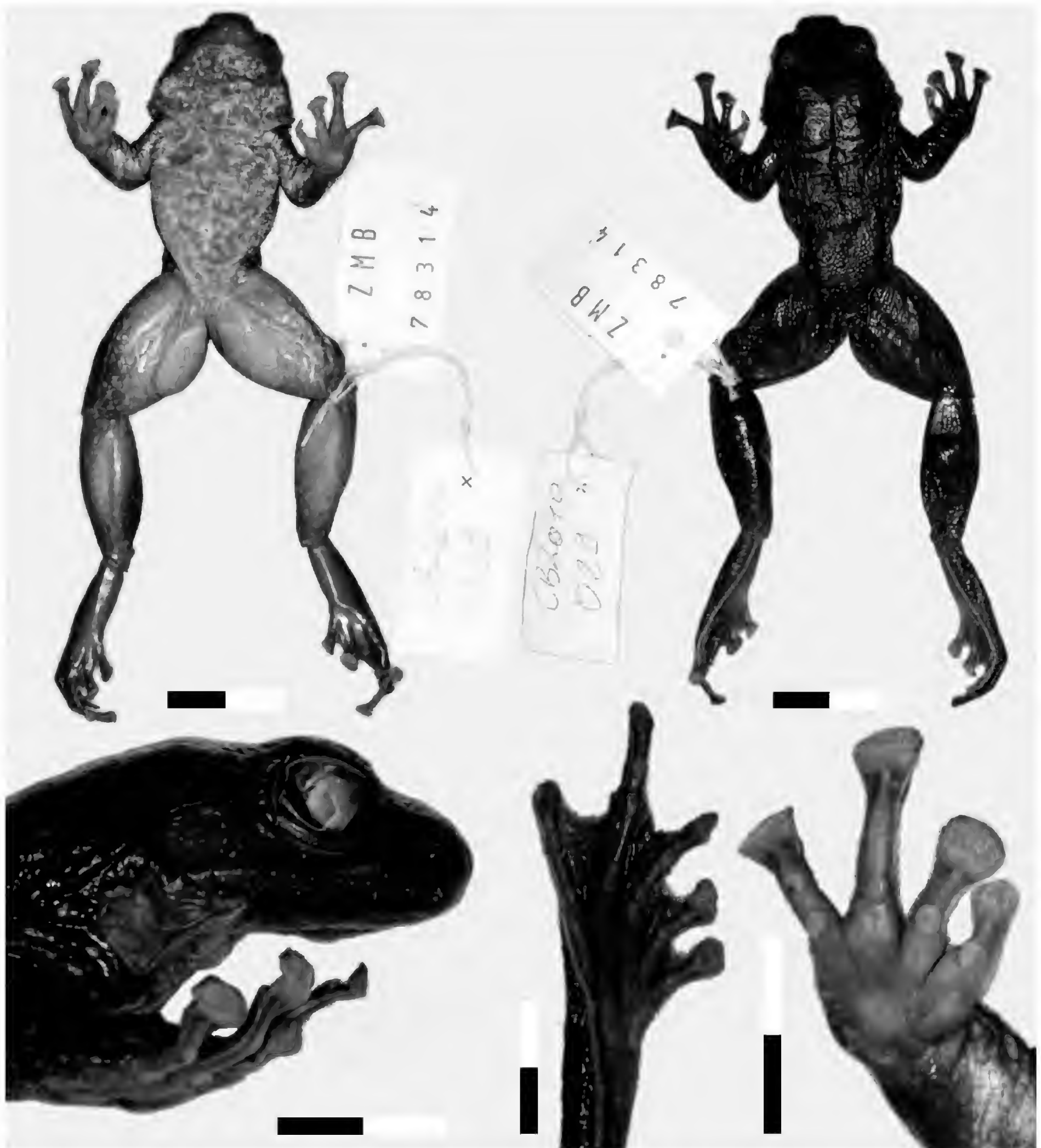
Figure 9. Male holotype of *Odontobatrachus fouta* sp. n. (ZMB 78314) in ventral and dorsal view (top from left to right, scale bar: 10 mm); head in lateral view, details of pedal webbing, details of hand (bottom left to right, scale bar: 5 mm).

flattened); venter somewhat rough and slightly granular; flank texture rough and granular as dorsum; webbing fully developed (0-0.75/0-1/0-1/1-0), skin fringe running along toe III, webbing between toes hardly concave. Damage of the male holotype: transverse cut at pectoral region (liver tissue sampled); glandular dorsal ridges partially not recognisable due to preservation.

Colouration of holotype in alcohol (Fig. 9). Dorsum dark brownish; hind limbs with dark blotches on upper

side, few pale lines recognisable; entire dirty blurred dark and pale, with several scratches (scars); venter as throat on the anterior part, more reticulated pattern on the belly; colouration between axillaries and elbows brighter; femoral glands pale, clearly silhouetted from femora, with blurred minuscule dark dots, posterior part darker; femora and tibia dark as belly.

Variation. Females ($N_{females}$ = 4) grow larger than males (N_{males} = 3), maximum SUL in females 62.5 mm and
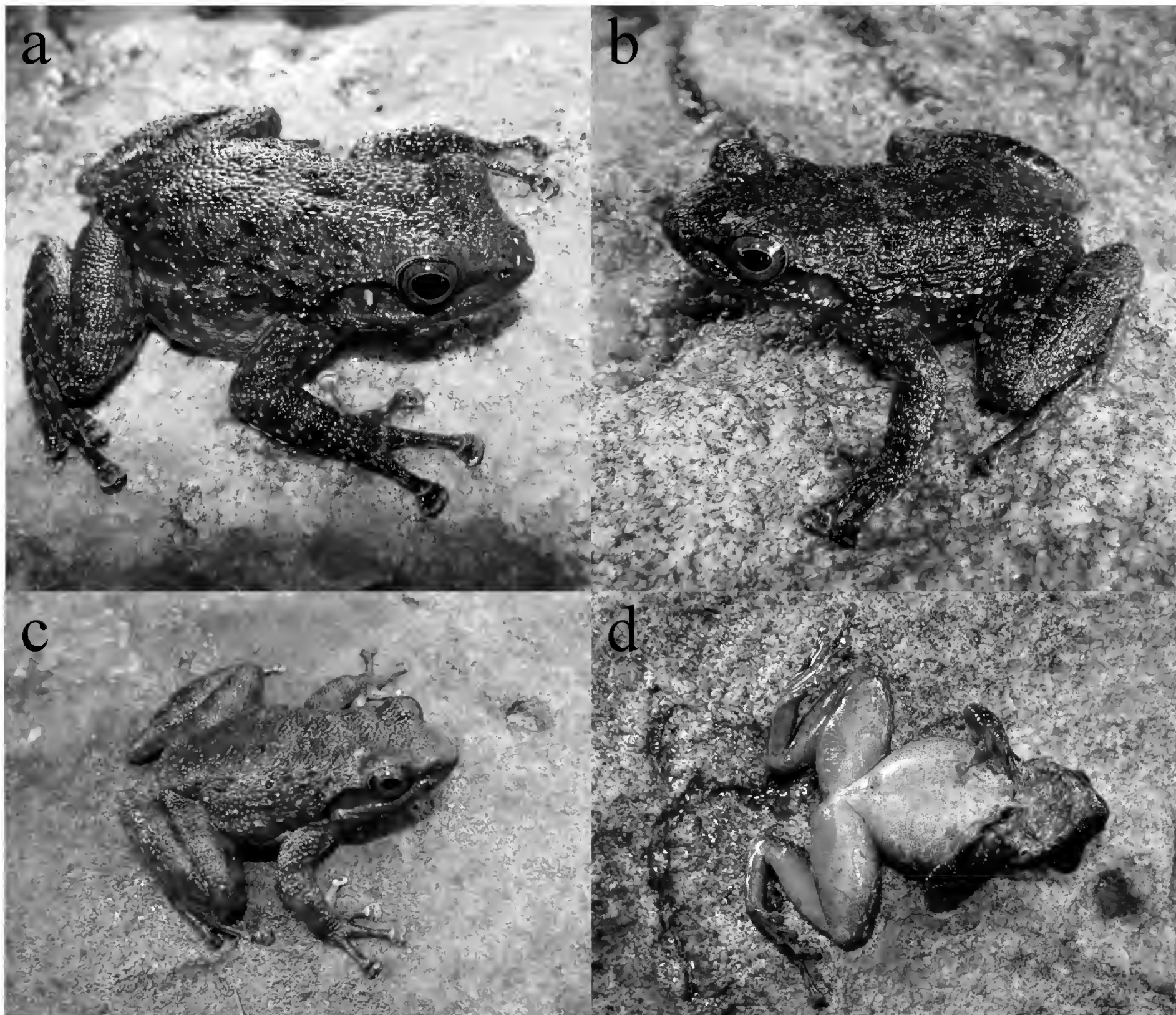
Figure 10. *Odontobatrachus fouta* sp. n. in life: **a**) female paratype ZMB 78315, Fouta Djallon: Labé, Sala, Guinea; **b**) male holotype ZMB 78314, Fouta Djallon: Labé, Sala, Guinea; **c**) male Dalaba\Chute de Ditinn, Guinea; **d**) colouration of femoral glands in the male holotype ZMB 78314.

57.0 mm in males, and absolute values for extremities are accordingly larger, too (Tables 1 and 2). However, males and females have similar ratios and mean values. Both sexes possess enlarged tusk-like prolongations in the lower jaw as well as the name-bearing 'teeth' on the upper jaw. Male secondary sexual characters are femoral glands, velvety nuptial excrescences on finger I and presence of vocal sacs. Webbing formulae showed little variance (Table 7). Dorsal ridges are short and knobbed (Fig. 10a) or elongated and slender (Fig. 10b). Number of distinct dorsal ridges (counted from spine to flank) ranges between three and six ridges per body site, usually four to five ridges per body site. However, this character was not recognisable due to preservation artefacts in all specimens. Glandular ridges on tibia usually are built of small to large conic glands and form rather interrupted lines (Fig. 10b, c). Dorsal colouration (in life) ochre coloured with dark brown markings along dorsal glandular ridges or almost uniform dark with few whitish markings along flanks and on dorsum. Male femoral glands are bright orange (Fig. 10d). Belly colouration (in alcohol) is mainly uniform dark, only few specimens possess paler markings or show a dirty smeared colouration, showing no sex-dependant differentiation.

Distribution. The distribution of *Odontobatrachus fouta* is restricted to isolated peaks in the central Fouta Djallon Highlands in western Guinea (Fig. 1). Localities of *O. natator* at the southern edge and of *O. smithi* close to western-central of the Fouta Djallon Highlands are in close proximity to *O. fouta*. However, *O. fouta* occurs in higher altitudes (southern edge: *O. natator* app. 500 m a.s.l. and *O. smithi* app. 92 m a.s.l. vs. *O. fouta* app. 650 m a.s.l.; western-central: *O. smithi* app. 510–650 m a.s.l. vs. *O. fouta* app. 750–900 m a.s.l.).

Etymology. The species epithet *fouta* is a noun in apposition, therefore invariable, referring to the species' type locality, the Fouta Djallon Highlands, in western Guinea.

Common name. We advise to use the term "Fouta Djallon torrent-frog" in English and "grenouilles des torrents de Fouta Djallon" in French.

Conservation status. Both, the EOO of 1318 km² and the AOO of 20 km² classify *O. fouta* as "Endangered (EN)" (Barej et al. 2015).

Odontobatrachus arndti **Barej, Schmitz, Penner, Doumbia, Sandberger-Loua, Emmrich, Adeba & Rödel, sp. n.**

http://zoobank.org/542C46CE-2B91-41AC-8314-5F84469AED04
OTU4 sensu Barej et al. (2015)

Holotype. ZMB 78355 (male), Republic of Guinea, Nimba Mts., River Mandey (Latitude: 7.64786; Longitude: -8.42397), 694 m a.s.l., 18 June 2009, coll. L. Sandberger-Loua and J. Doumbia.

Paratypes. Guinea: MHNG 2731.49 (male), ZMB 78356 (female), N'Zérékoré Region (7.65; -8.42), 670 m a.s.l., 18 June 2009; MHNG 2731.50, ZMB 78357 (2 females), N'Zérékoré Region (7.63; -8.41), 1121 m a.s.l., 4 November 2011; ZFMK 95467 (female), ZFMK 95468 (male), N'Zérékoré Region (7.65; -8.42), 674 m a.s.l., 2 January 2011; ZMB 78354 (female), same data as holotype.

Additional material. Côte d'Ivoire: ZMB 78326, ZMB 78329 (3 females), ZMB 78327-8 (2 males), Dix-Huit Montagnes Region (7.85; -7.39), app. 500 m a.s.l. Liberia: ZMB 78332 (male), Nimba County (7.56; -8.64), 647 m a.s.l.; ZMB 78333-35 (3 males), Nimba County (7.48; -8.58), 513 m a.s.l. Guinea: ZMB 78336 (female), ZMB 78337-39 (3 males), N'Zérékoré Region (7.61; -8.27), 400 m a.s.l.; ZMB 78340-41 (2 females), N'Zérékoré Region (7.61; -8.26), 460 m a.s.l.; ZMB 78342 (juvenile), N'Zérékoré Region (7.70; -8.40), 751 m a.s.l.; ZMB 78343, ZMB 78345 (2 females), ZMB 78344, ZMB 78346 (2 males), N'Zérékoré Region (7.70; -8.40), 760 m a.s.l.; ZMB 78347 (male), N'Zérékoré Region (7.71; -8.41), 518 m a.s.l.; ZMB 78348 (male), ZMB 78349 (female), N'Zérékoré Region (7.70; -8.40), 764 m a.s.l.; ZMB 78350-1 (2 females), ZMB 78352 (male), N'Zérékoré Region (7.68; -8.39), 1027 m a.s.l.; ZMB 78353 (juvenile), N'Zérékoré Region (7.65; -8.42), 670 m a.s.l.; ZMB 78358-59 (2 males), N'Zérékoré Region (7.65; -8.34), 577 m a.s.l.; ZMB 78360 (female), ZMB 78361 (male), N'Zérékoré Region (7.65; -8.36), 815 m a.s.l.; ZMB 78362 (female), ZMB 78363 (male), N'Zérékoré Region (7.63; -8.35), 652 m a.s.l.; ZMB 78364, ZMB 78367 (2 females), ZMB 78365-6 (2 males), N'Zérékoré Region (7.65; -8.37), 949 m a.s.l.; ZMB 78368 (female), ZMB 78369 (male), N'Zérékoré Region (7.67; -8.37), 1317

m a.s.l.; ZMB 78370 (male), ZMB 78371 (female), N'Zérékoré Region (7.67; -8.37), 1234 m a.s.l.; ZMB 78372 (female), ZMB 78373 (male), N'Zérékoré Region (7.62; -8.42), 1154 m a.s.l.; ZMB 78374 (female), ZMB 78375 (male), N'Zérékoré Region (7.62; -8.45), 701 m a.s.l.; ZMB 78376 (female), ZMB 78377 (male), N'Zérékoré Region (7.63; -8.44), 750 m a.s.l.; ZMB 78378 (female), ZMB 78379 (male), N'Zérékoré Region (7.67; -8.35), 786 m a.s.l.; ZMB 78380 (female), ZMB 78381 (male), N'Zérékoré Region (7.67; -8.40), 998 m a.s.l.

Diagnosis. Medium to large sized frogs, robust body shape; head narrow, highest eye diameter/eye-naris-distance ratio in the family, low mean tympanum diameter orbita diameter ratio, webbing almost fully developed, leaving 0.25–0.5 of the distal phalange free at the inner side of toe II, leaving 0.75–1 of the distal phalange free at toe IV, belly pattern very variable, glandular lines on tibia contain mean conic glands forming frequently interrupted lines. Genetically *O. arndti* differs by a minimum of 2.89% in the mitochondrial 16S gene from its congeners.

Differential diagnosis. *O. arndti* can be distinguished from its congeners by a combination of characters (characters distinguishing *O. arndti* vs. *O. ziama*, *O. smithi* and *O. fouta* see above; for all significant differences see Table 5): male *O. arndti* differ from *O. natator* by the following ratios (Table 1): larger TI/SUL, FL/SUL, O/EN and smaller HW/SUL, TD/O, IT/FL, TD/SUL in *O. arndti* than in *O. natator*; female *O. arndti* differ from *O. natator* by the following ratios (Table 2): smaller FM/TI, HW/SUL and O/EN larger in *O. arndti* than in *O. natator*. Webbing formulae are very similar in the two species (Table 7). Glandular lines on tibia contain small to mean conic glands forming hardly interrupted lines in *O. arndti* (Fig. 12b, c), while similar to *O. fouta* small to large glands form more or less interrupted lines in *O. natator* (Fig. 4a–e).

Genetics. The species is genetically well differentiated from all congeners and known populations form a well-supported and monophyletic clade (Barej et al. 2015). Uncorrected 16S p-distances between *O. arndti* and other *Odontobatrachus* species range from 2.89–5.55%, while maximum intrataxon differences of *O. arndti* add up to 0.58% (mean value 0.05%; N = 861; Appendix 1: Table A).

Holotype description. The male holotype has been assigned to this taxon in both DCA analyses (absolute values and ratios). The holotype is an adult male with a slightly robust body (Fig. 11): snout-urostyle length of 48.8 mm; head width 17.1 mm; head slightly longer than broad; snout in lateral view short, flattened and rounded at the snout tip; snout in dorsal view triangular, rounded; lower jaw with sharp tusk-like prolonga-
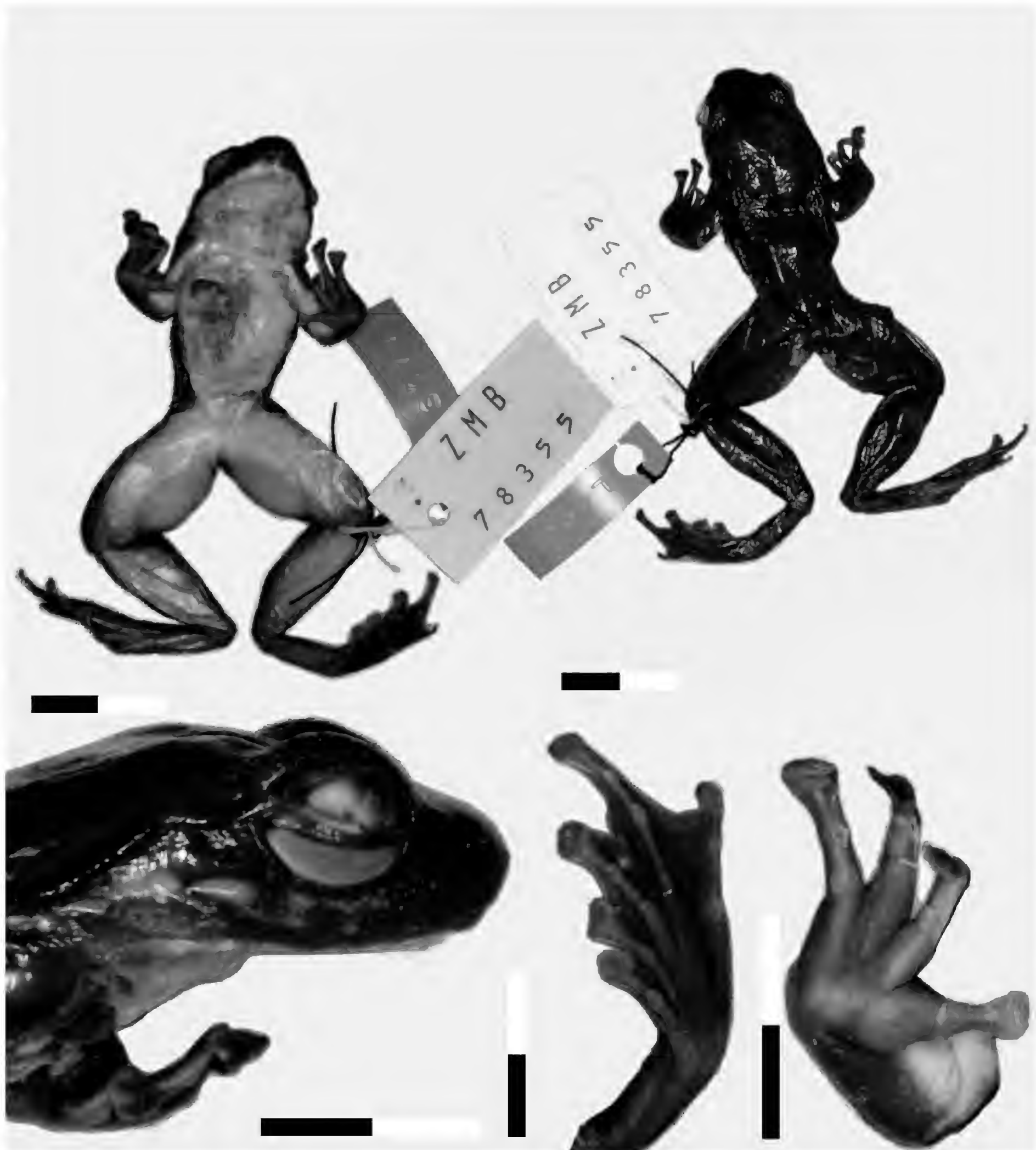
Figure 11. Male holotype of *Odontobatrachus arndti* sp. n. (ZMB 78355) in ventral and dorsal view (top from left to right, scale bar: 10 mm); head in lateral view, details of pedal webbing, details of hand (bottom left to right, scale bar: 5 mm).

tions protruding the skin and single triangular knob at lower jaw symphysis, corresponding socket in between premaxillae; upper premaxillae and maxillae with numerous teeth, posteriorly curved; vomerine teeth present, arranged in two small odontophores, closer to each other than to choanae; tongue broadly heart shaped; horizontal eye diameter 7.6 mm; interorbital distance 5.2 mm; pupil horizontally elliptical; eye diameter distinctly larger than tympanum diameter; tympanum distinct (horizontal diameter 2.7 mm); nares closer to snout than to eye; snout shorter than eye diameter; *canthus rostralis* rounded; loreal region concave; paired lateral vocal sacs; forelimbs robust, forearms hypertrophied, fingers slender; prepollex absent; relative finger lengths III>IV>II>I; velvety nuptial excrescences weakly developed on finger I; subarticular tubercles large, subconical; supernumerary tubercles absent; fingertips dilated, triangular, notched in the middle; femur length 24.6 mm; tibia length 26.3 mm; femoral glands large (length × width: left: 12.9 × 8.0 mm, right: 13.1 × 7.8
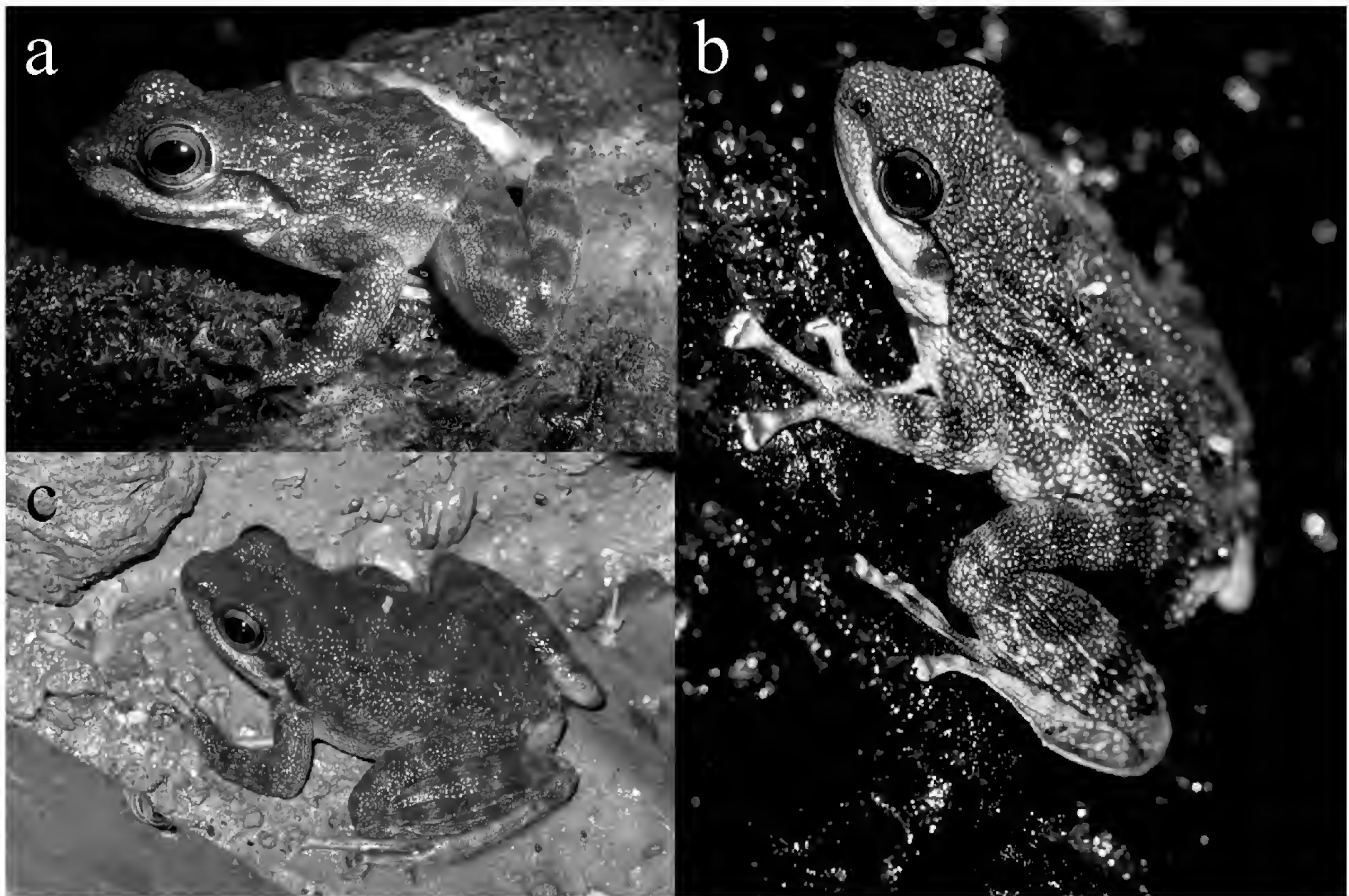
Figure 12. *Odontobatrachus arndti* sp. n. in life: **a**) and **c**) Nimba Mts, Guinea; **b**) Mt. Sangbé, Côte d'Ivoire. Non vouchered specimens. Note parasitic mites (minuscule red dots) close to the cloaca in (**c**).

mm); femoral glands positioned on the posterior part of the ventral side of femur; relation femoral gland length to femur length: 0.53; minuscule circular glands running along upper side of tibia; foot length (incl. longest toe) 34.9 mm; relative toe lengths IV>III>V>II>I; shortest toe 6.4 mm; inner metatarsal tubercle elliptical; toe tips broadened forming triangular dilated discs; inner metatarsal tubercle prominent (3.8 mm); number of sub-conical subarticular tubercles on toes I-V: 1, 1, 2, 3, 2; supernumerary tubercles absent; prominent skin fold on posterior side of feet; dorsal skin texture heterogeneous; dorsum and flanks covered with slender dorsal ridges of app. 2.5-3.5 mm (partially flattened); venter with fine granulation; flank texture rough and granular as dorsum; webbing fully developed (0-0/0-1/0-0.75/0.75-0); webbing between toes hardly concave. Damage of the male holotype: cut at pectoral region (liver tissue sampled) and skin cut on right lumbar region (in ventral view); glandular dorsal ridges partially not recognisable due to preservation.

Colouration of holotype in alcohol (Fig. 11). Dorsum dark brownish, few paler marblings; hind limbs coloured as dorsum; throat pale with few darker marblings; venter pale, area around incision darker; pale colouration between axillaries and elbows, femoral glands pale, clearly silhouetted from femora, with blurred minuscule reticulation; femora and tibia pale as belly.

Variation. Females are significantly larger than males (SUL: $Z = -4.933$, $p < 0.001$, $N_{males} = 26$, $N_{females} = 24$), max SUL in females 64.0 mm and 53.6 mm in males, and consequently possess longer extremities (FM: $Z = -3.894$, $p < 0.001$; TI: $Z = -4.458$, $p < 0.001$; FL: $Z = -4.264$, $p < 0.001$), broader heads (HW: $Z = -4.090$, $p < 0.001$), longer snouts (EN: $Z = -2.678$, $p < 0.01$; ES: $Z = -2.906$, $p < 0.01$) and larger eyes ($Z = -2.779$, $p < 0.01$), larger TD ($Z = -2.214$, $p < 0.05$). However, ratios are predominantly similar between the two sexes, although males show higher values in FL/SUL ($Z = -2.214$, $p < 0.05$), FM/SUL ($Z = -2.932$, $p < 0.01$), FM/TI ($Z = -3.010$, $p < 0.01$) and HW/SUL ($Z = -4.136$, $p < 0.001$). Both sexes possess enlarged tusk-like prolongations in the lower jaw as well as the name-bearing 'teeth' on the upper jaw. Male secondary sexual characters are femoral glands, velvety nuptial excrescences on finger I and presence of vocal sacs. Webbing formulae showed little variance (Table 7). However, some specimen possess a more extensive webbing on toe IV (0.5/0.5). Dorsal ridges are elongated and slender (Fig. 12a–c). Number of distinct dorsal ridges (counted from spine to flank) ranges between three and six, usually four to five ridges per body site. Glandular ridges on tibia usually are built of small to mean conic glands and form hardly interrupted lines (Fig. 12b, c). However, both characters were not recognisable due to preservation artefacts in many specimens. Dorsal colouration (in life) varies from almost black, beige with reddish-brown spots ar-
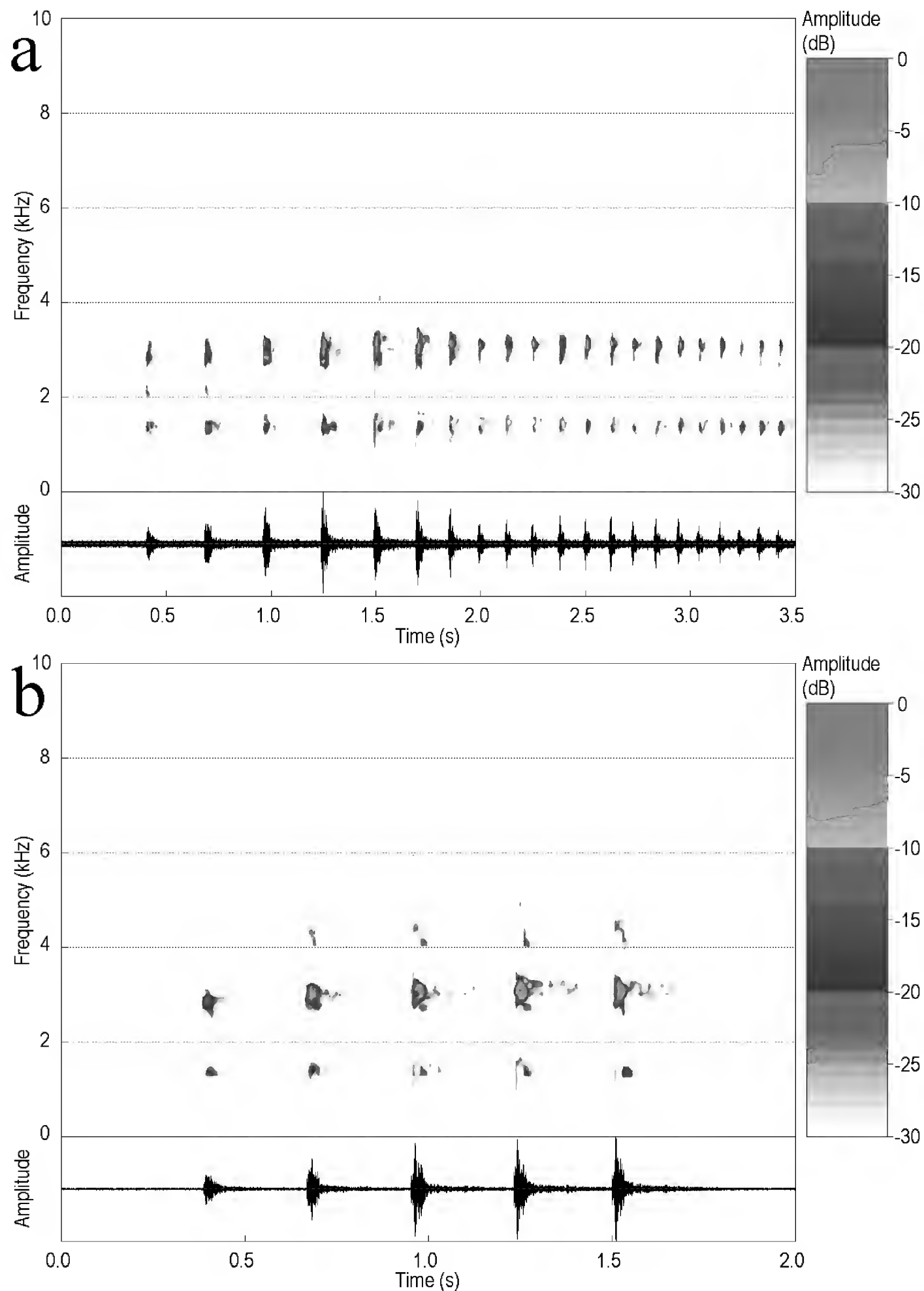
Figure 13. Spectrogram and oscillogram of two calls of *Odontobatrachus arndti* sp. n. from Nimba Mts., Guinea with a dominant frequency of app. 2800-3400 Hz, a fundamental frequency of app. 1400-1700 Hz and 22 notes showing decreasing pause duration between notes (**a**) and 5 notes with constant pauses between notes (**b**).

ranged in longitudinal lines (Fig. 12; Guibé and Lamotte 1958; Rödel and Bangoura 2004). Male femoral glands are yellow (Rödel 2003). Belly colouration (in alcohol) is very variable, ranging from completely whitish, dirty whitish, a distinct reticulation pattern, dark throat with marbling on belly, marbling on throat and belly blurring to paler colouration posteriorly, to completely dark throat and belly, showing no sex-dependant differentiation.

Acoustics. Three calls of *Odontobatrachus arndti* were recorded from specimens in terraria. Calls sound like a repeat of "chucks", consisting of several tonal notes. Two

harmonics were visible (Fig. 13), the second harmonic being the dominant frequency (2842.4–3359.2 Hz), the first being the fundamental frequency (1421.2–1679.6 Hz). The call duration ranged from 1.2 to 3.0 s. One call comprised 22 notes (Fig. 13a) and the other two comprised five notes each (Fig. 13b). Each note had a duration of 34.7 ± 0.01 ms (N = 32). The notes were separated from each other by pauses of 238.0 ± 0.01 ms (N = 12) with the two calls comprising five notes (Fig. 13b). Pause duration in the third call (22 notes) was decreasing from 160.0 to 67.0 ms from the beginning towards the end of the call (Fig. 13a). Rödel's (2003) anecdotal report of a

torrent frog call from Mt. Sangbé (Côte d'Ivoire) comprising a series of click sounds with ever-shorter intervals corresponds to the second call type (22 notes).

Distribution. *Odontobatrachus arndti* is known to occur on the Nimba Mts. in Guinea and Liberia, the adjacent areas at Mt. Gangra (Liberia) and Déré (Guinea), as well as the Mt. Sangbé in western Côte d'Ivoire (Fig. 1). This taxon represents the easternmost representative of the family. Localities at the southern end of the Nimba Mts. and along Mt. Gangra are in very close proximity to *O. natator*. Both species inhabit similar altitudes at the foot of the Nimba Mts. However, at present no differing habitat requirements or ecological adaptations are known (Barej et al. 2015), which could explain their spatial separation.

Etymology. The species epithet *arndti* was chosen in order to honour Prof. emerit. Dr. Rudolf G. Arndt, New Jersey USA, for his trust in young academics and his invaluable support of this study.

Common name. We advise to use the term "Arndt's torrent-frog" in English and "grenouilles des torrents d'Arndt" in French.

Conservation status. Both, the EOO of 2595 km² and the AOO of 156 km² classify *O. arndti* as "Endangered (EN)" (Barej et al. 2015).

Conclusive summary

Only recently, biogeographic separation of molecular lineages identified the monospecific West African torrent-frog family Odontobatrachidae as a complex of cryptic species (Barej et al. 2015). In contrast to studies that predominantly rely on genetics in diagnoses of new species (Jörger and Schrödl 2013; Satler et al. 2013; Petzold et al. 2014), no taxonomic actions were conducted in the case of the *Odontobatrachus natator*-complex. Phylogeographic insights formed the basis for our assessment of morphological characters which led to the formal description of four new species with distinguishing morphological characters.

Odontobatrachus species are phenetically very similar and show an overlap in their morphometrics. Nonetheless, males and females of all species are statistically distinguishable in their metrics and following McLeod et al. (2012), we could confirm that DCAs provided a reasonable method to assign individuals to single species when morphometrics and ratios show no obvious differentiation.

The application of qualitative characters for species differentiation was difficult and previously used diagnostic characters to distinguish *Odontobatrachus* populations (see Rödel and Bangoura 2004; Rödel et al. 2004a), which are appropriate and important in non-related genera, like shape arrangement of dorsal glandular

ridges in *Ptychadena* (Guibé and Lamotte 1957; Perret 1979; Rödel 2000) or ventral colouration in *Phrynobatrachus* (Rödel et al. 2012b; Zimkus and Gvoždík 2013), are not applicable in this family. Specimens belonging to one species from a single locality for example, already show high character variability (see e.g. Fig. 6c, d for glandular ridges in *O. ziama*). Yet, despite problems due to preservation artefacts, a few qualitative diagnostic characters (e.g. shape of the glandular line on tibia, differences in webbing formulae) could be successfully used (Table 7). All subtle differences between *Odontobatrachus* species are supported genetically, with interspecies differences of 2.89–5.55% in 16S rRNA uncorrected p-distance (Barej et al. 2015; Appendix 1: Table A) corresponding to species-level in non-related taxa and additionally, *Odontobatrachus* species are geographically isolated.

Thus, knowledge of the origin of vouchers can narrow down the potential species assignment, because only *O. natator* is widely distributed, from western Guinea to eastern Liberia and southeastern Guinea, while *O. fouta* and *O. smithi* occur only in the westernmost range of that distribution and *O. arndti* and *O. ziama* occur only in the easternmost range. Still, it would be somewhat unsatisfactory, if solely genetics provided a warranted identification of single specimens in areas of distributional overlap between morphologically rather indistinguishable species (Real et al. 2005). Fortunately, following an integrative approach, the consideration of molecular data, distribution patterns, and morphology, rendered recognition of different *Odontobatrachus* species comprehensible despite their superficial similarity. The similarity in morphology of *Odontobatrachus* species likely results from speciation lacking distinct external changes (Bickford et al. 2007), probably because the most conspicuous characters are all adaptations to the habitat of fast flowing streams. The *Odontobatrachus* species all exhibit a torrent-frog'-habitus as likewise independently developed in various non-related taxa (e.g. Petropedetidae: *Petropedetes, Arthroleptides*, Barej et al. 2010, 2014a, b; Hylodidae: *Hylodes*, Haddad and Giaretta 1999; Heleophrynidae: *Heleophryne, Hadromophryne*, Minter et al. 2004; Ranidae: *Staurois*, Matsui et al. 2007). Adults usually possess a rather flattened body shape and head, allowing them to hide between crevices and under rocks. Their extremities are long, terminal phalanges enlarged and digits on hands and feet spatulated offering a larger contact area with the slippery substrate (Minter et al. 2004; Scott 2005; Kamermans and Vences 2009). Torrent-frog tadpoles have a streamlined habitus with distinct tail musculature and a sucker-like mouth which are used to cling or climb on rocks (Barej et al. 2010; Minter et al. 2004). It seems likely that any radical deviation from that morphotype could have negative effects on species survival.

Recognition and description of species is just a first step which provides the baseline for subsequent studies to gather further data on the ecology or behaviour – or

Table 7. Important morphological features and measurements (in mm) that can be applied for species identification in West African torrent-frogs *Odontobatrachus*. See material and methods section for abbreviations.

	O. natator	O. ziama sp. n.	O. smithi sp. n.	O. fouta sp. n.	O. arndti sp. n.
OTU sensu Barej et al. (2015)		OTU1	OTU2	OTU3	OTU4
distribution	western to eastern Upper Guinea	eastern Upper Guinea (Simandou Mtn. Range)	western Upper Guinea (Fouta Djallon, Boffa)	western Upper Guinea (Fouta Djallon)	eastern Upper Guinea (Nimba Mts., Mt. Sangbé)
femoral glands in males	present	present	present	present	present
tusk-like odontoids	present	present	present	present	present
skin texture	heterogeneous, granular	heterogeneous, granular	heterogeneous, granular	heterogeneous, granular	heterogeneous, granular
typical glandular line on tibia	small to large conic glands, more or less interrupted lines	minuscule to small conic glands, almost continuous lines	small to mean conic glands, predominantly interrupted lines	small to large glandular conic glands, rather interrupted line	small to mean glandular conic glands, hardly interrupted line
ventral colouration	uniform pale, dirty whitish, dark with pale markings, uniform dark	uniform pale, dirty whitish, reticulated, uniform dark, dark with paler markings	uniform dark or few paler markings	uniform dark, rarely paler markings or dirty smeared	uniform pale, dirty whitish, reticulated, fading posteriorly from throat to belly, uniform dark
colouration of male femoral glands	rose-coloured	dark orange	pale orange	bright orange	unknown
typical webbing formula	0-0.25/0-0.75/0-0.5/0.5-0.75/0.75-0 0-0.5/0-1/0-1/1-0	0-0/0-0.5/0-0.5/0.5-0 0-(0.25 to 0.5)/0-1/0-0.75/0.75-0	0-(0.75 to 1)/0-1/0-1/1-0	0-0.75/0-1/0-1/1-0	0-0.5/0-1/0-1/1-0 0-0.25/0-1/0-0.75/0.75-0
typical number of dorsal glandular ridges	3 to 5	4 to 5	5 to 6	4 to 5	4 to 5
max SUL (m / f)	52.5 / 61.1	50.3 / 60.3	60.4 / 61.9	57.0 / 62.5	53.6 / 64.0
GL/FM	0.46	0.51	0.46	0.46	0.51
GL/GW	1.96	1.83	2.12	1.72	1.86
HW/SUL (m / f)	0.36 / 0.35	0.35 / 0.34	0.38 / 0.37	0.37 / 0.37	0.35 / 0.34
TD/O (m / f)	0.40 / 0.40	0.34 / 0.34	0.44 / 0.44	0.43 / 0.47	0.37 / 0.38
O/EN (m / f)	1.82 / 1.80	2.16 / 2.03	1.60 / 1.69	1.78 / 1.71	2.05 / 2.04

simply: naming does not mean knowing a species. Our knowledge on the family is still incomplete, as calls of four species remain unknown and data on tadpole morphology is lacking. Lamotte and Zuber-Vogeli (1954) published a detailed description on tadpoles collected in Liberia and the Nimba Mts., thus we cannot rule out that their description is based on material from two different species (*O. natator* and *O. arndti*). Solely Guibé and Lamotte (1958) described a series of tadpoles from the River Zougue on the Nimba Mts. (described as *O. arndti* in the present work) and provided a short and superficial description of the habitus. Our insights on the ecology of adult *Odontobatrachus* are scarce, too. While streams with rapids in primary forests are the preferred habitat of torrent-frogs, detailed ecological studies are missing. Differences in habitat requirements and ecology have been recognised as further characters of importance to distinguish species in other genera (e.g. *Amietophrynus regularis* vs. *A. maculatus* Amiet 1976; Böhme 1994a; *Phrynobatrachus guineensis* and *P. phyllophilus* Rödel and Ernst 2002).

'West African Forests' are recognised as one of the world's biodiversity hotspots (Myers et al. 2000; Bakarr et al. 2001) and biogeographically demarcated from the Central African forest block (Penner et al. 2011). Although West Africa, defined as ranging from Senegal to Nigeria, is regarded as one of the better known regions on the continent, more than ten new amphibian species have been described in the last decade (e.g. Blackburn et al. 2008; Ernst et al. 2008; Hillers et al. 2008b; Rödel 2007; Rödel and Bangoura 2004; Rödel et al. 2003, 2009a, b, c, 2010, 2011, 2012a, b) and more await formal description (Rödel et al. unpubl. data.). We agree with McLeod et al. (2012) that it is crucial to identify the "true" biodiversity and although species that are morphologically difficult to tell apart become condemned, their scientific recognition is an indispensable tool for conservation management.

In the past, only a single torrent-frog species, *O. natator*, was known to occur in West Africa and it has been listed as "Near Threatened (NT)" according to the IUCN Red List (IUCN 2011). However, we herein demonstrated the presence of four new *Odontobatrachus* species with dramatically constrained distribution ranges. According to Barej et al. (2015) all five species (therein treated as OTUs) require the IUCN category "Endangered (EN)" resulting from analyses of the range criteria Extent of Occurrence and Area of Occupancy. Environmental Niche Modelling (Barej et al. 2015) confirmed large distributional gaps and thus justify the use of AOO as criterion. Both sub-clades of *O. natator* should be treated distinctly with regard to conservation concern as evolutionary significant units (Moritz 1996, 2002; Ennos et al. 2005) and if recognised as distinct species, the Freetown Peninsula population demands the category "Critically Endangered (CR)". As habitat loss is ongoing due to forest fragmentation and

conversion (Chatelain et al. 1996; FAO 2006; Norris et al. 2010), conservation efforts need to be made soon. In the case of West African torrent-frogs, only *O. natator*, *O. ziama* and *O. arndti* occur in protected areas like National Parks and Biosphere Reserves (Barej et al. 2015) and even if lower priority areas are considered, they fail to protect all five species. Distribution patterns in *Odontobatrachus* cover ranges of various endemic species in the Fouta Djallon and the Nimba Mts.-Massif du Ziama-Simandou Mountain Range (Angel 1943; Porembski et al. 1994, 1995). Furthermore, these areas are assumed to have played an important role as forest refugia in Upper Guinea (Maley 1987; Porembski et al. 1994; Sosef 1994). Barej et al. (2015) suggested that the Loma Mts. and Tingi Hills in Sierra Leone, as the highest elevation occurrences in the Guinea Highlands, could represent a refugium in central Upper Guinea for *O. natator* and any conservation effort within these areas could consequently be beneficial in many non-related taxa and assure their long-term survival.

In summary, the diversity in the family Odontobatrachidae has been raised to five species. While our knowledge on this West African endemics is far from complete, nomination of OTUs recognised by Barej et al. (2015) is of importance for promoting immediate conservation actions as all species require the IUCN category "Endangered (EN)". West African torrent-frog species are at risk of becoming extinct because of habitat loss in the Upper Guinean biodiversity hotspot, whose "true" biodiversity is still far from being completely known.

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Appendix 1

Table summarising uncorrected p-distances within and between *Odontobatrachus* spp.

Table A. Uncorrected p-distances within (first column) and between *Odontobatrachus* spp. based on 567bp of the 16S rRNA gene. Minimum to maximum values (lower left corner), mean values with standard deviation and sample size (upper right corner) are given. For seemingly high intra-species differences in *Odontobatrachus natator* see Barej et al. (2015).

Taxon	intraspecies	*O. natator*	*O. ziama* sp. n.	*O. smithi* sp. n.	*O. fouta* sp. n.	*O. arndti* sp. n.
O. natator	0.00–1.98; 0.42 ± 051 (703)		4.36 ± 0.21 (1216)	4.88 ± 0.19 (1216)	4.34 ± 0.20 (418)	4.82 ± 0.27 (1596)
O. ziama sp. n.	0.00–0.72; 0.27 ± 0.21 (630)	3.74–4.87		5.03 ± 0.14 (320)	4.25 ± 0.13 (352)	3.36 ± 0.22 (1344)
O. smithi sp. n.	0.00–0.54; 0.20 ± 0.19 (45)	4.50–5.40	4.86–5.41		4.01 ± 0.11 (110)	5.21 ± 0.17 (420)
O. fouta sp. n.	0.00–0.36; 0.15 ± 0.15 (55)	3.97–4.88	3.99–4.53	3.79–4.15		4.52 ± 0.16 (462)
O. arndti sp. n.	0.00–0.58; 0.05 ± 0.11 (861)	3.40–5.40	2.89–3.97	4.60–5.55	4.17–4.98	

Appendix 2

Table summarising voucher specimens, and additional GenBank accession numbers.

Table B. List of additionally generated *Odontobatrachus* sequences and respective GenBank accession numbers. *Odontobatrachus* sequences analysed in Barej et al. (2015) refer to the following GenBank numbers and publications ([1]Barej et al. 2015; [2]Barej et al. 2014; [3]Loader et al. 2013; [4]Rödel et al. 2005): 16S: KP005071–124[1], KF693390–5[2], JX546953–4[3]; AY902379[4]; 12S: KP005195–243[1], KF693286–91[2], JX546938–9[3]; cytb: KP005418–32[1], KF693670–5[2], JX546968–9[3]; BDNF: KP005312–26[1], KF693488–93[2]; SIA: KP005377–KP005396[1], KF693550–5[2]; RAG1: KP005345–59[1], KF693610–5[2].

taxon	voucher	country	16S	RAG1
O. natator	ZMB 80505	Liberia	KP284862	...
O. natator	ZMB 80504	Liberia	KP284863	...
O. ziama sp. n.	MHNG 2731.45	Guinea	...	KP284864
O. ziama sp. n.	MHNG 2731.46	Guinea	...	KP284865
O. ziama sp. n.	ZFMK 95465	Guinea	...	KP284866
O. ziama sp. n.	ZMB 78299	Guinea	...	KP284867
O. ziama sp. n.	ZMB 78300	Guinea	...	KP284868
O. smithi sp. n.	ZMB 78311	Guinea	...	KP284869
O. fouta sp. n.	MHNG 2731.48	Guinea	...	KP284870
O. arndti sp. n.	MHNG 2731.50	Guinea	...	KP284871
O. arndti sp. n.	ZFMK 95467	Guinea	...	KP284872

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Appendix 3

Table summarising Kruskal-Wallis test statistics.

Table C. Kruskal-Wallis test statistics for *Odontobatrachus* male and female absolute values (A) and ratios (B). χ^2-value, degree of freedom (DF) and the asymptotic significance (Asymp. Sig.) are given. See material and methods section for abbreviations.

A		SUL	HW	FM	GL	GW	TI	FL	IT	TD	O	ID	EN	ES
male	χ^2	22.350	22.253	25.490	15.559	16.667	32.377	30.048	6.008	24.781	13.177	15.413	40.756	9.201
	DF	4	4	4	4	4	4	4	4	4	4	4	4	4
	Asymp. sig.	<0.001	<0.001	<0.001	<0.01	<0.01	<0.001	<0.001	0.20	<0.001	<0.05	<0.01	<0.001	0.06
female	χ^2	8.675	20.808	19.819	...	...	29.519	28.719	8.745	53.107	11.852	14.913	40.751	4.604
	DF	4	4	4	...	...	4	4	4	4	4	4	4	4
	Asymp. sig.	0.07	<0.001	<0.01	...	...	<0.001	<0.001	0.07	<0.001	<0.05	<0.01	<0.001	0.33
B		TI/SUL	FM/TI	FL/SUL	GL/FM	GL/GW	HW/SUL	TD/O	FM/SUL	IT/FL	O/EN	ES/O	TD/SUL	
male	χ^2	13.864	0.493	19.752	9.968	14.135	20.020	22.370	9.054	13.709	34.315	12.905	14.156	
	DF	4	4	4	4	4	4	4	4	4	4	4	4	
	Asymp. sig.	<0.01	0.98	<0.01	<0.05	<0.01	<0.001	<0.001	0.06	<0.01	<0.001	<0.05	<0.01	
female	χ^2	5.743	11.946	7.465	...	...	30.977	49.876	1.398	8.857	41.359	12.499	45.160	
	DF	4	4	4	...	...	4	4	4	4	4	4	4	
	Asymp. sig.	0.22	<0.05	0.11	...	...	<0.001	<0.001	0.85	0.07	<0.001	<0.05	<0.001	

Description and morphology of the "Juan de Fuca vent mussel", *Benthomodiolus erebus* sp. n. (Bivalvia, Mytilidae, Bathymodiolinae): "Phylogenetically basal but morphologically advanced"

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http://zoobank.org/D2E0E6B8-EFAB-4D25-93E6-64B9C212679D

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Abstract

The Juan de Fuca vent mussel first found in 1990 is formally described as *Benthomodiolus erebus* **sp. n.** Comparisons are made with the three other species previously assigned to *Benthomodiolus* namely *B. lignocola*, *B. geikotsucola* and *B. abyssicola*. The anatomies of all four species are examined and are shown to share the arrangement of pedal and byssus musculature, having largely un-fused mantle margins and a hind gut with a very short loop. The ctenidia were examined using both light and scanning electron microscopy. *B. erebus* and *B. geikotsucola* were found to have lamellar filaments with extensive abfrontal expansion and fusion of the ascending and descending arms. In this there is similarity with *Bathymodiolus*. *B. lignocola* and *B. abyssicola* were found to have linear filaments with narrow abfrontal surfaces with little fusion. All four species were shown to have the abfrontal surfaces covered by polygonal cushions of microvilli although these were much less apparent in *B. lignocola* and *B. abyssicola*. Although *Benthomodiolus* was shown by a number of previous studies, using molecular data alone, to be phylogenetically basal to all other Bathymodiolinae the anatomy is highly adapted for chemosymbiosis. Species of *Benthomodiolus* are found on wood-falls, whale-falls and vent sites and thus mirror the habits of the *Bathymodiolus/Idas* clade.

Introduction

In a forthcoming paper, Thubaut et al. (in press) rightly highlight the mismatch between molecular data and morphological taxonomy within the Bathymodiolinae. They cite, in particular, the case of the mussel found at the Juan de Fuca vent site in 1990 and listed as *Adipicola* sp. by Juniper et al. (1992). It was later considered to be an un-described species of *Bathymodiolus* by McKiness et al. (2005) then as an un-described species of *Adipicola* by Southward (2008) and as a code number only by Fontanez and Cavanaugh (2013). It has most recently been linked with the clade that includes *Benthomodiolus* (Lorion et al. 2013, Thubaut et al. 2013, Rodrigues et al. 2015).

None of the published literature illustrates this mussel with the exception of the micro-structure of the ctenidi-

um by Southward (2008). In the above molecular studies *Benthomodiolus*, including the Juan de Fuca mussel consistently appears as basal to all other Bathymodiolinae. Fontanez and Cavanaugh (2013) show that the sister group to the bathymodiolines consists of species of *Modiolus* sensu stricto. Gustafson et al. (1998) following Kenk and Wilson (1985) noted that the gill morphology of *Benthomodiolus abyssicola* (Knudsen, 1970) was like that of a typical *Modiolus* suggesting that it was a filter feeding species.

This paper will give a proper taxonomic status to the Juan de Fuca mussel through a description of the shell and anatomy and will make comparisons with other species assigned to Benthomodiolus. The habitat range of *Benthomodiolus* species includes the three major ecotypes, wood-fall, whale-fall and vent; this paper will investigate

whether there are any associated morphological characters. Comparisons with other members of the Bathymodiolinae will attempt to elucidate whether the basal position of *Benthomodiolus* is reflected in the morphology.

Materials and methods

The materials examined in this paper are listed under their respective sections in the results section.

The photographic images were made using a Leica Z6 macroscope and image stacking using Helicon Focus™ software. For some anatomical images the tissues were stained with methyl green to enhance contrast. For scanning electron microscopy gill tissues were excised and cut transversely and longitudinally using a thin razor blade. Tissues were dehydrated in 100% ethanol overnight and critically point dried with liquid CO_2 as the intermediate fluid in a Quorum K850 critical point dryer. Dried samples were mounted and gold coated before examination using a Jeol Neoscope™ SEM.

Institutional abbreviations

CMN ML   Canadian Museum of Nature, Mollusca
MNNZ   Museum of New Zealand Te Papa Tongarewa
NMW.Z.   National Museum Wales, Zoology
NSMT   National Science Museum Tokyo
ZMUC   Zoological Museum University of Copenhagen

Anatomical abbreviations

aa anterior adductor muscle, **an** anus, **apr** anterior pedal retractor muscle, **au** auricle, **bys** byssus, **ct** ctenidium, **ddd** duct to the digestive diverticula, **eav** exhalant aperture valve, **f** foot, **f(hl)** heel of foot, **f(t)** toe of foot, **fme** fused mantle edge, **h** heart, **hgl** hind gut loop, **ifj** inter filamentar junction, **ilp** inner labial palp, **imf** inner mantle fold, **lp** labial palps, **lps** suspensor muscle for labial palps, **me/ct** mantle edge ctenidial junction, **mef** mantle edge folds, **mmf** middle mantle fold, **oe** oesophagus, **olp** outer labial palp, **omf** outer mantle fold, **ot** oral tube, **pa** posterior adductor muscle, **pbr1** posterior pedal/byssus retractor muscle, **pbr2** anterior pedal/byssus retractor muscle, **per** pericardium, **pms** posterior mantle septum, **ppr** posterior pedal retractor muscle, **psf** polygonal surface, **rt** rectum, **sppr** secondary posterior pedal retractor muscles, **ss/mg** style sac and mid gut, **st** stomach, **vfg** ventral food groove, **vg** visceral ganglion, **vt** ventricle

Results

Class Bivalvia Linnaeus, 1758
Superfamily Mytiloidea Rafinesque, 1815
Family Mytilidae Rafinesque, 1815
Subfamily Bathymodiolinae Kenk & Wilson, 1985

Genus *Benthomodiolus* Dell, 1987

Type species. *Benthomodiolus lignocola* Dell, 1987

Definition. To 43 mm, Thin, umbonate, narrow modioliform, weakly arcuate with beaks about ¼ distance from the anterior, anterior margin rounded only a little narrower than rounded posterior margin. Median area slightly sulcate, widest part behind the umbos. Hinge margin lacking crenulations, ligament sunken, very long. Periostracum persistent, smooth or with sparse hairs. Pedal/byssus musculature in two groups, a posterior set close to the posterior adductor muscle and a median set attached to rear of the umbo. Mantle edge mostly free, poorly frilled, posterior junction short. Hind-gut with a short or very short loop. Ctenidial filaments linear or laminar. Symbiotic bacteria are extra-cellular.

Species included. *B. lignocola* Dell, 1987; *B. geikotsucola* Okutani & Miyazaki, 2007; *B. erebus* this paper; *B. abyssicola* (Knudsen, 1970).

Distribution. Described species are restricted to the Pacific Ocean from New Zealand, Japan, British Columbia and Panama at bathyal to abyssal depths. An undescribed species from the South Atlantic is reported by Thubaut et al. (2013)

Remarks. *Benthomodiolus lignocola, B. geikotsucola* and *B. erebus* are regarded as congeneric, based on a combined analysis of COI mtDNA and 28S rRNA (Thubaut et al. in press). In the tree by Thubaut et al. (in press) *B. lignocola* is shown as the sister taxon to *B. erebus* with *B. geikotsucola* and an un-named species from the South Atlantic as sister taxa on a separate branch. Kyuno et al. (2009) and Lorion et al. (2013) show *B. erebus* and *B. geikotsucola* as sister taxa with *B. lignocola* on a separate branch. The tree by Kyuno et al. (2009) is based on the mitochondrial ND4 gene while that of Lorion et al. (2013) is based on combined COI, NADH4, 16S, nuclear 28S and histone 3 data. *Modiolus abyssicola* lacks supporting molecular data and is placed in *Benthomodiolus* primarily on the disposition of the pedal/byssus musculature (Dell 1987, Thubaut et al. in press). Bouchet in WoRMS (2015) also includes *M. abyssicola* in *Benthomodiolus*. However Gustafson et al. (1998) and Kenk and Wilson (1985) reported that the gill of *M. abyssicola* was like that of *Modiolus* and thus probably a filter feeder and suggested that it should be placed in *Modiolus*. This species is revisited below and shown to have a more typical wedge shaped modioliform shell with sparse hairs. The gill filaments are linear and unlike the laminar filaments in *Bathymodiolus* as reported by Kenk and Wilson (1985). However it is shown below that the filaments of *B. lignocola* are also linear but that in both *B. lignocola* and *B. abyssicola* that the abfrontal surface bears polygonal microvillar structures typical of species with symbiotic bacteria. Regardless of the shell form it seems most likely that *M. abyssicola* does belong in *Benthomodiolus* but it remains tentative awaiting confirmation from molecular data.

Consequently the generic diagnosis is based on a combination of characters from *B. lignocola, B. geikotsucola* and *B. erebus*. If *M. abyssicola* is included, then the shell form needs to be expanded to include the more wedge shaped form.

Taxon	Max shell length AL/TL	Bathymetric range	Distribution	Habitat
B. lignocola	18.1 mm 0.23	810–2670m	Chatham Isds, SW Pacific	Wood
B. geikotsucola	42.5 mm 0.28	4020m	Torishima Seamount, W. Pacific	Whale bone
?B. abyssicola	17.2 mm 0.22	3670–3270m	Gulf of Panama, CE. Pacific	Wood
B. erebus	39.9 mm 0.23	2195m	Juan de Fuca Ridge, NE Pacific	Vent

Benthomodiolus erebus sp. n.

http://zoobank.org/9979FC77-8E46-45E5-908C-7452BD83A435
Adipicola sp. (n. sp.?) Juniper et al. 1992: 1797–1799, shell not illustrated
Bathymodiolus sp. JdeF McKiness et al. 2005: 109–116, shell not illustrated.
Adipicola MV Southward, 2008: 139–146, shell not illustrated

Material examined. *Holotype*. 1 specimen, ROV ROPOS dive R682, Clam Bed, Endeavour Segment, Juan de Fuca Ridge, 47°57.8'N 129°05.5'W, 2195m, 19/August/2002. CMNML 097165

Dimensions in mm. Length 39.9; Height 15.4; Width 13.2; Anterior length 9.5

Paratype. 1 broken and partly dissected specimen, Alvin dive 2803, clam bed, Kini's Site, Middle Valley, Juan de Fuca Ridge, 48°27.40'N 128°42.52'W, 2416m, 24/July/1994. NMW.Z.2015.013.1. This is the remainder of the material used by Southward (2008) in her description of the fine structure of the ctenidium..

Dimensions in mm. Length 39.2; Height 12.9; Width 10.3; Anterior length 10.1

Shell. Holotype, (Fig. 1a–d). Thin but not fragile (0.45–0.55 mm at margins). Equilateral. Umbos prominent. Inequilateral, beaks towards the anterior, just under ¼ the total length from the anterior. Outline modioliform elongate, posterior a little deeper than anterior, ventral margin slightly concave, dorsal margin gently curved. Median area sulcate, widest behind the beaks. Ligament sunken very long, 17.7 mm, no trace of underlying crenulations visible. Anterior hinge margin extending posteriorly beyond the beaks for a short distance. A small flattened lunule under the beaks. Sculpture of fine commarginal ridges increasing in size towards the margins, periostracum persistent of a golden amber shade and lacking hairs. Internally shiny, pearlescent, muscle scars indistinct. The prodissoconch and juvenile shell are too eroded to give any details here.

Paratype, (Fig. 1e). The shell is of the same proportions as the holotype, but is very much thinner (0.25–0.35 at margins) and more fragile.

Pedal byssus musculature. (Fig. 2a–c). The foot has a long toe and a greatly reduced heel. The byssus gland opens at the base of the heel and a longitudinal groove runs the entire length of the sole. The pedal/byssus retractor is divided into two widely separated bundles; the posterior bundle (pbr1) is greatly elongate, of two primary strands and attaches to the shell just above the posterior adductor muscle; the median bundle (pbr2) is relatively

short and extends dorsally attaching in the rear of the umbonal cavity. The posterior pedal retractor is slender and runs anterior and parallel to the median pedal/byssus retractor; associated are three fine strands or secondary posterior pedal retractor muscles that coalesce with median pedal/byssus retractor; the anterior pedal retractor consists of two primary bundles, is elongate and attaches above and separate from the anterior adductor muscle. A slender labial palp suspensor muscle rises from the base of the anterior pedal retractor and is inserted into the anterior adductor muscle.

Adductor muscles. The adductor muscles are of almost equal size, the posterior is circular in section while the anterior is oval (Fig. 2a).

Ctenidium and labial palps. The ctenidia (Figs 2a, 4a–e) run almost the entire length of the mantle cavity, are thick and fleshy but not deep (Fig. 2a). The inner demibranch is a few filaments longer than the outer demibranch, both have reflected filaments the ascending arms slightly shorter than the descending arms and these fused for over half their length. The filaments are extended abfrontally, appearing as lamellae (Fig. 4a). There is a groove along the ventral edge of each demibranch (Fig. 4a). The filaments are held together by a single row of large ciliary junctions.

Scanning electron microscopy reveals that the abfrontal surfaces are extensive giving a triangular plate like form to the largely fused ascending and descending arms of each filament (Fig. 4b, c). The inter filamental junctions are very prominent formed of a large bundle of cilia (Fig. 4d). The frontal surface is ciliated while the abfrontal surface is covered by an epithelium of microvilli arranged in a polygonal structure (Fig. 4c, e). The ultrastructure was described my Southward (2008) where the ctenidia were shown to harbor symbiotic bacteria extra–cellulary among dense epithelial microvilli.

The labial palps (Fig. 3f) are small, triangular with 15 sorting ridges on each, there is a short oral tube extending to the mouth.

Mantle edge and apertures. The mantle edges are free for their entire length (Fig. 2a) except for a narrow septum (Fig. 3b and c, pms) separating the exhalant aperture from the inhalant/pedal aperture. The mantle edge is composed of three major folds (Fig. 3c–e); the outer mantle fold (omf) is smooth and never fused. The middle mantle fold (mmf) is narrow and weakly frilled (Fig. 3d), the stronger frills border the posterior inhalant aperture and a short median section (Fig. 3e), probably marking the pedal aperture. The inner mantle fold is finely digitate
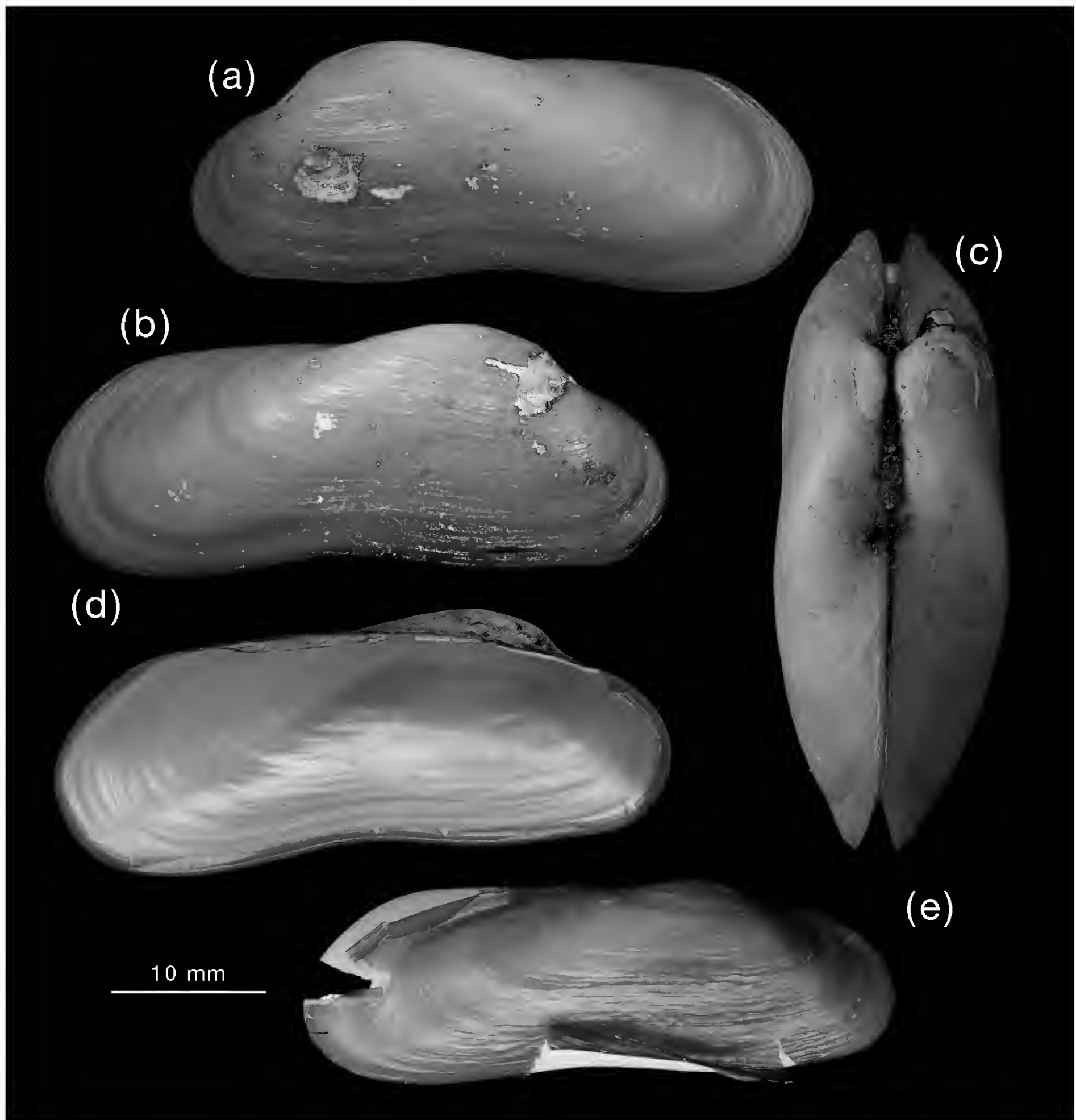
Figure 1. Shells of *Benthomodiolus erebus* sp. n. **a–d** holotype, **a** external of left valve, **b** external of right valve, **c** internal of left valve, **d** dorsal (CMNML 097165), **e** paratype external of right valve (NMW.Z.2015.013.1a).

along the entire length of the pedal/inhalant aperture (Fig. 3d). The exhalant aperture is muscular and smooth edged (Fig. 3b); a pair of folds visible on the inner face probably function as a valve (Fig. 3a, eav).

Alimentary system. The alimentary system (Fig. 3g) runs along the dorsal surface of the visceral mass. The oesophagus (oe) is flattened, relatively short, about the same length as the stomach (st). The mid gut and hind gut run in a straight line except for a very short, tight, loop (hgl) just anterior of the heart ; the hind gut runs through heart and the rectum (rt) runs over the posterior adductor muscle with the simple anus (Fig. 3c) opposite the exhalant aperture.

Stomach. The stomach (Fig. 3g) (st) is situated beneath the umbos, in front of the median pedal/byssus retractors (pbr2) and slightly to the left side. Externally it is elongate somewhat triangular in form, the oesophagus (oe) entering on the anterior face and the conjoined style sac/mid gut (ss/mg) leaving posteriorly, the right side is expanded posteriorly, there is a very slight division into anterior and posterior chambers with a shallow dorsal caecum (ca) projecting on the left anterior dorsal face and the thickening of the dorsal hood behind this (dh). Ducts to the digestive gland are prominent on the right dorsal side (ddd1–3) while a large duct exits on the lower mid left and a smaller
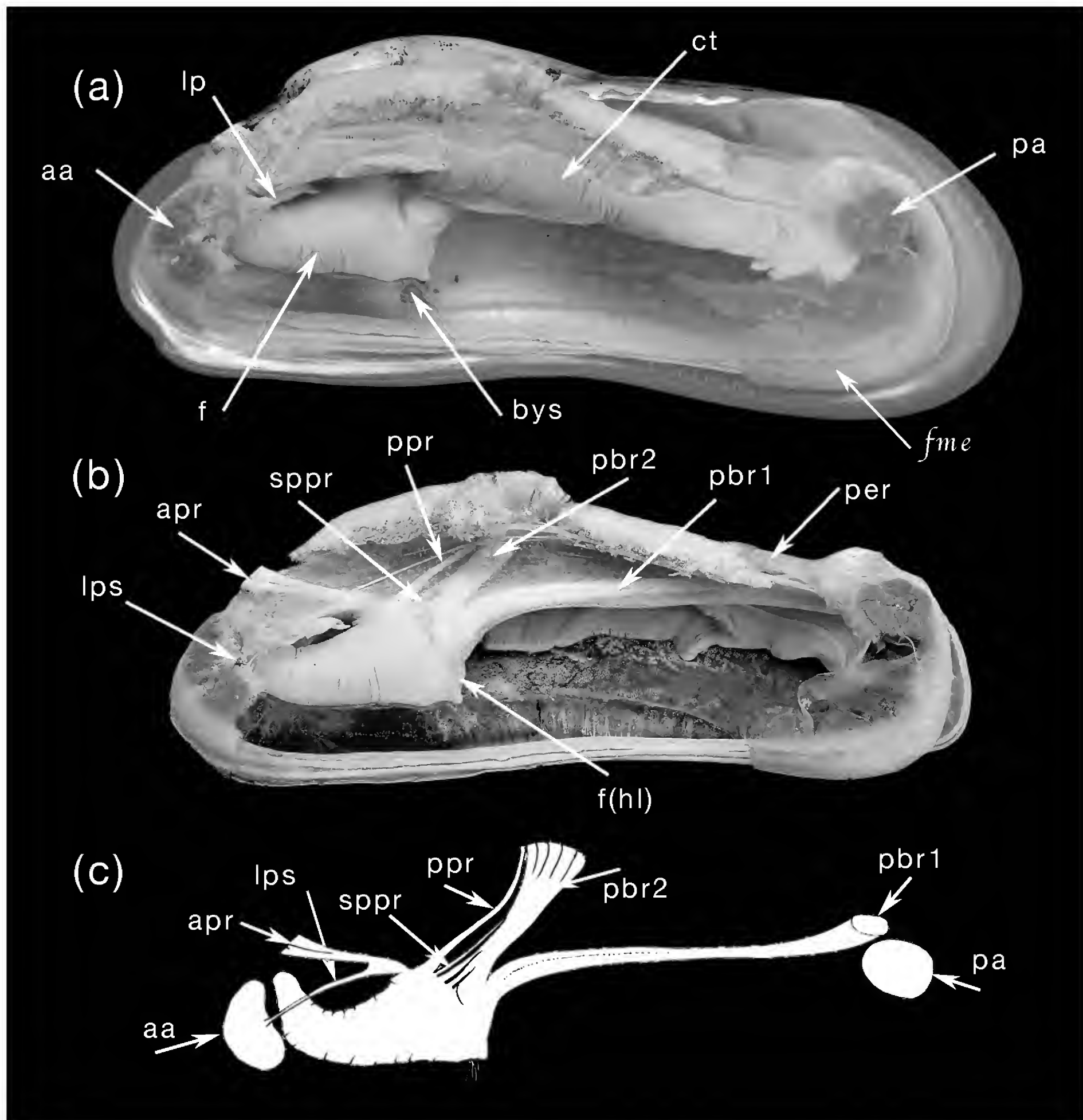
Figure 2. Gross anatomy of *Benthomodiolus erebus* sp. n. **a** after removal of left valve and mantle, **b** after further removal of the ctenidium, **c** diagram of the adductor, pedal and byssal musculature.

on the ventral anterior left, the latter are not visible in Fig. 3g.

Internally the major typhlosole runs longitudinally across the floor, to its right side there is a smooth depression and leading off this are tracts to the right side ducts. There is a deep embayment running from the anterior floor and up the anterior left side into the shallow caecum and sharply curving into the dorsal hood. A small duct exits this trough on the anterior floor and medially it opens into the left pouch where a large duct exits. The gastric shield is shaped into the dorsal hood and extends posteriorly of the left posterior dorsal face. Sorting ridges are nowhere apparent except for a well

defined but small area on the posterior edge of the left pouch.

Pericardium. The pericardium (Figs 2b, 3g) (per) is situated immediately anterior of and between the posterior byssus retractor muscles (pbr1). The ventricle (vt) is muscular while the auricles (au) are relatively large and thin walled. The arrangement of the auricular veins could not be elucidated.

Etymology. After *Erebus* (Greek), Noun in apposition, "place of darkness between earth and Hades" alluding to the abyssal, hydrothermal vent, type locality.
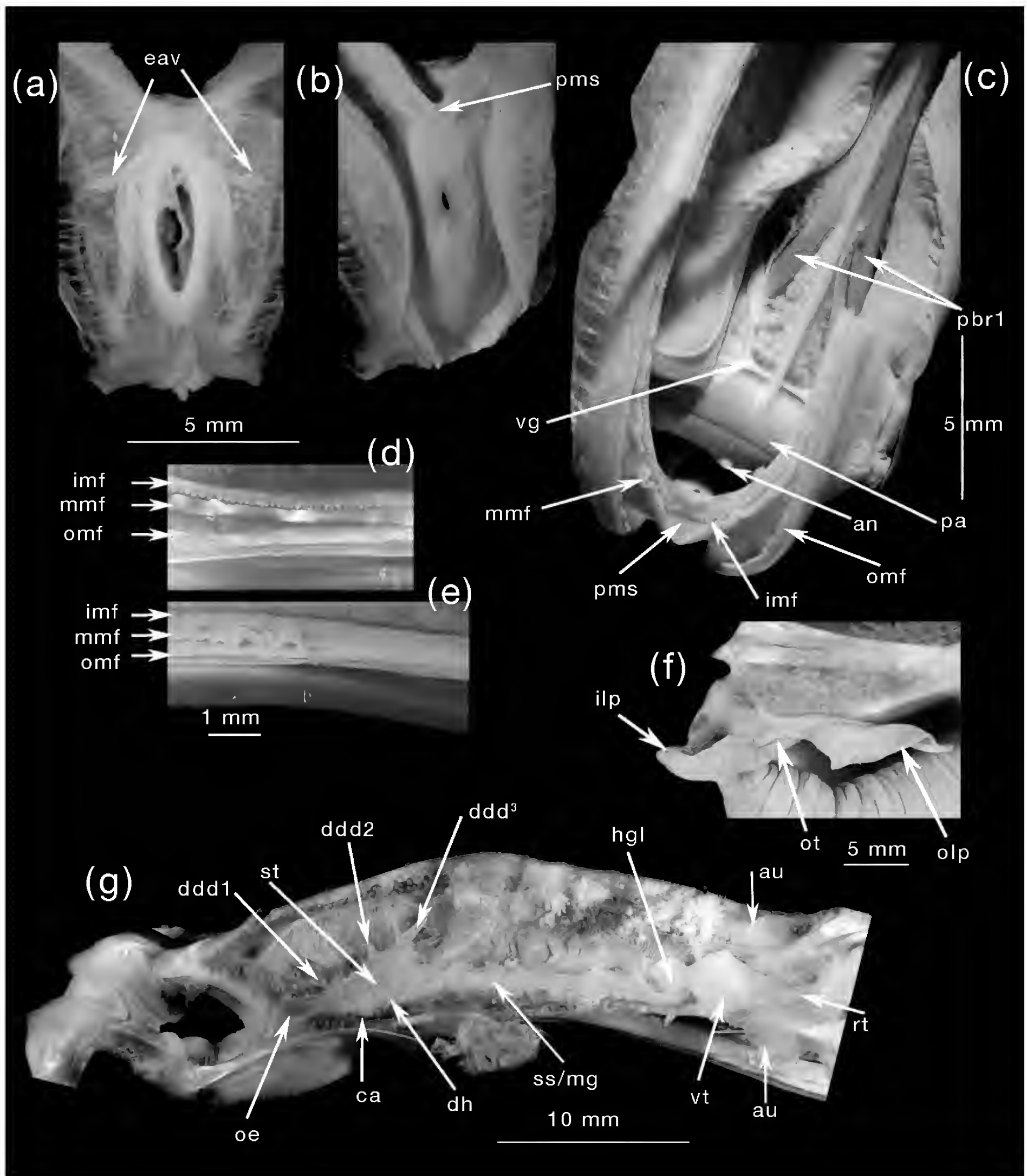
Figure 3. Anatomical details of *Benthomodiolus erebus* sp. n. **a** exhalant aperture interior, **b** exhalant aperture exterior, **c** posterior viewed from the ventral, **d** anterior mantle edge, **e** middle mantle edge, **f** labial palps, **g** the alimentary system and heart.

Habitat. Located in low temperature vent flows (<20 °C) associated with other vent endemic animals but is sparse and rarely collected. At Endeavour, it was recovered in a grab of the siboglinid *Ridgeia piscesae* at the base of the tubeworm cluster. At Middle Valley, it was recovered nestled in crevices of a sulphide block, also colonized by *R. piscesae*. Overall setting at both sites featured sulphide deposits with limited high temperature venting surround-ed by ponds of sediment where vesicomyid clams also occurred (Juniper et al. 1992). Although *B. erebus* sp. n. is not visible on ROV imagery the typical habitats are illustrated here (Fig. 5a–b).

Comparative taxa. In the following section the other species assigned to *Benthomodiolus* are described with reference to *B. erebus* rather than in full detail.
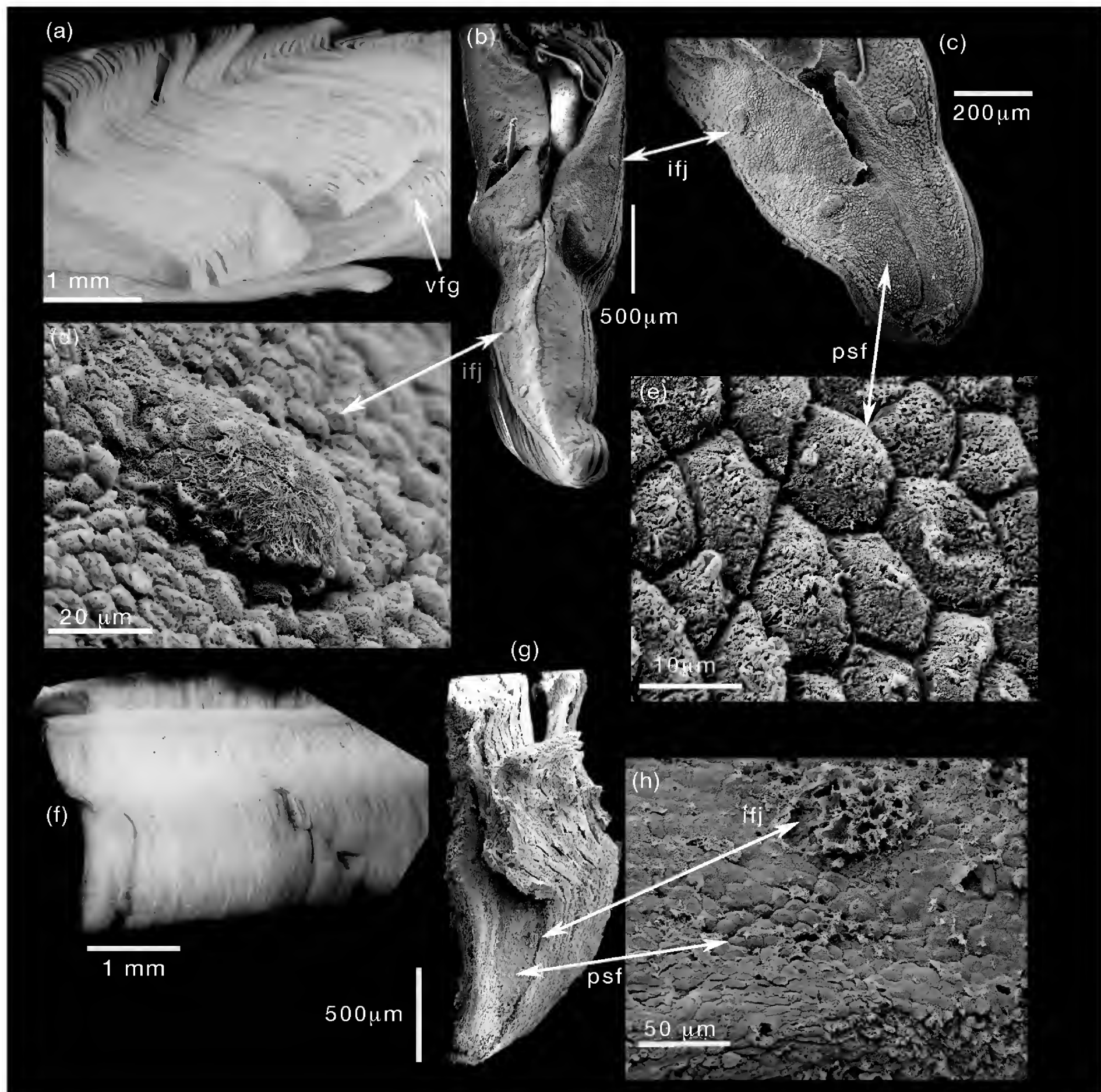
Figures 4. a–e The ctenidium of *Benthomodiolus erebus* sp. n. **a** gross anatomy, **b** SEM of whole filament, **c** SEM of tip of a single filament, **d** SEM of inter filamental junction, **e** SEM of polygonal surface of microvilli. **f–h** The ctenidium of *B. geikotsucola*. **f** gross anatomy, **g** SEM of a whole filament, **h** SEM of the polygonal surface.

Benthomodiolus geikotsucola Okutani & Miyazaki, 2007

Benthomodiolus geikotsucola Okutani & Miyazaki, 2007: 49–55, figs 2–3.

Material examined. *Holotype.* 1 shell, NSMT-Mo-76703, Summit of Torishima Seamount, 30°55'N 141°49'E, 4020m. Not examined, image courtesy of NSMT.

 *Paratype.* 1 specimen, NSMT Mo-76704j as holotype.

Shell. The holotype (Fig. 6a) is slightly larger than any of the shells of *B. erebus* reaching 42.5 mm. The paratype dissected here (Fig. 6b–d) was 28.1 mm in length. The shell is umbonate, narrowly arcuate in outline and medially sulcate; in these there is strong similarity to *B. erebus*.

The beaks are rather distant from the anterior margin, more so than in B. erebus with a total length /anterior length of 0.28 compared with a value of 0.24 for *B. erebus*. As in *B. erebus* the periostracum is smooth and devoid of hairs.

Pedal byssus musculature. The arrangement of the pedal and adductor muscles (Fig. 7b–c) is almost identical to that in *B. erebus* in that the byssal retractors (pbr2 and pbr1) are widely separated with pbr2 attached in the rear of the umbonal cavity. Both the anterior pedal retractor (apr) and posterior byssal retractor (pbr1) are slender. The posterior protractor muscle (ppr) in *B. geikotsucola* is very slender and simple and lacking the secondary muscles (sppr) seen in *B. erebus*.
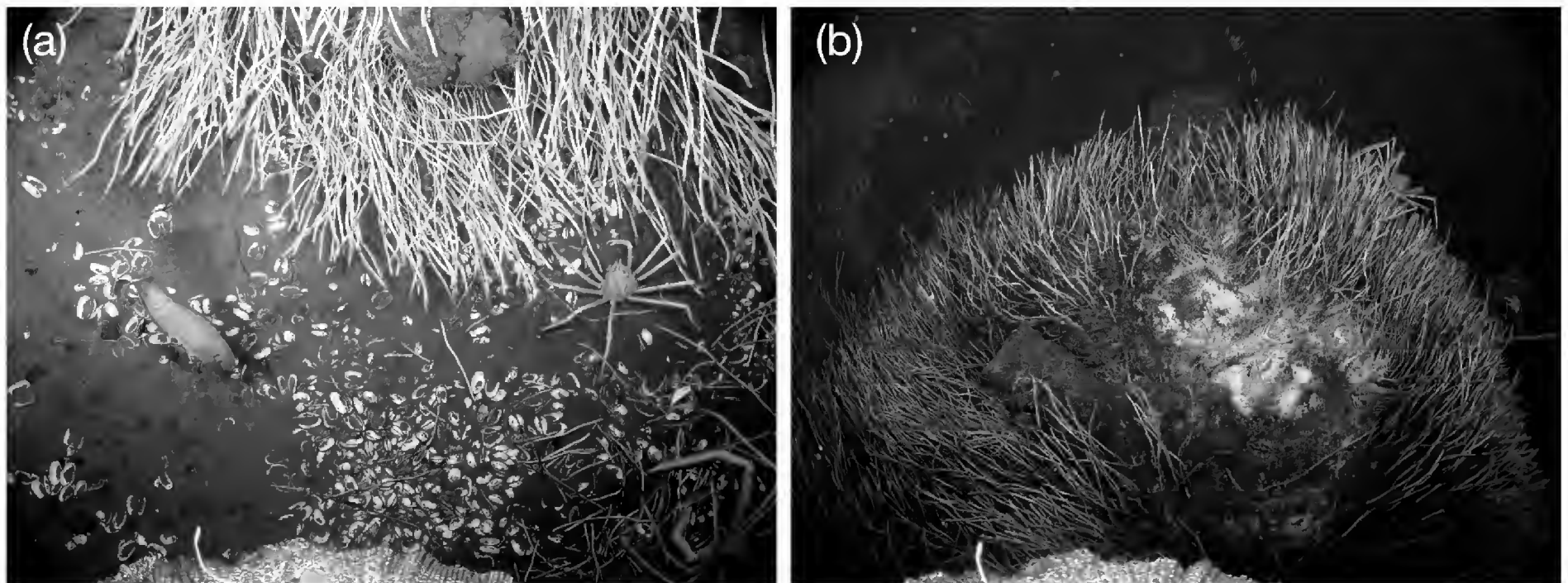
Figures 5. a–b Images of the benthic habitat at the Endeavour segment and Middle Valley. **a** showing a clump of *Ridgeia* and many dead vesicomyid clams, **b** a clump of *Ridgeia* around a sulphide block
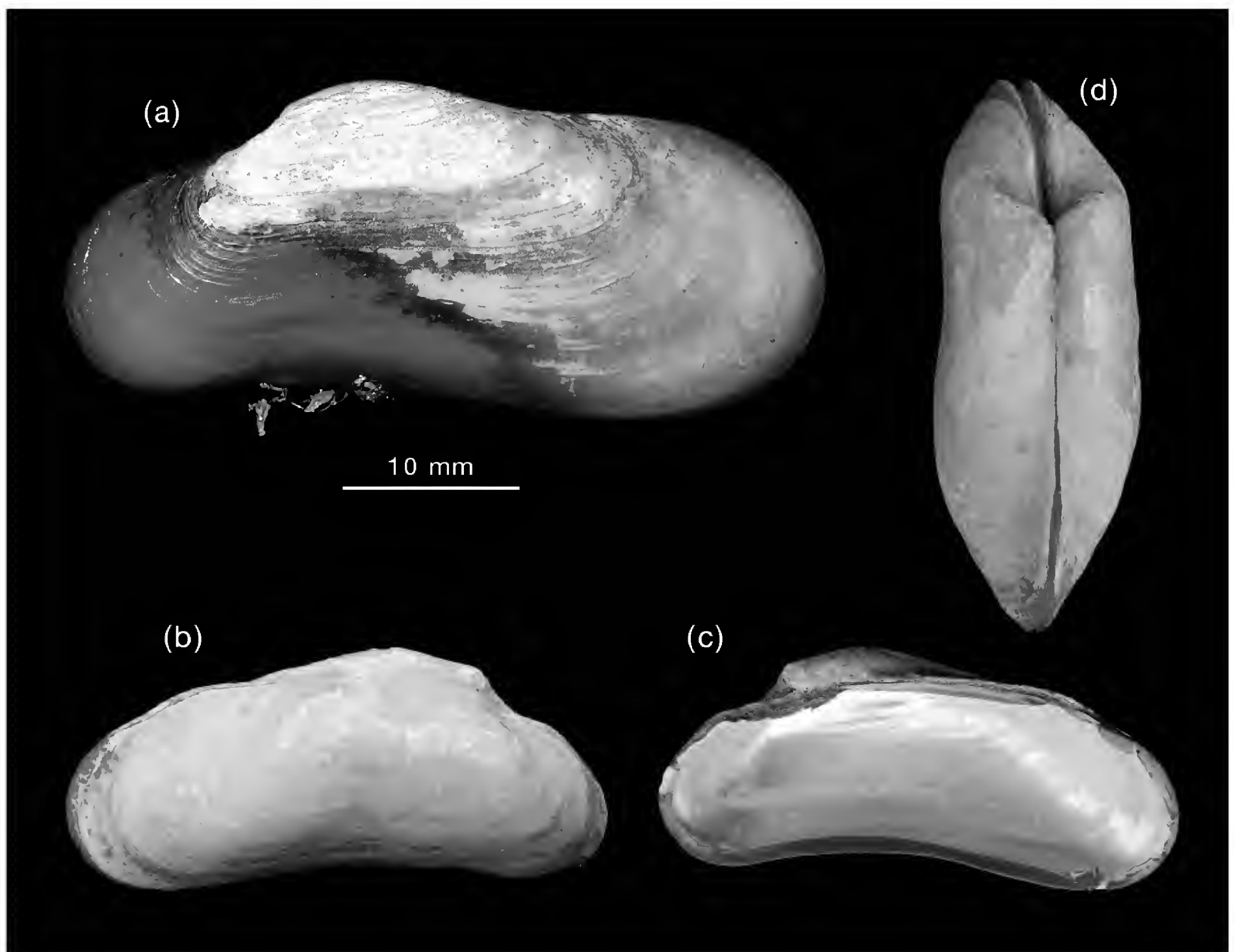
Figures 6. a–d Shells of *Benthomodiolus geikotsucola* Okutani and Miyazaki, 2007. **a** exterior of left valve of holotype (NSMT-Mo-7670349), **b–d** of the dissected paratype (NSMT Mo-76704j).

Ctenidium and labial palps. The ctenidium consists of both demibranchs and as in *B. erebus* the filaments are relatively short (Fig. 7a, 4f). The ascending and descending arms of the filaments are fused for over half their lengths and the abfrontal surface is extending cre-ating a triangular laminar form to each filament (Fig. 4g). The filaments have a single row of ciliary junctions on the ascending and descending arms (Fig. 4g–h). The abfrontal surface is composed of polygonal cushions of microvilli.
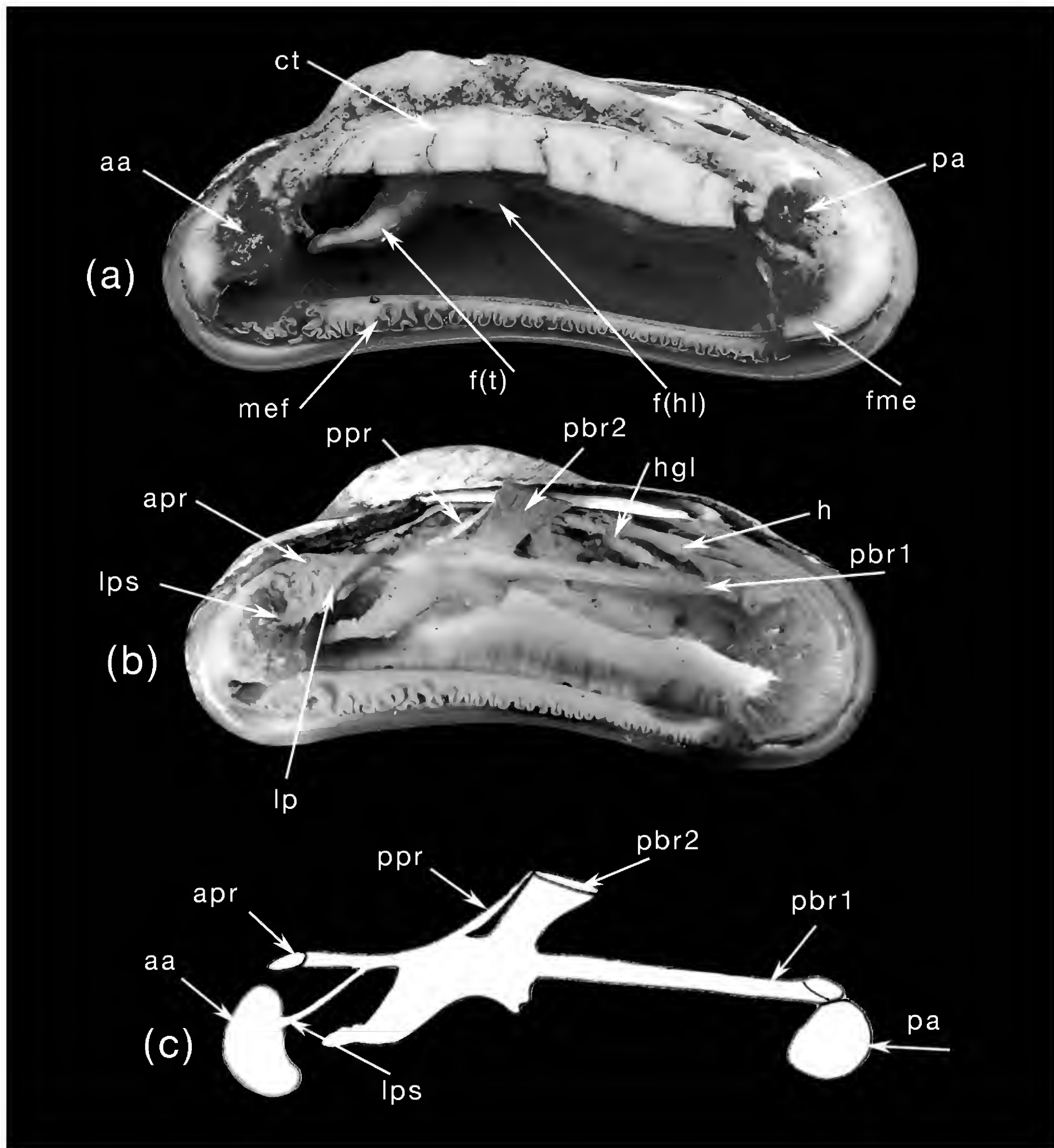
Figure 7. Gross anatomy of *Benthomodiolus geikotsucola* stained with methylene green. **a** after removal of left valve and mantle, **b** after further removal of the ctenidium, **c** diagram of the adductor, pedal and byssal musculature.

Mantle edge and apertures. The mantle edge is free for most of its length and fused only posteriorly to separate the ventral gape for the exhalant aperture. The entire length of the ventral gape the middle fold is thrown into a dense series of folds (Fig. 7a, mef) and in this unlike the almost lack of folding seen in *B. erebus*.

Alimentary system. The stomach was not dissected. The gut follows a similar path to that in *B. erebus* but the hindgut loop is distinct with a short reversed portion (Fig. 7b, hgl).

Benthomodiolus lignocola Dell, 1987

Benthomodiolus lignocola Dell, 1987: 33–34, figs. 44, 45, 48, 49, 52, and 53.

Material examined. *Holotype.* 1 shell, NW of Orete Point, White Island, 37°23.7'S 177°39.5'E, 1075–1100m, 23/Nov/1981; Museum of New Zealand Te Papa Tongarewa M.075023. Not examined, image courtesy of NMNZ.

Paratype. 5 specimens, NE of Chatham Islands, 42°47.10' 175°45.60'W, 1174–1180m, 22/Aug/1984; NMNZ M–075248/1.
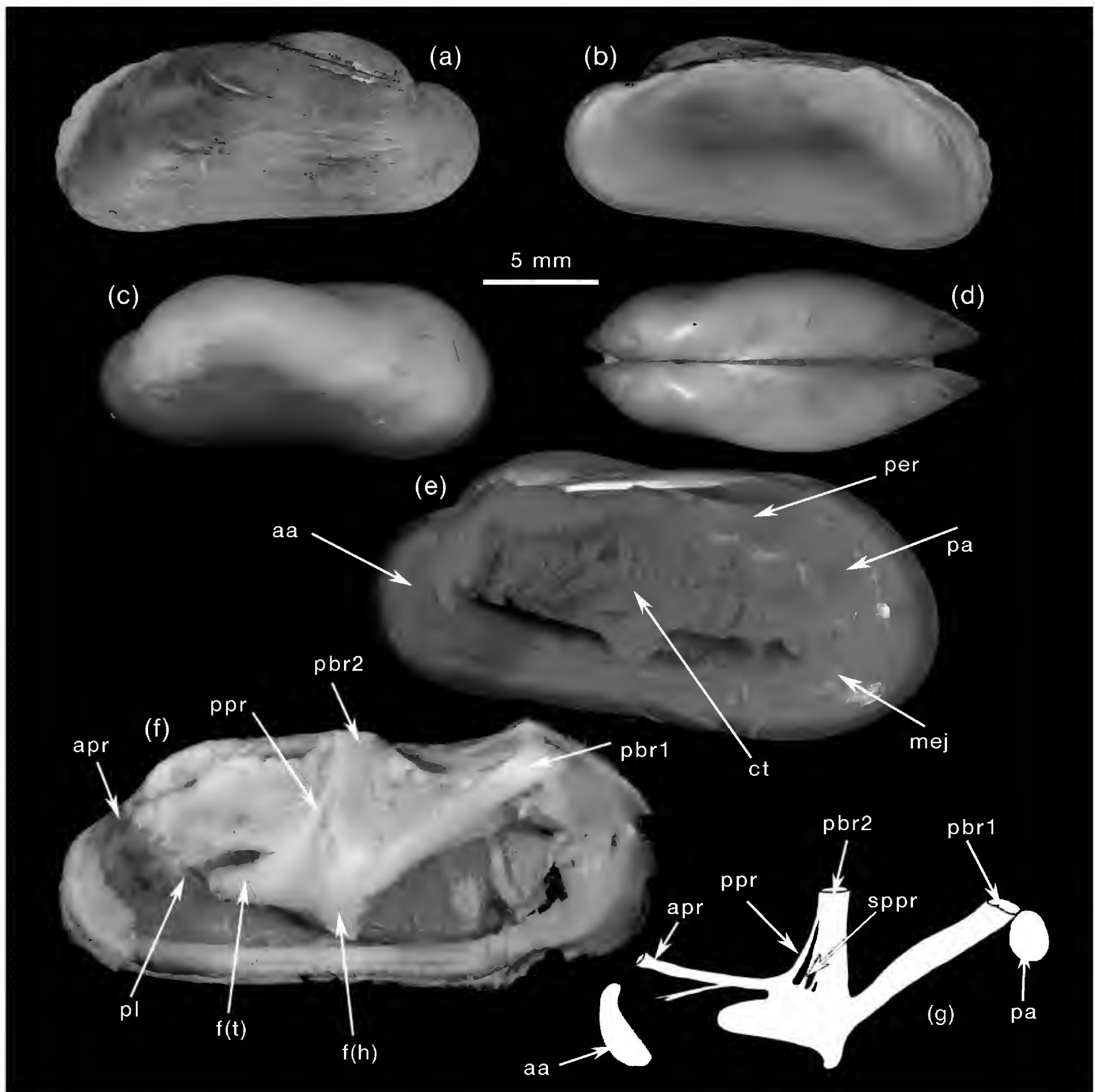
Figure 8. Shells and anatomy of *Benthomodiolus lignocola* Dell, 1987. **a** exterior of the right valve of the holotype (MNNZ M.075023), b-d the shell of the dissected paratype MNNZ M-075248/1, **e–g** gross anatomy stained with methylene green. **e** after removal of left valve and mantle, **f** after further removal of the ctenidium, **g** diagram of the adductor, pedal and byssal musculature.

Shell. The shells have a maximum length of 18.1 mm (Holotype, Fig. 8a–b) and the dissected specimen was 17.4 mm in length (Fig. 8c–d). The shell is umbonate, weakly arcuate with the posterior distinctly deeper than anterior in outline and medially sulcate. The beaks are rather distant from the anterior margin with a total length /anterior length of 0.23. The sculpture is weak of commarginal lines only except over the sulcate zone where faint fine irregular radial creases are present. Fine periostracal hairs are present but worn off in the larger specimens.

Pedal byssus musculature. The byssal retractors (Fig. 8f, g pbr2 and pbr1) are widely separated with pbr2 at-tached in the rear of the umbonal cavity. The angle between the byssal retractors is acute. The anterior pedal retractor (apr) is more slender than the posterior byssal retractor (pbr1). The posterior protractor muscle (ppr) is slender with secondary muscles (sppr).

Ctenidium and labial palps. The ctenidium consists of both demibranchs with relatively short strap-like filaments and has a flimsy appearance with weak inter-filamentar junctions (Fig. 10a). The ascending and descending arms of the filaments are fused for only a short distance (Fig. 10b, d). The filaments have a single row of ciliary junctions on the ascending and descending arms

(Fig. 10c). The frontal cilia are present but the abfrontal surfaces are largely devoid of epithelium, probably a consequence of poor fixation. Where present a weak polygonal pattern can be seen (Fig. 10e).

Mantle edge and apertures. The mantle edge (Fig. 8e) is free for most of its length and fused only posteriorly to separate the ventral gape for the exhalant aperture. The mantle edge along the ventral gape is smooth.

Alimentary system. The stomach was not dissected. The gut follows a similar path to that in *B. erebus* but the hind gut loop is distinct with a short reversed portion.

Benthomodiolus abyssicola (Knudsen, 1970)

Modiolus abyssicola Knudsen, 1970: 92–94, text figs 55–57, pl. 14, fig. 6.
Benthomodiolus abyssicola (Knudsen) — Dell 1987: 31-33.
Benthomodiolus abyssicola (Knudsen) — Coan et al. 2000: 162, pl. 24.
Benthomodiolus abyssicola (Knudsen) — Coan and Valentich-Scott 2012, 123, pl. 39.

Material examined. *Holotype.* Gulf of Panama, E. Pacific, 05°49'N 78°52'W, 3670-3270m. ZMUC, not examined.
  Paratype. From type locality, 10 specimens examined, ZMUC-BIV-30.

*Illustration.*Paratypes were dissected by Vita Kenk and her drawing was published in Coan and Valentich-Scott (2012). A copy of the original drawing was sent to me by Paul Valentich-Scott and is reproduced here (Fig. 9f).

Shell. The shells have a maximum length of 17.2 mm and the dissected specimen was 17.1 mm in length (Fig.9a–b). The shell is weakly umbonate, wedge shaped with the posterior distinctly deeper than anterior in outline and medially narrowed but weakly sulcate. The beaks are close the anterior margin with a total length /anterior length of 0.22. The sculpture is weak of commarginal lines only. Fine periostracal hairs are present (Fig. 9c) but worn off in the larger specimens.

Pedal byssus musculature. The byssal retractors (pbr2 and pbr1) are widely separated with pbr2 attached immediately behind the umbonal cavity (Fig. 9e–f). The angle between the byssal retractors is acute. The anterior pedal retractor (apr) is more slender than the posterior byssal retractor (pbr1). The posterior protractor muscle (ppr) is slender and incorporated into the anterior byssal retractor (Fig. 9e–f).

Ctenidium and labial palps. The ctenidium consists of both demibranchs with relatively short strap-like filaments and has a flimsy appearance with weak inter-filamentar junctions (Figs 9d, 10f). The ascending and descending arms of the filaments are fused for only a short distance (Fig. 10f). The filaments have a single row of ciliary junctions on the ascending and descending arms (Fig. 10g). The frontal cilia are present but the abfrontal surfaces are largely devoid of epithelium, probably a con-

sequence of poor fixation. Where present a weak polygonal pattern can be seen (Fig. 10h).

Mantle edge and apertures. The mantle edge is free along its entire length except for a small junction with the gill axis of the ctenidium to form a small exhalant aperture (Fig. 9d, f). The mantle edge bears weak folds posteriorly but smooth ventrally and anteriorly.

Alimentary system. The gut follows a simple route with the oesophagus, stomach and mid gut in a line running across the dorsal surface of the visceral mass (Fig. 9f). The hindgut descends into the visceral mass and returns to form a distinct loop (Fig. 9f, hgl) before passing through the heart.

Discussion

Benthomodiolus erebus is regarded as a distinct species but does bear close resemblance to *B. geikotsucola,* which differs in having the shell more extended anteriorly, having a distinct hind-gut loop and strongly folded mantle edge. The molecular data also confirm *B. erebus* and *B. geikotsucola* to be sister taxa (Lorion et al. 2013).

The molecular data link *B. erebus, B. geikotsucola* and *B. lignocola* but there are some distinct morphological difference that sets the latter apart from the former two. *Benthomodiolus lignocola* is much smaller; the shell is not quite so umbonate and bears periostracal hairs. The byssal retractor muscles although widely separated are less so than in the other species and reflects the more expanded posterior of the shell.

The ctenidial filaments are linear, strap-like, and unlike the triangular laminar forms seen in *B. erebus* and *B. geikotsucola.* In this, the ctendium of *B. erebus* and *B. geikotsucola* resembles that of *Bathymodiolus* and is highly adapted to host symbiotic bacteria. Although laminar the symbionts are extracellular unlike the intracellular condition of many species of *Bathymodiolus* and *Gigantidas* (Lorion et al. 2013)

Kenk and Wilson (1985) suggested that the ctenidium of *M. abyssicola* was unlike that of *Bathymodiolus* and thus retained *abyssicola* in *Modiolus.* Here the difference in filament form is confirmed but the polygonal epithelium of the abfrontal surface does suggest that *B. abyssicola* is chemosymbiotic. The ctenidial structures of *B. lignocola* and *B. abyssicola* are very similar and both are known from wood-falls. The pedal/byssus musculature is also similar and this suggests that they are indeed congeneric as listed by Bouchet in WoRMS (2015).

Morphologically one might propose *B. erebus* and *B. geikotsucola* to be in a separate genus from *B. lignocola* and *B. abyssicola,* based on the shell shape, lack of periostracal hairs and laminar gill filaments. However, there is currently no molecular support for any generic distinctions within the *Benthomodiolus* clade and there is as yet no molecular data for *B. abyssicola.*
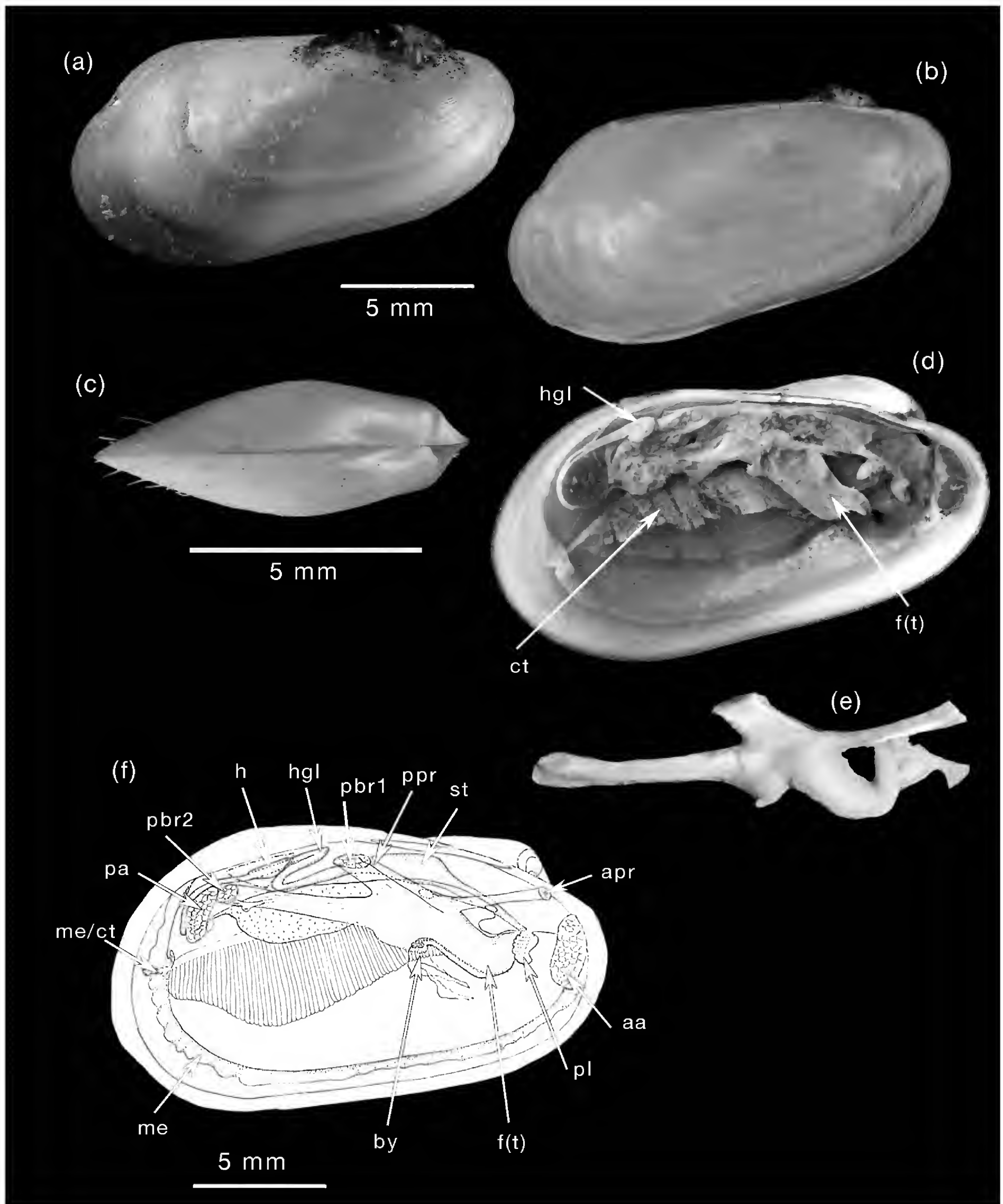
Figure 9. Shells and anatomy of paratypes *Benthomodiolus abyssicola* Knudsen, 1970 (ZMUC-BIV-30). **a** exterior of the right valve of the dissected specimen, **b** interior of left valve, **c** dorsal view of a small specimen with well preserved periostracal bristles, **d** gross anatomy, stained with methylene green after removal of right valve, mantle and ctenidium, **e** pedal and byssal musculature, **f** diagram of the gross anatomy of a paratype after Kenk in Coan and Valentich-Scott 2012.

The morphological basis to the generic systematics of the Bathymodiolinae has been shown by Thubaut et al. (in press) to be unreliable but the number of detailed morphological studies is few. The recent paper on *Idas argenteus* (Rodrigues et al. 2015) shows that quite different feeding strategies can be adopted by sister taxa, *I. argenteus* lacking symbionts and its sister taxon *I. washingtonia* being chemosymbiotic (Southward 2008). With
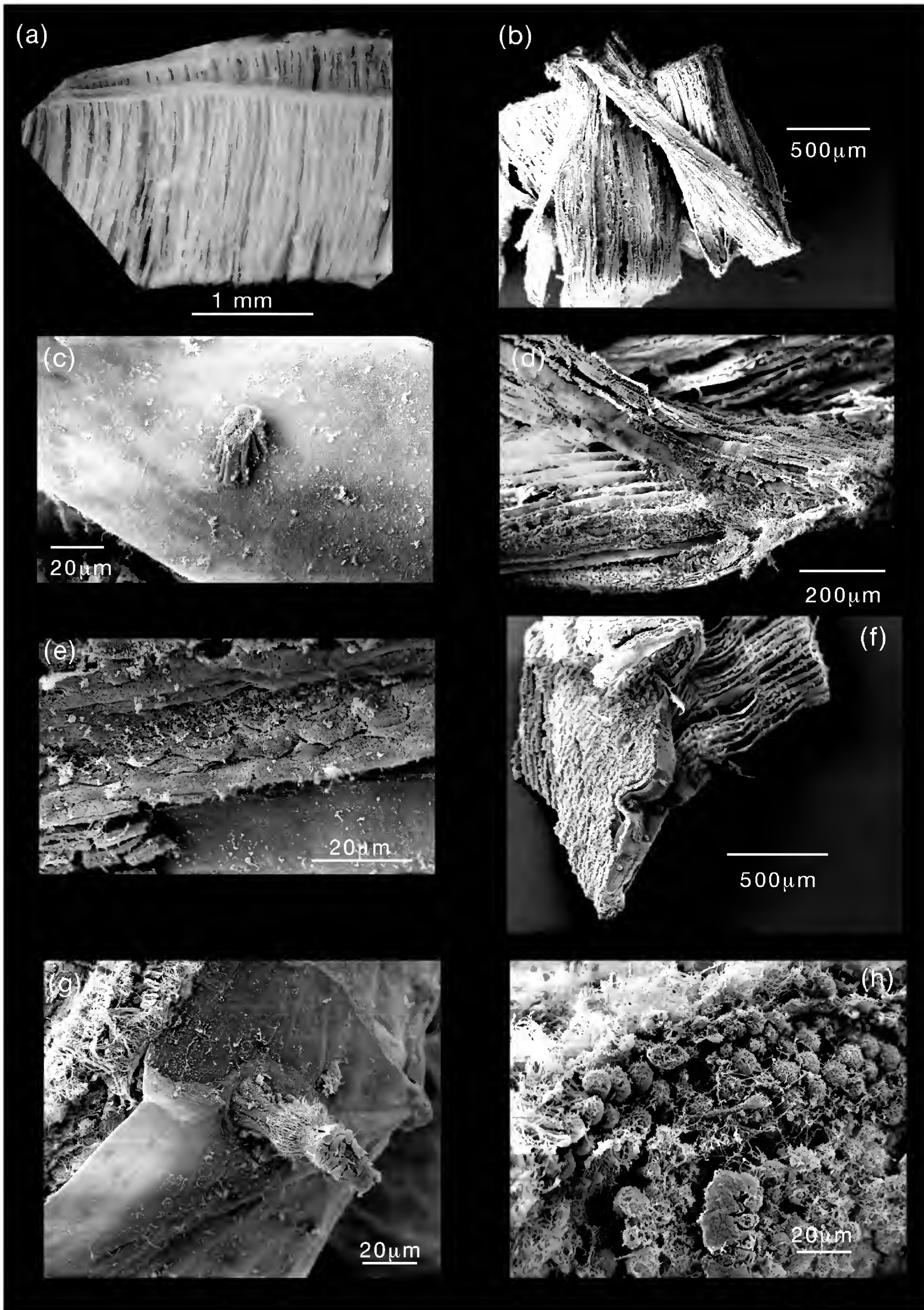
Figures 10. a–c The ctenidium of *Benthomodiolus lignocola* **a** gross anatomy, **b** SEM of whole filaments, **c** SEM of inter filamentar junction, **d** SEM of tip of filament, **e** SEM of polygonal surface of microvilli (Knudsen 1970). **Figures 4f–h.** The ctenidium of *B. geikotsucola*. **f** SEM of a whole filaments gross anatomy, **g** SEM of inter filamentar junction, **h** SEM of the polygonal surface.

so few morphological studies and the known variance in feeding strategies, no new generic taxa are proposed here. From morphology there is no single affinity with other bathymodioline genera although the pedal byssus musculature and largely unfused mantle margins are most similar to that seen in *Bathymodiolus*.

The shell form *B. erebus* and *B. geikotsucola* is rather unusual with the narrow arcuate outline and relatively long anterior portion, as such they can scarcely be described as modioliform where the outline is more wedge shaped. Although scarce this shell form occurs in other genera notably *Terua* but also in some species of *Bathymodiolus* and *Gigantidas*. The smaller *B. lignocola* and *B. abyssicola* are more modioliform and resemble *Idas*. Shell shape has no consistency within *Benthomodiolus* a situation mirrored in the other clades such *Bathymodiolus* and *Gigantidas* (Thubaut et al. in press).

The *Benthomodiolus* clade is considered to be the sister to the entire remainder of the bathymodiolines and occupies a basal position in all published phylogenetic trees (Thubaut et al. in press, Rodrigues et al. 2015, Lorion et al. 2013, Kyuno et al. 2009). Despite an apparent early origin in the late Cretaceous the origins of the known species are more recent in the Miocene (Lorion et al. 2013). By this time the clade had radiated into the three major ecotypes of wood-fall, whale-fall and vent suggesting a parallel radiation to the remainder of the bathymodiolines but lacking any species associated with cold seeps. The majority of species inhabiting vents and seeps have intra-cellular symbionts and belong to the *Bathymodiolus* and *Gigantidas* clades, the exceptions are *Benthomodiolus erebus* (Southward 2008) and *Vulcanidas insolitas* (Cosel and Marshall 2010) both having rather basal positions in the phylogeny but illustrating the widespread capacity for any clade to inhabit a wide range of chemosynthetic settings. All the morphological data suggest that the affinity of *Benthomodiolus* is with the Bathymodiolinae and not with the Modiolinae as indicated by Kyuno et al. 2009. All *Benthomodiolus* are chemosymbiotic, some with more highly adapted ctenidia than others, but there is no indication that they are in any way more morphologically primitive than other Bathymodiolinae, except perhaps in having extra cellular symbionts. Here again however this condition is found throughout the various clades (eg. *Idas*, see Rodrigues et al. 2015) and is perhaps more related to feeding ecology than phylogeny.

Acknowledgements

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A new species of *Parategastes* Sars, 1904 from the Thale Noi Lake, southern Thailand (Copepoda, Harpacticoida, Tegastidae)

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Abstract

Parategastes pholpunthini **sp. n.** is described and illustrated based on material collected in the Thale Noi Lake, Phatthalung province, southern Thailand. This species can be distinguished from its congeners by the number segments of female antennule, the lengths of rami and basis of P1, the shape of middle inner seta of P4 exp-3, shape of P5, and relative lengths of spine at apically of baseoendopod of P5. The differences among *Parategastes* species are pointed out and they are compared with the new species. An identification key to species of the genus *Parategastes* are proposed.

Key Words

Introduction

The family Tegastidae Sars, 1904 is characterised as being laterally compressed, amphipod-like, strongly chitinous and well sculptured (Huys et al. 1996). To date, this family comprises approximately 60 species in six genera. Each genus can be easily distinguished by the number of segments in P2-P4 and the length of caudal ramus (Gollner et al. 2008). This family is widely distributed, and has been recorded in areas such as the Andaman Islands, India (Fiers 1986); southwestern Australia (Bartsch 1995); the Mid-Atlantic Ridge (Ivanenko and Defaye 2004); Spitsbergen in the Arctic Ocean (Ferrari et al. 2007); a hydrothermal vent site on the East Pacific Rise (Gollner et al. 2008); the Gulf of Mexico (Plum and Arbizu 2009); and a hydrothermal vent in the Okinawa Trough, Japan (Back et al. 2010). One of the six genera, *Parategastes*, was created by Sars (1904) who proposed this genus base on the number segments of female antennule (6-7 segments), maxilliped and shape of P5. In addition, *Parategastes sphaericus* was proposed

as a type species of the genus (Sar 1904). Later, Huys et al. (1996) proposed the number of segments in P2-P4 to distinguish genus *Parategastes* from other genus in family Tegastidae. Currently, this genus has so far accommodated six valid species and one species incertae sedis (*Parategastes haphe*) (Wells 2007). Only one species, *P. sphaericus* (Claus, 1863) had been found in many localities such as from the Mediterranean, the North American Atlantic coast, European Atlantic coast (Lang 1948) Naples, Italy (Claus 1863), Chilka Lake, India (Sewell 1924) and Tunis, Tunisia (Monard 1935) whereas other species had been recorded in one locality, *P. chalmersi* (Thompson & Scott, 1903) from Ceylon, Sri Lanka (Thomson and Scott 1903); *P. haphe* Leigh-Sharpe, 1936 was described from Naples (Leigh-Sharpe 1936); *P. herteli* Jakobi, 1953 from Santa Catarina, Brazil (Jakobi 1953); *P. caprinus* Wellershaus, 1970 from Cochin backwater, South India (Wellershaus 1970); *P. coetzeei* Kunz, 1980 from the Wilderness Lakes, South Africa (Kunz 1980); and *P. conexus* Humes, 1984 from Moluccas, Indonesia (Humes 1984). However, this

is the first time record of genus *Parategastes* in Thailand. Thus, this paper will provide detailed description and illustrations of both female and male of the new species collected from Thale Noi Lake, southern Thailand and propose a key to species in genus *Parategastes*.

Material and methods

Samples were collected from Thale Noi Lake, Phatthalung province, southern Thailand, using a 60 μm mesh sized plankton net, every two months from February 2013 to February 2014. Samples were immediately preserved in 70% ethanol. Then specimens were sorted using an Olympus SZ-40 stereo microscope and each specimen was dissected and mounted on a slide in glycerine, and then sealed using nail varnish. The morphological characters were examined using an Olympus CH-2 compound microscope, and drawings were made of both complete and dissected specimens using a *camera lucida* connected to the Olympus CH-2 compound microscope. Descriptive terminology proposed by Huys et al. (1996) was adopted; abbreviations used in the text are: **A1** antennule; **A2** antenna; **P1-P5** swimming legs 1-5; **enp-1** (2, 3), proximal (middle, distal) segment of endopod; and **exp-1** (2, 3), proximal (middle, distal) segment of exopod. Holotypes and paratypes were deposited in the reference collection of the Princess Maha Chakri Sirindhorn National History Museum, Prince of Songkla University, Songkhla, Thailand.

Taxonomy

Order Harpacticoida Sars, 1903
Family Tegastidae Sars, 1904

Genus *Parategastes* Sars, 1904

P. sphaericus (Claus, 1863)
P. chalmersi (Thompson & Scott, 1903)
P. caprinus Wellershaus, 1970
P. coetzeei Kunz, 1980
P. conexus Humes, 1984
P. herteli Jakobi, 1953
P. pholpunthini sp. n.
P. haphe Leigh-Sharpe, 1936 (*incertae sedis*)

Type species. *Parategastes sphaericus* (Claus, 1863).

Generic diagnosis. *Parategastes* Sars, 1904. antennules with 6-7 segments in female and 8 segments in males; exopod of antenna with one segment and with two or three setae; anterior maxilliped with the 2 proximal lateral lobes replaced by simple setae, outermost lobe less broad and provided with only two setae at the tip, terminal joint produced at the tip to a long digitiform process. P2 and P3 with 2-segmented endopod and 3-segmented exopod; and P4 with 3-segmented endopod and exopod, whilst enp-1 was not swollen. P5 in female, baseoendopod very large, with inner expansion to broad and vaulted, exopod somewhat dilated towards the end, with a single short apical seta.

Parategastes pholpunthini sp. n.

http://zoobank.org/1260D5FC-FA5E-4375-9E3F-E08463CAB275

Type material. Holotype: adult female, dissected and mounted on 4 slides, (PSUZC-PK2001-01-PSUZC-PK2001-04); Paratype 1: undissected female, mounted on 1 slide, (PSUZC-PK2001-05); Paratype 2: undissected adult male, mounted on 1 slide, (PSUZC-PK2001-06); Paratype 3: adult female, dissected on 4 slides, (PSUZC-PK2001-07-PSUZC-PK2001-10); Paratype 4: adult male, dissected on 4 slides, (PSUZC-PK2001-11-PSUZC-PK2001-14). All specimens were collected from the type locality on 23 October 2013.

Additional materials. 8 females and 8 males from the type locality are stored in 70% ethanol.

Type locality. Klong Ban Klang canal, Thale Noi Lake, Phatthalung province, southern Thailand (07°46'30.47"N, 100°9'31.68"E). The canal is connected to Songkhla Lake. Water temperature ranged between 28.3 to 28.5 °C, pH of 5.71–6.07, salinity 1.1 ppt, depths between 0.8 m to 1.35 m, transparency of 0.2–1.35 m, and dissolved oxygen levels of 3.06–4.24 mgO$_2$/L. This area was covered with aquatic plants, such as *Neptunia oleracea* Lour., *Eichhornia crassipes* (C. Mart.) Solms, and *Nympheas* sp.

Description of the adult female. Body laterally compressed with sensilla, surface of whole body pitted (Fig. 1A). Total length, measured from the anterior margin of the cephalic shield to the posterior margin of the caudal rami, 280–340 μm (mean = 310 μm, n = 11). Prosome comprising of cephalothorax and three somites bearing P2 to P4. Urosome 5-segmented (Fig. 1D), first urosomite with P5, genital double somite and three abdominal somites. Caudal rami (Fig. 1C) as wide as long, with seven setae of different lengths. All setae smooth.

Egg sac (Fig. 2G) round, containing four eggs, located ventrally between fifth pair of legs.

Rostrum (Fig. 1B) a rounded prominence.

Antennule (Fig. 2A) 6-segmented, aesthetasc on 4th and 6th segments. Armature formula 1-[1], 2-[8], 3-[8], 4-[3+aesthetasc], 5-[5], 6-[10+acrothek]. Aesthetasc on 4th segment fused basally to one smooth seta. Apical acrothek consists of an aesthetasc fused basally with two slender smooth setae. Only seta on first segment bipinnate, all other setae smooth.

Antenna (Fig. 2B) 4-segmented, comprising coxa, basis and 2-segmented endopod. Coxa and basis without ornamentation, exopod 1-segmented with three setae. Enp-1 with one median seta; enp-2 with eight setae (two laterals and six apical).

Mandible (Fig. 2C) gnathobase with four teeth. Coxa-basis with row of long spinules and with two setae; endopod 1-segment with three setae; exopod represented by one seta.
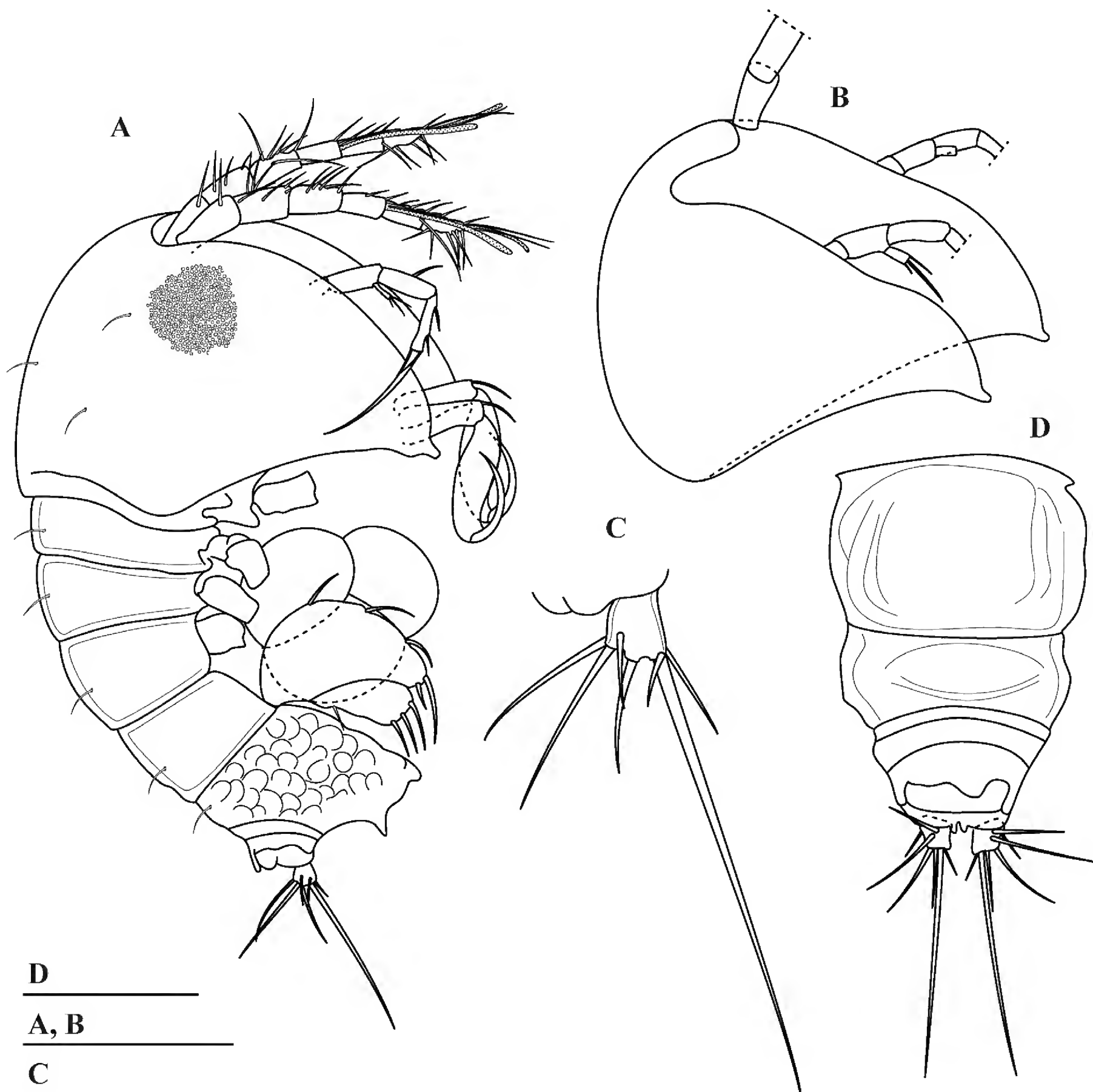
Figure 1. *Parategastes pholpunthini* sp. n., holotype female. **A** habitus, lateral view **B** rostrum **C** caudal ramus, lateral view **D** urosome. Scale bars: **A, B** = 100 μm; **C, D** = 50 μm.

Maxillule (Fig. 2D) precoxal arthrite bearing seven elements. Coxal endite with one seta. Endopod elongated with a row of spinules, five terminal setae, and two setae at outer margin.

Maxilla (Fig. 2E) syncoxa with transverse row of spinules proximo-laterally and with two endites. Precoxal endite with four setae, one seta elongated; coxal endite with three setae. Allobasis with three setae, middle seta forming a strong claw, and three setae at outer margin representing endopod.

Maxilliped (Fig. 2F) subchelate, 2-segmented, comprising syncoxa and basis. Syncoxa with one pinnate seta at the inner distal corner. Basis with row of spinules, one pad-like process ornamented with short spinules on its surface. Endopod 1-segmented, forming a strong claw with one seta proximally.

P1 (Fig. 3A) coxa with a row of setules along inner margin. Basis with a row of spinules along inner margin, one inner seta, and one outer seta. Both rami with 1-segmented endopod wider than exopod. Endopod with a row of setules along outer margin; with one inner proximal biplumose seta, one inner middle modified spine, one inner distal bipinnate spine, two apical bipinnate spines, and one outer bipinnate spine. Exopod with a row of spinules along inner margin; with two apical bipinnate spines, three outer setae, the middle of which being bare, and the others bipinnate.

P2 and P3 (Figs 3B, 4A) coxa with a set of spinules on pronounced disto-lateral corner. Basis elongated with one small outer seta and row of spinules along medial margin. Rami with 3-segmented endopods and 2-segmented exopods; all segments of endopod with a row of setules
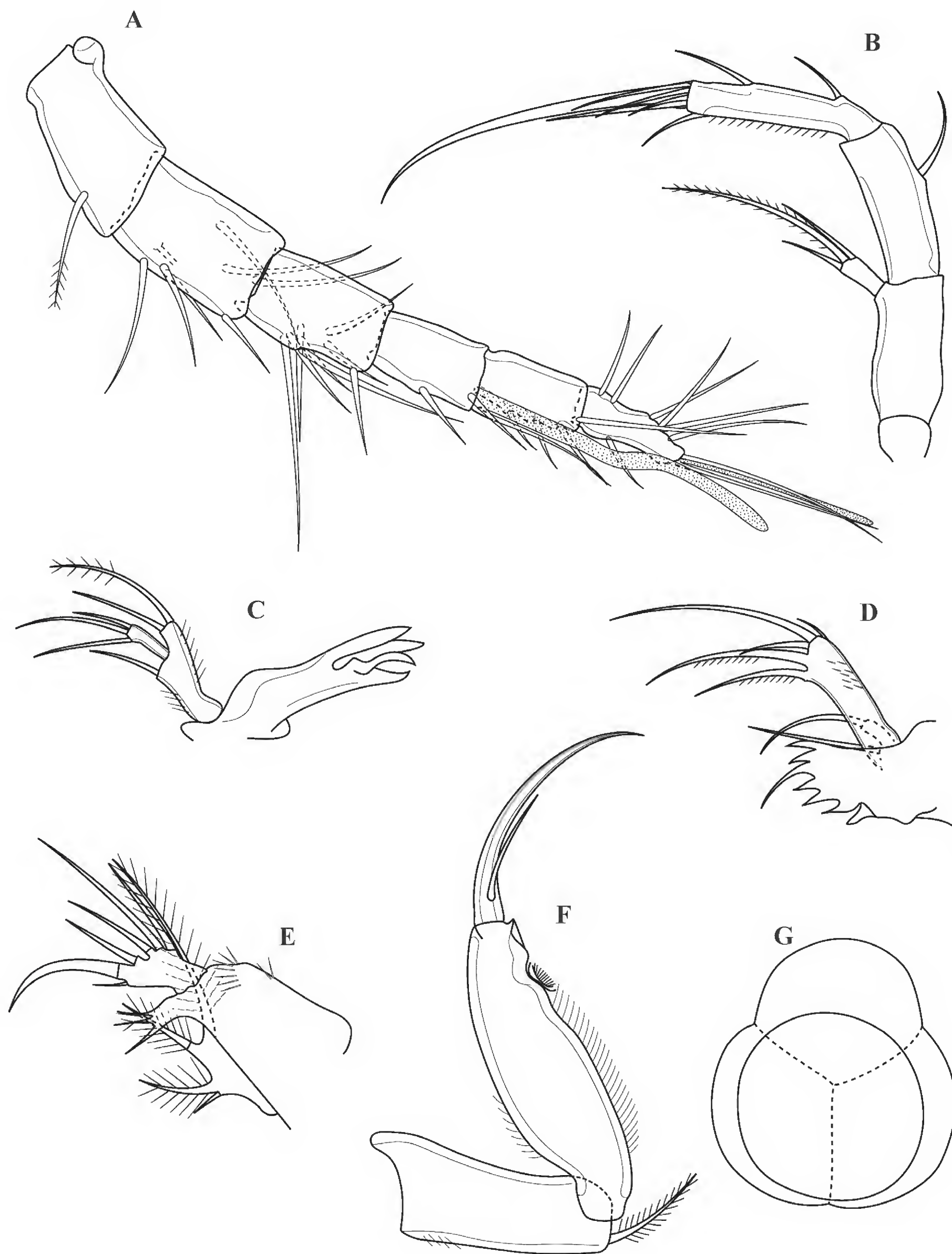
Figure 2. *Parategastes pholpunthini* sp. n., holotype female. **A** antennule **B** antenna **C** mandible **D** maxillule **E** maxilla **F** maxilliped **G** egg. Scale bars: **A–E** = 50 μm; **G** = 100 μm.

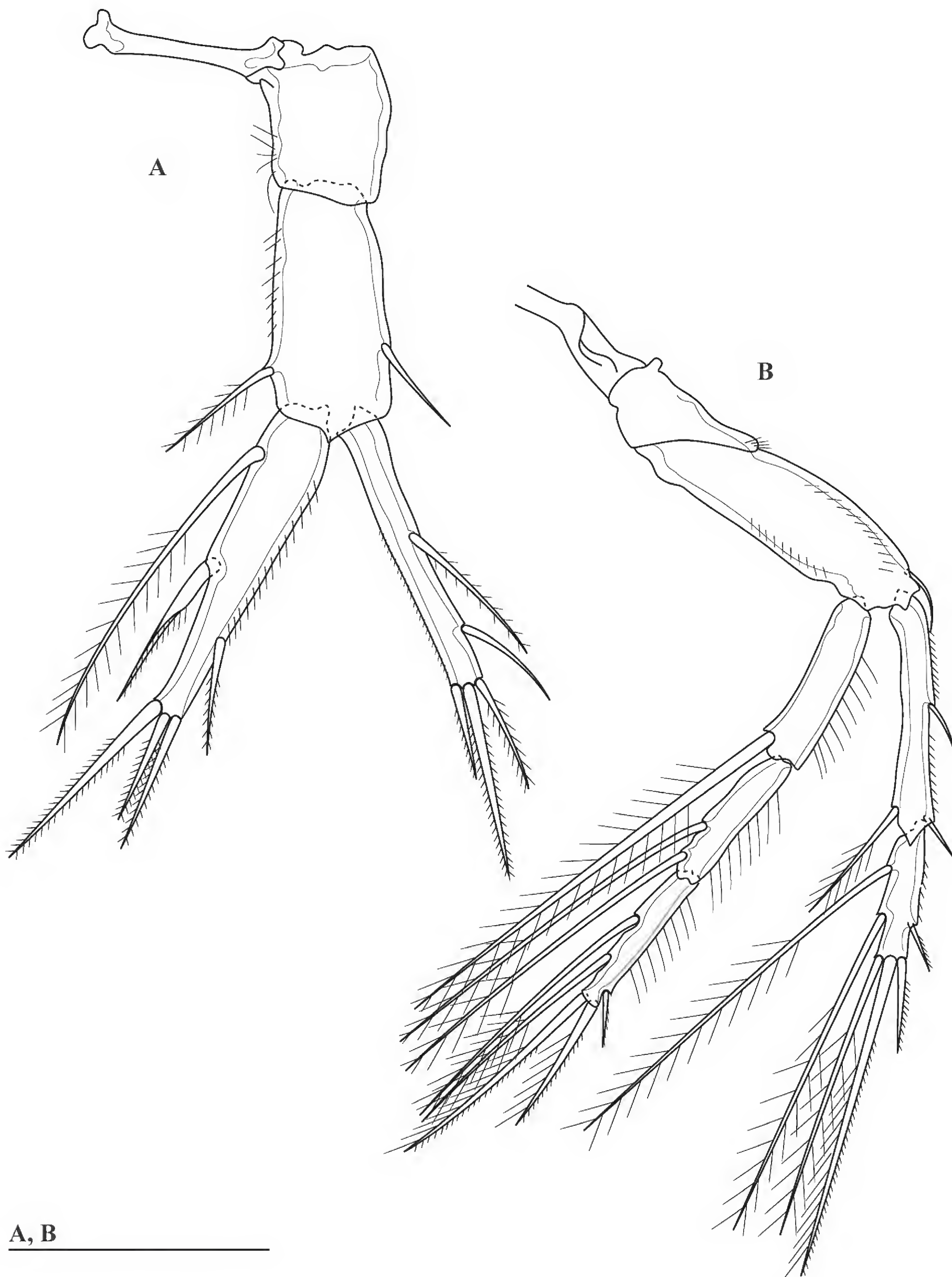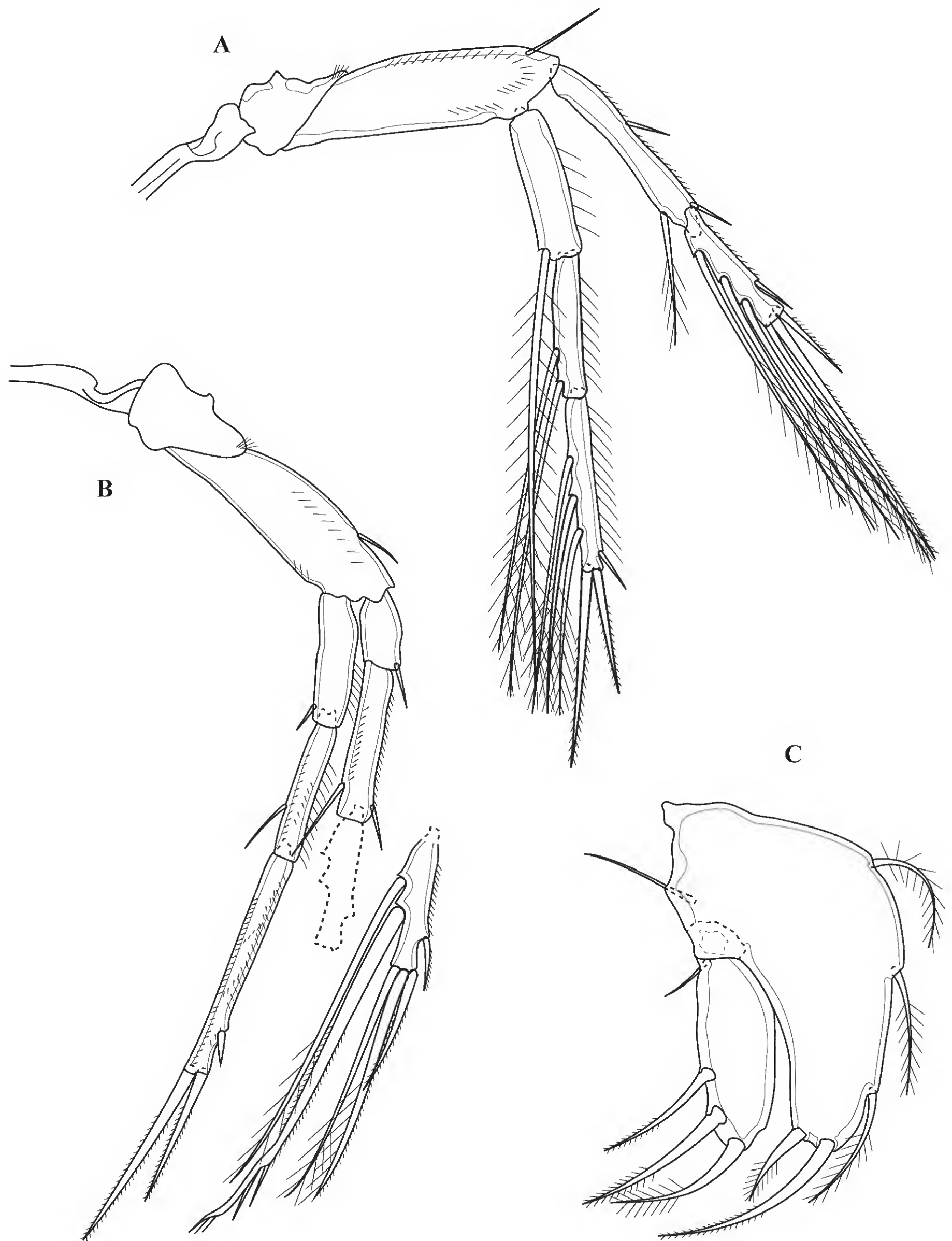**Figure 3.** *Parategastes pholpunthini* sp. n., holotype female. **A** P1 **B** P2. Scale bar: **A, B** = 50 μm.

Figure 4. *Parategastes pholpunthini* sp. n., holotype female. **A** P3 **B** P4 **C** P5. Scale bar: **A–C** = 50 μm.

Figure 5. *Parategastes pholpunthini* sp. n., paratype male. **A** habitus, lateral view **B** antennule **C** P5. Scale bars: **A** = 100 μm; **B, C** = 50 μm.

along outer margins, all segments of exopod with a row of spinules along outer margin.

P4 (Fig. 4B) coxa with a set of spinules on pronounced disto-lateral corner. Basis elongated with one small outer seta and a row of spinules on the inner and outer margins. Both rami 3-segmented. Enp-1 and enp-2 equal in length but shorter than enp-3; enp-1 and enp-2 with inner setae and with a row of setules along outer margins; enp-3 with two apical pinnate spines, and one outer seta. Exp-1 small, with one outer spine; exp-2 with one inner seta and one outer spine, and with a row of spinules on the inner and outer margins; exp-3 with a row of spinules on the outer margin, with one inner pinnate seta, middle inner seta enlarged, this enlarged seta has a peculiar shape with two long pinnae at the distal third, with the bending, and with three shorter pinnae distal end, two apical setae, one seta plumose, another seta inner margin plumose and outer margin pinnate, and two outer pinnate spines.

Armature formula of P1-P4 as in Table 1.

P5 (Fig. 4C) with baseoendopod and exopod; baseoendopod with three lateral biplumose setae, and one inner seta along proximolateral, apically with one inner bipinnate seta and one outer biplumose spine. Exopod with one apical biplumose seta and two outer bipinnate setae, one small seta basally.

Description of the adult male. Body laterally compressed (Fig. 5A). Total length, 290–310 μm (mean = 290 μm, n = 10). Spermatophore reservoir produced ventrally in a large, elongated prominence bearing distally asymmetrical genital flaps.

Antennule (Fig. 5B) 8-segmented, aesthetasc on 3rd, 4th and 8th segments. Armature formula 1-[1], 2-[9], 3-[7+aesthetasc], 4-[7+aesthetasc], 5-[1], 6-[2], 7-[1], 8-[10+acrothek]. Aesthetasc on 3rd and 4th segment fused basally to one naked seta. Apical acrothek consists of an aesthetasc fused basally with two slender naked setae. Only seta on first segment bipinnate, all others smooth.

Rostrum, antenna, mandible, maxillule, maxilla, maxilliped, P1-P4 (not shown) as in female.

P5 (Fig. 5C) 2-segmented with baseoendopod and exopod; baseoendopod small with long spinule at lateral margin; exopod elongate with one proximal outer seta, one subterminal outer spine and two apical spines.

Etymology. This species named after Dr. Pornsilp Pholpunthin, who has studied the freshwater copepods in Southern Thailand since last twenty years.

Table 1. Armature formula of P1-P4 of *Parategastes pholpunthini* sp. n.

Swimming legs	Coxa	Basis	Endopod	Exopod
P1	0-0	1-1	1, II, III	2, 1II, 0
P2	0-0	1-0	0-1; 0-2; I, II, 2	[I-0, I-1]; I, II1, 2
P3	0-0	1-0	0-1; 0-2; I, II, 3	[I-0, I-1]; I, II1, 3
P4	0-0	1-0	0-1; 0-1; I, II, 0	I-0; I-1; I, II1, 2

Note: roman numerals representing spines and arabic numerals representing setae

Discussion

Four *Parategastes* species have been recorded in the Oriental region (Sewell 1924, Thompson and Scott 1903, Humes 1984, Wellershaus 1970). They comprises of *Parategastes sphaericus*, *P. chalmersi*, *P. conexus* and *P. caprinus*. However, this is the first record of the genus *Parategastes* in Thailand.

Parategastes pholpunthini sp. n. is clearly distinguished from other *Parategastes* species in the following characters: (1) number of antennule segments of female, 7-segmented in most species of *Parategastes*, except *P. coetzeei* and *P. pholpunthini* sp. n. with 6-segmented; (2) P1, length of rami and basis in this genus can be separated into three groups: the first group, rami shorter than basis, comprises of *P. chalmersi* and *P. conexus*, the second group, rami approximately as long as basis, comprises of *P. herteli* and *P. sphaericus* and the third group, rami longer than basis, comprises of *P. caprinus*, *P. coetzeei* and *P. pholpunthini* sp. n. (Table 2). However, in the latter group, inner middle spine of P1 modified except *P. caprinus*; (3) P4, middle inner seta of exp-3 enlarged in various forms, in *P. sphaericus* and *P. caprinus*, this seta thickened and elongated with serrated margin at the middle to the distal end (see fig. 2 P4, Sewell 1924; plate XLIII, Sars 1903; fig. 54, Wellershaus 1970), *P. chalmersi*, thickened, inner and outer margins not serrated (plate IV, fig. 21, Thompson and Scott 1903), *P. conexus*, inner margin of this seta not serrated and outer seta with three slender teeth at distal end (see fig. 8f, Humes 1984), *P. herteli*, this seta enlarged (see fig.1, Jakobi 1953), *P. pholpunthini* sp. n., this seta has a peculiar shape with two long pinnae at the distal third, with the bending, and with three shorter pinnae distal end, and slender seta in *P. coetzeei* and (4) P5, baseoendopod, only *P. conexus* with sub-triangular shape, other species with sub-oval shape; all species in this genus, apically with inner seta as long as outer spine, except *P. pholpunthini* sp. n. length of inner seta twice of outer spine, and *P. conexus* length of inner seta twice of outer seta.

From the comparisons, it was found that *Parategastes pholpunthini* sp. n. resembles *P. coetzeei*, but differs from the latter in the following characters: (1) P1, proximal inner seta of endopod 1.08 times as long as the endopod in *P. pholpunthini* sp. n. (Fig. 3A) yet equal in length to the endopod in *P. coetzeei* (see Fig. 9, Kunz 1980), (2) P4 of *P. pholpunthini* sp. n. has short setae at the inner edge of enp-1 and outer edge of exp-1 (Fig. 4B); in *P. coetzeei* is without seta at the inner seta of enp-1 and outer seta of exp-1 (see Fig. 13, Kunz 1980), (3) *P. pholpunthini* sp. n. has one seta of enp-2 of P4 (Fig. 4B); in *P. coetzeei* it has two setae (see Fig. 13, Kunz 1980), (4) P5, length of the inner apical seta of baseoendopod is twice of outer apical spine in *P. pholpunthini* sp. n. (Fig. 4C), and length of the medial apical seta is as long as the lateral apical seta *P. coetzeei* (see Fig. 15, Kunz 1980), (5) the exopod of P5 of *P. coetzeei*

Table 2. Comparison of characters of female of *Parategastes* species (modified from Wells 2007).

Species/ characters	A1	Exp of A2 (setae)	P1				P2-P4			P4 exp-3		P5		
			rami/ basis	setae		inner middle spine of enp	inner setae		enp-3	middle inner seta	outer setae	baseoendopod		
				enp	exp		enp-1	enp-2				shape	inner/outer spine (seta)	
P. sphaericus	7	2	m	6	5	slender	1:1:1	2:2:1	5:6:3	thickened , with serrated	2	sub-oval	A	
P. chalmersi	7	-	s	5	4	slender	1:1:1	2:2:2	5:6:6	thickened, without serrated	3:wd: B	sub-oval	A	
P. caprinus	7	3	l	6	5	slender	1:1:1	2:2:2	5:6:3	thickened, with serrated	3:rud	sub-oval	A	
P. conexus	7	3	s	6	5	slender	1:1:1	2:2:2	5:6:5	enlarged, 3 teeth at the distal end	3:wd: A	sub-triangular	A	
P. herteli	7	3	m	6	5	slender	1:1:1	2:2:2	5:6:3	enlarged	2	sub-oval	A	
P. coetzeei	6	3	l	6	5	thickened	1:1:0	2:2:2	5:7:3	slender	2	sub-oval	B*	
P. pholpunthini sp. n.	6	3	l	6	5	thickened	1:1:1	2:2:1	5:6:3	enlarged, bending, pinnae at the distal third and distal end	2	sub-oval	B**	

Note: not include *P. haphe*. - = no data; P1, lengths of rami relative to lengths of basis; l = exopod or endopod longer than basis; m = exopod or endopod approximately as long as basis; s = exopod or endopod shorter than basis; P4 exp-3, number and type of setae on inner edge; 3:wd:A = 3 setae, distal seta well developed, about as long as both proximal setae; 3:wd:B = 3 setae, distal seta well developed, about as long as the proximal seta but much shorter than the middle seta; 3:rud = 3 setae, distal seta very thin and weak, barely reaching to the end of the segment; 2 = setation of inner edge apparently represented by only the two very long proximal setae; P5, lengths of inner spine(seta) relative to lengths of outer spine(seta); A= inner seta as long as outer seta, B* = inner seta is twice of outer seta, B** = inner seta is twice of outer spine.

(see Fig. 15, Kunz 1980) has a distance between the two terminal setae of the exopod relatively wider than that of *P. pholpunthini* sp. n. (Fig. 4C).

At present, the taxonomic status of members of the genus *Parategastes* is still unclear. Original descriptions and illustrations of type specimens of some species are poor. It seems that this genus is in need molecular data of each species.

Key to species of female of *Parategastes* Sars, 1904 (modified from Kunz 1980)

1   Endopod and exopod of P1 with 5 and 4 setae.. *P. chalmersi* (Thompson & Scott, 1903)
–   Endopod and exopod of P1 with 6 and 5 setae... 2
2   Exopod of antenna with two setae...*P. sphaericus* (Claus, 1863)
–   Exopod of antenna with three setae 3
3   Rami of P1 shorter than basis .... *P. conexus* Humes, 1984
–   Rami of P1 equal or longer than basis 4
4   A1 with 7 segmented, middle inner spine of endopod of P1 is slender.. 5
–   A1 with 6 segmented, middle inner spine of endopod of P1 is thickened 6
5   P1, endopod and exopod approximately as long as basis, proximal seta of inner edge of endopod longer than segment. Exp-3 of P4 with 2 outer seta..*P. herteli* Jakobi, 1953
–   P1, endopod and exopod longer than basis, proximal seta of inner edge of endopod shorter than segment. Exp-3 of P4 with 3 outer setae .... *P. caprinus* Wellershaus, 1970
6   Length of inner apical seta of baseoendopod of P5 as long as outer apical seta. Enp-1 of P4 without inner seta............ .... *P. coetzeei* Kunz, 1980
–   Length of inner apical seta of baseoendopod of P5 is twice of outer apical seta. Enp-1 of P4 with 1 inner seta.............. .... *P. pholpunthini* sp. n.

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Juan Cristóbal Gundlach's collections of Puerto Rican birds with special regard to types

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Abstract

The German naturalist Juan Cristóbal Gundlach (1810–1896) conducted, while a resident of Cuba, two expeditions to Puerto Rico in 1873 and 1875–6, where he explored the southwestern, western, and northeastern regions of this island. Gundlach made representative collections of the island's fauna, which formed the nucleus of the first natural history museums in Puerto Rico. When the natural history museums closed, only a few specimens were passed to other institutions, including foreign museums. None of Gundlach's and few of his contemporaries' specimens have survived in Puerto Rico. We located 191 bird specimens (43 species) collected there by Gundlach, all of which are in foreign institutions, especially Museum für Naturkunde Berlin. Here we list all located specimens and include data associated with them. Six new species were described out of the Gundlach collections from Puerto Rico, three of which are still taxonomically recognized today. Information about the types of those taxa is given.

Zusammenfassung

Der deutsche Naturforscher Johann Christoph Gundlach (1810–1896), auf Kuba lebend, führte 2 Expeditionen (1873 und 1875–6) nach Puerto Rico durch, auf denen er die südwestlichen, westlichen und nordöstlichen Regionen dieser Insel erforschte. Gundlach fertigte repräsentative Sammlungen der Fauna an, welche den Kristallisationskern des ersten naturkundlichen Museums Puerto Ricos bildeten. Die naturkundlichen Museen wurden später geschlossen und nur wenige Präparate wurden an andere Einrichtungen einschließlich ausländischen Museen übergeben. Kein Präparat von Gundlach und nur wenige aus dieser Zeit überdauerten auf Puerto Rico. Wir lokalisierten 191 von Gundlach gesammelte Vogelpräparate (43 Arten), welche sich heute alle an ausländischen Institutionen, insbesondere am Museum für Naturkunde Berlin, befinden. Eine Liste gibt Übersicht über alle nachgewiesenen Präparate. Aus der ornithologischen Sammlung Gundlachs von Puerto Rico wurden 6 Arten neu beschrieben, von denen 3 heute noch taxonomisch relevant sind. Informationen zu den Typen für diese Taxa werden angegeben.

Introduction

Dr. Juan Cristóbal (Johann Christoph) Gundlach (1810–1896) arrived in Cuba from his native Germany in January 1839, intending to stay only a short time. Instead, he remained in Cuba until the end of his long and productive life, with the exception of short trips to exhibit his collections and visit family and colleagues in Europe, and two extended expeditions to Puerto Rico from June to December 1873 and from September 1875 to July 1876. Starting

from the western part he explored the northern region to San Juan, as well as the south western region of Puerto Rico. Additionally he made two trips to the central parts of the island (Lares and Gaguana/Jayuya).

His zoological interests were manifold, and he published his observations widely, not only within Cuba, but also in several international journals, in Spanish, German, and English. Gundlach amassed superb representative collections especially insects, molluscs, reptiles, mammals, and birds. He maintained an active exchange of specimens with foreign institutions and friends, so a considerable part of his collection was dispersed among different countries from the very beginning. This resulted in single specimens collected by Gundlach housed in several diverse collections today. Our purpose here is to list those vouchers of the avifauna of Puerto Rico available for research on this island as well as for international taxonomists. We also hope to stimulate interest of museum curators to search for additional specimens in their collections.

A summary of Gundlach's lifework, especially of his two expeditions (including a map) as well as his influence on the development of natural history in Puerto Rico is presented in Wiley et al. (2014).

Collections of Puerto Rican Birds made by Gundlach and his colleagues

Even as a boy, Gundlach was interested in studying animals and got his introduction to taxidermy by helping his older brother preserve anatomical and biological specimens. After his university studies, Gundlach decided to make an expedition to Suriname. To finance the travel, Robert Wilhelm Bunsen (1811–1899), second director of the "Verein für Naturwissenschaften Kassel", initiated an arrangement wherein shares were issued to support Gundlach's 1838 expedition to Suriname. Some 105 shareholders subscribed for 203 shares with a value of 1218 Thaler (Dathe and Gonzales Lopez 2002). This financial support of Gundlach's expedition was made on the condition that he sends scientific objects back to Germany to reimburse the travel expenses. Although Gundlach made it only as far as Cuba, he sent large shipments of scientific material to Germany (Anonymous 2011; Dathe and Gonzales Lopez 2002). Eduard Sezekorn (1796–1869), managing director of the Verein für Naturwissenschaften Kassel, organized the sale of the collected materials.

This was the start of Gundlach's intensive long-term collecting activities in the Caribbean. The specimens gathered there were widely distributed from the very beginning. But Gundlach also retained extensive collected materials for his own museums of natural history in Cuba. In his autobiography, Gundlach (1896) wrote that one specimen of *Mellisuga helenae* which he collected in 1844 was the first specimen of his personal collection. He realized that the hummingbird was an unknown species and therefore he decided to keep it rather than to send it

to Germany. Thereafter, Gundlach kept one specimen of each species, except of fish and large reptiles, for his own collection (Gundlach 1896). A third part of his collected specimens was sent to his friends and colleagues in Cuba as well as all over the world for exchange or to serve as the basis for scientific discussions (e.g. Ramón M. Forns (La Habana, *fl.* 1858), Tomas Blanco (San Juan), Agustin Stahl (Mayagüez), George N. Lawrence (New York), and Wilhelm Peters and Jean-Luis Cabanis (Berlin)). In principle, the specimens which Gundlach collected in Puerto Rico were intended for comparison with the specimens from his Cuba collecting and suffered the same fate as the specimens from Cuba. By the 1870s, Gundlach's travel expenses should have been recouped by his investors, so the Puerto Rican specimens remained in Gundlach's collection or were sent only to friends and scientific colleagues, and none should have been sent to Germany to settle his 1838 account. Finally, in gratitude for their patronage and benefaction over the years, Gundlach gave many of his bird specimens to friends as salon decorations.

Today, Gundlach's personal collection is housed in the Instituto de Ecología y Sistemática (IES, La Habana, Cuba). The IES collections have received little evaluation since the catalogues of Gundlach (1895) and Valdes Ragués (1914), with the exception of recent reviews of Cuban birds (Aguilera Román and Garrido 2000; Aguilera Román et al. 2002; Wiley et al. 2008), reptiles and amphibians (Moreno García et al. 2002), and Noctuoidea (Lepidoptera) (Becker 2002). We examined 18 Cuban collections (Wiley et al. 2008), but found bird specimens from Gundlach's Puerto Rican expeditions only at the IES, although several of his Cuban bird specimens were dispersed among other Cuban institutions. Most of the specimens collected in Puerto Rico by Gundlach were sent to institutions in the United States and Europe, especially Germany. We found Puerto Rican specimens collected by him in seven institutions (Table 1; see Suppl. material 1 for names of all institutions we visited or queried regarding holdings of Gundlach's Puerto Rican bird specimens). Most of these specimens were direct donations from Gundlach, whereas a few arrived at those institutions as part of the collections of others, such as the Henry W. Bryant collection containing specimens given by Gundlach to G. N. Lawrence.

Most of the Gundlach specimens received by the U. S. National Museum of Natural History (USNM) came as direct donations (~12 separate acquisitions) from him. There was one accession in 1868 of ten specimens from Puerto Rico, and an accession in 1877 of 9–10 birds from the West Indies (in litt. James Dean to JWW; 10 November 2006; USNM).

Today, the most comprehensive collection of Gundlach's specimens from Puerto Rico is housed in the Museum für Naturkunde Berlin. Once he settled in Cuba, Gundlach sent his ornithological observations to Eduard Sezekorn, who transformed Gundlach's notes into manuscripts and sent them to Jean Louis Cabanis (1816–1906), the bird curator of the Zoological Museum Berlin (ZMB, today Museum für Naturkunde). Cabanis published the

Table 1. Numbers of bird specimens (skins and mounts) collected by Juan Gundlach in Puerto Rico.

Institution[†]	No. of specimens	Acquisition (source)
MfN	102[‡]	Gundlach
IES	44	Gundlach
AMNH	18	Lawrence
FMNH	14	.
USNM	11	Gundlach, Lawrence
BMNH	1	.
MCZ	1	AMNH
Total	191	

[†]Full museum names: MfN = Museum für Naturkunde Berlin; IES = Instituto de Ecología y Sistemática, La Habana; AMNH = American Museum of Natural History, New York; FMNH = Field Museum of Natural History, Chicago; USNM = US National Museum of Natural History, Washington; BMNH = The Natural History Museum, Tring; MCZ = Museum of Comparative Zoology, Cambridge.

[‡]Gundlach collected 95 specimens; another 7 were collected by Agustin Stahl and presented to MfN by Gundlach.

articles with remarks in his *Journal für Ornithologie*. The first specimens from Cuba arrived at ZMB in 1862 through Sezekorn and were most probably part of the remuneration of Gundlach's travel expenses to the "Verein für Naturwissenschaften Kassel". A direct relation between the ZMB (Wilhelm Peters, Jean-Luis Cabanis) and Gundlach was established in 1861 (Peters) and 1862 (Cabanis) at the latest, more than 10 years before his first trip to Puerto Rico (Museum für Naturkunde, Historische Bild- u. Schriftgutsammlungen, Zool. Mus. [hereafter MfNHBSZM], Signatur ZMB S I, Gundlach, I.). Following his participation in the Exposition Universelle in Paris, Gundlach visited the ZMB in 1867 for a week, where he met with the museum's curator Wilhelm Peters (1815–1883). Further specimen donations to the ZMB followed (MfNHBSZM Signatur ZMB S I, Gundlach, I)

The material collected by Gundlach in Puerto Rico was sent in three shipments to the ZMB (arrival dates March 1874, June 1874, October 1876). Birds from other collectors (Blanco, Krug) in Puerto Rico were included in those shipments to the ZMB (in litt. JCG to Wilhelm Peters [hereafter WP]; 15 February 1874; from Fermina, Cuba; MfNHBSZM, Signatur ZMB S I, Gundlach, I., p. 219a). All, however, were registered with Gundlach as collector, and the determination of the actual collector is difficult or impossible today because the original labels were not conserved. Further, it seems that not all specimens Gundlach sent were maintained in the Berlin collection. Thus, a hummingbird mentioned by Gundlach in a letter to Peters dated 15 February 1874 is not available today and could not be traced in museum catalogues (MfNHBSZM, Signatur ZMB S I, Gundlach, I., p. 129a). This, and other specimens, may have been lost, destroyed, or exchanged with other collections.

The first of Gundlach's three shipments included 34 bird specimens that arrived at ZMB in March 1874. A second shipment, with 28 birds (according to the cata-

logue) arrived in June 1874. All of those birds were collected during Gundlach's 1873 expedition to Puerto Rico. The month of collection was recorded for most birds in the second shipment, whereas it was not for most specimens in the first shipment.

A third shipment of three boxes (including one of birds) of specimens collected during Gundlach's 1875–76 expedition to Puerto Rico arrived at ZMB in October 1876. Gundlach mentioned that among these birds were specimens from Krug's collection (in litt. JCG to WP; 17 August 1876; from Mayagüez, Puerto Rico; MfNHBSZM, Signatur ZMB S I, Gundlach, I., p. 259.). Cabanis confirmed that 30 stuffed birds, 67 eggs, and 7 nests arrived in October 1876 (MfNHBSZM, Signatur ZMB S I, Gundlach, I., p. 266). Twenty-eight of those birds were catalogued in October 1876. After this date seven additional specimens from Puerto Rico arrived in June 1878, January 1879, December 1880, and June 1881. Those specimens were probably part of Stahl's collection, which Gundlach had received at that time (in litt. JCG to Jean Louis Cabanis; 29 September 1878; MfNHBSZM, Signatur ZMB S I, Gundlach, I., p. 294). It cannot be excluded, however, that Gundlach sent some specimens of his own expeditions which had been maintained in his personal collection.

Upon their arrival in Berlin, the specimens were assigned entrance numbers (B-numbers) and, after preparation, inventory numbers. Only five specimens of the second shipment (1874) were inventoried later. All of Gundlach's specimens for the ZMB collection were mounted when received. Collectors labels were removed usually and new museum labels were created. Of the inventoried specimens, 70% have survived (98% of the eggs), and 85% of those have been transformed to skins. The transformation from mounts to skins was initiated by Erwin Stresemann (1889–1972) in the middle of the 20th century to better protect the specimens.

In general, Gundlach seemed to collect for taxonomical purposes only, and he used the specimens as vouchers and for determination. But the exact collecting locality and the collecting date are never mentioned. For some specimens a month is given on the label as Gundlach mentioned that several birds are to be found in different seasons only. The lack of more data is unfortunate, because locality and date data could provide a more useful baseline in assaying what has been the dramatic change of biodiversity that has occurred in Puerto Rico, including several species that have been extirpated from in the island or are now extinct (Wiley 1985, Snyder et al. 1987).

Puerto Rican Bird Specimens Collected by Juan Gundlach

We present an annotated inventory of Gundlach's bird specimens (skins and mounts, as well as eggs and nests) from Puerto Rico in the two largest collections: the collection at the MfN, Berlin, and the collection at the IES, La Habana, with notes on Gundlach's Puerto Rican spec-

imens encountered in other institutions. Both the IES and MfN specimens have been unreported, except for Valdes Ragués' (1914) incomplete catalogue of the Museo cubano "Gundlach" when it was housed at the Instituto de Segunda Enseñanza, La Habana. The IES specimens were "discovered" among Gundlach's collection of Cuban skins, mounts, eggs, and nests during our recent inventory. The MfN specimens remained more or less unknown or international scientists had thought they had been destroyed in WWII. Our 2011 inventory, however, revealed that although the MfN collections were damaged during the Allied bombing of Berlin, many of Gundlach's specimens and their records survived. Figures 1 to 3 are examples of how the specimens appear today in the MfN.

The list, as well as the species names (Latin, English) including current subspecies names, follow Dickinson and Remsen (2013) and Dickinson and Christidis (2014). The scientific name used by Gundlach (1874, 1878a) is added in brackets. For each species, institution and catalogue number(s), sex, date(s) of collection, and locality, as available, are presented. As already written, none of the IES Puerto Rican specimens has detailed data. Nevertheless, some of the specimens can be matched to Gundlach's published accounts of his collecting activities, and thus further data were derived from interpretation. For specimens housed in MfN, we use the original institution name of ZMB. The ZMB specimens could generally be assigned to the different collecting expeditions due to the accession date. We present information included by Valdes Ragués in his 1914 catalogue of Gundlach's collection in the Instituto de Segunda Enseñanza. Puerto Rican specimens in the Instituto de Segunda Enseñanza museum were identified by a yellow label, but Valdes Ragués (1914) did not always mention that information in his catalogue. Full names and acronyms for institutions are presented in Table 1. Expanded specimen data are available from the authors.

Following the specimen list, we provide a list of the types belonging to the Gundlach collection from Puerto Rico. Six species were described from that collection. Except for *Asio portoricensis*, all of those species were detected by Gundlach himself, but in some cases they were "officially" (in the sense of the ICZN) published by other authors. This was true for G. N. Lawrence (New York), who received a bird collection from Gundlach and J. L. Cabanis (Berlin).

Localities

Mayagüez (Mayaguez: town on the west coast of Puerto Rico [18°12'03"N; 67°08'22"W (DMS)]; Gundlach collected there several times in both expeditions.

Lares: town in the moutains of western Puerto Rico [18°17'40"N; 66°52'37"W (DMS)]; Gundlach collected there from July to October 1873.

Cueva de Pajita: cave near Callejones Barrio, western Puerto Rico [18° 19'38"N; 66° 50'56"W (DMS)]; Gundlach collected there in June and July 1873.

Figure 1. ZMB 21494: *Melanerpes portoricensis*, female, collected by Gundlach on Puerto Rico in November 1873, mounted and labelled in ZMB (photo Hwa Ja Götz, MfN).

Quebradillas: town in western Puerto Rico [18°28'25"N; 66°56'18"W (DMS)]; Gundlach collected in the area in October and November 1873, and winter and spring 1876.

Vega Baja: town in in north-central Puerto Rico [18°26'39" N; 66°23'15" W (DMS)].

Figure 2. ZMB 21632: *Mimus polyglottos*, female, collected by Gundlach on Puerto Rico in October 1873, mounted and labelled in ZMB, remounted in the 20th century (photo Hwa Ja Götz, MfN).

Figure 3. Original label of Gundlach of the holotype of *Chlorestes gertrudis* ZMB 21628 (photo Hwa Ja Götz, MfN).

List of specimens

The following list summarizes the information on the specimens of Puerto Rico originally belonged to the collection of Gundlach (Gundlach mainly given as collector).

The catalogue is structured as follows:

Scientific species name, subspecies name, **English name** [scientific name used by Gundlach]

Number of specimens: ♂ ♀ + unsexed. Registration number. Registered dates.

Number of eggs: Registration number. Registered dates.

Number of nests: Registration number. Registered dates.

Anatidae
Dendrocygna arborea, **West Indian Whistling Duck** [*Dendrocygna arborea*]

1 egg: ZMB 2000.30251 Date: 1875/1876.

Oxyura jamaicensis jamaicensis, **Ruddy Duck** [*Erismatura rubida*]

4 eggs: ZMB 2000.30255–30257 (clutch?), ZMB 2000.30374. Dates: 1873, 1875/1876.

Columbidae
Patagioenas inornata, **Plain Pigeon** [*Chlorœnas inornata*]

1: ♀. ZMB 25226. Lares. Collector probably Stahl.

Zenaida aurita zenaida, **Zenaida Dove** [*Zenaida amabilis*]

1: ♂. ZMB 22676. Date: 1875/1876.

2 eggs: ZMB 2000.30249–30250 (clutch?). Date: 1875/1876.

Phaethontidae
Phaethon lepturus catesbyi, **White-tailed Tropicbird** [*Phaëton flavirostris*]

1: ZMB 22677. Date: Aug 1876.

3 eggs: ZMB 2000.30268–30270 (3 clutches?). Date: 1875/1876.

Apodidae

***Cypseloides niger niger*, Black Swift** [*Nephocætes niger*]
3: 2♂ 1♀. ZMB 22657–22659. Dates: 1875/1876.

Trochilidae

***Anthracothorax dominicus aurulentus*, Antillean Mango** [*Lampornis aurulentus*]
13: 8♂ 5♀. ZMB 21471–21473 (21471 missing), 21625 (missing), 21626, 27054, 27055; IES 2560, 2562, 2567; AMNH 46356, 46360; USNM 353410. Dates: Mar, Apr, Sep, Oct; 1873, 1876.

***Anthracothorax viridis*, Green Mango** [*Lampornis viridis*]
9: 9♂. ZMB 21474, 21627, 22668–22671; IES 2563; FMNH 42399; USNM 087706. Dates: Sep, Oct; 1873,1875,1876.

***Chlorostilbon maugaeus*, Puerto Rican Emerald** [*Chlorolampis Maugæus*]
6: 3♂ 1♀ + 2. ZMB 21628 (Holotype for *Chlorestes gertudis* Gundlach 1874), 22672–22674; IES 2578; AMNH 38784. Dates: Nov; 1873, 1875/1876.

Cuculidae

***Crotophaga ani*, Smooth-billed Ani** [*Crotophaga ani*]
1 egg: ZMB 2000.30296 (missing). Date: 1875/1876.

***Coccyzus americanus americanus*, Yellow-billed Cuckoo** [*Coccyzus americanus*]
1: ♂. ZMB 21496. Date: 1873 (missing).

***Coccyzus minor*, Mangrove Cuckoo** [*Coccyzus minor*]
1: ♂. ZMB 21497. Date: Jul 1873.

***Coccyzus vieilloti*, Puerto Rican Lizard Cuckoo** [*Saurothera Vieilloti*]
Valdes Ragués (1914) listed one specimen in his catalogue of the Gundlach collection.
5: 5♂. ZMB 21495, 21600; IES 2569, FMNH 41320, 41321. Dates (4): Feb, Sep, Nov; 1873, 1876; "5 March 1892" [likely the date of acquisition].
1 egg: ZMB 2000.30295. Date: 1875/1876.

Rallidae

***Rallus longirostris caribaeus*, Clapper Rail** [*Rallus crepitans*]
1: ZMB 22679. Date: 1875/1876.

***Gallinula galeata cerceris*, Common Gallinule** [*Gallinula galeata*]
1 egg: ZMB 2000.30376. Date: 1873.

***Fulica americana americana*, American Coot** [*Fulica americana*]
6 eggs: ZMB 2000.30259–30263 (clutch?), ZMB 2000.30375 (missing). Dates: 1873, 1875/1876.

Procellariidae

***Puffinus lherminieri lherminieri*, Audubon's Shearwater**
Valdes Ragués (1914) noted inexplicably "15 exemplars de Puerto Rico" under *Puffinus auduboni*, but Gundlach made no mention of the species elsewhere. We did not find specimens in any collection.

Ardeidae

***Ixobrychus exilis exilis*, Least Bittern** [*Ardetta exilis*]
2 eggs: ZMB 2000.30253–30254 (clutch?). Date: 1875/1876.

***Nycticorax nycticorax hoactli*, Black-crowned Night Heron** [*Nyctiardea Gardeni*]
1 egg: ZMB 2000.30252. Date: 1875/1876.

Strigidae

***Asio flammeus portoricensis*, Short-eared Owl** [*Brachyotus Cassinii*]
4: 2♂ + 2. AMNH 44768, 44769; USNM 086039; MCZ 96647 (Syntypes for *Asio portoricensis* Ridgway 1882). Date (3): 1873–1876 (if really collected by Gundlach).

***Megascops nudipes nudipes*, Puerto Rican Screech Owl** [*Gymnoglaux nudipes*]
In his 1874 catalogue, Gundlach listed this species as "*Gymnoglaux Krugii* Gundl. n. sp." (in honor of Krug), but as "*Gymnoglaux nudipes* (*Strix*) Daud." in his 1878(b) publication. The Valdes Ragués (1914) catalogue lists two specimens, so one is now missing from the IES collection.
11: 8♂ 3♀. ZMB 21596, 21597 (Syntypes for *Gymnoglaux Krugii* Gundlach 1874); ZMB 22654, 22655 (missing), 25223 (collected by Stahl), 25224 (collected by Stahl); IES 2584; AMNH 44792–44795. Dates: Jan, May, Nov; 1873, 1875/1876, 1877.
1 egg: ZMB 2000.30245. Date: 1875/1876.

Picidae

***Melanerpes portoricensis*, Puerto Rican Woodpecker** [*Melanerpes portoricensis*]
6: 4♂ 2♀. ZMB 21493, 21494, 21601; IES 2559, 2591; AMNH 44132. Dates: Oct, Nov; 1873, 1876.

Todidae

***Todus mexicanus*, Puerto Rican Tody** [*Todus hypochondriacus*]
5: 2♂ 1♀ + 2. ZMB 21490 (missing), IES 2222, AMNH 43105, FMNH 41736, USNM 055110. Date: Sep; 1873.
4 eggs: ZMB 2000.30281 (missing)–30283 (clutch?), ZMB 2000.30365. Dates: 1873, 1875/1876.

Falconidae

Falco sparverius caribaearum, **American Kestrel** [*Tinnunculus dominicensis*]

Gundlach was particularly interested in the variation in color among kestrels and collected a good series of specimens of *F. s. dominicensis* and *sparverioides* from Cuba, as well as at least 14 specimens of *F. s. caribaearum* from Puerto Rico. Valdes Ragués (1914) listed two kestrel specimens from Puerto Rico in the ISE collection, so one is now missing.

13: 7♂ 6♀. ZMB 21498–21501, 22652, 22653, 2000.17994; IES 2561, AMNH 45001–45005. Dates: 1873, 1875/1876.

2 eggs: ZMB 2000.30246–20347 (clutch?). Date: 1875/1876

Psittacidae

Amazona vittata vittata, **Puerto Rican Parrot** [*Chrysotis vittatus*]

Gundlach collected parrots near Lares in 1873, noting that he was able to retrieve three of four shot (in litt. JCG to Felipe Poey; 1 August 1873; from Mayagüez, Puerto Rico; AhULH). He must have collected at least a fourth specimen at some point in 1876 (see below). Valdes Ragués (1914) listed *Chrysotis leucocephala* [Cuban Parrot *Amazona leucocephala*] among the Gundlach specimens, but noted it had a yellow tag, which designated a bird collected in Puerto Rico. The specimen was identified by IES staff as *A. vittata*. It was stolen from the collection *c.* 1990. The parrot disappeared from western Puerto Rico within 50 years of Gundlach's visit, surviving only in the easternmost part of the island (Snyder et al. 1987). The parrot is currently considered to be Critically Endangered (IUCN 2014).

4: 1♂ 1♀ + 2. ZMB 22675; IES 2589 (missing); FMNH 40353, 40354. Dates: Jul; 1873, 1875/76.

Psittacara chloropterus maugei, **Hispaniolan Parakeet**

2: 2 wings: ZMB 2000.35468-35469 (missing)(Syntypes of *Conurus Gundlachi* Cabanis 1881a), collected by C. F. Block on Mona Island.

Tyrannidae

Tyrannus dominicensis dominicensis, **Gray Kingbird** [*Melittarchus griseus*]

1: USNM 055109.

Tyrannus caudifasciatus taylori, **Loggerhead Kingbird** [*Tyrannus Taylori*]

4: 3♂ 1♀. ZMB 21638, 21639, 22656; IES 2587. Dates (4): Feb, Jun, Sep; 1873, 1876.

1 egg: ZMB 2000.30273. Date: 1875/1876.

Myiarchus antillarum, **Puerto Rican Flycatcher** [*Myiarchus antillarum*]

2: 2♂. ZMB 21491 (missing); FMNH 31069. Date (1): 1873.

2 eggs: ZMB 2000.30271–30272 (clutch?). Date: 1875/1876.

Contopus latirostris blancoi, **Lesser Antillean Pewee** [*Blacicus Blancoi*]

Cabanis (1875) described the endemic Puerto Rico subspecies as *Blacius blancoi* in 1875 from a specimen sent by Gundlach, but probably collected by Tomás Blanco.

2: 1♂ 1♀. ZMB 21492 (Holotype for *Blacius blancoi* Cabanis 1875); IES 2576; Date: Dec; 1873.

Vireonidae

Vireo latimeri, **Puerto Rican Vireo** [*Vireo Latimeri*]

1: ♂. ZMB 21629. Date: Oct 1873.

2 eggs: ZMB 2000.30358 (missing), ZMB 2000.30359 (clutch?). Date: 1875/76.

1 nest ZMB 2000.30312 (missing). Date: 1875/76.

Vireo altiloquus altiquus, **Black-whiskered Vireo** [*Phyllomanes calidris*]

1: ♂. ZMB 21480. Date: 1873.

2 nests ZMB 2000.30310–30311 (missing). Date: 1875/76.

Corvidae

Corvus leucognaphalus, **White-necked Crow** [*Corvus leucognaphalus*]

Gundlach shot eight crows at Cueva de Pajita, Lares, where he found large numbers, in July 1873 (in litt. JCG to Felipe Poey; 1 August 1873; from Mayagüez, Puerto Rico; AhULH). Gundlach also saw the crow at Utuado, and south of Quebradillas. He was told that the crow was more abundant in the island's interior than in the east. Despite its abundance during Gundlach's visit, the crow was extirpated from Puerto Rico by the 1960s (Raffaele 1989; Wiley 2006). The specimen listed as "cuervo" in the catalogue of Valdes Ragués (1914) is probably the IES individual, because it has a yellow label, indicating a specimen collected in Puerto Rico.

5: 1♂ 2♀ + 2. ZMB 22666, 23347 (collected by Stahl), 25225 (collected by Stahl), 2000.736; IES – without number in the actual catalogue: (O.C.#6140; "E190"). Dates: Jul (JCG in litt., see above); 1873, 1875/76.

Estrildidae

Estrilda melpoda, **Orange-cheeked Waxbill** [*Habropyga melpoda*]

Gundlach was the first naturalist to record the species in Puerto Rico; he noted (1878a) it was living free near Mayagüez, Añasco, and Cabo Rojo. Wetmore (1927) believed it had become established in Puerto Rico during the period of slave trade, whereas Raffaele (1983) suggested it arrived at the end of that period, in the mid-19th century.

3: 1♂ 1♀ + 1. ZMB 21486 (Mayagüez), 2000.19661; IES 2577. Dates: Jan; 1873, 1876.

***Spermestes cucullata*, Bronze Mannikin** [*Spermestes cucullatus*]

The mannikin is thought to have been introduced to Puerto Rico during the era of slave trafficking (Wetmore 1927; Danforth 1936), well before Gundlach's visits. Gundlach (1878a) reported it as very abundant in several locations, including Mayagüez, Lares, Quebradillas, and Vega Baja.

6: 2♂ 3♀. ZMB 21484 (missing), 21485, 21635; IES 2573, 2579, 2583. Dates (6): Feb, Mar, Jul, Aug; 1873, 1876.

Fringillidae

***Euphonia musica sclateri*, Antillean Euphonia** [*Euphonia Sclateri*]

8: 5♂ 2♀ + 1. ZMB 21475 (missing), 21476 (missing), 22678; IES 2564, 2558; AMNH 40514; FMNH 27047; USNM 054929. Dates: Jul, Dec; 1873, 1876.

Phaenicophilidae

***Nesospingus speculiferus*, Puerto Rican Tanager** [*Chlorospingus speculiferus*]

2: 1♂ + 1. ZMB 24887 (collected by Stahl); USNM 075331 (Holotype of *Chlorospingus speculiferus* Lawrence 1875) [given to Gundlach by Blanco].

***Spindalis portoricensis*, Puerto Rican Spindalis** [*Spindalis portoricensis*]

7: 3♂ 4♀. ZMB 21477, 21615 (missing), 21616; IES 2555, 2572; FMNH 27622, 27623. Dates : Mar, Sep; 1873, 1876.

Parulidae

***Setophaga petechia bartholemica*, Yellow Warbler** [*Dendroica petechia*]

4: 3♂ 1♀. ZMB 21482, 21614 (missing); USNM 54924, 54925. Dates : Aug, Sep; 1873.

2 eggs: ZMB 2000.30354 (missing), ZMB 2000.30355 (clutch?). Date: 1875/1876.

1 nest: ZMB 2000.30308. Date: 1875/1876.

***Setophaga adelaidae*, Adelaide's Warbler** [*Dendroica Adelaidae*]

Both IES specimens were included in the catalogue of Valdes Ragués (1914).

5: 3♂ 2♀. ZMB 21469, 21470; IES 2565, 2568; FMNH 26119. Dates : 1873.

Icteridae

***Icterus icterus ridgwayi*, Venezuelan Troupial** [*Icterus vulgaris*]

Gundlach noted (on label), "Es de costa firma — introducida," whereas Danforth (1936: 163) suggested it was indigenous to Puerto Rico. Raffaele (1989) and Raffaele and Kepler (1992) considered it introduced to the island.

1: IES 2336. Date: Oct.

***Icterus portoricensis*, Puerto Rican Oriole** [*Xanthornus portoricensis*]

8: 4♂ 2♀ + 2. ZMB 21487, 21488 (missing), 21623, 21624 (missing), 22661 (missing); IES 2429, 2588, 2571. Dates: Feb, Mar, Aug, Sep, Oct; 1873, 1875/1876.

1 egg: ZMB 2000.30291 (missing). Date: 1875/1876.

***Agelaius xanthomus xanthomus*, Yellow-shouldered Blackbird** [*Agelaius chrysopterus*]

The Yellow-shouldered Blackbird is now considered endangered and decreasing (IUCN 2014), in large part because of the activities of the Shiny Cowbird (*Molothrus bonariensis*), a brood parasite, which arrived in Puerto Rico some 75 years after Gundlach's visits (Post and Wiley 1976, Wiley et al. 1991). The blackbird's range has been greatly reduced since Gundlach's time (Post and Wiley 1977, Post 1981).

6: 3♂ 2♀ + 1. ZMB 21489, 22662, 22663; IES 2570; USNM 106119, 106120. Dates: Feb; 1873, 1875/1876.

3 eggs: ZMB 2000.30292–30393 (missing), ZMB 2000.30294. Date: 1875/1876.

1 nest: ZMB 2000.30313 (missing). Date: 1875/1876.

***Quiscalus niger brachypterus*, Greater Antillean Grackle** [*Chalcophanes brachypterus*]

5: 1♂ 3♀ + 1. ZMB 21636 (missing), 21637, 22664 (missing), 22665; IES 2585. Dates: Jan, Aug; 1873, 1875/1876.

7 eggs: ZMB 2000.30284–30290 (clutch?). Date: 1875/1876.

Thraupidae

***Coereba flaveola portoricensis*, Bananaquit** [*Certhiola portoricensis*]

Valdes Ragués (1914) listed two specimens in his catalogue of the ISE collection.

7: 3♂ 2♀ + 2. ZMB 21478, 21479, 21633, 22667 (all missing); IES 2554, 2556; FMNH 9762. Dates: Feb, May, Oct; 1873, 1875/1876.

5 eggs: ZMB 2000.30360, ZMB 2000.30361–30364 (missing) (clutch?). Date: 1875/1876.

1 nest: ZMB 2000.30309. Date: 1875/1876.

***Melopyrrha portoricensis portoricensis*, Puerto Rican Bullfinch** [*Pyrrhulagra portoricensis*]

3: 1♂ + 2. ZMB 22660 (missing); IES 2557; FMNH 23988. Dates (2): Jul; 1875/76.

***Melanospiza bicolor omissa*, Black-faced Grassquit** [*Euethia bicolor*]

5: 3♂ 1♀ + 1. ZMB 21483 (missing), 21634; IES 2580, 2582; AMNH 41310. Dates: Feb, Mar, Jul; 1873, 1876.

7 eggs: ZMB 2000.30323–30324, ZMB 2000.30389 (missing) (clutch?), ZMB 2000.30331–30332 (clutch?), ZMB 2000.30352–30353 (missing) (clutch?). Date: 1875/1876.

Hirundinidae
***Petrochelidon fulva puertoricensis*, Cave Swallow** [*Petrochelidon fulva*]

> **3**: 1♂ 1♀ + 1. ZMB 21481 (missing), 21618; IES 2151. Dates: Mar, Sep; 1873, 1876.
> **1 egg**: ZMB 2000.30274. Date: 1875/1876.

***Progne dominicensis*, Caribbean Martin** [*Progne dominicensis*]

> **5**: 3♂ 2♀. ZMB 21617; IES 2566, 2574, 2575; BMNH 84.11.21.119. Date: Aug 1873.
> **5 eggs**: ZMB 2000.30275–30279 (clutch?). Date: 1875/1876.

Mimidae
***Margarops fuscatus fuscatus*, Pearly-eyed Thrasher** [*Margarops fuscatus*]

It is remarkable that this now-abundant and obvious species (Snyder et al. 1987; Arendt 2006) was rare in Puerto Rico during Gundlach's time. He wrote (1878a): "Solamente en dos ocasiones he observado esta especie, y la creo poco común, porque pocas personas la conocían." He collected one in a coffee plantation, and observed a pair near Utuado in July.

> **2**: 1♂ 1♀. ZMB 23642 (collected by Stahl); IES 2411. Quebradillas. Dates: July, Nov (both in Gundlach 1878a).

***Mimus polyglottos orpheus*, Northern Mockingbird** [*Mimus polyglottus*]

> **3**: 2♂ 1♀. ZMB 21630–21632. Dates (3): Oct, Nov; 1873.
> **2 eggs**: ZMB 2000.30298–30299 (clutch?). Date: 1875/1876.

Turdidae
***Turdus plumbeus ardosiaceus*, Red-legged Thrush** [*Mimocichla ardosacea*]

> **5**: 4♂ + 1. ZMB 21468, 21598, 21599; IES 2586; FMNH 26782. Dates (4): Mar, Sep; 1873, 1876.
> **2 eggs**: ZMB 2000.30356–30357 (clutch?). Date: 1873.

List of type specimens of the collection of Gundlach from Puerto Rico

Chlorospingus speculiferus Lawrence

Chlorospingus speculiferus Lawrence 1875: 383.

Now. *Nesospingus speculiferus* (Lawrence 1875): 383. See Cory (1889: 86).

Type series. Lawrence (1875) described this species and attributed its discovery to Gundlach, but Gundlach (1878b) corrected this, stating that the specimen was collected by Tomás Blanco y González in Puerto Rico. Gundlach most probably received the specimen from Blanco in 1868. He later transferred it to the Smithsonian Institution in Washington, D. C. (today USNM) and there Gundlach was given as collector. As Lawrence (1875) wrote "Type in National Museum, Washington", the type series consists of only one specimen which is the only available specimen of that species at the USNM "collected by Gundlach" and is therefore the holotype. An additional specimen of this species from the Gundlach collection is housed at the ZMB (ZMB 24887), but it was collected by Stahl between 1878 and 1880 and was sent directly from Gundlach to Berlin. It is therefore not part of the type series. LeCroy (2012) noted that a further specimen at the American Museum of Natural History (AMNH 3262) has a type label for *Chlorospingus speculiferus*. But she did not consider it as a type specimen because it is not dated and Gundlach is not indicated clearly as the collector. We follow this argument. It appears that Gundlach never collected this species himself.

Holotype. USNM 75331, skin, unsexed, collected by Tomás Blanco y González on Puerto Rico [in the period 1863–1868].

Type locality. Porto Rico [today Puerto Rico], no further details available.

Remarks. The specimen was catalogued at the USNM on 18 October 1878, but Lawrence previously wrote in the description in 1875 that the specimen was housed in the National Museum, Washington. We do not know when the specimen arrived at the USNM. It may be that it remained there uncatalogued until the discussion about the type with Gundlach started in 1877. As it is the only specimen of this species collected by "Gundlach" at the USNM, we believe that USNM 75331 should be the type specimen.

Blacius blancoi Cabanis

Blacius blancoi Cabanis 1875: 244.

Now. *Contopus latirostris blancoi* (Cabanis 1875): 224. See Cory (1889: 129), Sclater (1888: 243), Traylor (1979: 135).

Type series. Not specified by Cabanis (1875). There is only one specimen noted in the inventory catalogue of the ZMB (ZMB 21492). But because the species was discovered and named by Gundlach all the specimens collected during the first expedition to Puerto Rico should be regarded as types according the ICZN (1999). Another specimen collected by Gundlach is housed at the IES (IES 2576). The date is given as "December" on the label. As Gundlach left Puerto Rico on 4 December 1873 after his first expedition to Puerto Rico it is more likely that he collected this specimen during his second expedition

(December 1875). Thus, this specimen cannot be included in the type series because the description of Cabanis was published in April 1875.

Syntype. ZMB 21492, skin, adult male, collected by J. C. Gundlach in Portorico [Puerto Rico] [in the period 06.–12.1873].

Type locality. Portorico [today Puerto Rico], no further details available.

Remarks. Gundlach discovered this new species and named it in honor of his friend Tomás Blanco y González (1840–1892), living in Puerto Rico. Gundlach (1874) included the species in his published list without description. Cabanis (1875) added the missing description, so he is officially the author of this species, following the ICZN (1999).

Gymnoglaux Krugii Gundlach

Gymnoglaux Krugii Gundlach 1874: 310, 315.

Now. *Megascops nudipes nudipes* (Gundlach 1874): 310, 315. See Gundlach (1878b: 164/165), Cory (1889: 192).

Type series. Not specified by Gundlach (1874). There are two specimens noted in the inventory catalogue of the ZMB. Therefore these are syntypes.

Syntype. ZMB 21596, skin, male, collected by J. C. Gundlach in Portorico [Puerto Rico] [in the period 06.–12.1873].

Syntype. ZMB 21597, skin, female, collected by J. C. Gundlach in Portorico [Puerto Rico] [in the period 06.–12.1873].

Type locality. Portorico [today Puerto Rico], no further details available.

Remarks. Gundlach discovered this new species and named it in honor of his patron Carl Wilhelm Leopold Krug (1833–1898), who served as the German and British Vice-consul in Mayagüez. In his 1878(b) publication, Gundlach synonymized it with "*Gymnoglaux nudipes* (*Strix*) Daud".

There are further specimens of this species from the Gundlach collection in the ZMB (ZMB 22654, 22655 [missing], 25223 [collected by Stahl], 25224 [collected by Stahl]), the IES (IES 2584 and without number [missing, fide Valdes Ragués (1914)]) and the AMNH (AMNH 44792–44795) which were (most probable for the IES and two of the AMNH specimens) collected during Gundlach's second expedition to Puerto Rico or even later by Agustin Stahl (1842–1917). Thus, due to missing exact information we do not regard any of these specimens as types.

Asio portoricensis Ridgway

Asio portoricensis Ridgway 1882: 366.

Now. *Asio flammeus portoricensis* (Cory 1889: 191; Peters 1940: 170).

Type series. Ridgway (1882) attributed the discovery of this species to Baird et al. (1874) when the existence of a new species of this genus from Puerto Rico was determined. Lacking further material for comparison, this species remained unnamed in 1874. In his description, Ridgway (1882) wrote that he had studied 4 specimens from Puerto Rico, one specimen in the USNM (USNM 39643 which was illustrated and described as aberrant by Baird et al. (1874) and three specimens which were collected by Gundlach and received for determination from George Newbold Lawrence. Ridgway reported that all four specimens were very similar and described the new species. Therefore, all four specimens are syntypes. Some of Gundlach's specimens passed to Lawrence. The specimen USNM 086039 came with the Lawrence collection to the USNM where it was catalogued in 1882 (in litt. James Dean to JWW, 3 June 2009). Two other specimens of *Asio flammeus* of the Lawrence collection came to the AMNH (AMNH 44768, AMNH 44769) in 1887 (in litt. Mary LeCroy 27 September 2013). One of them (AMNH 44768) was later (c. 1920, in litt. Jeremiah Trimble 25 October 2013) given to the MCZ, where it is housed today (MCZ 96674).

Syntype. USNM 086039, skin, adult male, collected by J. C. Gundlach in Portorico [Puerto Rico].

Syntype. USNM 39643, skin, adult, collected by G. Latimer on the north side of Portorico [Puerto Rico] [in 1864/65 according to catalogue USNM], catalogued at the USNM on 10 November 1865.

Syntype. AMNH 44769, skin, male, collected by J. C. Gundlach in Portorico [Puerto Rico].

Syntype. MCZ 96674 (former AMNH 44768), skin, collected by J. C. Gundlach in Portorico [Puerto Rico].

Type locality. Porto Rico [today Puerto Rico], no further details available.

Chlorestes gertrudis Gundlach

Chlorestes gertrudis Gundlach 1874: 312, 315.

Now. *Chlorostilbon maugaeus* (Gundlach 1878b, Cory 1889: 154, Salvin 1892: 58, Peters 1955: 39).

Type series. Not specified by Gundlach (1874). In 1878, Gundlach (1878b) wrote that he collected one male of this new species which was sent to Berlin later and so it is the holotype.

Holotype. ZMB 21628, skin, adult male, collected by J. C. Gundlach in Portorico [Puerto Rico] in November [1873].

Type locality. Porto Rico [today Puerto Rico, western part (Wiley et al. 2014)].

Remarks. Gundlach was not aware of the description of *Sporadinus maugaeus* Viell. 1817 when he described *Chlorestes gertrudis*. In his second publication on the birds of Puerto Rico (1878b), Gundlach suspected synonymy with this species but he was uncertain.

Gundlach (1874) named *Chlorestes gertrudis*, a synonym of *Chlorostilbon maugaeus*, as a new species, in honor of Gertrud Krug, wife of his friend Leopold Krug (Gundlach 1878a: 182; in litt. JCG to Felipe Poey; 1 August 1873; from Mayagüez, Puerto Rico; AhULH). Blanco (1969), however, claimed it was named in recognition of doña Gertrudis Gonzalez de la Parte, mother of Tomás Blanco. Valdes Ragués (1914) listed five unnamed specimens of "zumbador" in his catalogue, probably a combination of Antillean mango (3 specimens), green mango (1), and Puerto Rican emerald (1).

Five additional specimens of this species were collected by Gundlach (ZMB 22672–22674; IES 2578; AMNH 38784), but all have been collected during his second expedition to Puerto Rico and, therefore, we do not regard them as types.

Conurus gundlachi **Cabanis**

Conurus gundlachi Cabanis 1881a: 5.

Now. *Psittacara chloropterus maugei* (Souancé 1856: 59, Cory 1889: 180, Salvadori 1891: 189, Ridgway 1916: 155, Peters 1937: 188, Wiley et al. 2014: 257, Olson 2015).

Type series. Cabanis mentioned in his description as well as in Cabanis (1881b) that he described the species based on two left wings, which means his description is based on two specimens.

Syntype (lost). ZMB 2000.35468, wing, [collected by Dr. Claudio Federico Block on Isla de Mona, Portorico [Puerto Rico] before 1874].

Syntype (lost). ZMB 2000/35469, wing, [collected by Dr. Claudio Federico Block on Isla de Mona, Portorico [Puerto Rico] before 1874].

Type locality. Insel Mona near Portorico [today Isla de Mona, Puerto Rico].

Remarks. Three wings of a parakeet from Isla de Mona were collected by Dr. Claudio Federico Block (or Bloch), a Danish physician and hunter from Mayagüez, who gave the wings to Gundlach probably in 1875. Two of the wings

arrived in Berlin in October 1876 (B 14144). But they did not get inventory numbers and the whereabouts of the wings are unclear, so they are apparently lost. The whereabouts of the third wing is unknown, too (Olson 2015).

Summary

In summary, we can account for 191 bird skin and mount specimens collected by Gundlach in Puerto Rico, representing 43 species. An additional 69 eggs (19 species) and 6 nests (5 species) were located, including eggs of 7 species not represented by skins. A total of 147 skin and mount specimens collected by Gundlach in Puerto Rico were found in foreign institutions, with the Museum fuer Naturkunde holding the largest number (102), including two holotypes and two syntypes (Table 1). The MfN collection contains 53.4% of all specimens, and 93.0% of the species in all collections examined. Further, MfN specimens represent 26.1% of the 153 species Gundlach reported from Puerto Rico (Gundlach 1878a, 1878b).

We found 44 specimens (41 extant), representing 28 species, collected by Gundlach in Puerto Rico in the IES collection. Valdes Ragués (1914) listed 18 Puerto Rican specimens by name or group in his analysis of Gundlach's collection, with another improbable 15 Audubon's Shearwaters. The IES collection contains 23.0% of all specimens, and 65.1% of the species in all examined collections (Gundlach 1878a, 1878b; Table 1). Further, the IES specimens represent 18.3% of the 153 species Gundlach reported from Puerto Rico.

Conspicuously missing from all collections of Gundlach's bird skins from Puerto Rico are specimens of shorebirds, waterfowl, and waders, even though he actively collected in several coastal and wetland areas and did take several eggs of waterbirds.

Unfortunately, none of Juan Gundlach's journals or field notes has been found. Such material might provide much-desired additional data on Gundlach's collected specimens, much of which would be valuable in further determination of types. Some information may be available in Gundlach's correspondence, and we urge others to search for all such materials.

Concluding remarks

The theft of the Puerto Rican Parrot specimen, sadly, was not the last of Gundlach's bird specimens to have been stolen from the IES collection. In 2007, thieves again raided the collection, this time making off with several Gundlach-collected birds. Tragically, the only Cuban Macaw (*Ara tricolor*) specimen in Cuba (and one of 19 known specimens worldwide; Wiley and Kirwan 2013) was taken by the thieves, who also stole air conditioners and other valuable items essential for maintaining the collections. The specimens were most likely taken not

for their scientific value or for their value to rogue private collectors, but rather for use in local spiritual rituals. Although substantial improvements have been made in security, the IES collections are still vulnerable to future raids. The IES collection of birds and other natural history specimens is an important record of Cuban and Puerto Rican biodiversity, but is threatened by a lack of funding to maintain the collections. Such funding is desperately needed to prevent the further degradation or loss of these important treasures.

We hope to stimulate interest of museum curators to search for additional specimens in their collections.

Acknowledgements

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Supplementary material 1

List of consulted collections

Authors: Sylke Frahnert; Rafaela Aguilera Román; Pascal Eckhoff; James W. Wiley

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Explanation note: Collections in Europe, United Kingdom, United States of America, and Caribbean queried or inventoried for presence of Puerto Rican bird specimens collected by Juan Gundlach. Where available, institution abbreviations follow standards set in AVECOL (http://www.museum.lsu.edu/~Remsen/AVECOLlections.html).

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A quadriocellar scoliid wasp (Hymenoptera, Scoliidae) from Mallorca, with a brief account of supernumerary ocelli in insects

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Abstract

A remarkable teratological female of *Megascolia* (*Regiscolia*) *maculata flavifrons* (Fabricius, 1775) (Scoliidae: Scoliinae: Scoliini) with a supernumerary median ocellus is described and illustrated. While supernumerary ocelli have been reported before from Diptera, Orthoptera, and Hymenoptera, this is the first record of such a malformation from a scoliid wasp. Four other teratological scoliid wasps have been reported in the literature but all were gynandromorphs. A brief summary of known records of supernumerary ocelli among insects is provided.

Key Words

Introduction

According to Nichols (1989) teratology is the "study of structural abnormalities, especially monstrosities and malformations." Most accounts of teratology in the literature derive from experimental manipulation of the developmental process, while naturally occurring malformations are less frequently reported. In the past, many of these malformations were often neglected or mentioned merely as footnotes. This is perhaps not surprising as most authors surely must have thought that no significant conclusions could be drawn from isolated aberrant individuals. Indeed, it is challenging to infer much from isolated cases (e.g., Glasgow 1925), but when teratologies are made known and eventually summed up with others, there exists a great potential for patterns to emerge and broader conclusions to be drawn. The description of individual cases serves to build up a larger data set from which explanatory hypotheses can be formulated, highlighting the great value of descriptive science (e.g., Grimaldi and Engel 2007). A great example is the occurrence of gynandromorphs among aculeates, where the gradual accumulation of often-isolated, published accounts over the last 125 years has amassed into a body of data sufficient for the exploration of generalized patterns (e.g., Wcislo et al. 2004, Michez et al. 2009, Hinojosa-Díaz et al. 2012), and has aided attempts to homologize traits across sexes (e.g., Michener 1944, Engel 2007). It is therefore worthwhile

to contribute to the accumulation of data on teratologies in the hopes that someday sufficient numbers will exist to permit more critical study.

There exists a relative abundance of reports concerning insects with partially fused antennomeres (e.g., Asiain and Márquez 2009, Popovici et al. 2014), or gynandromorphs (e.g., Hinojosa-Díaz et al. 2012), particularly as these relate to understanding more standard morphologies, such as whether a reduction in total antennomere count results from fusion or loss. Less often are other structural anomalies reported, such as supernumerary appendages or organs like legs (e.g., Cockayne 1937), antennae (e.g., Cockayne 1938), ocelli (e.g., Engel et al. 2014), or even compound eyes (e.g., Banerjee and Kevan 1959).

The first major review of teratologies among Hymenoptera was by Dalla Torre and Friese (1899) who focused their attention on gynandromorphs. Balazuc (1958), however, was the first to provide a comprehensive account of teratological Hymenoptera, including all kinds of malformations. Among the aculeate Hymenoptera, malformed specimens are exceptionally well reported for bees (Anthophila), with gynandromorphs alone reported for more than 110 species in 29 genera (Hinojosa-Díaz et al. 2012). With respect to Scoliidae, however, the number of peculiar malformed specimens reported in the literature is limited to four records of gynandromorphs (De Romand 1835, Krombein 1949, Wolf 1989, Osten 1993). In the course of sorting several boxes of unidentified scoliid wasps VL recognized a female of *Megascolia* (*Regiscolia*) *maculata flavifrons* (Fabricius 1775) with four ocelli (Figs 1, 2). The aim of the present contribution is to describe and illustrate this specimen and to provide a short account on supernumerary ocelli in pterygote insects.

Materials and methods

Measurements were taken using a Keyence VHX 5000 Digital Microscope. The morphological terminology for the description of the specimen is adopted from Betrem (1971). The photographs were captured with a Nikon D800 digital camera with a Nikon AF-S Micro-NIKKOR 60 mm 1:2,8G ED lens in combination with the software programs Helicon Remote, Adobe Lightroom und Helicon Focus Pro. The identification of the specimen, which is deposited in the entomological collection of the Übersee-Museum Bremen (UMB), is based on the key provided by Osten (2000).

Systematics

Megascolia (*Regiscolia*) *maculata flavifrons* (Fabricius, 1775)

'Quadriocellar Deformity'
Figs 1–4

Material. ♀; E [Spain], Mallorca, Finca bei Polença, 09. 06. 2010, leg. D. Pawelek (UMB).

Measurements. Total body length: 38.0 mm; head width: 7.0 mm; forewing length: 32.5 mm; hind wing length: 16.0 mm; mesoscutal width: 5.8 mm.

Descriptive notes. The female specimen, which seems to be normal in every other respect, has four ocelli instead of the three which is the common state in the family and generally so across Aculeata. In this specimen the posterior ocelli are normal in position, form, and size whereas the anterior ocellus is represented by two, perfectly-formed ocelli that are disposed symmetrically, one on each side of the fissura frontalis by which they are separated (Figs 3, 4). The two aberrant anterior ocelli, which are located together in a single ocellar depression, are of the same size (maximum diameter) as the posterior ocelli whereas the anterior ocellus is about 1.1 times larger than the posterior ocelli in normal individuals (Figs 5, 6). This specimen shows no other malformations nor any traces of stylopisation.

Comments. This particular subspecies is represented in the collection of the UMB by an additional 10 males and 17 females from Italy (Liguria, South Tyrol, Apulia, and Sardinia), Spain (Catalonia and Ibiza), and France (Corsica). None of them has been collected at the same locality as the above female, nor does any show a similar malformation. Scoliids are moderately diverse, with approximately 560 species in 143 genera (Aguiar et al. 2013), and are often robust and large insects such that if teratologies are discovered they should be readily spotted.

Discussion

Among the recorded wild forms of pterygote insects with supernumerary ocelli, two different kinds of teratology are known – those resulting from duplication of the anterior ocellus, such as reported here, or of the lateral ocelli (Table 1). Whereas most records report a supernumerary anterior ocellus, Engel et al. (2014) reported an augochlorine bee with five ocelli – a single median ocellus and two sets of posterior paired ocelli. The only other account of malformed posterior ocelli is that of Ashmead (1880) who described a new species of the aphelinid genus *Aphytis* Howard with the type specimens having "three ocelli triangularly arranged, with two smaller red ones back of these". However, according to Rosen and DeBach (1979), "Ashmead apparently mistook the pigment spots, commonly seen in dry or slide-mounted specimens of small Chalcidoidea, for supernumerary ocelli." While their assessment is likely accurate, it remains unclear whether Ashmead might have found a true malformation given that his type material for the species in question has been lost (Rosen and DeBach 1979; and sources cited therein). These two records aside, all other accounts pertain to modifications of the anterior ocellus and result either from an apparent division of the structure or for its reappearance.

Figures 1–6. *Megascolia* (*Regiscolia*) *maculata flavifrons* (Fabricius). **1–4.** Quadriocellar female from Mallorca. **1.** Habitus in lateral view. **2.** Habitus in dorsal view. **3.** Head in dorsal view. **4.** Ocellar area. **5–6.** Normal female from Ibiza. **5.** Head in dorsal view. **6.** Ocellar area. Photos: Matthias Haase.

Supernumerary ocelli have been reported from Diptera, Orthoptera, and Hymenoptera (Table 1), but since these particular deformations are not as abundant as gynandromorphs a proper name has never been established for them. Thus they appear under quite different 'labels'

in the literature. It was perhaps the naturalist explorer Henry Walter Bates (1825–1892) who first recorded an observation of supernumerary ocelli, which he dubbed a "twin ocellus" (Bates 1863; the same term was used later by Brent 1886). Other authors described this unusu-

Table 1. List of recorded aberrant insect specimens with supernumerary ocelli from nature. Records are for each kind of aberration within a sex for a given species or subspecies. Thus, records of multiple or additional individuals with an identical teratology for a given sex and species are combined (citations for the individual accounts provided), while different teratologies for a species are listed individually. The numbering system for ocellar counts is formatted as: total # of ocelli (# of anterior ocelli + # of lateral ocelli) typical # of ocelli. All formicid records were for the worker caste (note that for some of the myrmecines listed the worker may have a normally reduced number of ocelli when compared with the gyne and so the total number listed is for the caste reported, and even major and minor workers may differ in their total number of ocelli). Generic and specific names have been updated to their current classification.

Order	Family	Species/subspecies	# of ocelli	Sex	References
Hymenoptera	Aphelinidae	*Aphytis flavus* (Ashmead) ‡	5 (1+2/2) 3	♀	Ashmead 1880
	Formicidae	*Acromyrmex coronatus* (Fab.)	2 (2+0) 0	♀	Weber 1947
		Atta cephalotes (L.) §	2 (2+0) 3	♀	Bates 1863, Wheeler 1936, Weber 1947
			3 (2+1) 3	♀	Wheeler 1936
			4 (2+2) 3	♀	Wheeler 1936, Weber 1947
		Atta laevigata (Smith)	4 (2+2) 3	♀	Weber 1947
		Atta sp.	2 (2+0) ?	♀	Brent 1886
		Cephalotes atratus (L.)	4 (2+2) 3	♀	Wheeler 1936
		Carebara diversus laotinus (Santschi)	2 (2+0) 1	♀	Wheeler 1936
	Halictidae	*Caenaugochlora inermis* (Vachal) ∣	5 (1+2/2) 3	♀	Engel et al. 2014
	Scoliidae	*Megascolia maculata flavifrons* (Fab.)	4 (2+2) 3	♀	Herein
	Tenthredinidae	*Tenthredo semirubra* (Norton)	4 (2+2) 3	♂	Smulyan 1923
		Hemichroa crocea (Geoffrey)	4 (2+2) 3	♀	Moller 1975
Orthoptera	Acrididae	*Melanoplus differentialis* (Thomas)	4 (2+2) 3	–	Glasgow 1925
		Melanoplus d. differentialis (Thomas)	4 (2+2) 3	♂	Slifer 1960
		Melanoplus d. nigricans Cockerell	4 (2+2) 3	♀/♂	King and Slifer 1965
		Melanoplus femurrubrum (DeGeer)	4 (2+2) 3	–	Blackman 1912
Diptera	Calliphoridae	*Calliphora grahami* Aldrich	4 (2+2) 3	♂	Hori et al. 1967
	Drosophilidae †	*Drosophila melanogaster* Meigen	4 (2+2) 3	–	Waddington et al. 1942, Baker et al. 1985

† The records of supernumerary ocelli in specimens of *Drosophila* Fallén are based on laboratory manipulations and not wild forms. Thus, we made no attempts for an exhaustive literature search for this genus and only report a couple here as examples.

‡ As discussed in the text, the supernumerary ocelli described by Ashmead (1880) are likely misinterpreted pigment spots (Rosen and DeBach 1979).

§ Weber (1947, footnote) discussed the taxonomic affinities of the specimens reported in Bates (1863). The following subspecies of *Atta* reported by Wheeler (1936) and Weber (1947), *Atta cephalotes integrior, A. c. isthmicola*, and *A. c. opaca*, are synonyms of *Atta cephalotes* (Borgmeier 1959). However, the subspecies "*Atta cephalotes gorgo*" (in Wheeler 1936), seems to be unavailable as the paper Wheeler alludes to for its formal description never appeared and the name was not treated by Borgmeier (1959) (Wheeler died in 1937 of a sudden heart attack and likely never had the opportunity to complete his work).

∣ This is the only validated record of a wild form where the lateral ocelli are affected, in all other listed records it is the median ocellus.

al development as either "supernumerary median ocelli" (Blackman 1912), "four [dorsal] ocelli" (Smulyan 1923, Glasgow 1925, Hori et al. 1967), "para-median ocelli" (Glasgow 1925), "binary anterior ocelli" (Wheeler 1936, Weber 1947), "two median ocelli" (Blackman 1912, Slifer 1960, King and Slifer 1965, Hori et al. 1967), or "abnormal median ocelli" (King and Slifer 1965). The term 'supernumerary ocelli' seems appropriate for the general class of teratologies involving the duplication of ocelli, with individual malformations dubbed by the number involved (e.g., quadriocellar, quintocellar). Once a more sizeable number of these deformations are documented a more systematic classification can be established.

A casual perusal of the list of occurrences of supernumerary ocelli (Table 1) would give the impression that some groups are more prone than others toward developing such teratologies. All records of ants with supernumerary ocelli are from the subfamily Myrmicinae, and particularly the leaf-cutting ants (Attini), whereas all recorded Orthoptera are from a single genus (*Melanoplus* Stål). While on the surface this is interesting, it perhaps reflects more the interest of those working on such groups. For example, species of *Melanoplus* are some of the most intensely studied of agricultural pests, and Weber was particularly interested in attines during the course of his myrmecological career, perhaps accounting for the fact that all of his records stem from that one tribe (Weber 1947). Indeed, even Wheeler's records stemmed largely from taxonomic work he was undertaking on the genus *Atta* Fabricius (Wheeler 1936, p. 188). Until more extensive studies into the occurrence of such teratologies are completed, it is impossible to say whether particular clades are more susceptible or not. In order to get an idea of the relative abundance of such malformed specimens we contacted several colleagues and asked for similar observations in their respective

groups of Hymenoptera: Stephan Blank, Symphyta; Andrew Polaszek, Platygastroidea and Chalcidoidea; Heinrich Wolf, general Aculeata; Celso O. Azevedo, Bethylidae; Denis J. Brothers, Mutillidae, Bradynobaenidae, Plumariidae, and Scolebythidae; James M. Carpenter, Vespidae; James P. Pitts, Mutillidae and Pompilidae; Lynn S. Kimsey, Tiphiidae, Chrysididae, and apoid wasps; Michael Ohl, apoid wasps; Fritz Gusenleitner, Andrenidae. Since none had come across a similar malformation, we assume that the low number of published records generally reflects the rarity of this kind of teratology in Hymenoptera.

The presence of supernumerary ocelli is almost certainly not the result of mutation, and therefore not present in the genetic makeup of the individual or heritable, instead resulting from errors in the developmental process and formation of adult tissues. That said, for at least one report in *Melanoplus*, King and Slifer (1965) found that their quadriocellar individuals successfully reproduced and that a "large proportion" of the offspring were similarly quadriocellar in condition. This suggests that for some cases there might be a genetic component, although the general rarity of supernumerary ocelli in other groups tends to suggest that such apparent heritability is far from the norm. What is remarkable is that these aberrant individuals are often captured while carrying on otherwise seemingly normal lives, a particularly remarkable fact given that ocelli are implicated in light responses and orientation (e.g., Taylor 1981a, 1981b, Schuppe and Hengstenberg 1993, Warrant 2006, Berry et al. 2006, Viollet and Zeil 2013). The scoliid wasp reported herein was perfectly developed in all other respects, and it does not seem that it had to deal with negative effects caused by the supernumerary ocellus. The consequences of malformations such as this are hardly predictable given that a complete understanding of ocellar function remains elusive (e.g., Wilson 1978, Goodman 1981, Stange et al. 2002).

Investigating the supernumerary median ocelli one is tempted to interpret such a malformation as an individual evolutionary throwback considering the hypothesis on the evolution of the median ocellus in insects. Snodgrass (1935) surmised that the median ocellus was formed by the fusion of two primitive anterior ocelli, and according to Paulus (1979) the anterior ocellus is homologous to the anterior pair of *Nauplius* eyes in Crustacea and the pair of median ocelli in *Limulus*. Indeed, the fusion hypothesis of Snodgrass has considerable support (Mizunami 1994), and in various groups the paired origin remains visible during ontogeny (e.g., Patten 1887, Viallanes 1887, Mobbs 1979). Even in adults the median ocellus retains its paired innervation, while the lateral ocelli are singly innervated (e.g., Leydig 1864, Hesse 1901, Imms 1948). It seems plausible that due to some unknown disruption during development the fusion has failed in those individuals with two anterior ocelli. However, it is quite obvious that significantly more information is needed, including extensive documentation of further occurrences of supernumerary ocelli. It is hoped that this account will spur

entomologists to pay greater attention to the occurrence of such teratologies and to put them on record. The scarcity of literature reports of aberrant insect specimens assuredly does not reflect their immense diversity nor their abundance in natural history collections.

Acknowledgements

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Two new species of the genus *Cophixalus* from the Raja Ampat Islands west of New Guinea (Amphibia, Anura, Microhylidae)

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Abstract

Based on morphological and bioacoustic traits, two new species of the microhylid genus *Cophixalus* Boettger, 1892 are described from the Raja Ampat Islands off the western tip of New Guinea. Both are small (SUL < 23 mm), slender, scansorial species that are morphologically most similar to *Cophixalus tetzlaffi* Günther and *C. monosyllabus* Günther, two congeners also known only from far western New Guinea. Their description brings the total number of *Cophixalus* known from New Guinea and surrounding islands to 46, and the total number from western New Guinea (Papua and West Papua Provinces including the Raja Ampat Islands) to 10. One *Cophixalus* specimen from Salawati Island is considered a hermaphrodite because it has a well-developed vocal sac and vocal slits, but also has an ovary containing eggs in an advanced developmental stage. This frog uttered advertisement calls that did not differ from calls of conspecific males. The first evidence of the genus *Cophixalus* from Misool Island is also documented.

Introduction

The frog genus *Cophixalus* comprises 63 recognised species (Frost 2014). Of these, 19 occur in north-eastern Australia, 35 are known only from Papua New Guinea (many of them described in recent years; for example from Kraus and Allison (2009) and Kraus (2012)), five are known only from western New Guinea (Papua and West Papua Provinces of Indonesia), three are recorded from both Papua New Guinea and Indonesian New Guinea, and one species seems to be endemic to the island of Halmahera about 300 km off the western tip of New Guinea. Although *Cophixalus montanus* (Boettger) from Halmahera has been known since 1895, five of the species known from the western part of New Guinea (on Yapen Island, on the Wandammen Peninsula, and on the

Bomberai Peninsula) were described only recently (Günther 2003, 2006, 2010, Kraus 2012) and many additional *Cophixalus* species probably await discovery in western New Guinea. Here we describe two new *Cophixalus* species from the Raja Ampat Islands off the western tip of New Guinea.

Material and methods

Frogs were generally located at night by tracking their advertisement calls, and selected specimens were photographed in life prior to preservation. Tissue probes from liver were taken from some specimens and stored in about 96% ethanol to enable DNA sequencing. All specimens were fixed in 10% formalin and transferred to 75% ethanol for permanent storage.

Measurements were taken with a digital calliper (> 10 mm) or with a binocular dissecting microscope fitted with an ocular micrometer (< 10 mm) to the nearest 0.1 mm:

SUL – snout-urostyle length: from tip of snout to distal tip of urostyle-bone. SUL is subject to lower measurement error than the traditionally used snout-vent length (SVL) (R. Günther, pers. obs.) so we have used it here. However both measurements are very similar, SUL being at most 0.5–1.0 mm shorter – if at all - than SVL in small frogs. We therefore directly compare SUL measurements reported here with SVL measurements of congeners presented in the literature. TL – tibia length: external distance between knee and ankle; TaL – length of tarsus: external distance, tarsal and ankle joints held at a right angle; T4L – length of fourth toe: from tip of toe to proximal end of inner metatarsal tubercle; T1D – transverse diameter of disc of first toe; T4D – transverse diameter of disc of fourth toe; F3L – length of third finger from tip to proximal margin of palmar tubercles; F3D – transverse diameter of disc of third finger; F1D – transverse diameter of disc of first finger; T1L – length of first toe: distal of inner metatarsal tubercle; MTL – length of inner metatarsal tubercle; HL – head length: from tip of snout to posterior margin of tympanum; HW – head width, taken in the widest point; SL – snout length: from an imaginary line that connects the centres of eyes to tip of snout; END – distance from anterior corner of orbital opening to centre of naris; IND – internarial distance between centres of external nares; ED – eye diameter: from anterior to posterior corner of orbital opening; TyD – horizontal diameter of tympanum.

Advertisement calls were recorded with a Sony™ WM D6C Professional Walkman tape recorder and a Sennheiser ME66 shotgun microphone and analysed with Avisoft-SAS Lab Pro software. All specimens are stored in the collection of the Museum Zoologicum Bogoriense (MZB) in Cibinong (Bogor), Indonesia and bear registration numbers of that institution.

All statistical calculations were done with the program Statgraphics Centurion Version 15.2.14 (Statpoint Technologies, Inc., Warrenton, Virginia, USA). All p-values in the running text and in the tables are calculated by the non-parametric Mann-Whitney (Wilcoxon) Test for comparison of medians. All mean values are arithmetic means. Box-whisker plots are used to illustrate comparative mensural data.

Voucher specimens, including types, of the genus *Cophixalus* that were studied for comparative purposes are listed in the papers by Richards et al. (1992), Günther (2003, 2006, 2010), Richards and Oliver (2007, 2010), Günther and Richards (2011), and Günther et al. (2014). Additional comparative information was taken from original species descriptions and recompiled treatises (Zweifel 1956a, 1956b, 1962, Tyler 1963, Menzies 2006, Kraus and Allison 2006, 2009, Kraus 2012).

Results

Cophixalus rajampatensis sp. n.

http://zoobank.org/7F007383-1C83-4395-A501-72F70A072514

Holotype. MZB Amph.12094 (field number, FN: SJR 7638); adult male collected at unnamed camp near Urbinasopen Village, Waigeo Island, Raja Ampat Islands, West Papua Province, Indonesia (00°20.231'S, 131°15.528'E) (Fig. 1) on 12/06/2005 by S. Richards, B. Tjaturadi and K. Krey.

Paratypes. MZB Amph.12097 (FN: SJR 7615), MZB Amph.12169 (FN: 7617), same data as holotype; MZB Amph.12098 (FN: SJR 7575), MZB Amph.12095 (FN: 7592) collected at Warinkabom Camp, Batanta Island, Raja Ampat Islands, West Papua Province, Indonesia (00°50.196'S, 133°43.318'E on 7 and 9/06/2005 respectively by S. Richards, B. Tjaturadi and K. Krey; MZB Amph.12092 (FN: SJR 7551), MZB Amph.12096 (FN: SJR 7559), MZB Amph.12163 (FN: 7516) collected at Waire Camp, Batanta Island, Raja Ampat Islands, West Papua Province, Indonesia (00°50.384'S, 130°31.534'E on 6-7/06/2005 by S. Richards, B. Tjaturadi and K. Krey. All seven paratypes are adult males.

Diagnosis. With an SUL of 17.6–19.5 mm in eight adult males, the new species is one of the smaller species of *Cophixalus*. Body slender, dorsum smooth except for occasional scattered tubercles and partly interrupted dorsolateral skin folds; legs moderately long (TL/SUL 0.48–0.52), third toe clearly longer than fifth. Toe and finger discs distinct, those of fingers slightly larger than, or equal in size to, those of toes (T4D/F3D 0.8–1.0), except that of first finger which is scarcely wider than penultimate phalanx. Call a short train of peeps or whistles, each with a mean duration of 178 milliseconds (ms). Number of notes (= peeps) per call 2–5 (mean 3.42), repeated at a rate of 3.3–4.6 notes/s (mean 3.96) and dominant frequency 3.7 kHz.

Description of the holotype (Fig. 2a–d). For measurements see Table 1. Head much broader than long (HL/HW 0.78), canthus rostralis rounded; loreal region flat; snout protruding in profile and nearly rounded in dorsal view; nostrils directed laterally; horizontal eye diameter greater than eye-naris distance; tympanic annulus scarcely visible, tympanum less than half eye diameter (TyD/ED 0.38), supratympanic fold short; internarial distance greater than distance between eye and naris (END/IND 0.75); tongue large, broadened posteriorly with a small notch, posterior and lateral margins free; prepharyngeal fold not serrated; long vocal slits on both sides of the tongue. Legs moderately long, no webbing between fingers or toes; disk of third finger wider than disk of fourth toe (T4D/F3D 0.89), disks of fingers II, III and IV about the same width as those of toes II, III and IV, first finger much smaller than other fingers, its disk only scarcely
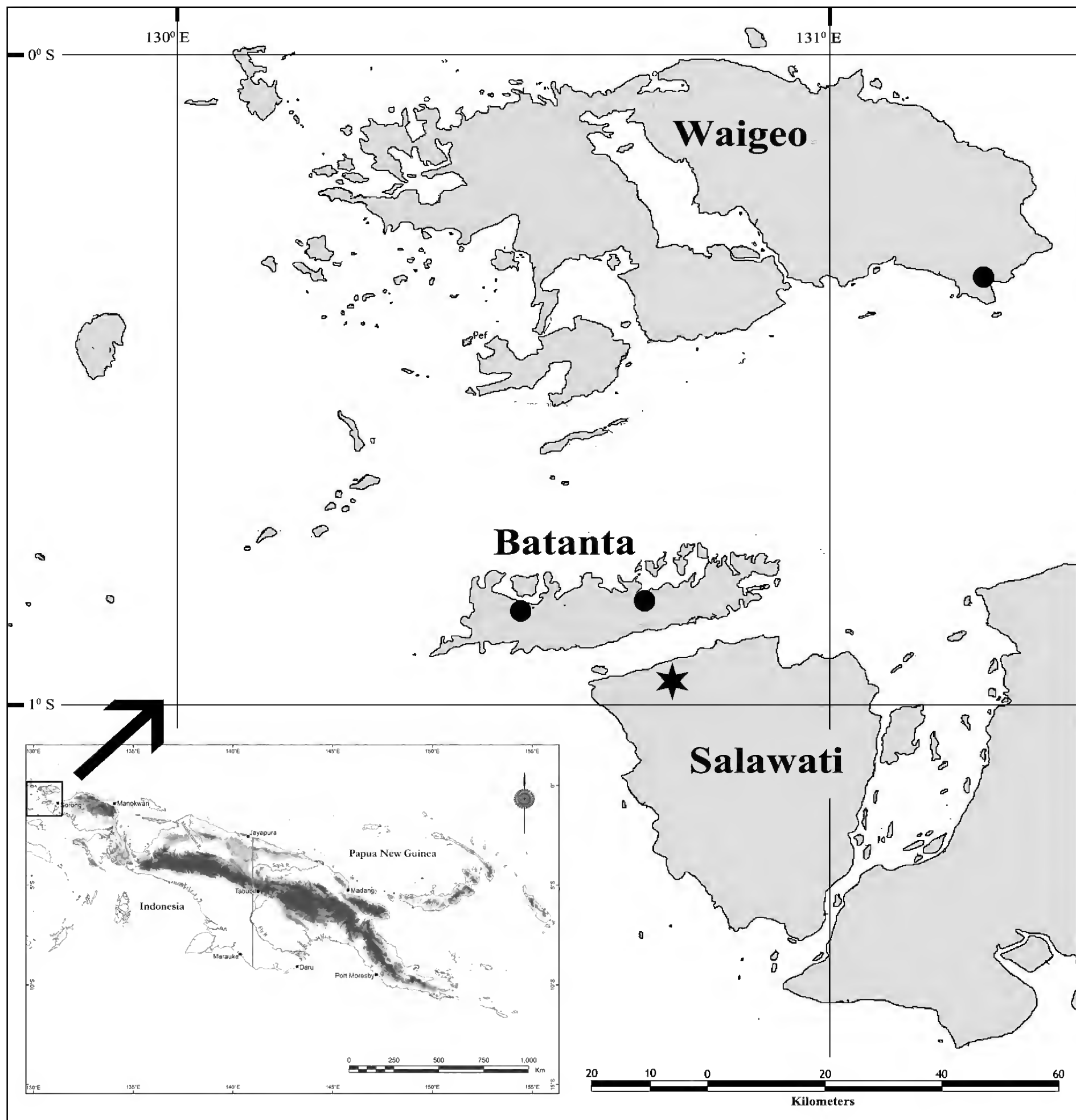
Figure 1. Distribution of *Cophixalus rajampatensis* sp. n. (circles) and *Cophixalus salawatiensis* sp. n. (stars) in the Raja Ampat Islands off western New Guinea.

wider than the penultimate phalanx; relative length of fingers 3>4=2>1; third toe clearly longer than the fifth, disk of first toe slightly smaller than disk of fifth toe, disks of remaining toes clearly wider than those of first and fifth toe; all finger and toe disks with terminal grooves; relative length of toes 4>3>5>2>1, subarticular, metatarsal and metacarpal tubercles not or only scarcely developed. Some tubercles on flanks, posterior back and dorsal shanks and irregular and partly interrupted dorsolateral skin folds from near eye to inguinal region. All remaining dorsal, lateral, and ventral surfaces smooth, except a gular fold between insertion of the fore limbs that indicates posterior margin of the vocal sac.

In preservative dorsal surfaces of head, body and extremities mid-brown, most tubercles with dark base and light tip, dorsolateral folds are accompanied by dark brown stripes. A fine light middorsal line from snout to anal opening. Body sides blotchy; a conspicuous whitish fleck extends from posterior of eye through tympanum to arm insertion. Ventral surfaces of extremities off-white with dense irregularly shaped brown spots; belly off-white with a few brown spots anteriorly, throat and chest intensely brown; region around anal opening blackish. In life dorsal surfaces bronze-brown with a few dark brown flecks laterally; tubercles pink. Conspicuous is an off-white stripe from tip of snout along canthus rostralis

Table 1. Body measurements and body ratios of the type series of *Cophixalus rajampatensis* sp. n. MZB 12094 is the holotype, all types are adult males, all measurements in mm. Inv.-No = Registration number; MZB = Museum Zoologicum Bogoriense; SD = Standard deviation; explanation of measurements in "Material and methods".

Inv.-No	MZB 12092	MZB 12094	MZB 12095	MZB 12096	MZB 12097	MZB 12098	MZB 12163	MZB 12169	Mean ± SD
SUL	18.5	18.6	17.6	17.8	19.5	17.9	18.3	18.1	18.3±0.56
TL	9.6	9.3	8.8	8.9	9.4	9.0	9.2	9.3	
TaL	5.9	6.0	5.3	5.7	5.8	5.9	5.9	5.8	
L4T	8.7	8.5	7.9	8.5	8.6	8.7	8.4	8.6	
T4D	0.8	0.8	0.8	0.8	0.9.	0.8	0.7	0.8	
L3F	4.5	4.3	3.8	4.2	4.1	4.2	4.3	4.6	
F3D	1.0	0.9	0..8	0.9	0.9	0.8	0.8	0.9	
F1D	0.3	0.25	0.25	0.3	0.2	0.25	0.2	0.3	
T1D	0.4	0.4	0.4	0.4	0.35	0.35	0.3	0.4	
HL	5.7	5.3	5.1	5.2	5.5	5.1	5.6	5.8	
HW	6.9	6.8	6.5	6.4	6.7	6.2	6.6	6.5	
SL	2.8	2.8	2.5	2.4	2.7	2.5	2.6	2.7	
END	1.7	1.5	1.5	1.3	1.4	1.5	1.6	1.5	
IND	2.0	2.0	1.8	1.7	2.1	2.0	1.9	2.0	
ED	2.2	2.4	1.9	2.1	2.3	2.1	2.0	2.2	
TyD	1.1	0.9	0.7	0.8	1.0	0.9	0.9	0..9	
L1T	1.4	1.3	1.5	1.2	1.2	1.2	1.3	1.4	
LMT	0.9	0.8	0.7	0.8	0.7	0.7	0.9	0.9	
TL/SUL	0.52	0.50	0.50	0.50	0.48	0.50	0.50	0.51	0.50±0.01
TaL/SUL	0.32	0.32	0.30	0.32	0.307	0.33	0.32	0.32	0.32±0.01
L4T/SUL	0.47	0.46	0.45	0.48	0.44	0.49	0.46	0.48	0.47±0.02
L3F/SUL	0.24	0.23	0.22	0.24	0.21	0.23	0.23	0.25	0.23±0.012
F3D/SUL	0.054	0.048	0.045	0.051	0.046	0.045	0.044	0.050	0.048±0.004
T4D/SUL	0.043	0.043	0.045	0.045	0.046	0.045	0.038	0.044	0.044±0.003
T4D/F3D	0.80	0.89	1.00	0.89	1.00	1.00	0.88	0.89	0.92±0.074
F1D/SUL	0.016	0.013	0.014	0.017	0.010	0.014	0.011	0.017	0.014±0.002
T1D/SUL	0.022	0.022	0.023	0.022	0.018	0.020	0.016	0.022	0.021±0.002
T1D/F1D	1.33	1.60	1.60	1.33	1.75	1.40	1.50	1.33	1.48±0.16
LMT/L1T	0.64	0.62	0.47	0.67	0.58	0.58	0.69	0.64	0.61±0.069
HL/SUL	0.31	0.27	0.29	0.29	0.28	0.28	0.31	0.32	0.29±0.018
HW/SUL	0.37	0.37	0.37	0.36	0.34	0.35	0.36	0.36	0.36±0.011
HL/HW	0.83	0.78	0.78	0.81	0.82	0.82	0.85	0.89	0.83±0.036
SL/SUL	0.151	0.150	0.142	0.135	0.138	0.140	0.142	0.149	0.143±0.006
END/IND	0.85	0.75	0.83	0.76	0.67	0.75	0.84	0.75	0.78±0.061
ED/SUL	0.119	0.129	0.108	0.118	0.118	0.117	0.109	0.122	0.118±0.007
TyD/ED	0.50	0.38	0.37	0.38	0.43	0.43	0.45	0.41	0.41±0.062

and margin of upper eyelid and reaching to posterior eye margin (this off-white stripe has nearly disappeared in fixative), and a dark brown "face-mask" covering loreal and gular region up to insertion of upper arm. Iris silvery with orange parts anteriorly and posteriorly and many irregular dark brown spots.

Variation in the type series. Mensural variation for the type series is shown in Table 1. Basic colour and colour pattern elements of all preserved paratypes are fairly uniform and closely resemble the holotype. Characteristic for most paratypes is a lighter brown dorsum which is bordered by irregular blackish dorsolateral lines and dark brown upper flanks. Three paratypes exhibit a dark mid-dorsal line; none have a light mid-dorsal line like the holotype. Abdomen from unspotted to various degrees of spotted; throat and chest uniform dark brown or heavily

spotted; loreal, subocular and gular region middle to dark brown in fixative and blackish in life. All specimens with a large off-white postocular fleck which is bordered antero-dorsally by a smaller dark spot and ventrally by the posterior "face-mask". Six specimens have a small blackish spot above insertion of fore leg. The conspicuous pale canthal stripe that was present in all living specimens is very inconspicuous in the fixed ones.

Vocalisation. Most calling activity occurred at night after rain. Calls of three males (MZB 12095, 12096 and 12163) recorded at temperatures of approximately 26 °C were analysed. Calls each contained 2–5 (mean 3.4, SD 0.7) finely pulsed notes which sound like peeps or whistles (Fig. 3), and 4–6 calls were uttered in succession (a call series) with variable intervals between individual calls (4–20 s) and between call series (20 s to some
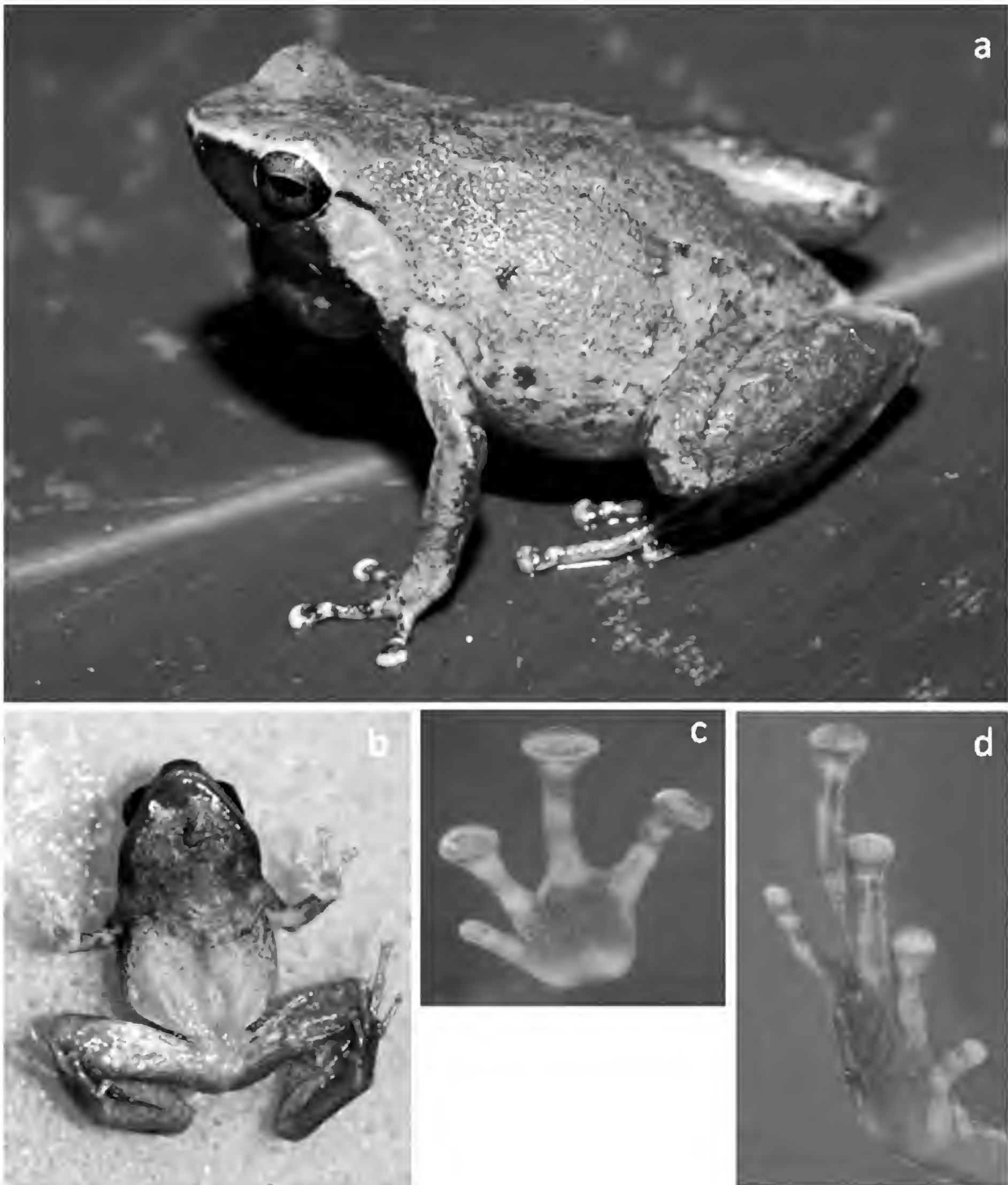
Figure 2. Holotype of *Cophixalus rajampatensis* sp. n. (**a**) dorsolateral view in life, (**b**) ventral view after preservation, (**c**) palmar view of left hand after preservation, (**d**) plantar view of right foot after preservation.

minutes). The shortest time between two successive calls within a series was 3.5 s. The call length was 0.40–1.54 s, mean 0.88 s, SD 0.22, n=52. Mean of means of note length was 178 ms, SD 10.4, range of means 158–202 ms, n=52; total range of note length 142–238 ms. Mean of means of internote length 115 ms, SD 34.9, range of means 68–172 ms, n=52; total range of internote intervals 63–179 ms. Mean note repetition rate was 3.96 notes/s, SD 0.37, range 3.3–4.6 notes/s, n=52. Frequencies are concentrated mainly between 3.4 and 4.0 kHz, with a dominant frequency of 3.7 kHz (Fig. 4). Most notes start explosively with nearly maximum amplitude, and volume may remain constant during the entire note but may also undulate, with the greatest volume mostly in the middle
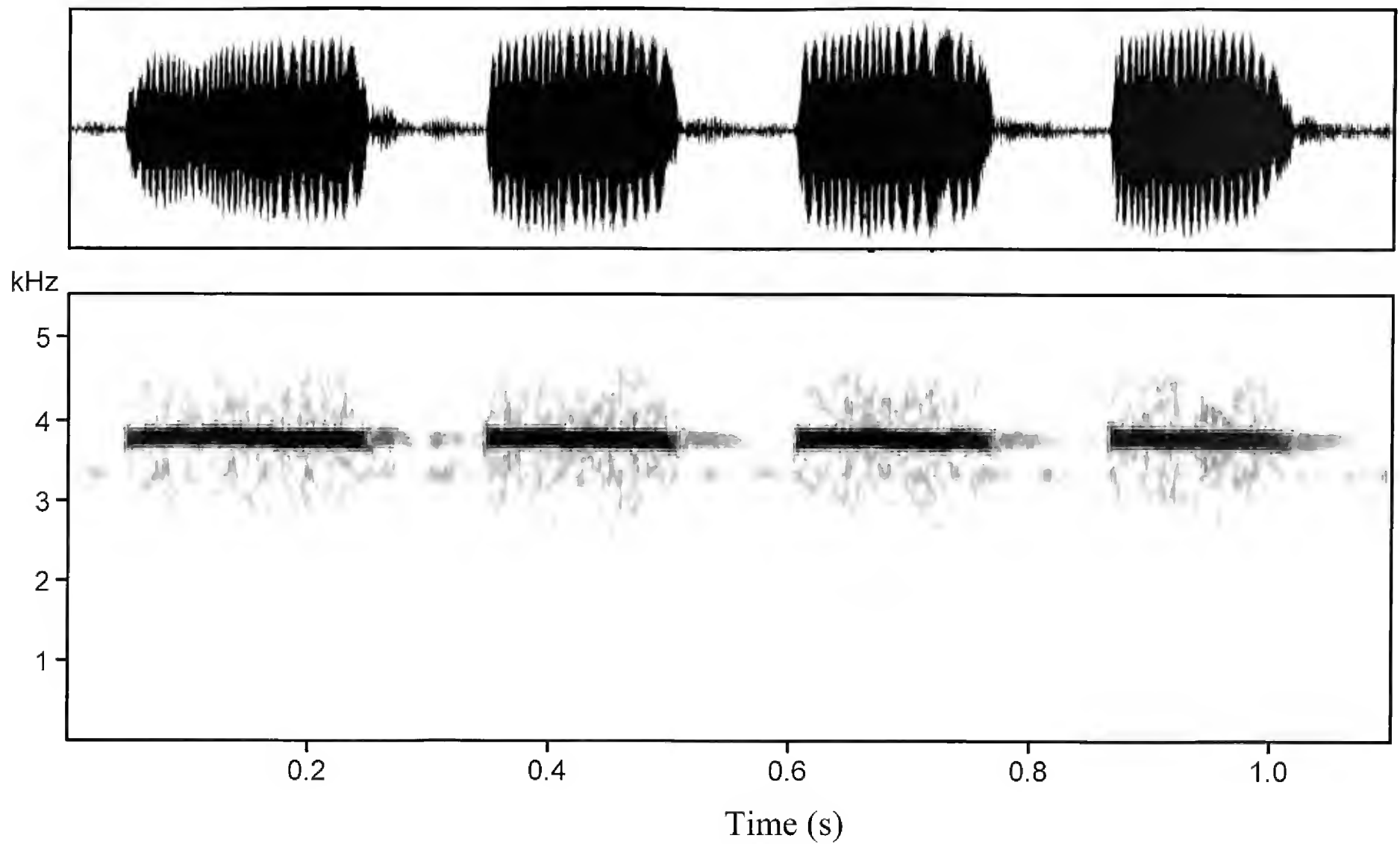
Figure 3. Wave form (above) and spectrogram (below) of an advertisement call of *Cophixalus rajampatensis* sp. n. consisting of four notes.
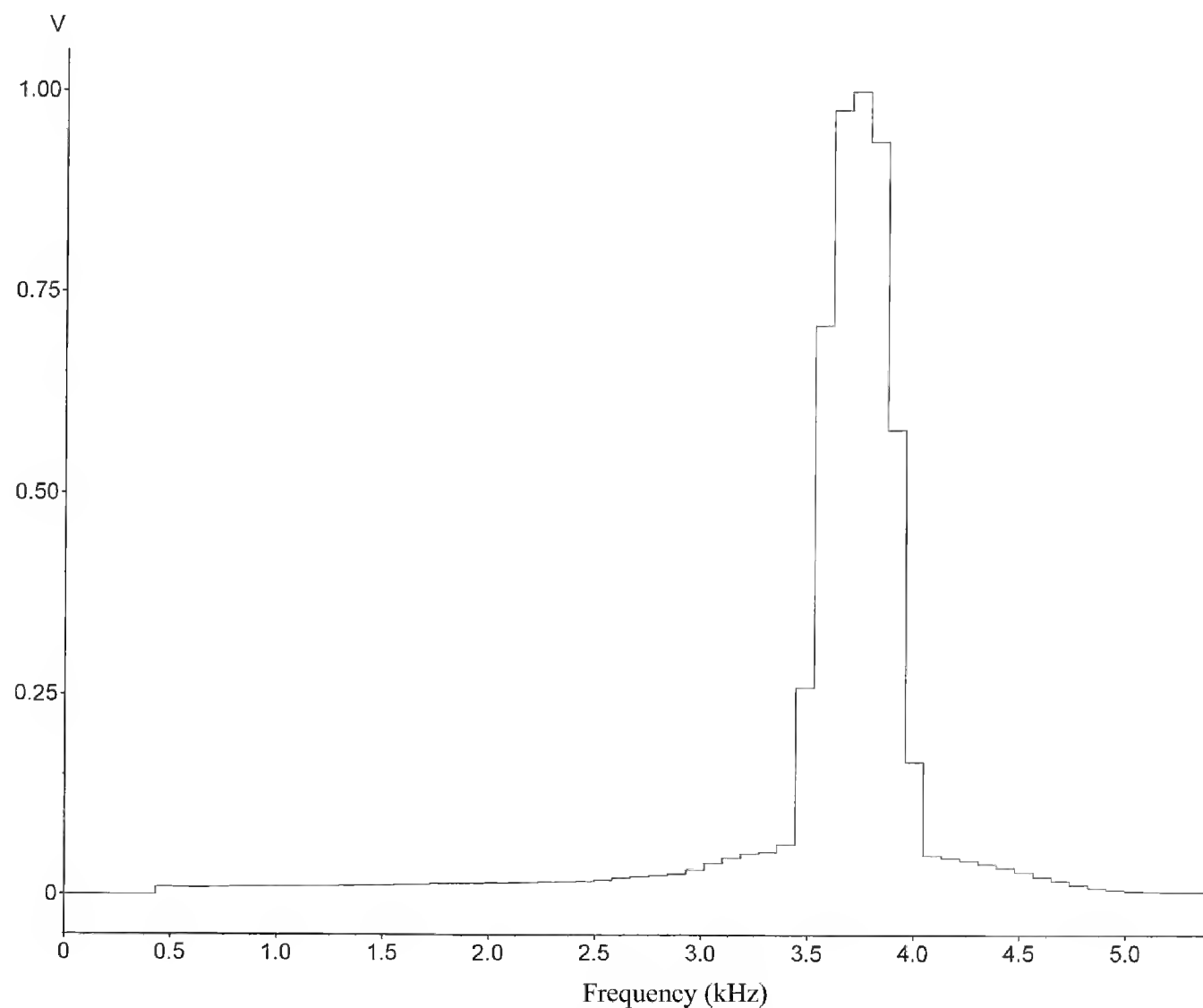

Figure 4. Power spectrum of an advertisement call of *Cophixalus rajampatensis* sp. n.

of the note. The end of the note has a less steep amplitude slope than the beginning. The first note of a call is nearly always the longest.

Distribution and ecological remarks. *Cophixalus ampatensis* sp. n. is currently known from two localities on Batanta Island and one locality on Waigeo Island, both

in the Raja Ampat Island group off western New Guinea (Fig. 1). It was found to be common in moderately to heavily logged lowland rainforest where males called from the surfaces of leaves in low foliage ~30 cm – 1 m above the ground after heavy rain at night. Intensive searches on nearby Salawati Island failed to detect this species there despite similar climatic conditions and high activity of other frogs on that island. A number of other faunal lineages show evidence of a disjunction between Salawati and Batanta (e.g. Oliver et al. 2008), reflecting the different geological histories of these islands within the Raja Ampat Island Group. Batanta shares much of its biogeographic history with Waigeo, and the deep Sagewin strait that separates them from Salawati appears to be a major barrier for at least some taxa. Together these observations suggest that *C. rajampatensis* sp. n. probably does not occur on Salawati, and may be endemic to Batanta and Waigeo Islands.

Etymology. The latinized specific epithet *rajampatensis* refers to the fact that the species occurs on the Raja Ampat Islands off the western tip of New Guinea.

Comparison with other species. *Cophixalus* species described from New Guinea and adjacent islands that can be immediately distinguished from *C. rajampatensis* (with males 17.6–19.5 mm) by their smaller adult size are (sizes presented below are for adult males unless otherwise indicated): *amabilis* Kraus (13.6–14.3 mm), *ateles* (Boulenger) (12–14 mm), *desticans* Kraus & Allison (13.1–16.2 mm), *humicola* Günther (14.5–16.2), *iovaorum* Kraus & Allison (13.2–16.0 mm), *kethuk* Kraus & Allison (12.4–13.5 mm), *linnaeus* Kraus & Allison (13.4–14.7 mm), *misimae* Richards & Oliver (15.5–16.1 mm), *phaeobalis* Kraus & Allison (15.3 mm), *timidus* Kraus & Allison (13.5–17.5 mm), *tomaiodactylus* Kraus & Allison (13.2–16.1 mm), *tridactylus* Günther (14.3–16.2), and *viridis* Günther, Richards & Dahl (15.8–16.2 mm). With an SVL of 15.7 mm the only known specimen of *Cophixalus pictus* Kraus is smaller than *C. rajampatensis* but its description was based on a rather poorly preserved (and presumed immature) male from the Bomberai Peninsula of West Papua Province (Kraus 2012) and, if immature, this distinction could disappear once adult material has been documented. However the new species also differs from *C. pictus* in a number of features that are unlikely to reflect the immature status of the holotype and only known specimen, including having (vs. lacking) a distinct supratympanic fold, in having longer legs (TL/SUL 0.48–0.52 vs. TL/SVL 0.47) and in having a very different ratio of internarial distance to eye-to-naris distance (END/IND 0.67–0.85 vs. 1.08 in *C. pictus*) (Kraus 2012). A further 15 species can be readily distinguished by their much larger size: *balbus* Günther (26–28 mm), *biroi* (Méhely) (to 27 mm; Zweifel 1979), *caverniphilus* Kraus & Allison (25.5.2–36.7 mm), *cheesmanae* Parker (to 31 mm; Zweifel 1979), *clapporum* Kraus (23.2–27.5 mm), *cryptotympanum* Zweifel (to 30 mm (both sexes)

at the type locality; to 40 mm elsewhere: Zweifel 1956, 1962, Menzies 2006), *cupricarenus* Kraus & Allison (23.4–28.7 mm), *kaindiensis* Zweifel (to 28 mm; Zweifel 1979), *montanus* (to 26 mm (sex not stated; Parker 1934), *nubicola* Zweifel (22.4–24.3 mm), *parkeri* Loveridge (to 30 mm), *riparius* Zweifel (to 45 mm) and *verrucosus* (Boulenger) (to 25 mm).

From the species with overlapping body sizes: *C. albolineatus* Kraus (16.8–20.5 mm), *C. interruptus* Kraus & Allison (16.6–18.7 mm), *C. melanops* Kraus & Allison (16.4–18.9 mm), *C. tagulensis* Zweifel (to 18 mm), *C. tenuidactylus* Günther & Richards (18.4–20.3 mm) and *C. verecundus* Zweifel & Parker (15–17 mm) the new species can be immediately distinguished by having finger discs of the same size or larger than toe discs (vs. smaller than the toe discs). *Cophixalus variabilis* Kraus & Allison (13.6–18.6 mm) has a tuberculate (vs. smooth) dorsum with extensive colour variation including longitudinal stripes in about 50% of specimens (lacking in *rajampatensis*). *Cophixalus bewaniensis* Kraus & Allison (15–17 mm) and *C. shellyi* Zweifel (~17 mm) have strongly reduced first fingers and *C. sphagnicola* Zweifel & Allison (15.8–18.5 mm), in contrast to *C. rajampatensis* sp. n., completely lacks discs on fingers and toes. *Cophixalus pipilans* Zweifel (16.1–18.5 mm) has longer legs (TL/SVL >0.53 vs. TL/SUL 0.48–0.52) and calls with 20–33 (vs. 2–5) peeping notes. *Cophixalus daymani* Zweifel (to 21.7 mm [females]) is distinguished by very short hind legs (TL/SUL less than 0.38 vs. 0.48–0.52) and occuring higher than 2200 m a.s.l. *Cophixalus nexipus* Kraus (18.9–22.7 mm) differs by having basal webbing on toes and advertisement calls consisting of a single, long note lasting more than one second (vs. 2–5 short, finely pulsed peeps). *Cophixalus wempi* Richards & Oliver (15.5–16.1 mm) has (vs. lacks) a distinct spiniform tubercle above the eyelid and has advertisement calls with 28–33 (vs. 2–5) peeping notes.

On the basis of external morphology *C. tetzlaffi* and *C. monosyllabus* exhibit most similarities to *C. rajampatensis* sp. n. and are compared in more detail. With an SUL of 20.0–22.7 mm, mean 21.3 mm, SD 0.92, n=8, *C. tetzlaffi* is larger than *C. rajampatensis* sp. n. with an SUL of 17.6–19.5 mm, mean 18.3, SD 0.60, n=8 (Fig. 5). Relative tibia length of *C. tetzlaffi* is significantly greater than that of *C. rajampatensis* sp. n. (TL/SUL 0.51–0.53 vs. 0.48–0.52; p=0.008, Fig. 6), although the values overlap. There are also significant differences between these species (no overlap in the values) in their advertisement calls; note length of *C. tetzlaffi* (347–518 ms) is longer than that of *C. rajampatensis* sp. n. (142–238 ms); internote interval in the former is longer (186–299 ms) than in the latter (63–179 ms); and note repetition rate in the former is slower 1.5–1.8 notes/s than in the latter 3.3–4.6 notes/s.

Ten male specimens of *C. monosyllabus* have a larger snout-urostyle length than eight males of *C. rajampatensis* sp. n. and there is no overlap: 20.6–24.3 mm, mean 22.9 mm (SD 1.04) vs. 17.6–19.5 mm, mean 18.3 mm
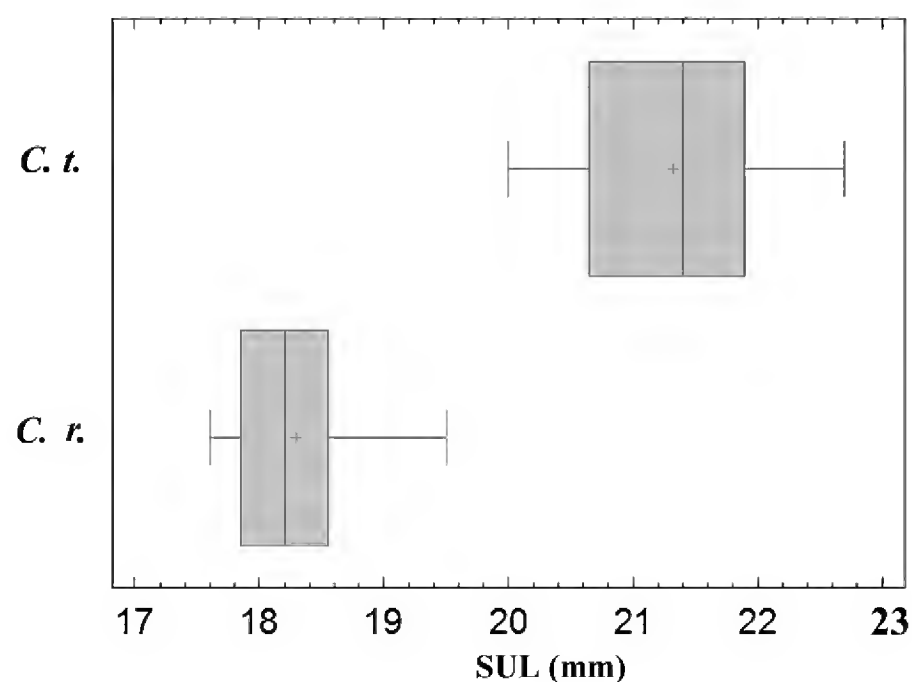
Figure 5. Box-Whisker-Plot of snout-urostyle length in mm (SUL) of eight males of *Cophixalus tetzlaffi* (*C.t.*) and eight males of *C. rajampatensis* sp. n. (*C. r.*). The horizontal blue line represents the range, the vertical blue line represents the median, the box represents the interquartile (50% of the values) and the red cross indicates the arithmetic mean.
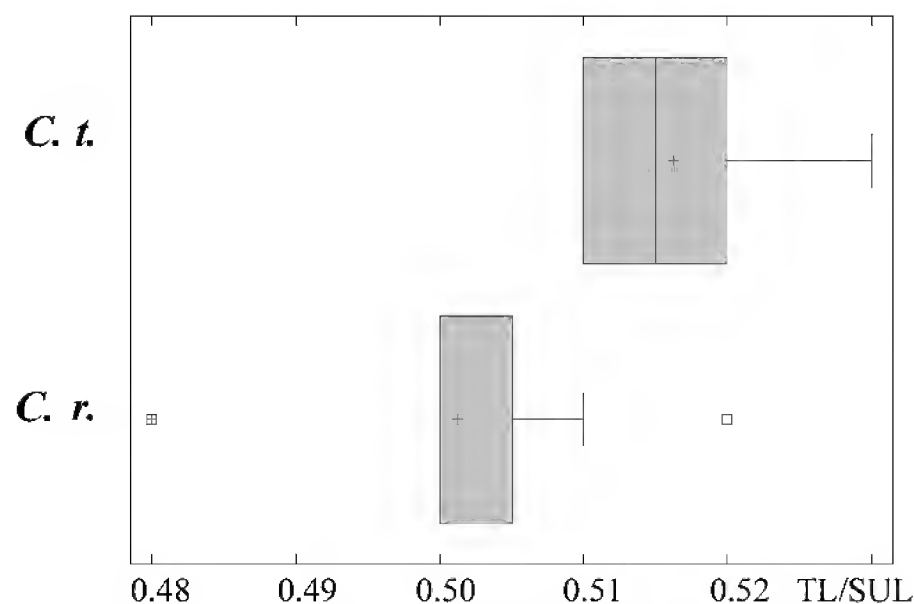

Figure 6. Box-Whisker-Plot of the ratio TL/SUL of eight males of *Cophixalus tetzlaffi* (*C.t.*) and eight males of *C. rajampatensis* sp. n. (*C. r.*). Small squares are so-called outliers.

Figure 7. Box-Whisker-Plot of the ratio F3D/SUL of ten males of *Cophixalus monosyllabus* (*C.m.*) and eight males of *C. rajampatensis* sp. n. (*C. r.*).

Figure 8. Box-Whisker-Plot of the ratio HW/SUL of ten males of *Cophixalus monosyllabus* (*C.m.*) and eight males of *C. rajampatensis* sp. n. (*C. r.*).

(SD 0.56) and also differ significantly in the following body ratios (*monosyllabus* vs. *rajampatensis*): F3D/SUL (0.055–0.067 vs. 0.044–0.054: Fig. 7), F3L/SUL (0.26–0.28 vs. 0.21–0.25), F1D/SUL (0.017–0.031 vs. 0.010–0.017), HW/SUL (0.37–0.42 vs. 0.34–0.37: Fig. 8) and END/IND 0.84–0.96 vs. 0.67–0.85, p=0.001). Moreover, *C. monosyllabus* and *C. rajampatensis* sp. n. differ in their advertisement calls. Calls of the former consist of single notes while calls of the latter always contain 2–5 notes, mean 3.4 notes, SD 0.7.

Cophixalus salawatiensis sp. n.

http://zoobank.org/66A11935-9819-4272-B3E1-CA2E04159C61

Holotype. MZB Amph.12165 (FN: SJR 7797); adult male collected at Weybya camp, Salawati Island, Raja Ampat Islands, West Papua Province, Indonesia (00°57.383'S,
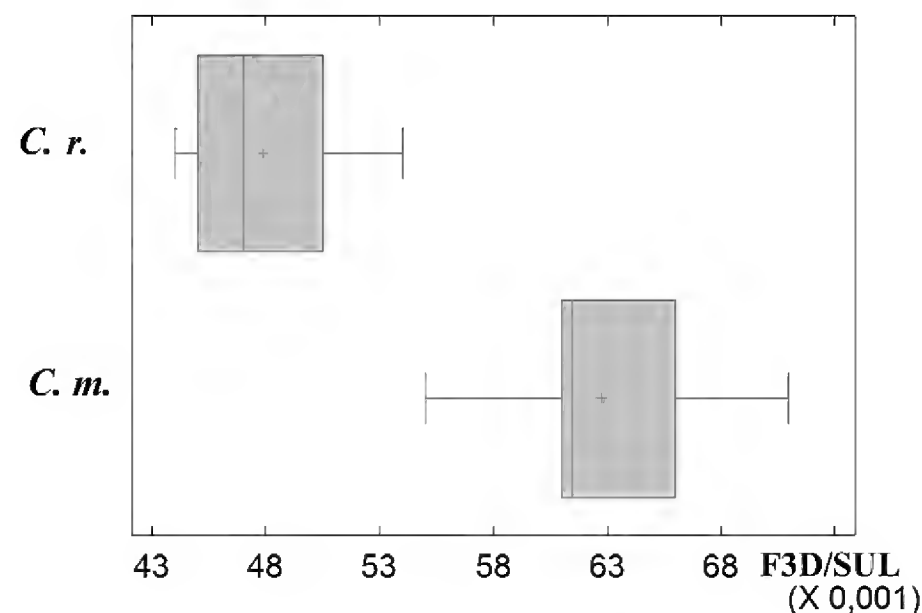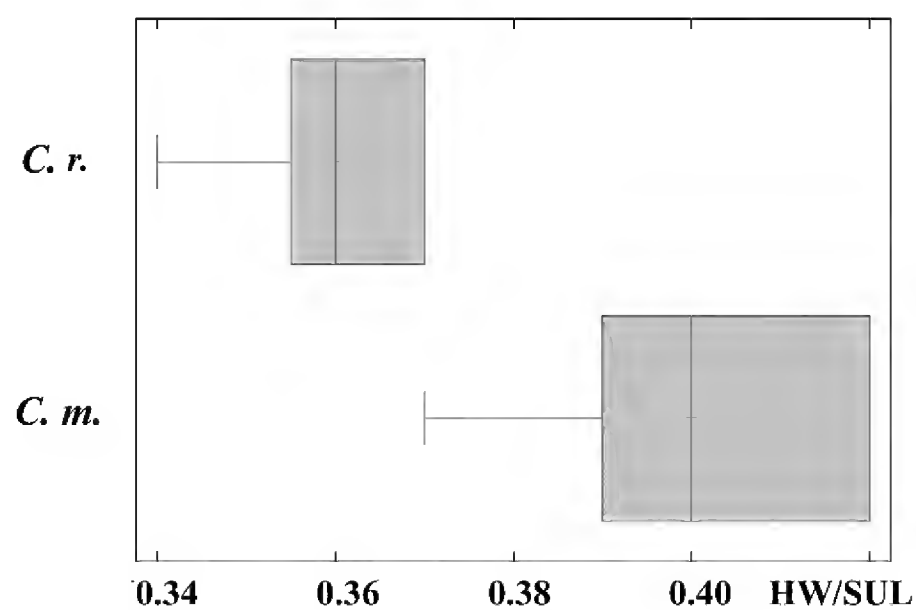
130°47.060'E on 27/06/2005 by S. Richards, B. Tjaturadi and K. Krey.

Paratypes. MZB Amph.12157 (FN: SJR 7731), MZB Amph.12159 (FN: 7756), MZB Amph.12161 (FN: SJR 7755), MZB Amph.12162 (FN: SJR 7730), MZB Amph.12166 (FN: SJR 7757), MZB Amph.12167 (FN: 7772), MZB Amph.12168 (FN: 7760), MZB Amph.12170 (FN: SJR 7795), and MZB Amph.12171 (FN: SJR 7796), same data as for holotype, collected between 24-27/06/2005. All specimens are adult males and MZB Amph.12166 is considered a hermaphrodite (see below).

Diagnosis. *Cophixalus salawatiensis* sp. n. can be distinguished from all congeners by a combination of the following characters: Body small (SUL of 10 males 19.6–22.5 mm), slender, dorsum smooth except for scattered tubercles, head laterally with a distinct dark 'face mask' (grey in life); legs moderately long (TL/SUL 0.49–0.53), third toe clearly longer than fifth, no webbing between digits. Toe and finger discs distinct, those of fingers
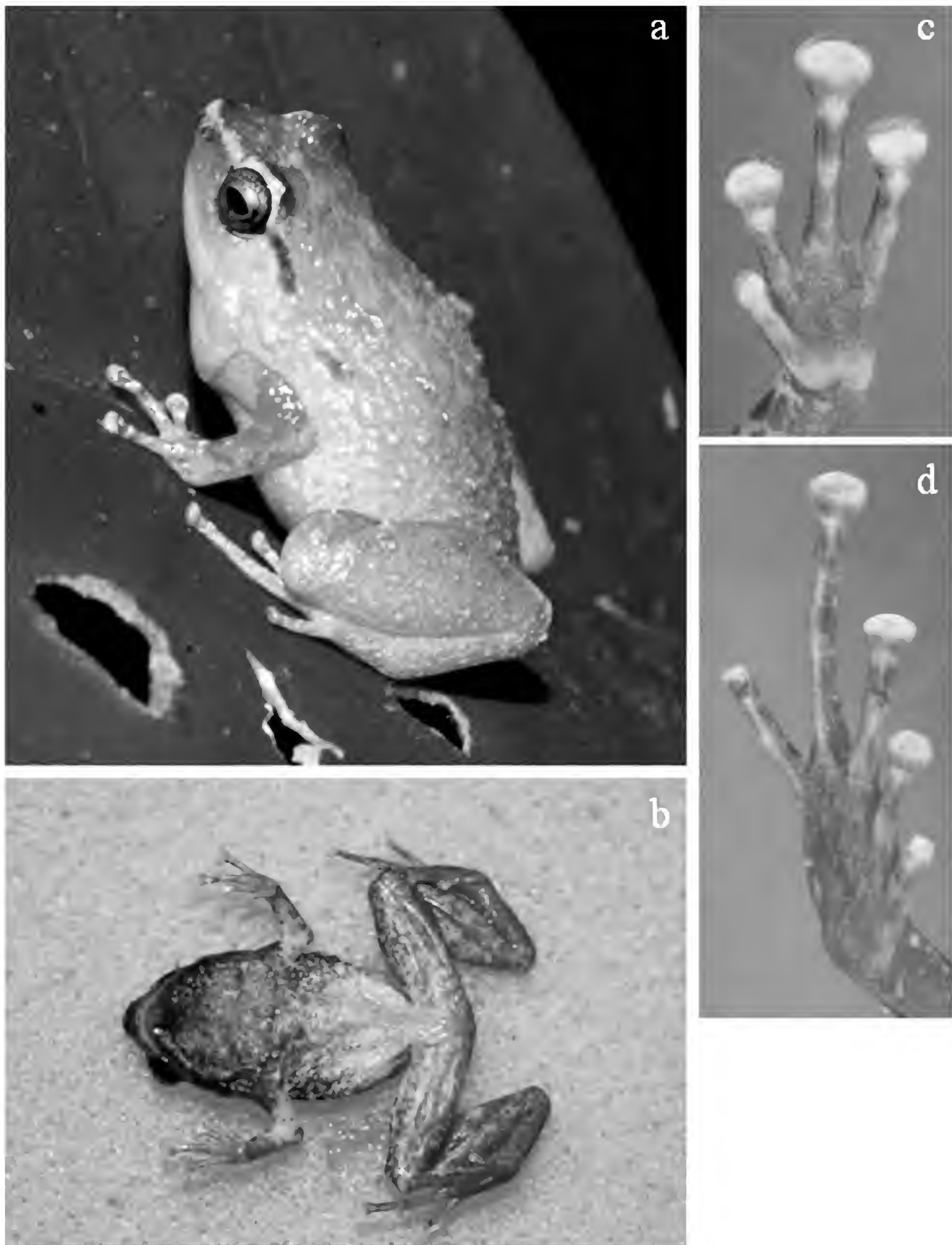
Figure 9. Holotype of *Cophixalus salawatiensis* sp. n. (**a**) dorsolateral view in life, (**b**) ventral view after preservation, (**c**) palmar view of left hand after preservation, (**d**) plantar view of right foot after preservation.

Table 2. Body measurements and body ratios of the type series of *Cophixalus salawatiensis* sp. n. MZB 12165 is the holotype, all types are adult males, MZB 12166 is a hermaphrodite; all measurements in mm. For explanation of abbreviations see Table 1.

Inv.-No.	MZB 12157	MZB 12159	MZB 12161	MZB 12162	MZB 12165	MZB 12166	MZB 12167	MZB 12168	MZB 12170	MZB 12170	Mean ± SD
SUL	20.8	21.5	22.5	21.2	20.2	21.7	19.6	20.1	20.3	19.8	20.8±0.94
TL	10.2	10.9	11.3	10.6	10.4	10.8	10.0	10.6	10.5	10.1	
TaL	6.5	6.4	7.0	6.7	6.1	6.8	6.4	6.7	6.7	6.4	
L4T	10.3	10.1	10.8	10.2	9.7	10.4	9.6	9.8	9.5	9.6	
T4D	1.0	1.1	1.1	0.9	1.1	1.0	1.0	0.9	1.0	1.0	
L3F	5.1	4.9	5.3	4.9	4.7	5.2	4.6	5.1	5.2	4.5	
F3D	1.2	1.1	1.2	1.1	1.1	1.2	1.1	1.1	1.0	1.0	
F1D	0.4	0.5	0.6	0.5	0.4	0.4	0.5	0.4	0.4	0.4	
T1D	0.5	0.5	0.6	0.6	0.5	0.5	0.6	0.6	0.5	0.5	
HL	7.0	6.9	6.4	6.3	6.1	6.5	6.2	5.9	6.3	5.8	
HW	8.1	8.2	8.5	8.0	7.7	8.0	8.0	7.5	8.0	7.6	
SL	3.2	3.1	3.0	2.9	3.1	3.0	3.0	3.1	2.9	2.7	
END	1.8	1.8	1.9	1.7	1.7	1.7	1.7	1.6	1.8	1.8	
IND	2.1	2.3	2.2	2.1	1.8	1.9	2.1	2.1	2.1	2.0	
ED	2.6	2.6	2.7	2.6	2.5	2.4	2.5	2.4	2.4	2.3	
TyD	0.7	1.0	0.9	0.8	0.7	0.9	0.7	1.0	0.8	0.7	
L1T		1.7	1.7	1.8	1.7	1.5	1.5	1.6	1.6	1.5	
LMT		0.9	0.8	0.8	0.7	1.0	0.9	0.8	0.9	1.1	
TL/SUL	0.49	0.51	0.50	0.50	0.51	0.50	0.51	0.53	0.52	0.51	0.51±0.01
TaL/SUL	0.31	0.30	0.31	0.33	0.30	0.31	0.33	0.33	0.33	0.32	0.32±0.01
L4T/SUL	0.50	0.47	0.48	0.48	0.48	0.48	0.49	0.49	0.47	0.48	0.48±0.009
L3F/SUL	0.25	0.23	0.24	0.23	0.23	0.24	0.23	0.25	0.26	0.23	0.24±0.01
F3D/SUL	0.058	0.051	0.053	0.052	0.054	0.055	0.056	0.055	0.049	0.050	0.053±0.003
T4D/SUL	0.048	0.051	0.049	0.042	0.054	0.046	0.051	0.045	0.049	0.050	0.049±0.003
T4D/F3D	0.83	1.00	0.92	0.82	1.00	0.83	0.91	0.82	1.00	1.00	0.91±0.08
F1D/SUL	0.019	0.023	0.027	0.024	0.020	0.018	0.026	0.020	0.020	0.020	0.022±0.003
T1D/SUL	0.024	0.023	0.027	0.028	0.025	0.023	0.031	0.030	0.025	0.025	0.026±0.003
T1D/F1D	1.25	1.00	1.00	1.20	1.25	1.25	1.20	1.50	1.25	1.25	1.22±0.14
LMT/L1T		0.53	0.47	0.44	0.41	0.67	0.60	0.50	0.56	0.73	0.55±0.11
HL/SUL	0.34	0.32	0.28	0.30	0.30	0.30	0.32	0.29	0.31	0.29	0.31±0.02
HW/SUL	0.39	0.38	0.38	0.38	0.38	0.37	0.41	0.37	0.39	0.38	0.38±0.01
HL/HW	0.86	0.84	0.75	0.79	0.79	0.81	0.78	0.79	0.79	0.76	0.80±0.03
SL/SUL	0.154	0.144	0.133	0.137	0.153	0.138	0.153	0.154	0.142	0.136	0.144±0.008
END/IND	0.86	0.78	0.86	0.81	0.94	0.89	0.81	0.76	0.86	0.90	0.85±0.06
ED/SUL	0.125	0.121	0.120	0.123	0.124	0.111	0.128	0.119	0.118	0.116	0.121±0.005
TyD/ED	0.27	0.38	0.33	0.31	0.28	0.41	0.28	0.42	0.33	0.30	0.33±0.05

slightly larger than, or equal in size to, those of toes (T4D/F3D 0.82–1.0). Call a train of 6–8 notes that sound like peeps or whistles; calls last for approximately 0.5 s, notes are less than 50 ms and produced at a rate of 13.5–15.6/s.

Description of the holotype (Figs 9a–d): For measurements see Table 2. Head wider than long (HL/HW 0.79), canthus rostralis straight and rounded; loreal region flat; snout protruding in profile and acuminate in dorsal view; nostrils directed laterally and near end of snout; horizontal eye diameter greater than eye-naris distance; tympanum scarcely visible, about one third of eye diameter (TyD/ED 0.28), supratympanic fold weakly expressed and S-shaped; internarial distance only slightly greater than distance between eye and naris (END/IND 0.94); tongue medium-sized, a little broadened posteriorly and lacking notch, with posterior and lateral margins free; prepharyngeal ridge not serrated; long vocal slits on both sides of the tongue. Legs moderately long (TL/SUL 0.51), no webbing between fingers or toes; disks of fingers II, III and IV about same width as disks of toes II, III and IV, disks of finger I as well as of toe I and V much smaller than that of other fingers and toes, relative length of fingers 3>4>2>1; third toe clearly longer than the fifth, all finger and toe disks with terminal grooves; relative length of toes 4>3>5>2>1, no clearly expressed subarticular tubercles on fingers and toes and no distinct palmar or plantar tubercles. Some low, pale-tipped tubercles on flanks, dorsally on shanks, and in two weakly-defined rows on posterior surfaces of back. All remaining dorsal, lateral, and ventral surfaces smooth, except a gular fold between insertion of the fore limbs that indicates the posterior margin of the vocal sac.

In preservative dorsal surfaces of head, body and limbs light grey-brown, flanks lighter than dorsum; most tubercles with dark base and light tip; irregular dark brown

flecks on limbs and flanks. A longish dark-brown post-ocular spot followed by a small dark-brown spot above arm insertion. A broad off-white fleck extends from posterior of eye through tympanum up to arm insertion. This fleck is bordered antero-dorsally by the dark postocular spot and ventrally by the posterior part of the dark brown "face-mask". The face-mask continues below the eyes, runs along the loreal region and reaches to the snout tip. Ventral surfaces of limbs and abdomen off-white with irregularly shaped brown spots that are often reticulated, throat and chest dark brown with a few off-white speckles; region around anal opening blackish.

In life dorsum grey-orange, central dorsum more intensely coloured than flanks, conspicuous is a big orange spot on foreleg and a whitish canthal stripe that continues on upper eyelid. Dorsal surface of head with a mixture of grey and orange spots, face-mask greyish. The small dark and the big off-white postocular flecks less intensely marked than in fixative. Dorsal and lateral tubercles more strongly expressed than in preservative, a dorsolateral row of inconspicuous tubercles present.

Variation in the type series (in preservative): Mensural variation for the type series is shown in Table 2. Basic colour and colour pattern elements of all paratypes are fairly uniform and resemble the holotype. Ground colour of dorsal surfaces light grey to light brown. Dorsolateral glandular ridges weakly developed and often interrupted - they extend in some specimens from eye to lumbar region, in others they are shorter and confined to middle and posterior back. Dorsolateral prior glandular ridges in most specimens indicated by rows of dark brown spots that vary from rare to numerous and can form shorter or longer stripes. Characteristic for most paratypes is a dark brown postocular spot as well as a spot of the same colour above insertion of fore arm. Tympanal region in all specimens off-white and clearly demarcated from the dark postocular spot and the dark "face-mask". Dark brown lateral colour of head merges into that of dark gular region. Lateral surfaces of body usually more strongly pigmented towards dorsal regions. One specimen (MZB Amph.12166) shows heavily spotted dorsal and lateral surfaces. Throat and chest in all specimens covered by a dense net of dark brown flecks, abdomen off-white with few small brownish flecks and ventral surfaces of limbs a little more mottled than abdomen. No specimen with either dark or light mid-dorsal line.

MZB Amph.12166 is considered a hermaphrodite because it has a well-developed vocal sac and two vocal slits in the floor of the mouth, but also has an ovary containing eggs in an advanced developmental stage (Fig. 10). This frog also uttered advertisement calls that were recorded and that did not differ from calls of conspecific males.

Vocalisation. Calling occurred at night, predominantly after heavy rain. The advertisement call of *Cophixalus salawatiensis* sp. n. consists of a short series of 6–8 peeps or whistles (Fig. 11) with fairly long and irregular intervals between the calls. Shortest inter-call interval 7 s

and longest 58 s. Thirty-nine calls of three males, (MZB Amph.12166–168), all recorded at 24 °C, were analysed. Mean duration of these calls was 0.53 s, SD 0.16, range 0.42–0.55 s. Mean number of notes/call 7.2, SD 0.77, range 6–8. Mean of means of note length 43.6 ms, SD 1.39, range 41–46 ms, total range of note length 32–50 ms. Mean of means of internote interval length 31.2 ms, SD 2.0, range of means 26–33 ms, total range 21–41 ms. Mean repetition rate 14.3 notes/s, SD 0.51, range 13.5–15.6 notes/s. First note of almost all calls the shortest, and last interval the longest of all inter-note intervals. All notes are composed of many dense pulses. Their amplitude rises rapidly up to maximal level, remains stable on this level over most of the note and then drops gradually to the end of the note. Frequencies scatter from 2.5 to 3.25 kHz with dominant frequency at 2.9 kHz (Fig. 12). Harmonics are very weakly expressed and there is no modulation of frequencies (Fig. 11 below).

Distribution and ecological remarks. *Cophixalus salawatiensis* sp. n. is currently known only from one location on Salawati Island in the Raja Ampat Island group off western New Guinea (Fig. 1). It was common in moderately logged lowland rainforest where males called from the surfaces of leaves in low foliage ~30 cm – 1.5 m above the ground after heavy rain at night. Intensive searches on nearby Batanta and Waigeo islands failed to detect this species there despite similar climatic conditions and strong activity of other frogs. This suggests that *C. salawatiensis* sp. n. probably does not occur there. A recently collected *Cophixalus* specimen (NME A2216/15 in the "Naturkundemuseum Erfurt, Germany") represents the first evidence of the genus *Cophixalus* from Misool Island. The specimen, a male, was collected by D. Telnov on 20 March 2009 from inside a rotten log where it guarded (at least) five rather well developed eggs in a "primeval moist lowland forest, district Misool Utara, Aduwey, valley of River Ifeyo, 01°58′41′′S and 129°55′18′′E". It measures 21.2 mm SUL and is morphologically most similar to *C. salawatiensis* but without knowledge of its advertisement calls we refrain from confirming the species' presence there. However it is biogeographically interesting to confirm that the genus *Cophixalus* also occurs on Misool. It is not known whether *C. salawatiensis* occurs on the nearby New Guinea mainland.

Etymology. The latinized specific epithet *salawatiensis* means that the new species occurs on Salawati Island off the western tip of New Guinea.

Comparison with other species (see species comparison section for *C. rajampatensis* for specific size ranges of all congeners discussed below). *Cophixalus amabilis, ateles, bewaniensis, desticans, humicola, interruptus, iovaorum, kethuk, linnaeus, melanops, misimae, phaeobalis pictus, pipilans, tagulensis, timidus, tomaiodactylus, tridactylus, variabilis, verecundus, viridis* and *wempi* all have adult
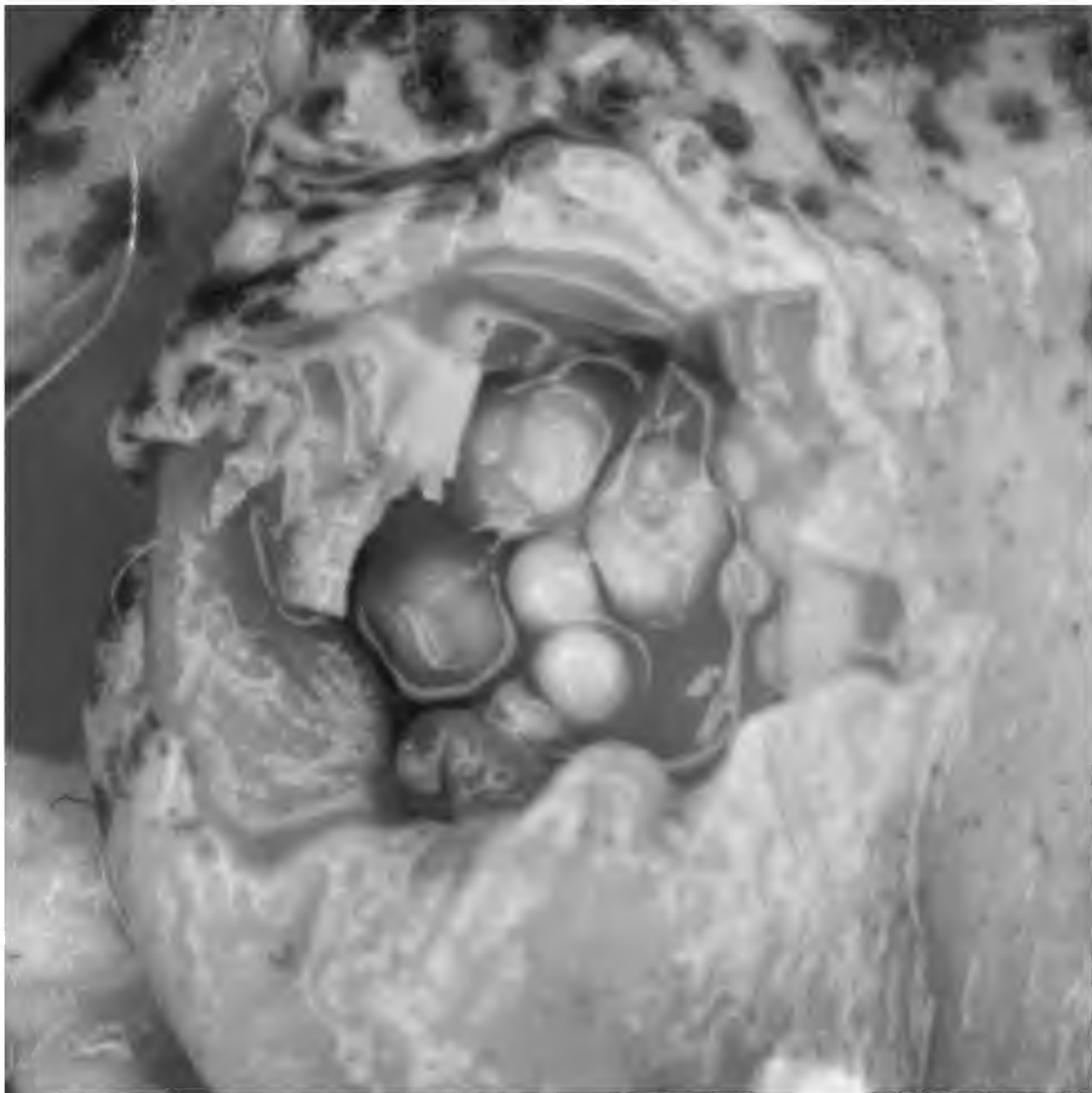
Figure 10. Ventral view of the opened up abdomen of *Cophixalus salawatiensis* sp. n., MZB Amph.12166, showing eggs of different developmental stages in a specimen with vocal slits that was recorded calling.

male SUL's of less than 19 mm and so can be immediately distinguished from *C. salawatiensis* (SUL 19.6–22.5 mm). With an SVL of 15.7 mm the only known specimen of *Cophixalus pictus* Kraus is smaller than *C. salawatiensis* but its description was based on a rather poorly preserved (and presumed immature) male from the Bomberai Peninsula of West Papua Province (Kraus 2012) and, if immature, this distinction could disappear once adult material has been documented. However *C. salawatiensis* also differs from *C. pictus* in a number of features that are unlikely to reflect the immature status of the holotype and only known specimen, including having (vs. lacking) a distinct supratympanic fold, in having longer legs (TL/SUL 0.49–0.53 vs. TL/SVL 0.47) and in having a very dif-

ferent ratio of internarial distance to eye-to-naris distance (END/IND 0.76–0.94 vs. 1.08 in *C. pictus*) (Kraus 2012). *Cophixalus balbus, biroi, caverniphilus, cheesmanae, clapporum, cryptotympanum, cupricarenus, kaindiensis, montanus, nubicola, parkeri, riparius* and *verrucosus*) all have minimum adult male body sizes > 23 mm and so can also be distinguished from *C. salawatiensis* sp. n. (SUL 19.6–22.5 mm). From the species of about the same size *C. albolineatus* has finger discs smaller than toe discs, while in the new species finger discs are of the same size or larger than toe discs. *Cophixalus shellyi* has a short, strongly reduced first finger (vs. normal length with moderately large disc in *salawatiensis*) and *C. sphagnicola* and *C. tenuidactylus*, in contrast to *C. salawatiensis*,
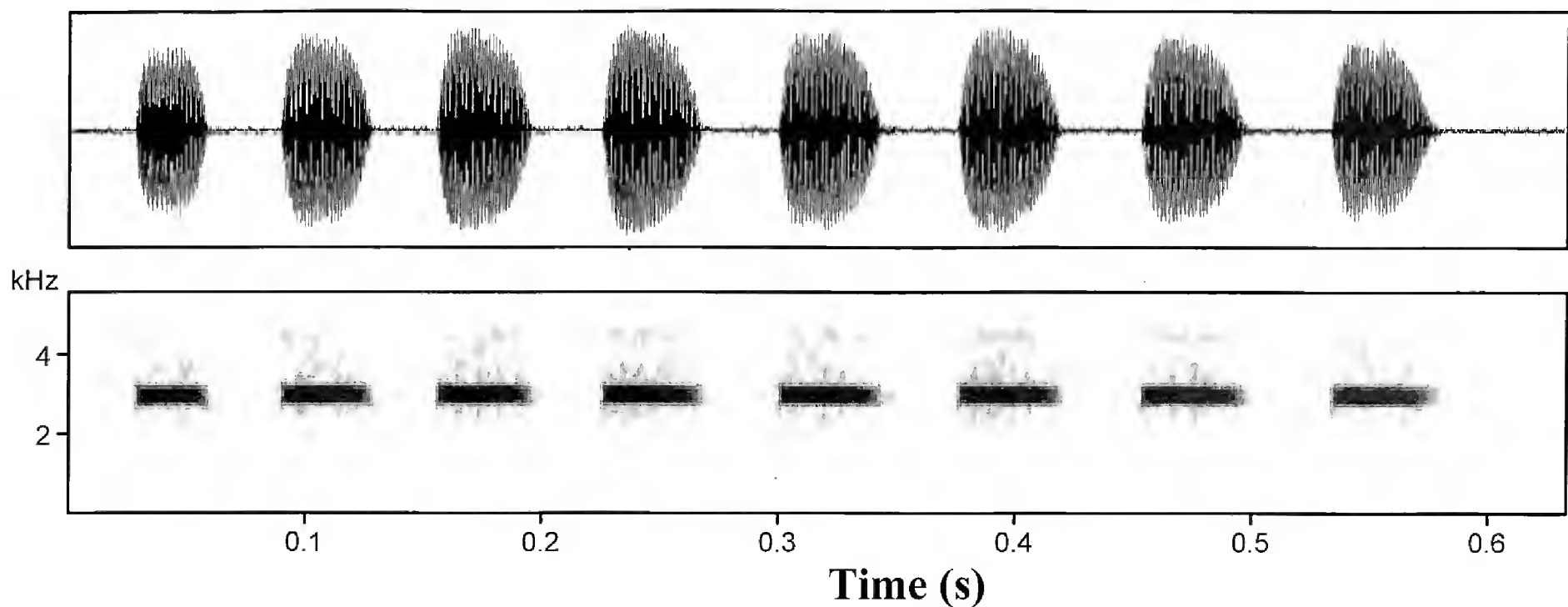
Figure 11. Wave form (above) and spectrogram (below) of an advertisement call of *Cophixalus salawatiensis* sp. n. consisting of eight notes.
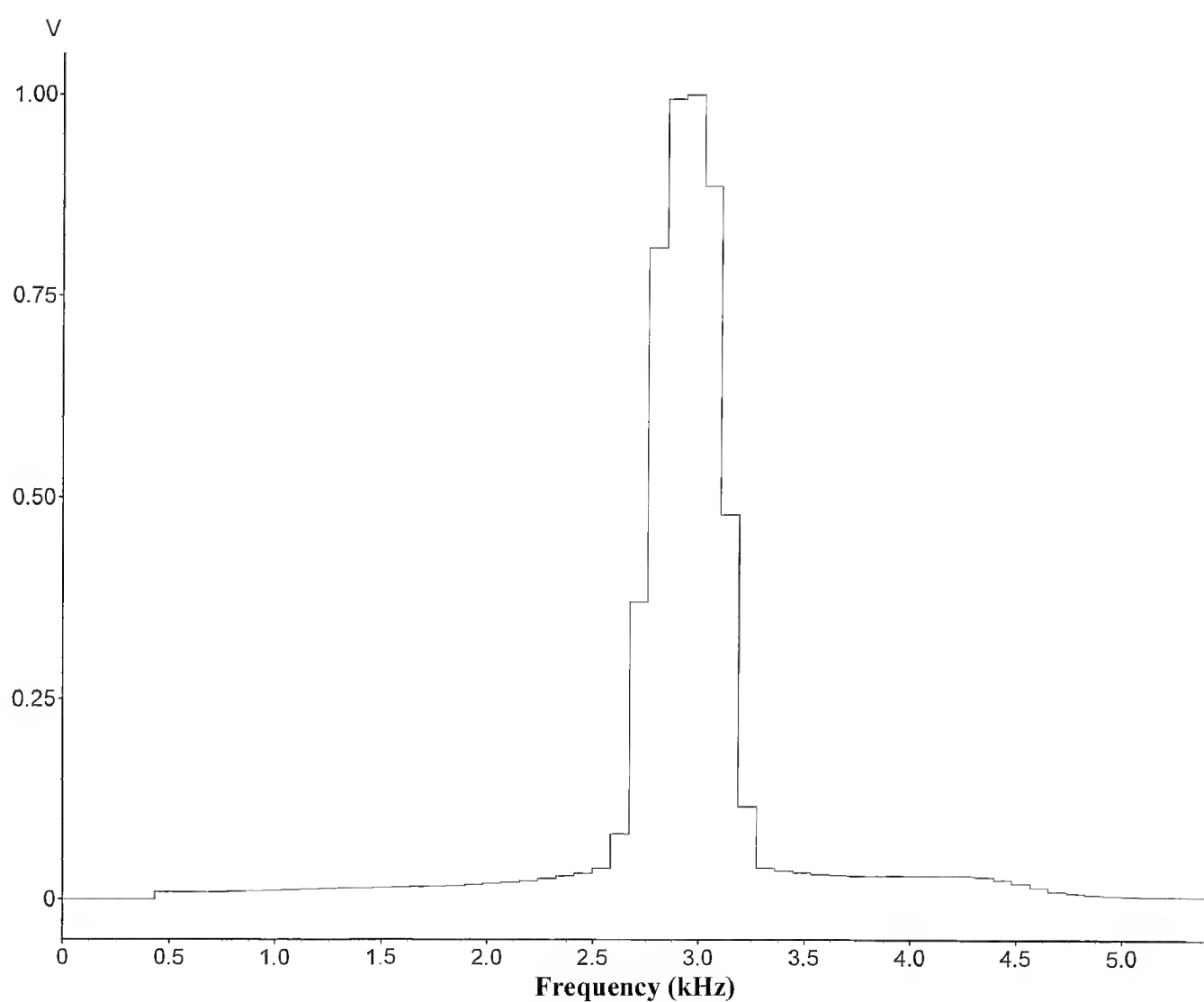

Figure 12. Power spectrum of an advertisement call of *Cophixalus salawatiensis* sp. n.

completely lack discs on fingers and toes. *C. daymani* and *C. nubicola* are distinguished by their very short hind legs (TL/SUL less than 0.48 vs. more than 0.49 in *C. salawatiensis* sp. n.). *Cophixalus nexipus* differs by having basal webbing on toes and advertisement calls consisting of a single, long note lasting more than one second (vs. 6–8 notes). *Cophixalus pulchellus* Kraus & Allison has a dorsum boldly blotched with black on a light grey background (vs. uniform in *C. salawatiensis*).

In external morphology, *C. tetzlaffi*, *C. monosyllabus* and the above described *C. rajampatensis* exhibit most similarities to *C. salawatiensis* sp. n.

Cophixalus tetzlaffi has clearly smaller discs on finger one and toe one than *C. salawatiensis* sp. n. – the ratio F1D/SUL in 8 specimens of *C. tetzlaffi* is 0.016, SD 0.0016, range 0.014–0.018; mean of the same ratio in 10 specimens of *C. salawatiensis* sp. n. is 0.022, SD 0.0031, range 0.018–0.027; mean of the ratio T1D/SUL in *C. tetzlaffi* is 0.020, SD 0.0019, range 0.018–0.023 and in *C. salawatiensis* sp. n. 0.026, SD 0.0028, range 0.023–0.031. These species also have different advertisement calls - calls of *C. tetzlaffi* consist of 3–4 notes with note lengths of more than 300 ms, those of *C. salawatiensis* sp n. consist of 6–8 notes with a note length of less than 50 ms.
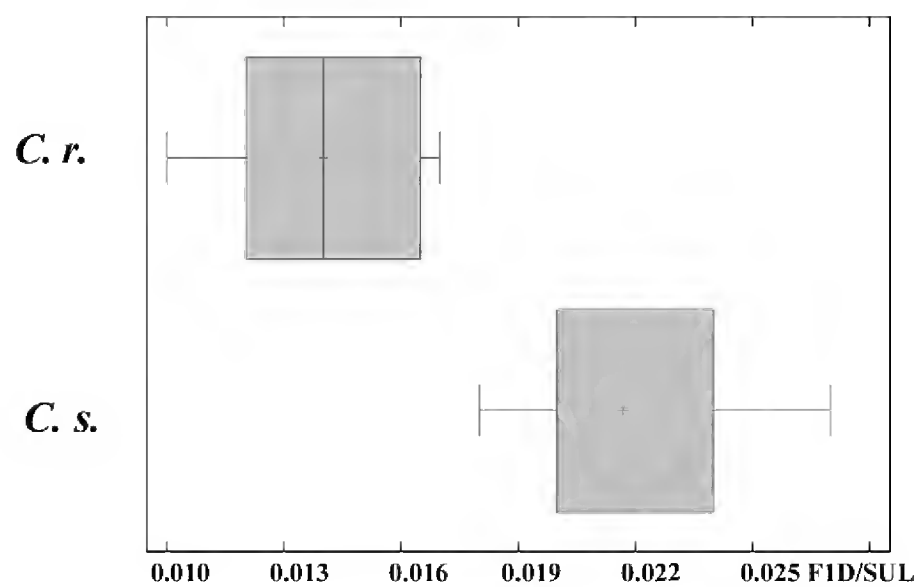
Figure 13. Box-Whisker-Plot of the ratio F1D/SUL of eight males of *Cophixalus rajampatensis* sp. n. (*C. r.*) and ten males of *C. salawatiensis* sp. n. (*C. s.*).
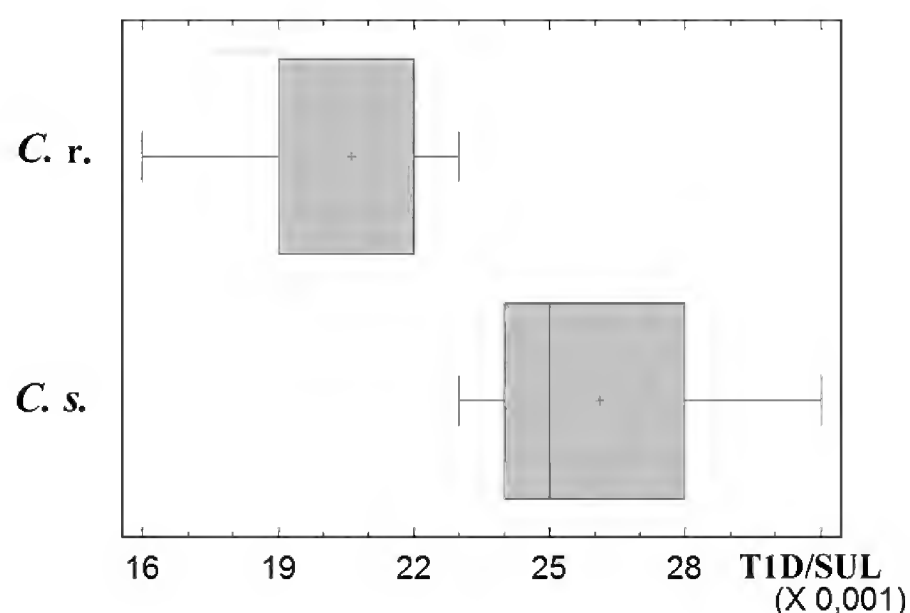

Figure 14. Box-Whisker-Plot of the ratio T1D/SUL of eight males of *Cophixalus rajampatensis* sp. n. (*C. r.*) and ten males of *C. salawatiensis* sp. n. (*C. s.*).
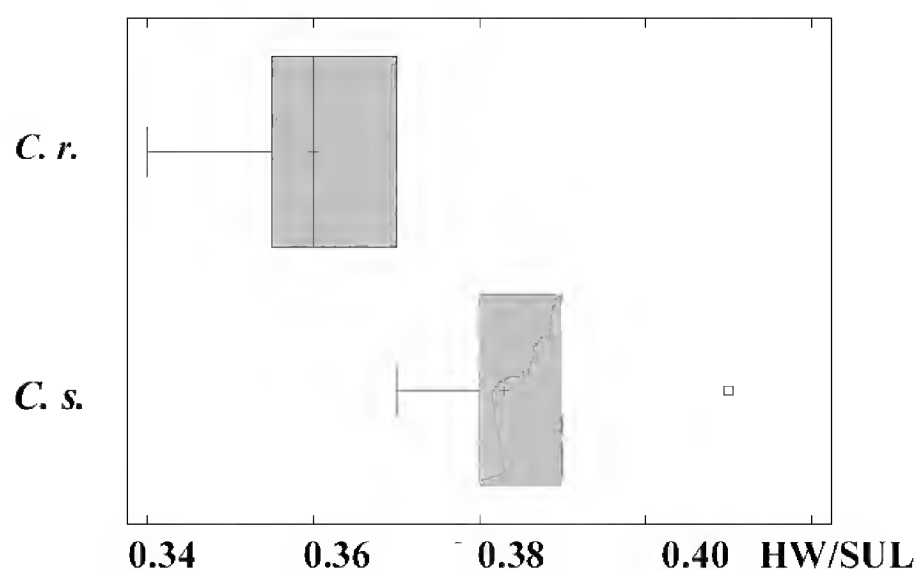

Figure 15. Box-Whisker-Plot of the ratio HW/SUL of eight males of *Cophixalus rajampatensis* sp. n. (*C. r.*) and ten males of *C. salawatiensis* sp. n. (*C. s.*).

Cophixalus monosyllabus is morphologically very similar to *C. salawatiensis* sp. n. and, although the species differ significantly in body size [mean SUL of the former (n=10 adult males) 22.9 mm, SD 1.04, range 20.6–24.3 mm and of the latter (n=10 adult males) 20.8 mm, SD

0.94, range 19.6–22.5 mm (p=0.001 for comparisons of medians)], there is substantial overlap in SUL. The species also differ significantly in size of disc of third finger - mean ratio F3D/SUL in *C. monosyllabus* 0.063, SD 0.004, range 0.055–0.071 and in *C. salawatiensis* sp. n. 0.053, SD 0.003, range 0.049–0.058 (p=0.0004 for comparison of medians) but again there is some overlap. However these species have consistently and strikingly different advertisement calls – in *C. monosyllabus* these consist of single notes with a duration of more than 140 ms vs. 6–8 notes with note duration not longer than 50 ms in *C. salawatiensis* sp. n. and we consider these differences sufficient to warrant their recognition as distinct species.

C. rajampatensis (n=8) and *C. salawatiensis* sp. n. (n=10) have non-overlapping body sizes (17.6–19.5 vs. 19.6–22.5 mm) and further differ in the following body ratios: F1D/SUL 0.010–0.017 (mean 0.014) in the former vs. 0.018–0.027 (mean 0.022) in the latter, (Fig. 13); T1D/SUL 0.016–0.023 (mean 0.021) vs. 0.023–0.031 (mean 0.26), p=0.0005 (Fig. 14); HW/SUL 0.34–0.37 (mean 0.36) vs. 0.37–0.41 (mean 0.38), p=0.0009 (Fig. 15), and T1D/F1D 1.33–1.75 (mean 1.48) vs. 1.00–1.50 (mean 1.22), p=0.002.

Cophixalus rajampatensis and *C. salawatiensis* sp. n. also differ in their advertisement calls; calls of the former consist of 2–5 notes per call, note length 142–238 ms, 3.3-4.6 notes/s vs. 6–8 notes per call, note length 32–50 ms, 13.5–15.6 notes/s in the latter.

Acknowledgements

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PENSOFT.

Stenothoids living with or on other animals (Crustacea, Amphipoda)

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http://zoobank.org/89B46F52-BA36-41A9-9D7F-B635CAF052C1

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Abstract

This paper describes new or little known species of Stenothoidae, collected from sea anemones, bivalves or hermit crabs. A key to world *Stenula* species is provided.

Key Words

Introduction

Associations between amphipods and other animals are probably not all that rare, but few have been recorded hitherto, mainly because collection methods earlier were too crude. With the advance of Scuba-diving, new associations are being discovered at a rapid pace. The present paper adds a few more examples from the family Stenothoidae.

Material and methods

All specimens in alcohol were studied under a Reichert and Wild M5 dissecting microscope, then drawn from preparations (dissected and stored in glycerine or Faure's medium) under a Wild M20 microscope. The pencil drawings were partly inked by hand, and partly with the software Adobe Illustrator CS 3, using a Wacom A4 drawing board. The material is lodged at the Museums of Verona (MVR, Italy), Copenhagen (ZMUC, Denmark), Smithsonian Washington (USNM, USA), see detailed indications at the descriptions.

Abbreviations

A1, 2, antennae 1, 2; art, article; Cx, coxa; Ep, epimeral plate; Gn 1, 2, gnathopods 1, 2; Md, mandible; Mx1, 2, maxillae 1, 2; Mxp, maxilliped; P 3–7, peraeopods 3–7; U 1–3, uropods 1–3.

In this paper the following terms are applied (see also Krapp-Schickel 2011: 1–2): tooth: non-articulated pointed ectodermal structure; spine: stout, articulated structure (synonymous with "robust seta"); seta: slender, flexible articulated structure.

Systematics

Stenothoidae Boeck, 1871
Stenothoinae Boeck, 1871

Genus *Stenothoe* Dana, 1852

Type species. *Stenothoe valida* Dana, 1852

The diagnosis of this genus is summarized, it shows few eminent features: Md palp lacking totally, P 5 basis linear, P 6–7 basis widened, T laminar. The genus contains many apparently free-living species and others that are living in symbiosis with other marine invertebrates; these latter in many cases show greatly diminished sexual dimorphism.

Stenothoe species are mainly found in the Atlantic and Mediterranean, while few are living in the Pacific, in the region of Australia-New Zealand or in the Indian Ocean (Krapp-Schickel 2015). There remains the strong suspicion that e.g. members living near Australia or in the Indian Ocean are not closely related to those from the Atlantic Ocean (urosome segments tend to lose their clear articulation in the former), but as the easier visible appendages such as legs or antennae match the generic definition, plus the fact that both have lost the Md palp (most probably an independent evolution), also these species are treated in the same genus *Stenothoe*, for the time being. Here we have examples of these different groups:

Stenothoe bartholomea sp. n.

http://zoobank.org/54165FFC-8D4D-4299-A613-6C12FCC1147C
Figures 1–5
Vader 1983: 146 sub *Stenothoe* n.sp.

Holotype. female ov. 3 mm, Florida, Monroe County, Cross Key: from *Bartholomea annulata* (Lesueur, 1817), 4/10/1971, J. Thomas coll. Slide MVRCr 7716, 7717.

Additional material. one female front part, slide MVR-Cr 7718; several hundreds, same locality, same date, in alcohol, deposited at MVRCr.

Type locality. Florida, Monroe County, Cross Key.

Etymology. after the host anemone *Bartholomea annulata* (Lesueur, 1817), from where it was collected.

Diagnosis. Sexual dimorphism lacking. Gn 1 and Gn 2 propodus similar in shape, propodus hind margin rounded, in Gn 2 without clear palmar corner. P 6, 7 basis posterodistal corner lengthened and rounded; merus posterodistally widened and lengthened, reaching half length of carpus. U3 peduncle < ramus. T with or without submarginal spines and marginal setae.

Description.
Length 2.5 – 3 mm.
Head. Eyes round, medium. Mouthparts: Md with acute hook on the place where a palp would have inserted; molar absent, incisor and lacinia mobilis strong, spine row present. Mx 1 palp with 2 arts, Mx 2 plates sitting upon each other. Mxp IP small, OP vanishing. Antennae: subequal, A1 flagellum about 15 arts, A2 flagellum about 10 arts.

Peraeon. Gn 1 basis about 3 times as long as wide, merus triangular, distally rounded and beset with spines, nearly reaching end of carpus; carpus triangular; propodus somewhat wider and clearly longer than carpus, anteriorly beset with long setae; palm well defined by a group of spines and palmar corner. Cx 2 tongue-shaped, posteriorly excavate. Gn 2 basis 3 –3.5 times longer than wide, rectangular; merus rectangular-trapezium-shaped, distally pointed, carpus triangular, distally with stiff short and long pectinate setae, propodus oval, posterior margin regularly rounded, palm defined by some robust spines but palmar corner lacking, with one or more small triangular elevations.

Peraeopods: Cx 3 rectangular, distal margin not much longer than proximal one. Cx 4 triangular, clearly wider than long. P 3, 4 similar, slender, dactylus longer than half propodus. P 5 basis rectolinear. P 6, 7 basis with posterior margin rounded, postero-distal lobe well developed; merus lengthened and widened, reaching about half length of carpus; propodus > carpus, dactylus > half of propodus.

Pleon. U 1 peduncle > subequal rami, all beset with a few spines. U 2 peduncle > longer than unequal rami; U 3 peduncle about 3 times as long as wide, peduncle < ramus, ramus art 1 < art 2, with few short spines.

Telson with or without pair of submarginal spines and small marginal setae.

Remarks. In Krapp-Schickel 2015 a key is offered for *Stenothoe* species world-wide, grouped after geographical regions. Among these species this new one is one of only two with a regularly rounded Gn 2 male propodus, lacking excavations, deep incisions or prominent elevations. The other one is *S. tergestina* (Nebeski, 1881), frequently found in the Mediterranean, free-living in algae.

Distribution. Florida, Atlantic Ocean.

Ecology. living with and on the sea-anemone *Bartholomea annulata* (Lesueur, 1817).

Stenothoe miersii (Haswell, 1879)

Figures 6, 7
Montagua Miersii Haswell, 1879: 323, pl. 24, fig. 4.
Montagua longicornis Haswell, 1879: 323, pl. 24, fig. 5.
Stenothoe miersi. —Stebbing 1906: 200 (in part). —Stebbing 1910: 637 (in part). Lowry and Stoddart 2003: 260. —Krapp-Schickel 2009: 873–875.
Stenothoe ?miersii. —J.L. Barnard 1974: 120, figs 75–76.
Stenothoe valida. —Sheard 1937: 21 (=*S. miersii*, but confused with other species).
not *Montaguana miersii.* —Chilton 1883: 79 (part =*S. moe*, J.L Barnard 1972).
not *Montagua miersii.* —Chilton, 1884: 1043 (part =*S. moe*, J.L Barnard 1972).
not *Probolium miersii.* —Thomson and Chilton 1886: 150 (=*S. valida*).

Type locality. Port Jackson, New South Wales, Australia.
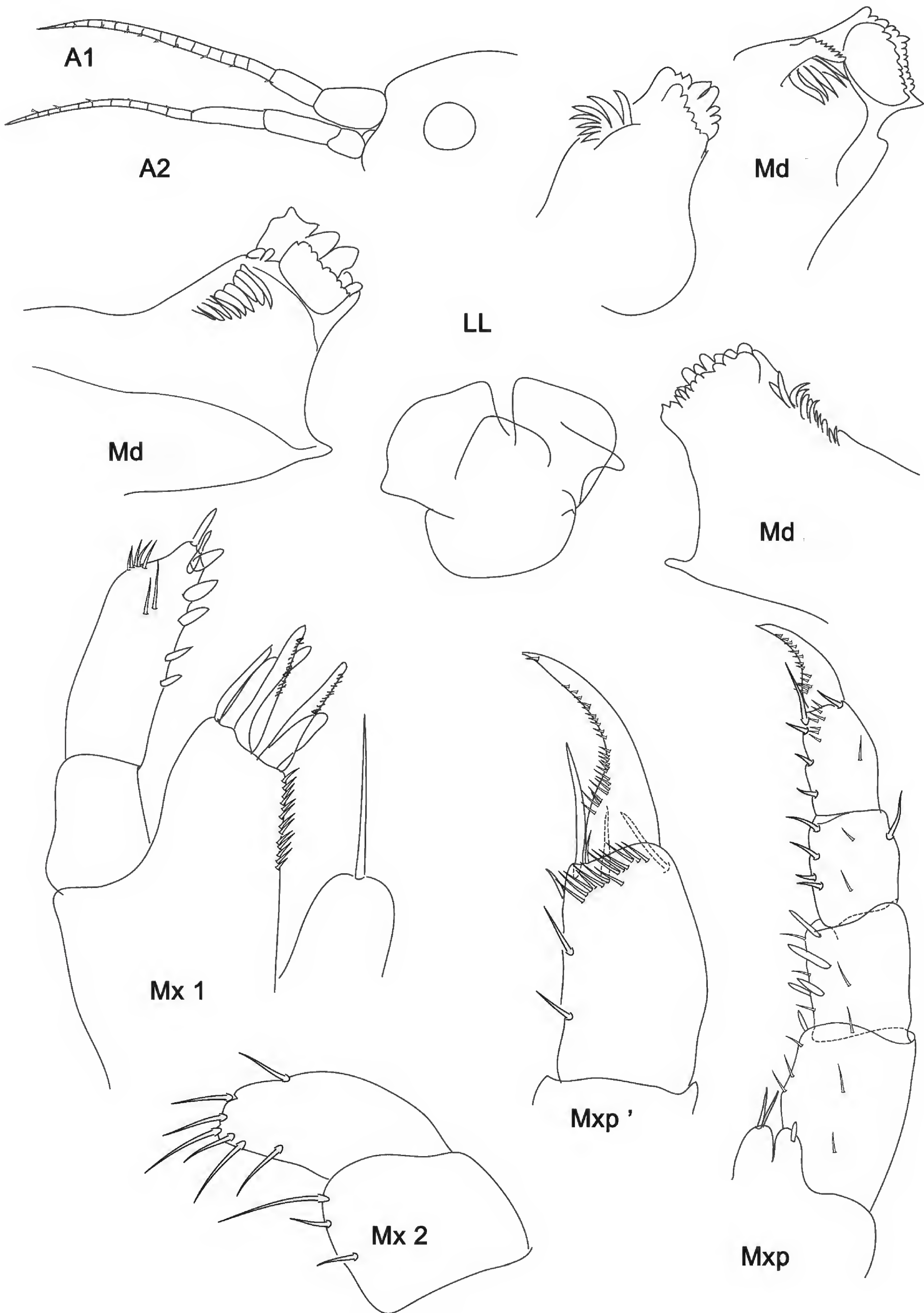
Figure 1. *Stenothoe bartholomea* sp. n. female 3 mm holotype. **A1, 2** antennae; **Md** mandibles from both sides; **LL** lower lip; **Mx1, 2** maxillae 1, 2; **Mxp** maxilliped; **Mxp'** maxilliped dactylus and propodus enlarged.
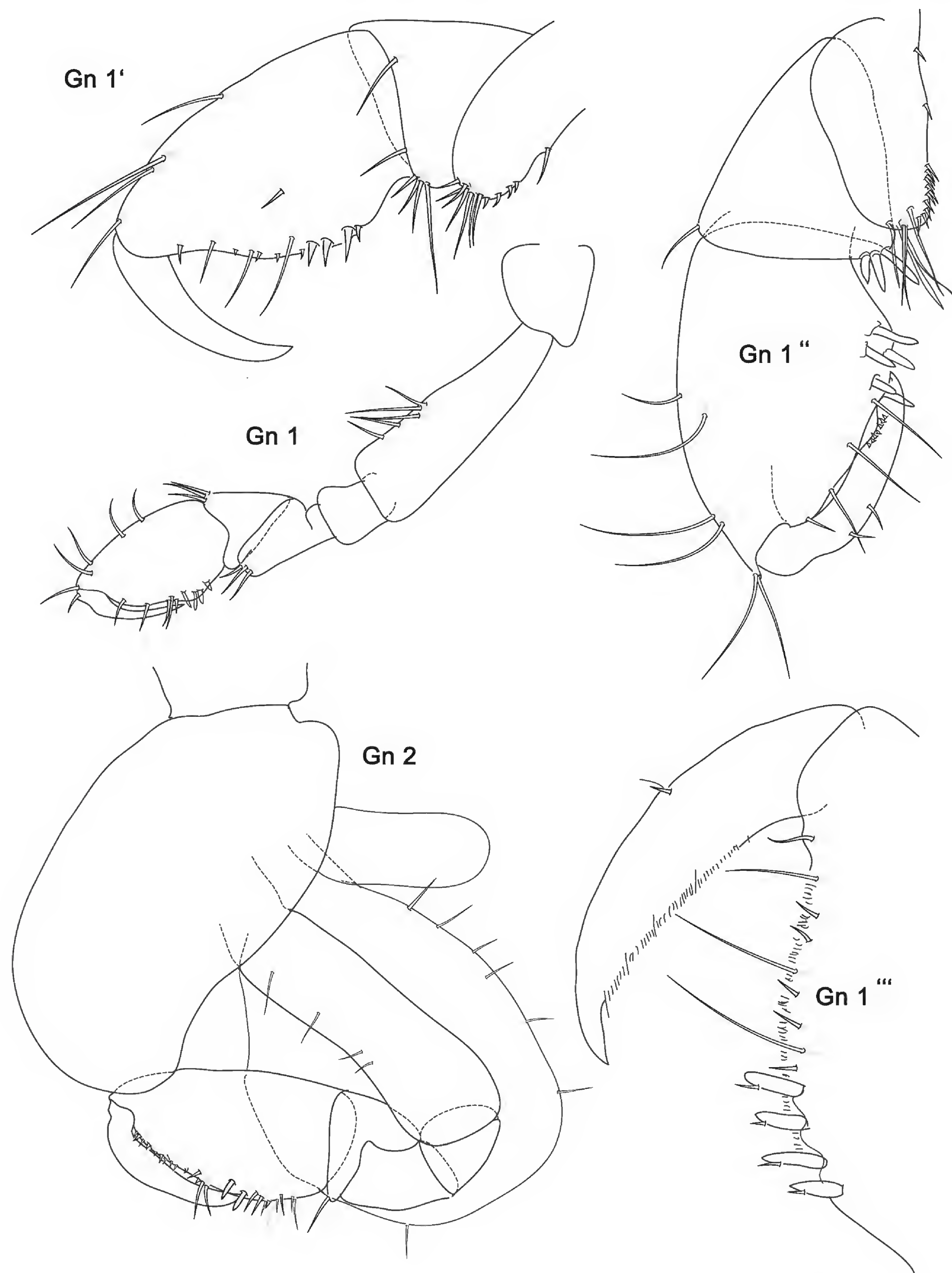
Figure 2. *Stenothoe bartholomea* sp. n. female 3 mm holotype. **Gn 1** gnathopod 1; **Gn 1', Gn 1 '', Gn 1 '''** palm and dactylus enlarged; gnathopod 1 dactylus and propodus enlarged; **Gn 2** gnathopod 2.
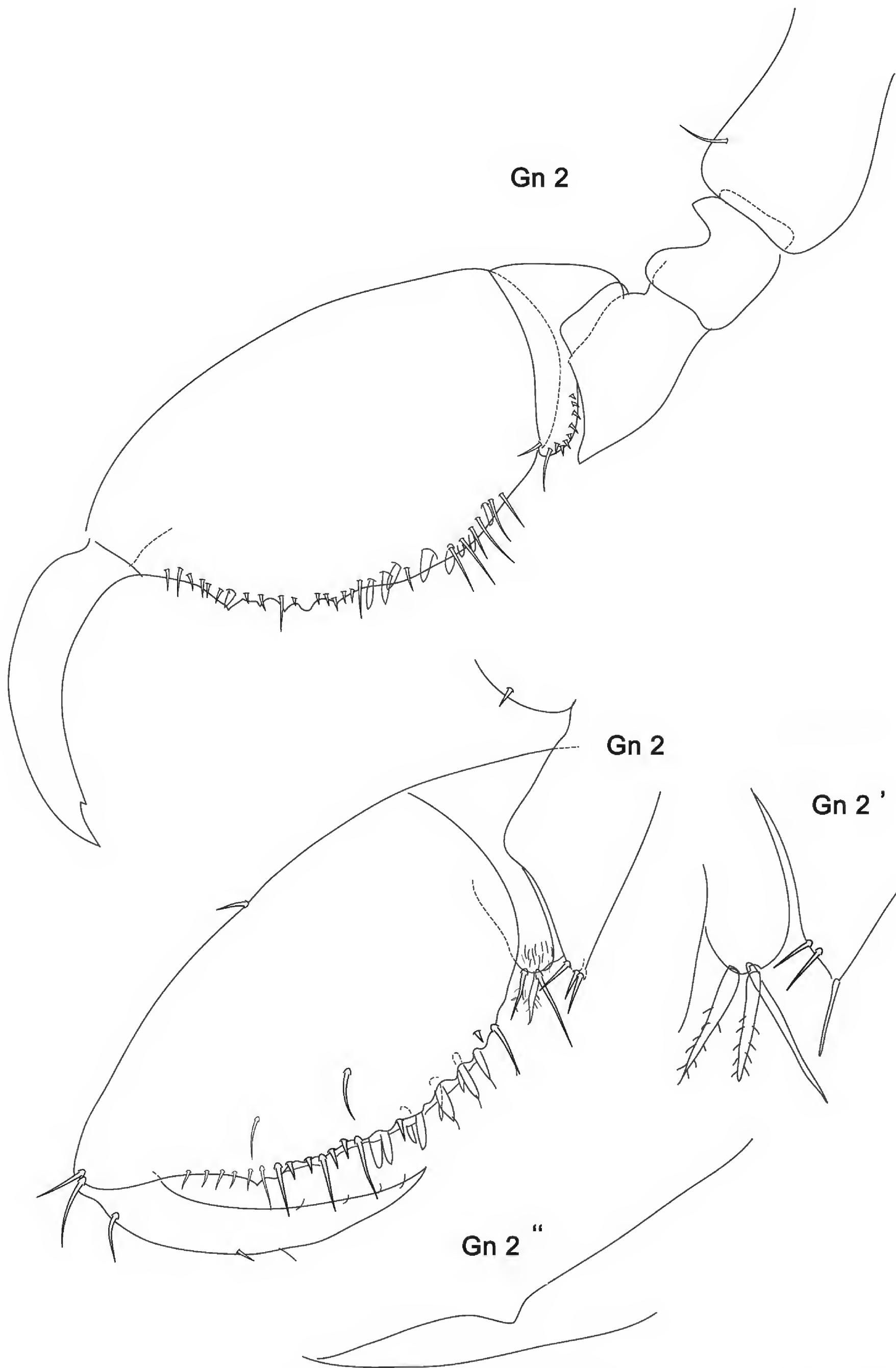
Figure 3. *Stenothoe bartholomea* sp. n. female 3 mm holotype. **Gn 2, Gn 2', Gn 2''** gnathopod 2 from both sides and tip of carpus + merus resp. palmar corner enlarged.
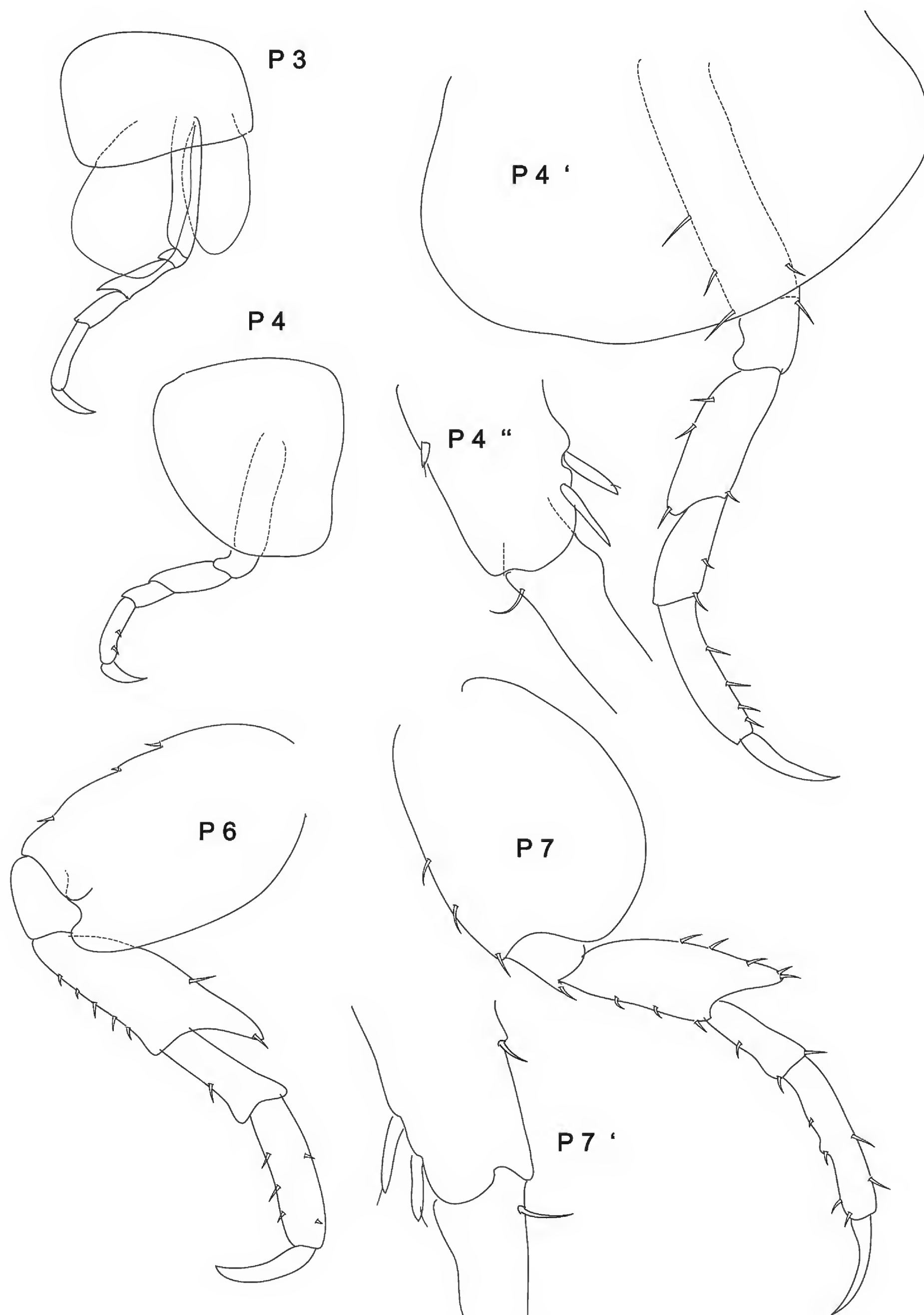
Figure 4. *Stenothoe bartholomea* sp. n. female 3 mm holotype. **P 3–4, P 6–7** peraeopod 3–4; peraeopod 6–7; **P 4', P 4''** entire leg with coxa resp. distal end of propodus **P 4** enlarged; **P 7'** distal end of propodus **P 7** enlarged.
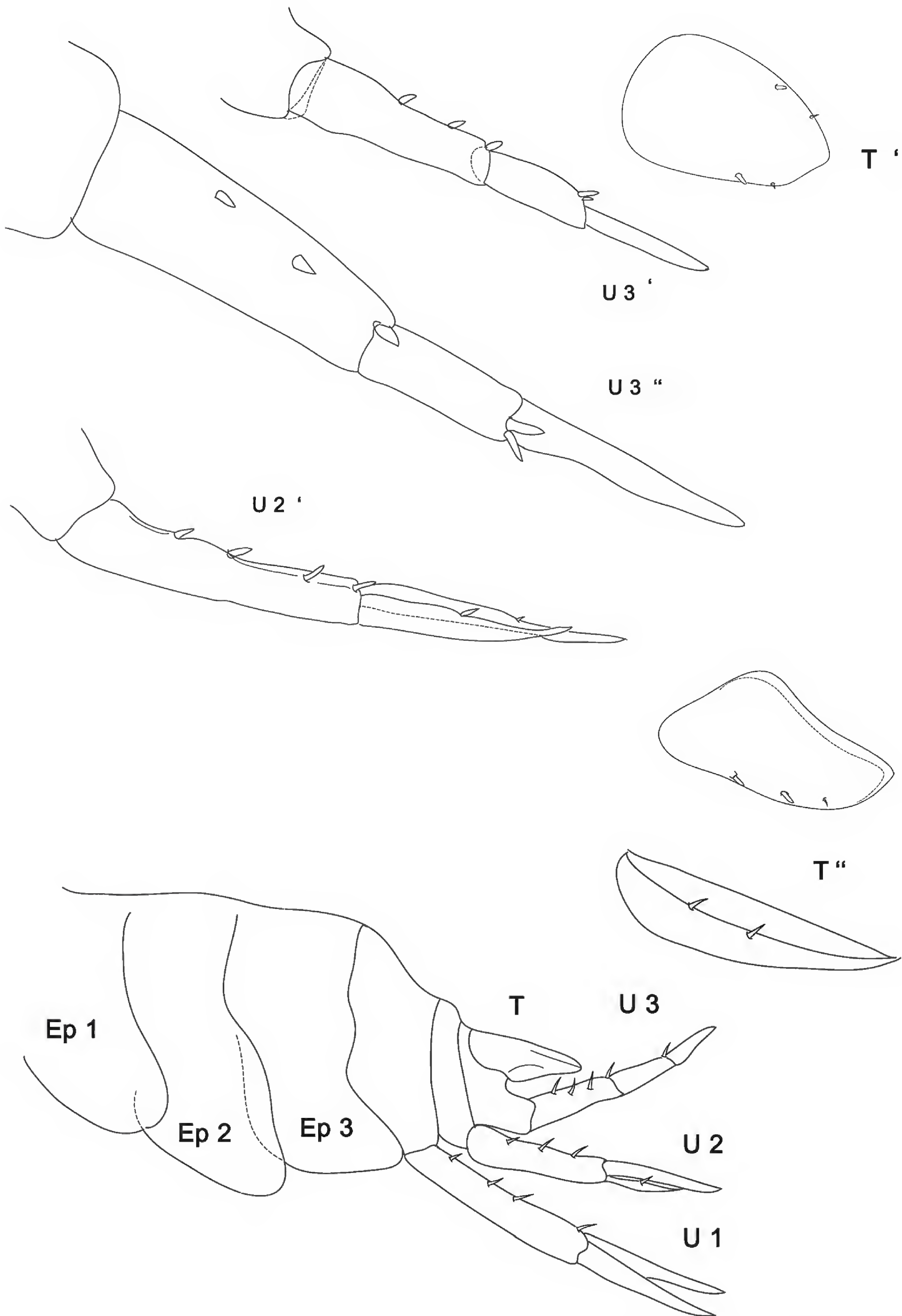
Figure 5. *Stenothoe bartholomea* sp. n. female 3 mm holotype. **Ep 1, 2, 3** epimeral plates 1–3. **U 1, 2, 3** uropod 1, 2, 3; **U 2'** uropod 2 enlarged; **U 3', U 3''** uropod 3 in two enlargements. **T', T''** telson enlarged in different positions.
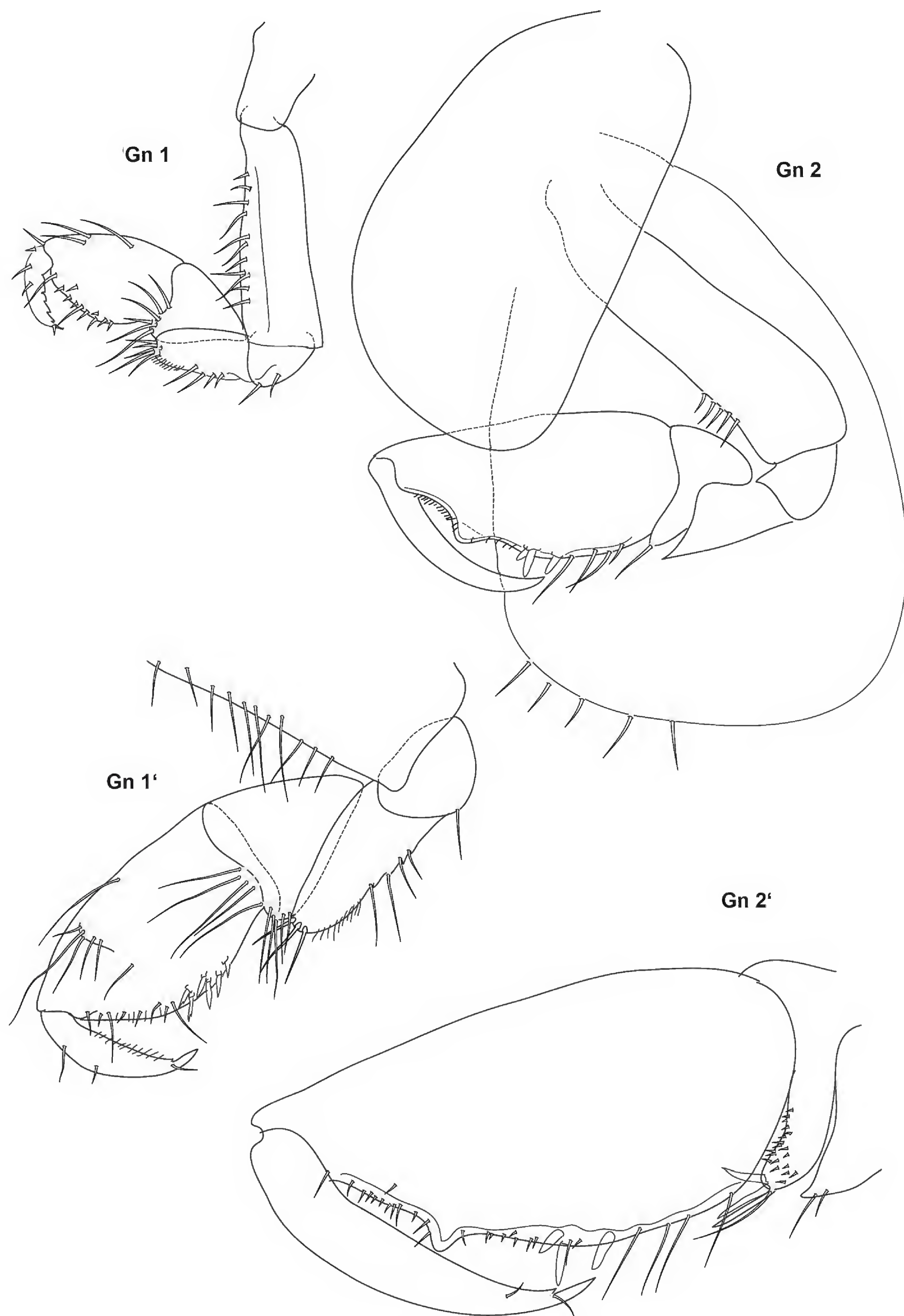
Figure 6. *Stenothoe miersii* (Haswell, 1879) male 3 mm; **Gn 1, 2** gnathopod 1, 2; **Gn 1', Gn 2'** gnathopods distally enlarged.
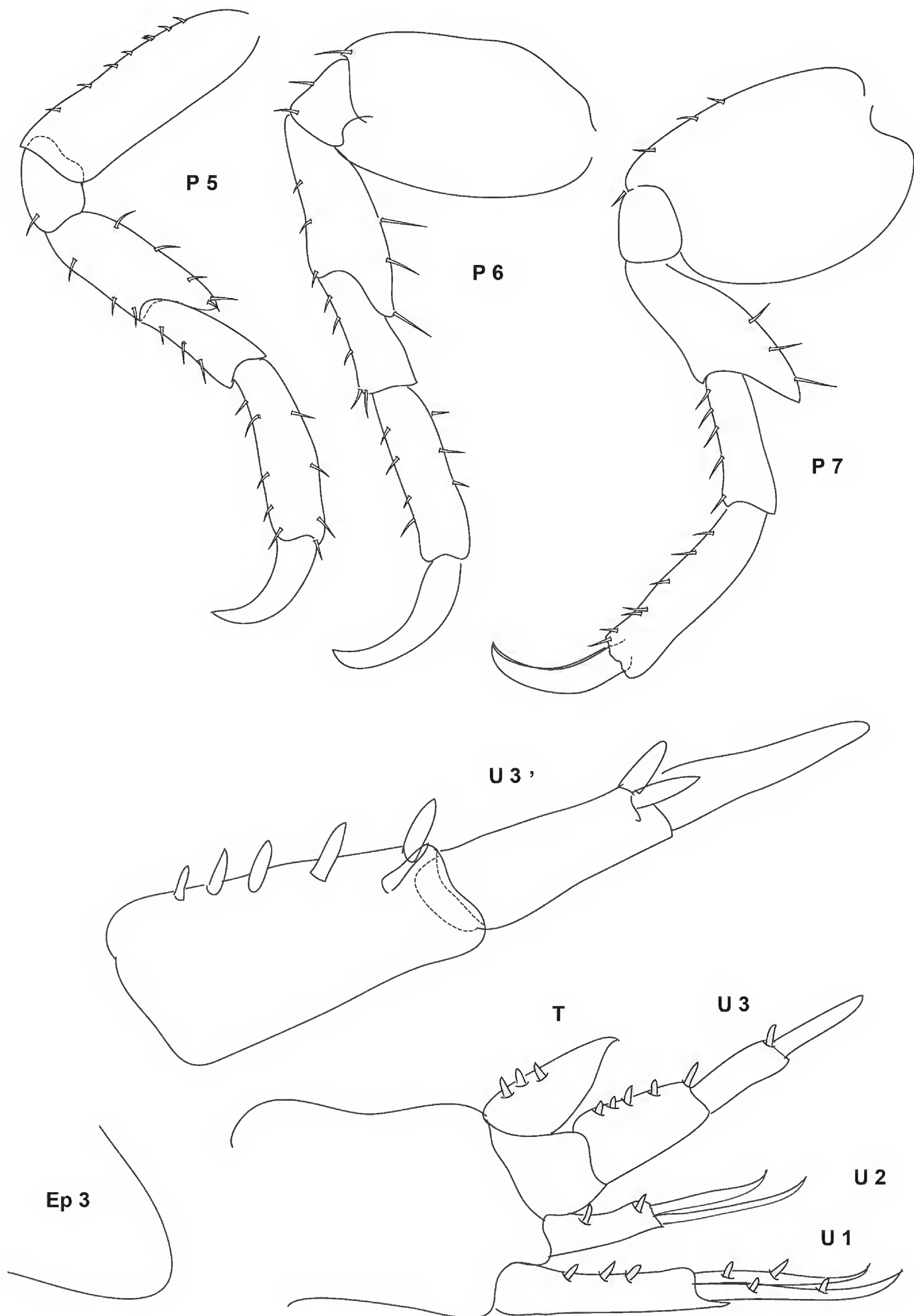
Figure 7. *Stenothoe miersii* (Haswell, 1879) male 3 mm; **P 5–7** peraeopod 5–7; **U 1–3** uropod 1–3; **U 3'** third uropod enlarged; **Ep 3** third epimeral plate; **T** telson.

Material examined. 1 spec. from Bermagui (400 km S of Port Jackson, Australia), 8/6/1989, Wim Vader collected on a hermit crab in shallow water. Stored at the Australian Museum (AM xxx) Sydney.

Remarks. In Haswell (1879) two species of *Montagua* were described from Port Jackson, Sydney: *M. miersii*, directly followed by *M. longicornis*. It appears that the first was the female, the latter the male of the same species, belonging to *Stenothoe*. J.L. Barnard (1974) described four species of *Stenothoe* from Australia. One of these he called *S. ?miersi*, doubting about the synonymy, as no type material is available. Barnard's description matches the different populations around most of the Australian coastline, also the Lizard Island specimens (Krapp-Schickel 2009: 873–875), and again the illustrations given here of a female.

Ecology. It may well be that this specimen lived among the encrusting hydroids and bryozoans growing on top of the hermit-crab-shell and thus had no direct association with the crab; it was the only *Stenothoe* specimen found among many hermit crabs.

Genera *Metopa* Boeck, 1871 and *Stenula* Barnard, 1962

Lincoln 1979 has synonymized *Stenothoides latipes* Chevreux & Fage, 1925, with *Metopa rubrovittata* Sars, 1883 and transferred both to the genus *Stenula*, a genus coined by Barnard 1962.

Members of *Metopa* are mainly distributed in the Atlantic and Arctic, only very few are living outside. They can be divided into three groups by looking at their Gn 1 palmar corner:

L   Gn 1 locking, palmar corner 120°, propodus widened

SI   simple, Gn 1 palmar corner absent, propodus hind margin straight

N   normal, Gn 1 palmar corner 150–160°, propodus hind margin rounded

a) Atlantic Ocean and Arctic:

M. abyssalis Stephensen, 1931 .... N
M. aequicornis Sars, 1879 .... N
M. affinis Boeck, 1871 ....SI/N
M. alderi (Bate, 1857)... N
M. boeckii Sars, 1892... N
M. borealis Sars, 1883 ....SI
M. bruzelii (Goës, 1866)..SI
M. clypeata (Krøyer, 1842) ....SI
M. cristata Gurjanova, 1955 L
M. eupraxiae Krapp-Schickel, 2009 L
M. gigas Just, 2013 ....SI
M. glacialis Krøyer, 1842 L
M. groenlandica Hansen, 1888...................................L
M. hearni Dunbar, 1954.. N
M. invalida Sars, 1892 ....SI.
M. latimana Hansen, 1888.. N

M. leptocarpa Sars, 1883............... L (Md palp lacking?)
M. longicornis Boeck, 1871...SI
M. longirama Dunbar, 1942...SI
M. normani Hoek, 1889 .... N
M. norvegica (Liljeborg, 1851)......................................L
M. palmata Sars, 1895 ....SI
M. propinqua Sars, 1892..SI
M. pusilla Sars, 1892 ....SI
M. quadrangula Reibisch, 1905....................................SI
M. robusta Sars, 1892 ....SI
M. rubrovittata Sars, 1883 .... N
M. samsiluna J.L. Barnard, 1966 .... N
M. sinuata Sars, 1892... N
M. solsbergi Schneider, 1884...................................... N
M. spinicoxa Shoemaker, 1955.................................... N
M. submajuscula Gurjanova, 1948 L
M. spitzbergensis Brüggen, 1907.................................SI
M. tenuimana Sars, 1892 ....SI
M. wiesei Gurjanova, 1933 ....SI

b) Pacific Ocean:

M. abyssi Pirlot, 1933 Pacific .... N
M. angustimana Gurjanova, 1948...............................SI
M. bulychevae Gurjanova, 1955 L
M. cistella J.L. Barnard, 1969.....................................SI
M. colliei Gurjanova, 1948 L
M. dawsoni J.L. Barnard, 1962...................................SI
M. exigua Krapp-Schickel, 2009 .... N
M. japonica Gurjanova, 1952 L
M. kobjakovae Gurjanova, 1955 L
M. koreana Gurjanova, 1952 ....SI
M. layi Gurjanova, 1948 .... N
M. majuscula Gurjanova, 1948....................................L
M. mirifica Gurjanova, 1952...L
M. samsiluna JL Barnard, 1966 L
M. timonovi Gurjanova, 1955 L
M. torbeni Krapp-Schickel, 2009.................................SI
M. uschakovi Gurjanova, 1948 .... N

Many authors have cited *Metopa rubrovittata*: Sars 1883: 90, 1895: 255, Reibisch 1905: 31, Chevreux and Fage 1925: 125, Stephensen 1929: 5, 1931: 189, 1938: 175, Schellenberg 1942: 120, Gurjanova 1951: 432, Oldevig 1959: 44, Moore 1984: 26. None of them gives illustrations of the mouthparts, only Lincoln 1979: 192 found a very short uniarticulate mandible palp in his material from the British coasts which he then called *Stenula rubrovittata* (Sars), confirmed by Vader (see Lincoln op. cit., p. 180) for a specimen from the Norwegian west coast, but we have no information about the mouthparts of the Norwegian material called *Metopa rubrovittata* collected by Sars.

Chevreux 1900 erected a new genus *Stenothoides* for stenothoid species with present, but reduced mandibular palp.

J.L. Barnard 1962 coined a new genus *Stenula* leaving the species with rectilinear basis of P 6 in *Stenothoides* and splitting those species which have P 6 basis widened (see Chevreux and Fage 1925: 130, Gurjanova 1938: 279 and 1951: 445). His diagnosis for *Stenula* is the following:

P 5 basis slender; P 6, 7 basis broad; Md palp with 1 article; Mx1 palp with 1 article.

Barnard 1962 included 10 species in *Stenula*: *S. angusta* (Shoemaker), *S. arctica* (Gurjanova), *S. bassarginensis* (Gurjanova), *S. beringiensis* (Gurjanova), *S. carinatus* (Gurjanova), *S. latipes* (Chevreux & Fage) (type), *S. modosa* J.L. Barnard, 1962, *S. ratmanovi* (Gurjanova), *S. serripes* (Gurjanova), *S. ussuriensis* (Gurjanova), nota bene 7 of 10 species described by Gurjanova from the Arctic Sea.

Just 1980: 52 transferred *Metopa nordmanni* Stephensen, 1931 to *Stenula*.

In the European register of marine species Bellan-Santini and Costello 2001 cited 15 *Stenula* species: the ten above specified by Barnard 1962, plus *S. alexanderi* Tzvetkova & Golikov, 1990, *S. nordmanni* (Stephensen, 1931), *S. peltata* (where they mistakenly cited Della Valle 1893 as author instead of Smith 1874) and – following Lincoln 1979 – they placed *Stenothoides latipes* Chevreux & Fage, 1925 in junior synonymy with *S. rubrovittata* (Sars, 1892), which according to them therefore should be the actual type at the moment. We do not think this is correct, as *Stenothoides latipes* remains in any case the type.

S. carinatus (Gurjanova) was transferred to *Metopa* and renamed *M. eupraxiae* sp. n. by Krapp-Schickel 2009b.

Thus at the beginning of this study 14 species belong to *Stenula*. Judging from the illustrations of the mandible in Tandberg 2011: fig. 25, *M. invalida* Sars, 1892 from N. Norway has to be added as 15th species. These species are mainly living in the far north region (N-Atlantic, N-Pacific, Arctic), only two of them were described by J.L. Barnard from California.

Tandberg & Vader could demonstrate in Tandberg (2011), that the character of Gn 2 palmar corner present/absent does not bring any clear results in a cladistic analysis. E.g. *Metopa clypeata* (the type species) or *M. palmata*, both with strongly rectangular palmar corner, strangely enough are not grouped together with *M. alderi* = *M. spectabilis* or *M. norvegica*, probably because of the strong allometry, which shows their members with very different palmar corners depending on age. It might therefore be more helpful to look at the shape of Gn 1, which shows normally much less allometry and which can be basic (the type of *Stenula* plus several other members of this genus and a lot of *Metopa*, with the carpus shorter than or equal to the propodus) or elongated (type of *Metopa* and some other *Stenula*, with Gn 1 simple and carpus, often also propodus, much lengthened and narrow).

Tandberg 2011 cites in her thesis at the beginning a letter from G.O. Sars to Sparre-Schneider, writing „I have advanced to the supposedly most difficult of all amphipod-families: *Stenothoidae*". There is no doubt that there is a great difference between having a fully developed mandibular palp (*Metopa*) or none (*Stenothoe*), but the genus *Stenula*, as presently conceived, gathers all transitions, and is with high probability heterogeneous.

There are also various transitions within the maxillae, having two (*Stenothoe*) or one (*Metopa*, *Stenula*) articles in Mx1 palp, where often one cannot clearly decide if and where an articulation is present; while the Mx2 plates may sit in tandem-position (many *Metopa* like *M. affinis*, *aequicornis*, *groenlandica*, *glacialis*, *clypeata*) or riding position (in some *Stenothoe* and *Stenula*), with all steps in-between.

In three species we have no information about the mandible palp at all: *S. rubrovittata*, *S. modosa*, *S. peltata*. The following have a short stump, about as long as the width of the mandible-incisor: *S. angusta*, *S. bassarginensis*, *S. ratmanovi*. All other species have a uniarticulate mandible palp which is clearly longer than the mandible-incisor: *S. alexanderi*, *S. arctica*, *S. beringiensis*, *S. incola*, *S. serripes*, *S. ussuriensis*, and also *Metopa derjugini* Gurjanova, 1948, which is therefore here also transferred to *Stenula* (see above). Just 1980: 52 looked at the mandible of *Metopa nordmanni* using the type specimen, and found again a uniarticulate palp longer than the mandible-incisor (also illustrated by Tzvetkova and Golikov 1990), while Shoemaker 1955: 127 found material from Point Barrow with strikingly similar legs but different antennae (A1>A2), a two-jointed Mx1 palp and a 3-articulated mandible palp. Although he cites *Metopa nordmanni* Stephensen, 1931 in the synonymy-list, his species belongs to *Proboloides* and thus is a different animal with nearly identical body but different mouthparts, an observation which can be made rather frequently within Stenothoidae.

Stenula species could also be divided by the ratios of articles in Gn 1, having propodus subequal to carpus, or clearly much longer resp. clearly shorter. The first group is formed by the majority: *S. beringiensis*, *S. derjugini*, *S. incola*, *S. latipes*, *S. modosa*, *S. peltata*, *S. ratmanovi*, *S. serripes*; propodus is longer than carpus in *S. angusta*; propodus is shorter than carpus in *S. arctica*, *S. bassarginensis*, *S. nordmanni*, *S. ussuriensis* and also

S. alexanderi (this species is very aberrant also in the shape of Gn 1 dactylus).

It is the great help of a cladistic analysis that one can test the states of many characters together, and if a group of characters is changing together, it is more probable that a naturally related clade is found. But in the above listed species there are A1 subequal A2 or much different, Gn 1 propodus simple, rounded or with strong palmar corner, Gn 1 carpus short or extremely lengthened, Gn 2 propodus regularly rounded or deeply excavated, P 6, 7 strongly rounded or with widened but parallel margins, telson spinose or naked. And even using more than 60 characters as in the very exhaustive phylogenetic analysis of Tandberg & Vader (Tandberg 2011), there remains the big danger that the character states are not homologous. As an example, several analyses bring *Stenula incola* J.L. Barnard, 1969 from the intertidal of California always closely together with *Stenula serripes* Gurjanova, 1955: both show a one-articulate mandibular palp of medium length, Gn 2 with a well defined palmar corner, P

7 basis about as wide as long and P 7 merus very much lengthened and widened, and they are thus „very similar" after the coded characters. But their biogeography, Gn 1 and P 6 are obviously quite different, and they are most probably not closely related at all.

At the moment there is nothing else to do than to continue „making order" within this complicated family of Stenothoidae in describing as completely as possible its single members.

First we tried to find material of *Metopa rubrovittata* Sars from the northern North Atlantic (type loc. Christiansund and Finnmark) for comparing it with material of *Stenula latipes* (Chevreux & Fage) from the English Channel (type loc. Saint Vaast la Hougue, see Chevreux 1908: 42, 1925: 130).

Metopa rubrovittata Sars, 1883

Figures 8–9, 10B, C
Sars 1883: 90, t. 4, fig. 2, 2a; 1892: 255, pl. 89, fig. 2; Reibisch 1905: 31; Chevreux and Fage 1925: 127, fig., 125; Stephensen 1929: 5; Stephensen 1931: 189; Stephensen 1938: 175; Schellenberg 1942: 120, fig. 98; Gurjanova 1951: 432, fig. 276; Oldevig 1959: 44.

Type locality. Christiansund (W Norway) and Vadsø (Finnmark)

Material examined.
–7 spec. in alcohol, 2 on slide: Surtsey (Vestmannaey-jar) off Iceland, 63.30 N, -20.60 W. NA 30 m 18/8/1971 leg. Sigurdsson, det. J. Just. ZMUC-CRU–4464.

–1 spec. 20/8/1971 same locality as above, ZMUC-CRU–4465.

– 4 spec. North Sea without date, 4 spec., 57.266667 N 5.5 E. ZMUC-CRU–4467.

Discussion. It seemed strange that the sharp eye of Chevreux would have overlooked the synonymy between *M. rubrovittata* (cited by him in the same work Chevreux and Fage 1925: 127, fig. 125) and his newly erected *Stenothoides latipes* (loc. cit.: 129, fig. 127, 128), later transferred to *Stenula* and finally synonymized as *Stenula rubrovittata* (Sars, 1883) by Lincoln 1979. But until now really nobody had looked at the mouthparts of *M. rubro-vittata*, an often cited species, which nevertheless is rarely found in Museum collections.

In fact, the studied material shows a classical mandible palp of *Metopa* species with 3 articles, though it has to be admitted that it was quite a difficult task to see always the articulations. But nevertheless, already the length of the mandible palp is very different in the material from the Channel (cf. Fig. 8 with Fig. 10B, C), thus it can be confirmed that Chevreux was right: both *Metopa rubro-vittata* Sars, 1883 as well as *Stenula latipes* (Chevreux & Fage, 1925) do exist, and they show extremely similar body morphology, colour pattern and even ecological niche, only the mouthparts are somewhat different. Thus, *Metopa rubrovittata* Sars, 1892 is herewith revalidated and *Stenothoides latipes* (Chevreux & Fage, 1925) re-

mains the type species of *Stenula*. It is not clear what the geographical distribution of the two species is, as all the numerous citations cannot be judged without examining their mandible.

For control Jean-Claude Sorbe sent us material from the Bay of Biscay, and the single specimen he had collected affirms this decision.

Stenula latipes (Chevreux & Fage, 1925)

Fig. 10A
Chevreux and Fage 1925: 130, fig. 127–129.

Type locality. Grandcamp-les-Bains (Calvados), on the shell of *Eupagurus bernhardus* (L.); very common in a dredge of 20m depth in Saint-Vaast-la-Hougue. English Channel. Chevreux and Fage 1925: 11 specified that the shell from which the amphipod was collected was *Buccinum undatum* inhabited by the hermit crab.

Material examined. 1 spec. 3 mm, Survey OXYBENT 9 STN OB9–B–TS04; 43.8175 N, 2.042 W; Bay of Biscay, Capbreton Canyon; 500–510 m; 22/06/1999; coll. Sorbe.

4 spec. Denmark, Anholt (Kattegat) without date, 17,5 fathoms = 31,5 m. ZMUC-CRU–4466.

?Stenula peltata (Smith, 1872)

Figures 11–13
Smith 1872 in: Smith and Harger 1872: 29, pl. 3, fig. 5–8; Della Valle 1893: 570; Stebbing 1906: 194–195.
? synonymous to Gurjanova 1948: 310 *S. ratmanovi*

Type locality. St. George's Banks, 55 m depth. Near Cultivator Shoal.

Material examined. one specimen USNM 35636, 41.5557 N, 68.1641 W, NA, 30 fathoms, sandy bottom, 29/8/1872.

As the original paper is not easily accessible and as there is some confusion about the authors, I repeat herewith the type-description by Smith:

Description.
Female. Eyes round and nearly white in alcohol. Antennulae (=A1) considerably shorter than epimera of the 4th segment (Cx 4); first article of the peduncle stout, subequal to head, the second shorter, the third very short and similar to the arts of the flagellum; flagellum scarcely longer than the peduncle, with 8 arts. Antennae (=A 2) slightly longer than antennulae; peduncle art 4, 5 about equal in length; flagellum subequal to flagellum of antennulae. Cx 2 (fig. 5) nearly ovate, twice as high as broad; Cx 3 somewhat rectangular, not wider than the second but considerably deeper; Cx 4 (fig. 6) very large, slightly deeper than Cx 3 and 1/3–1/4 longer than deep, being about as long as the first five segments of the thorax, the inferior margin regularly curved and the posterior convex in outline. Gn 1 (fig. 7) small and slender; merus
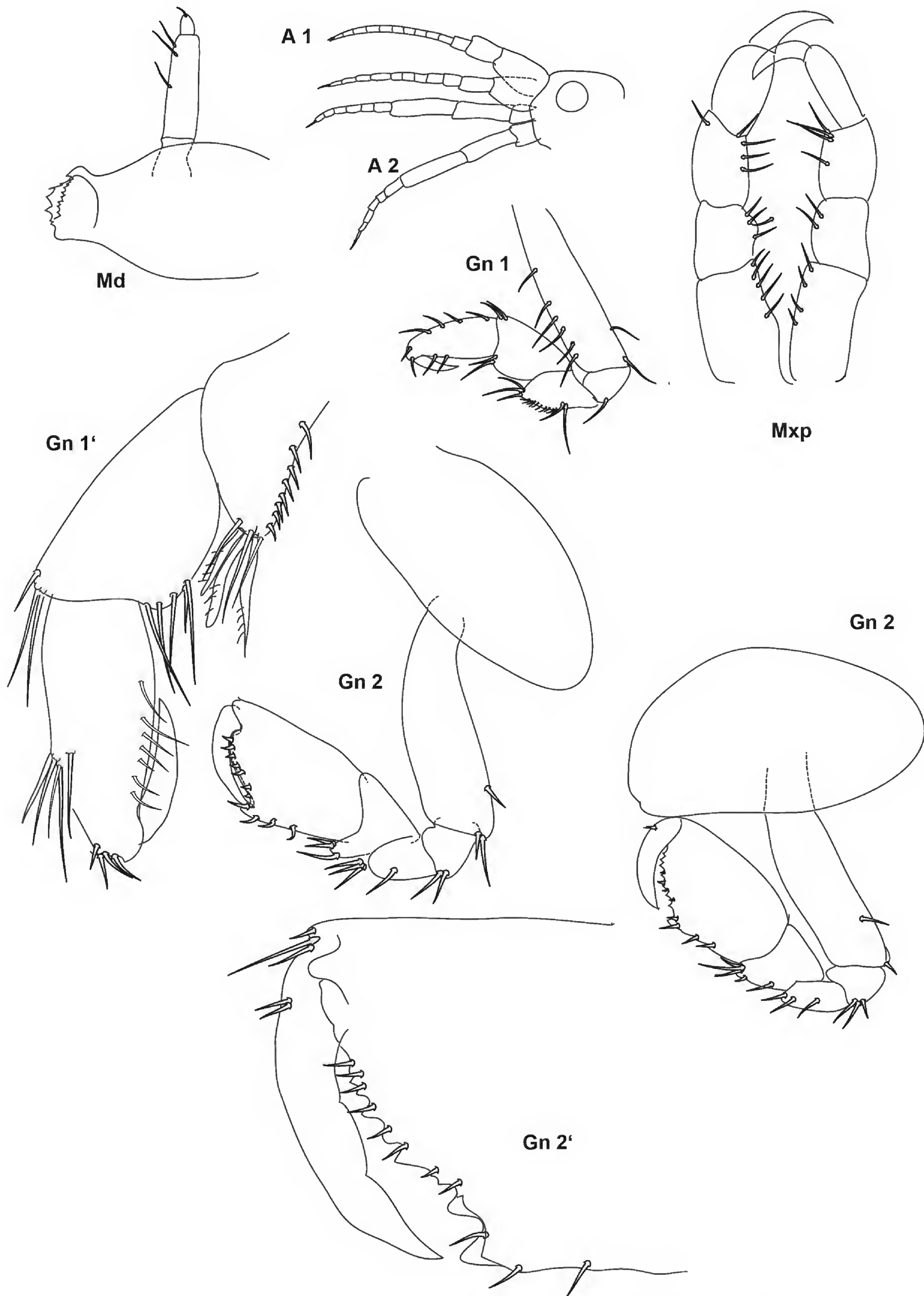
Figure 8. *Metopa rubrovittata* Sars, 1883 male 3 mm; **Md** mandible; **Mxp** maxilliped; **A 1, 2** antenna 1, 2; **Gn 1, 2** gnathopod 1, 2; **Gn 1', Gn 2'** gnathopod 1, 2 distally enlarged.
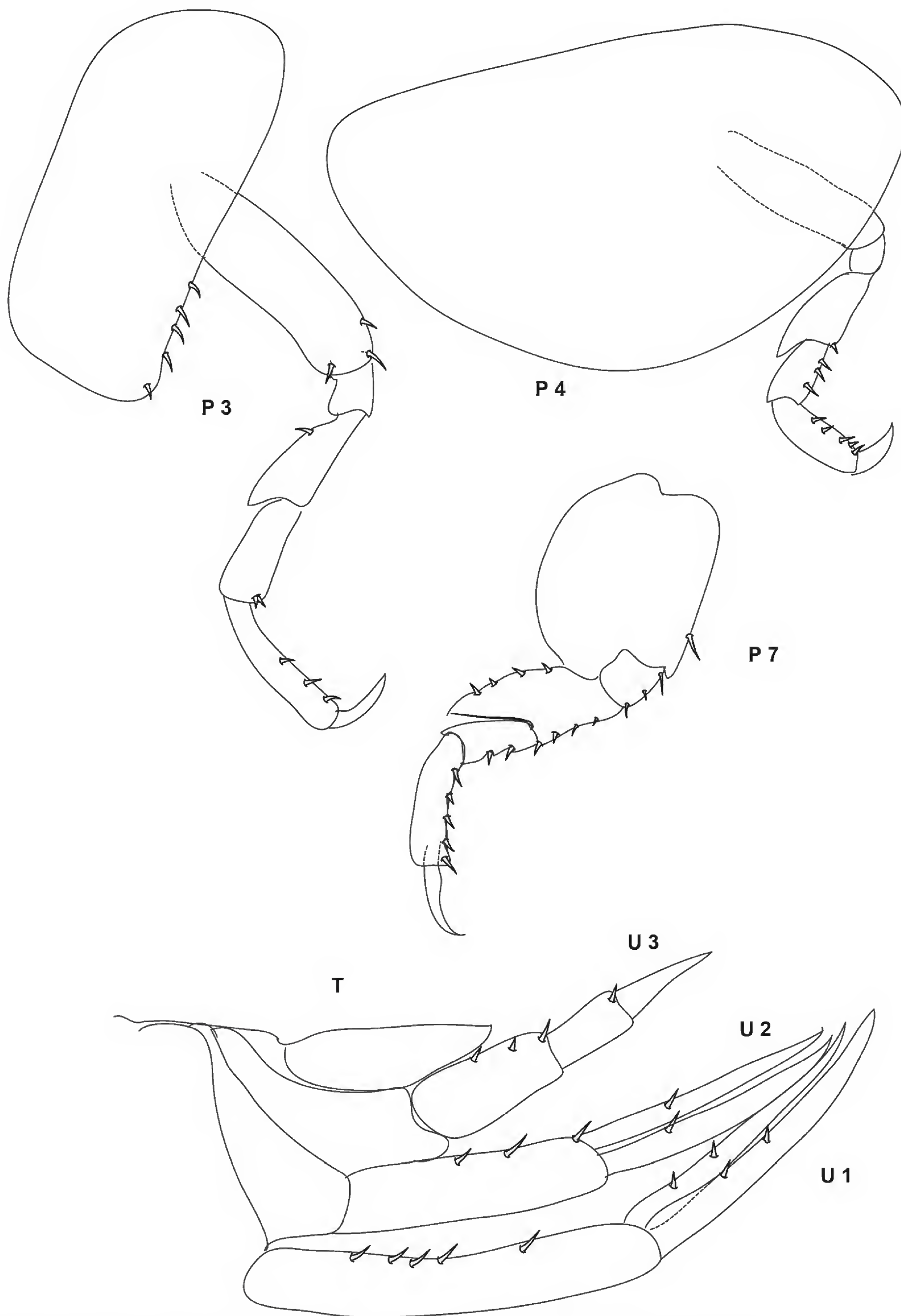
Figure 9. *Metopa rubrovittata* Sars, 1883 male 3 mm; **P 3, 4, 7** peraeopod 3, 4, 7; **U 1–3** uropod 1–3; **T** telson.
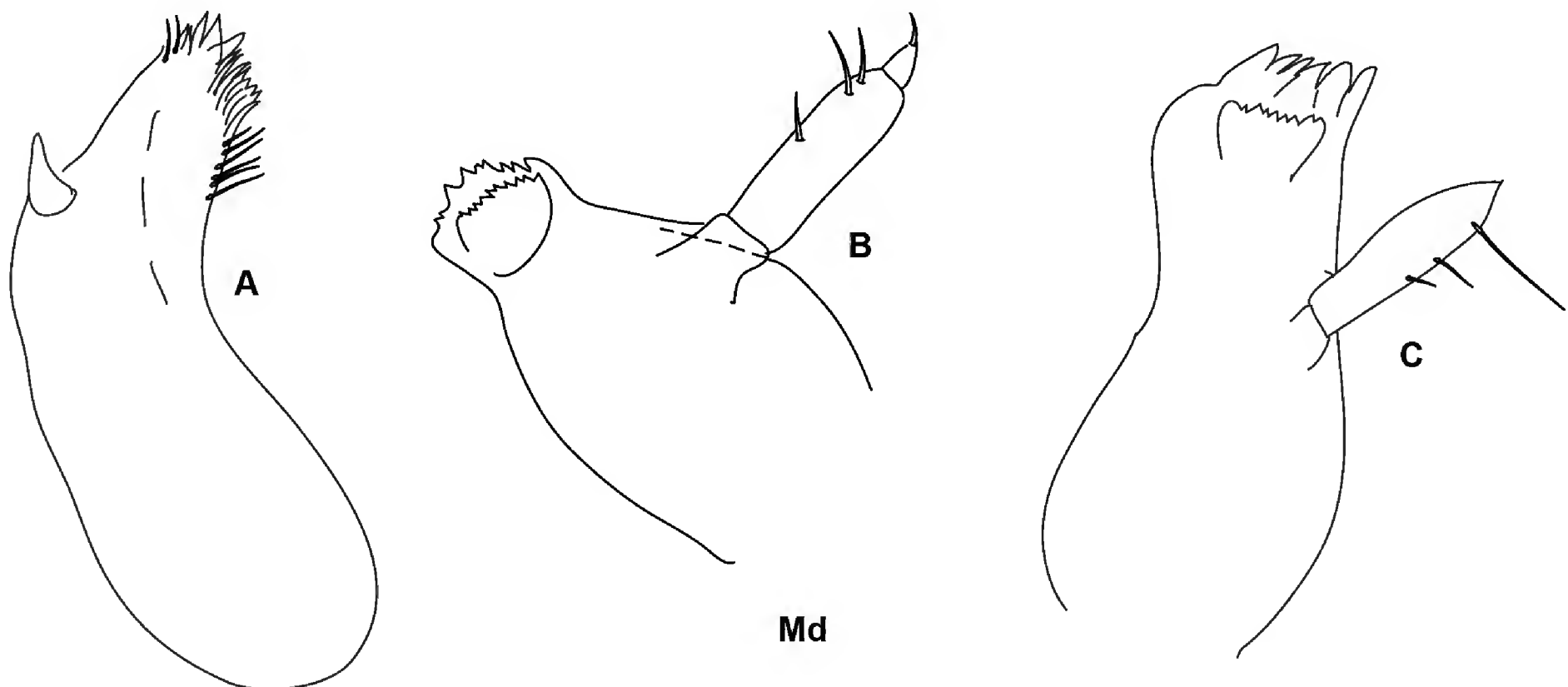
Figure 10. *Stenula latipes* Chevreux & Fage, 1925: **Md A** mandible A; *Metopa rubrovittata* Sars, 1892: **Md B, C** mandible B, C.

triangular and distally broader than the carpus, which is not quite twice as long as broad and has the lateral margins parallel; propodus narrower but slightly longer than the carpus and narrowed distally; dactylus about half as long as the propodus. Gn 2 (Fig. 5) stouter; merus short triangular, carpus much broader than long and only slightly produced beneath the propodus; propodus about as long as the breadth of Cx 2, nearly twice as long as broad; palmar margin (Fig. 8) convex in outline, slightly oblique, with an acute lobe and a spine at the posterior angle, within which the top of the dactylus closes. P 4, 5 slender and nearly naked, P 5 basis slender, four times as long as broad, not wider than the merus. P 6, 7 slightly shorter than P 5, basis posteriorly dilated and squamiform in both pairs, but broader in P 7. U 3 ramus slightly longer than the peduncle.

Length of largest specimen, from front of head to tip of telson, about 6 mm.

The mandibles are without palp or molar tubercles, and in all other characters the species agrees with the genus *Stenothoe* as restricted by Boeck, but it seems to be very distinct from either of the European species.

Discussion. The hint after the original description, that this species should belong to *Stenothoe* as it has no mandible palp, was not convincing: no *Stenothoe* is described from the region off Massachusetts or Connecticut, nor from the entire Atlantic, with gnathopods similar to the ones illustrated.

The incomplete illustrations of *S. ratmanovi* (Gurjanova, 1948) are very similar to what little we know about ?*Stenothoe peltata*, and the two species may well be synonymous, in spite of the large geographic distance between the type localities. In that case the older name *Stenula peltata* (Smith, 1874) would become the valid name of the taxon.

We hoped to get more information by studying the single type specimen (see Fig. 11, 12) and illustrate here all what we could see; but there were no mouthparts except the maxilliped, and we still don't know anything about the shape of the mandibular palp.

A sample in the collections of the Smithsonian Inst. (Washington) raised new hope to shed light in this situation: there could exist a *Stenula* sp. from the coelenteron of *Haliactis arctica*. Will this be *S. peltata*?

Stenula pugilla sp. n.

http://zoobank.org/F3F651B1-E8C4-430E-8184-219956DFE464
Figures 14–18
Vader 1983: 146, sub *Stenothoe* sp.

Holotype. male 3 mm in alcohol; USNM 1241824; M/v"John N.Cobb", cruise 43, sta. 45, Project Chariot Cruise, Vessel John N Cobb R/V; Chukchi Sea: 67°31'N, 167°12'W coll. Spark. 27 fm = 49 m depth, 19/8/1959. Gear dredge. From coelenteron of *Haliactis arctica* T. Bowman Acc.No.234238.

Additional material. male, female on 2 slides, both 3 mm.

Type locality. Chukchi Sea, Arctic. From coelenteron of *Haliactis arctica*.

Etymology. the epitheton should remind on the shape of the propodus Gn 2, which looks somewhat like a small fist, in Latin „pugilla"; it is used as noun in apposition.

Description.
Length 3 mm.
Head. Eyes round, normal. Mouthparts: Md with very short palp, length about half of width of incisor. Mx 1
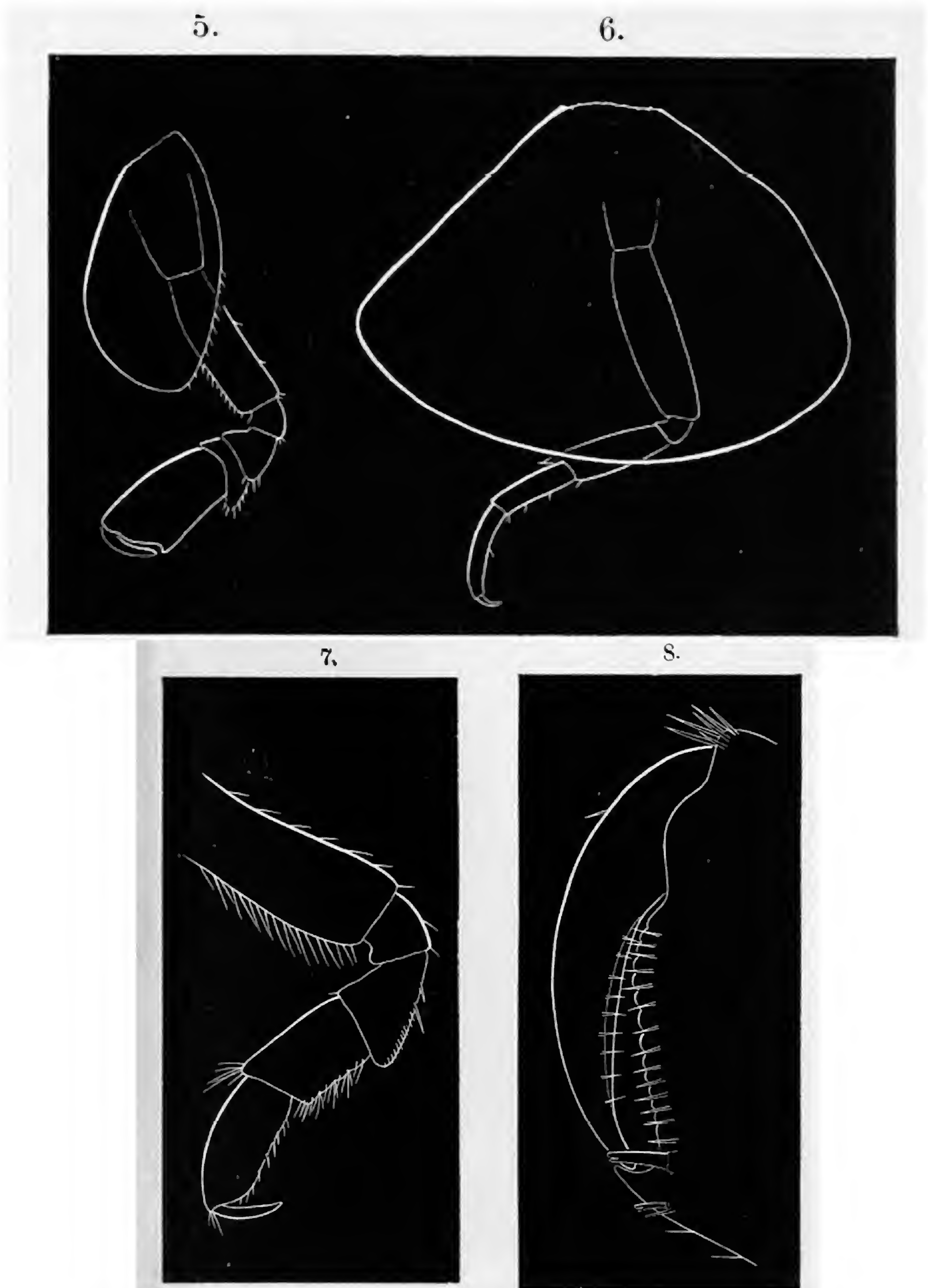
Figure 11. *Stenula peltata* (Smith, 1872): original illustrations of **5** gnathopod 2; **6** ♀ peraeopod 4; **7** gnathopod 1; **8** dactylus and propodus of gnathopod 2 distally.
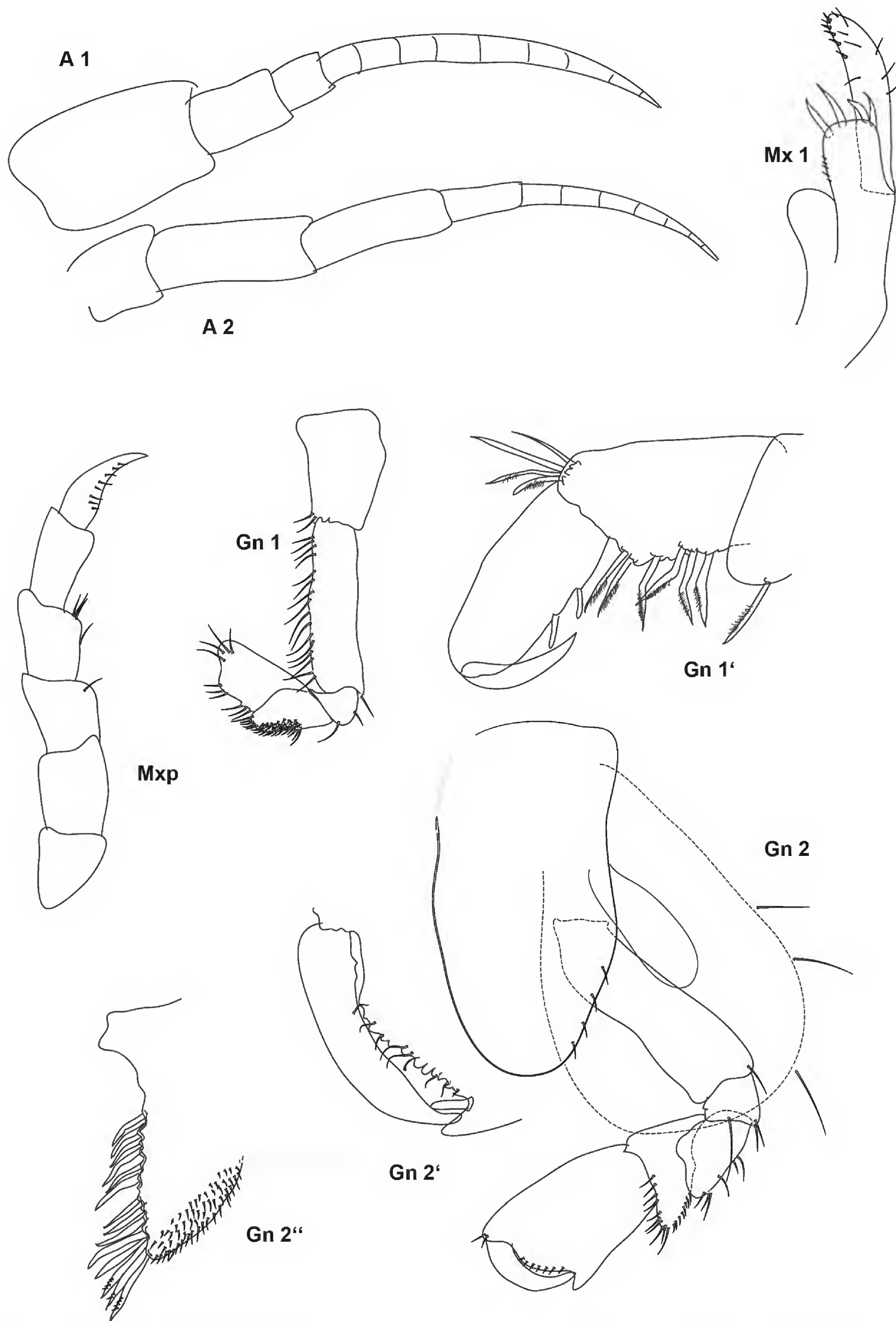
Figure 12. *Stenula peltata* (Smith, 1872): illustration of the single type specimen; **A 1, 2** antenna 1, 2; **Mx 1** maxilla 1; **Mxp** maxilliped; **Gn 1** gnathopod 1 without propodus and dactylus; **Gn 1'** dactylus, propodus and carpus enlarged; **Gn 2** gnathopod 2; **Gn 2'** dactylus and propodus enlarged; **Gn 2''** gnathopod 2 tip of carpus enlarged.
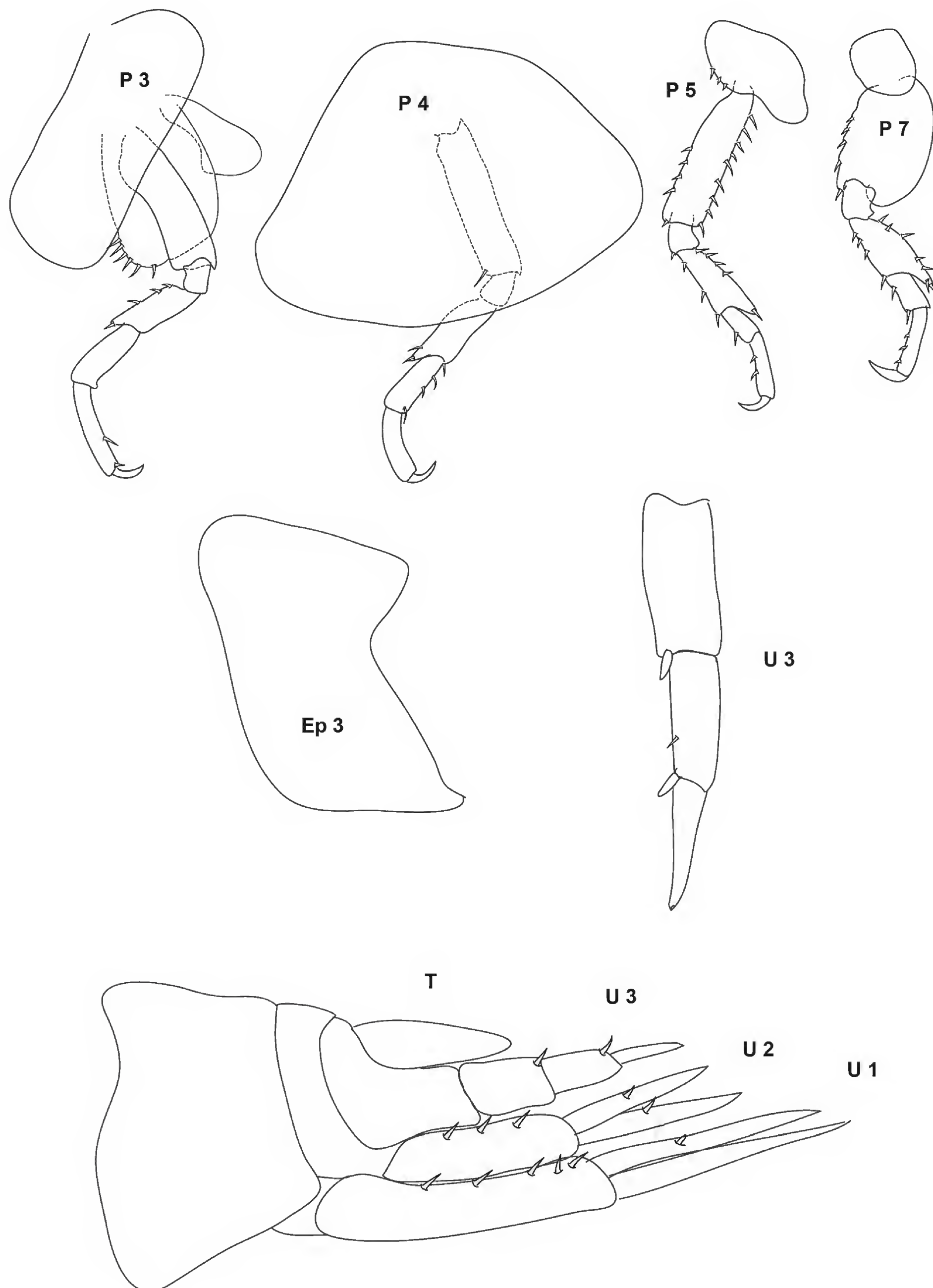
Figure 13. *Stenula peltata* (Smith, 1872): illustration of the single type specimen; **P 3, 4 5, 7** peraeopod 3, 4, 5, 7; **Ep 3** third epimeral plate; **U 1, 2, 3** uropod 1, 2, 3; **T** telson.
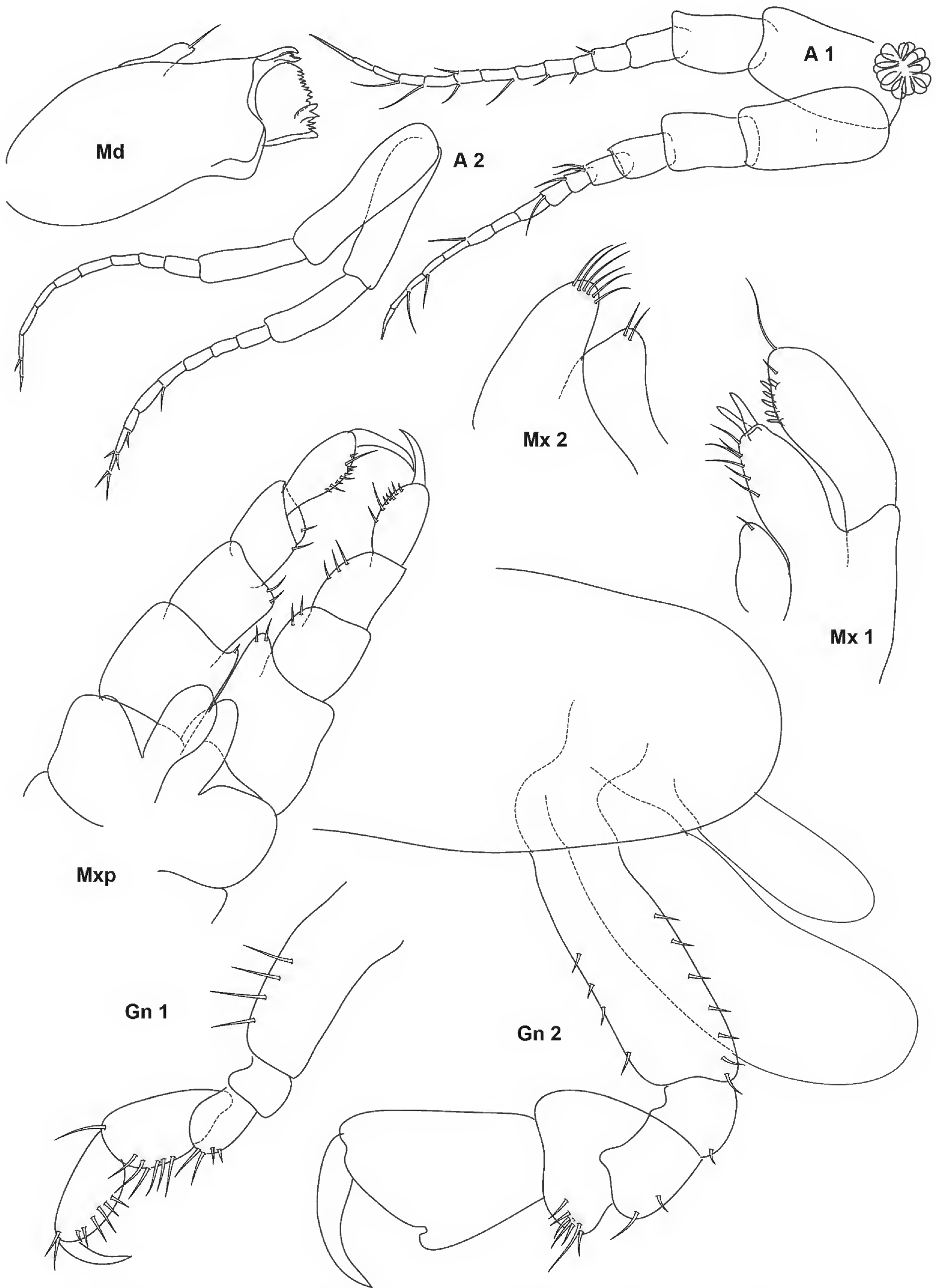
Figure 14. *Stenula pugilla* sp. n. female 3 mm: **A 1, 2** antenna 1, 2; **Md** mandible; **Mx 1, 2** maxilla 1, 2; **Mxp** maxilliped; **Gn 1, 2** gnathopod 1, 2.
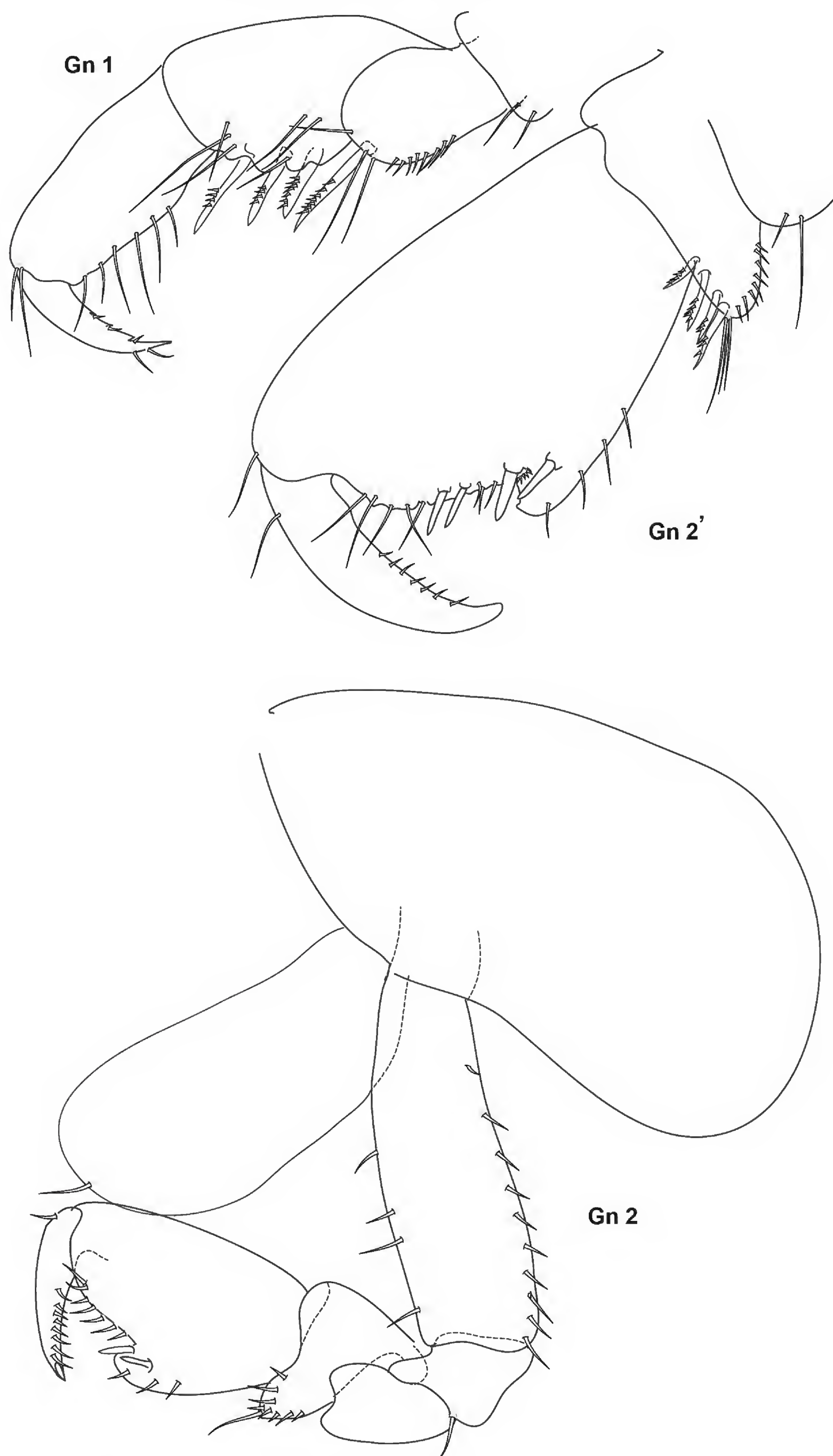
Figure 15. *Stenula pugilla* sp. n. female 3 mm: **Gn 1** gnathopod 1 distal arts; **Gn 2** gnathopod 2; **Gn 2'** gnathopod 2 distally enlarged.
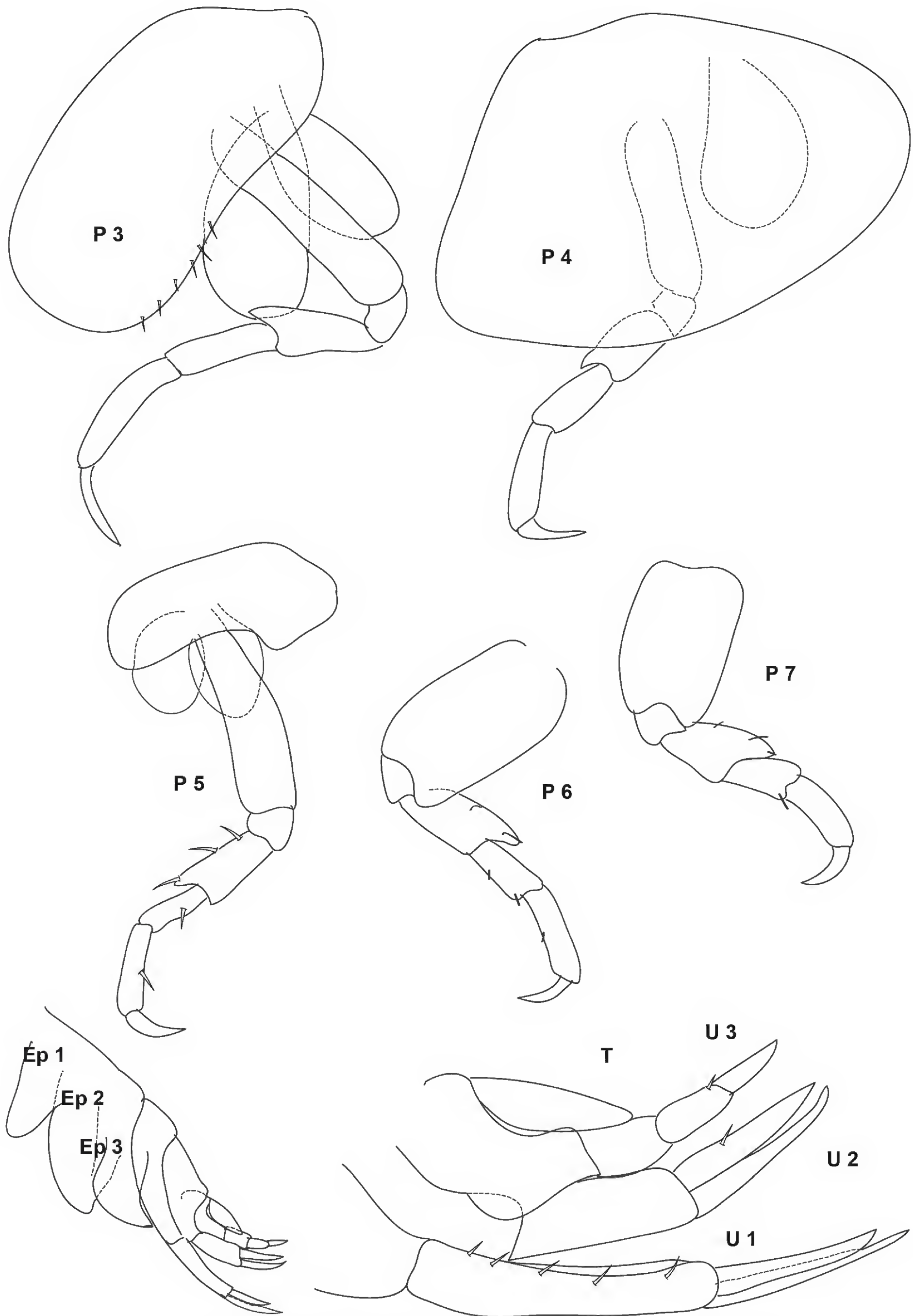
Figure 16. *Stenula pugilla* sp. n. female 3 mm: **P 3–7** peraeopod 3–7; **Ep 1–3** epimeral plate 1–3; **U 1–3** uropod 1–3; **T** telson.

Gn 1

Gn 2

Gn 2'

Figure 17. *Stenula pugilla* sp. n. male 3 mm: **Gn 1** gnathopod 1; **Gn 2, Gn 2'** gnathopod 2 from both sides.

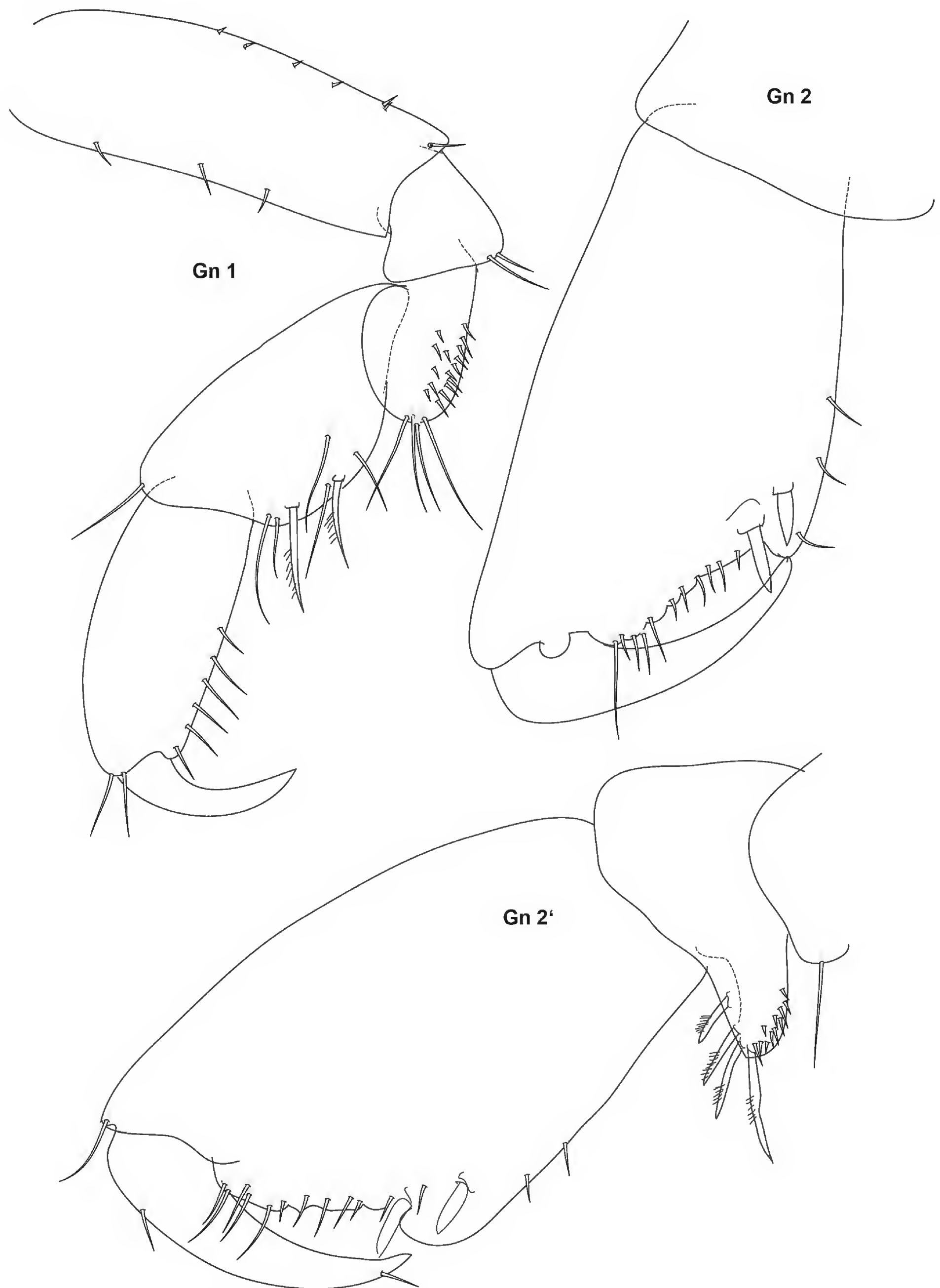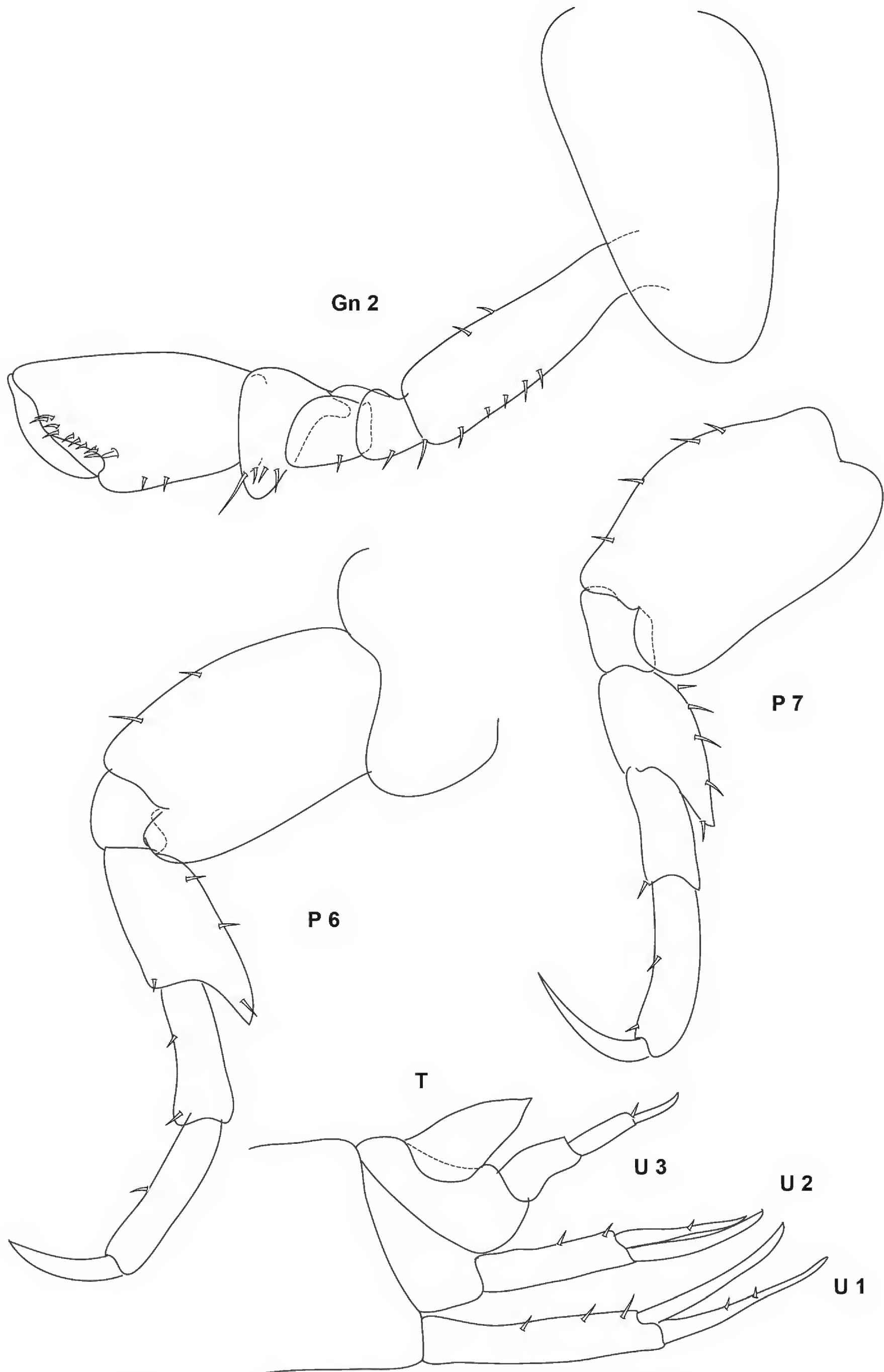**Figure 18.** *Stenula pugilla* sp. n. male 3 mm: **Gn 2** gnathopod 2; **P 6, 7** peraeopod 6, 7; **U 1–3** uropod 1–3; **T** telson.

palp with one art, Mx 2 plates in tandem – position. Antennae: subequal, A 1 and A 2 flagellum about 10–12 arts. Mxp length of inner plate about half length of ischium, outer plate about 1/3–1/2 of merus.

Peraeon. Gn 1 basis on anterior margin setose, merus with short stiff setae, carpus with long setae and pectinate spines, propodus hind margin setose, somewhat rounded to nearly straight.

Cx 2 tongue-shaped. Gn 2 male and female basis on both margins beset with setae; merus rectangular, naked, carpus triangular, with stiff setae posterodistally, propodus medially widest with setose palm, defined by thumb-shaped protrusion.

Peraeopods: Cx 3 narrow, distally rounded, posteriorly with some setae; Cx 4 distally about 3× wider than Cx 3. P 3 dactylus clearly longer than in other peraeopods; P 3–7 merus somewhat widened and not much lengthened; P 6, 7 basis widened, with parallel margins.

Pleon. U 1 peduncle spinose, > slightly unequal rami; U 2 rami subequal; U 3 peduncle = ramus art 1 = ramus art 2.

Telson naked, triangular.

Female: subsimilar to male.

Remarks. The note ‚from the coelenteron of *Haliactis*' on the label of this sample may as well just have meant that the sea anemones had contracted on collection.

Discussion. Within the above discussed criteria of dividing *Stenula* species into groups, the new species belongs to the majority having Gn 1 propodus subequal to carpus (together with *S. solsbergi*, see below, here transferred to *Stenula*) and to the few members having a very short stump of mandible palp. The shape of Gn 2 palm male and female defined by a thumb-like hump is unique and quite helpful in identifying this species.

Stenula solsbergi (Schneider, 1884)

Figures 19, 20
Metopa solsbergi Schneider, 1884: 71; Sars 1892: 266, t. 94; Lincoln 1979: 186, fig. 84.

Material examined. one specimen, 3 mm, from the vicinity of Tromsø, N Norway (without date), collected together with *Chlamys*. The material was dredged in an area with much *Metridium*.

Type locality. Malangenfjord, Norway; 18 m depth.

Vader 1983 reported already *Metopa solsbergi* from the North Atlantic: Elmhirst 1925 found this species present on *Metridium senile* L. on pier piling in western Scotland, Fenwick and Steele 1983 off the coast of Newfoundland, Canada, again on *Metridium*.

This seems to be the very first time that the mouthparts were checked, and a reduced, uniarticulate mandibular palp could be illustrated, moving also this species to *Stenula*.

At the end of our study, we now know 16 members of the genus *Stenula*, as *S. invalida*, *S. solsbergi* and *S. pugilla* sp. n. are added, *S. arctica* given in synonymy with *S. nordmanni*, *S. latipes* revived and *S. rubrovittata* put back into *Metopa*:

S. alexanderi Tzvetkova & Golikov, 1990; Siberia
S. angusta (Shoemaker, 1955); Alaska, N Pacific
S. bassarginensis (Gurjanova, 1948); Arctic
S. beringiensis (Gurjanova, 1948); Bering Sea, N Pacific
S. derjugini (Gurjanova, 1948); Bering Sea, N Pacific
S. incola J.L. Barnard, 1969; California, Pacific
S. invalida (Sars, 1892); Atlantic
S. latipes (Chevreux & Fage, 1925) (type); Atlantic
S. modosa J.L. Barnard, 1962; California, Pacific
S. nordmanni (Stephensen, 1931); Greenland, ?SW-North Sea (fide Schellenberg 1942: 120) ?
 (syn. with *S. arctica* (Gurjanova, 1951); Arctic)
?*S. peltata* (Smith, 1872); Atlantic (generic allocation doubtful)
S. pugilla sp. n.; Chukchi Sea, Alaska
S. ratmanovi (Gurjanova, 1948) (could be junior synonym of *S. peltata*)
S. serripes (Gurjanova, 1955); Kurile Isl., NE Pacific
S. solsbergi (Schneider, 1884); N. Norway, N. Atlantic
S. ussuriensis (Gurjanova, 1948); Japan Sea, NE Pacific

Key to world *Stenula* s. l.

The amphipod genus *Stenula* is probably not a monophyletic entity (cf. Tandberg and Vader 2011, this paper), and it is at present not possible to decide which taxa belong to it. In this key we have therefore cast our nets widely, and we include all species in the *Metopa-Stenula* complex with a uniarticulate mandible palp. This palp is very short in what we might call ‚typical *Stenula*', a bit longer, but still shorter than the incisor of the mandible, in a number of other species, also traditionally placed in *Stenula*, and still a bit longer, but clearly uniarticulate, in a few *Metopa* species: *M. hearni*, *M. palmata* and *M. sinuata*. Just's (1980) „*Stenula* sp." is in our opinion identical with *M. sinuata*, as that author himself already suspected.

The task has been made more difficult by several factors: many species have only been partly illustrated, and at least for the species *S. angusta*, *S. invalida*, *S. modosa* and ?*S. peltata*, as well as possibly some of Gurjanova's species, males are still unknown.

We treat here the 16 *Stenula* species mentioned above, plus three species which are closely related, but until now still left unchanged in *Metopa*:

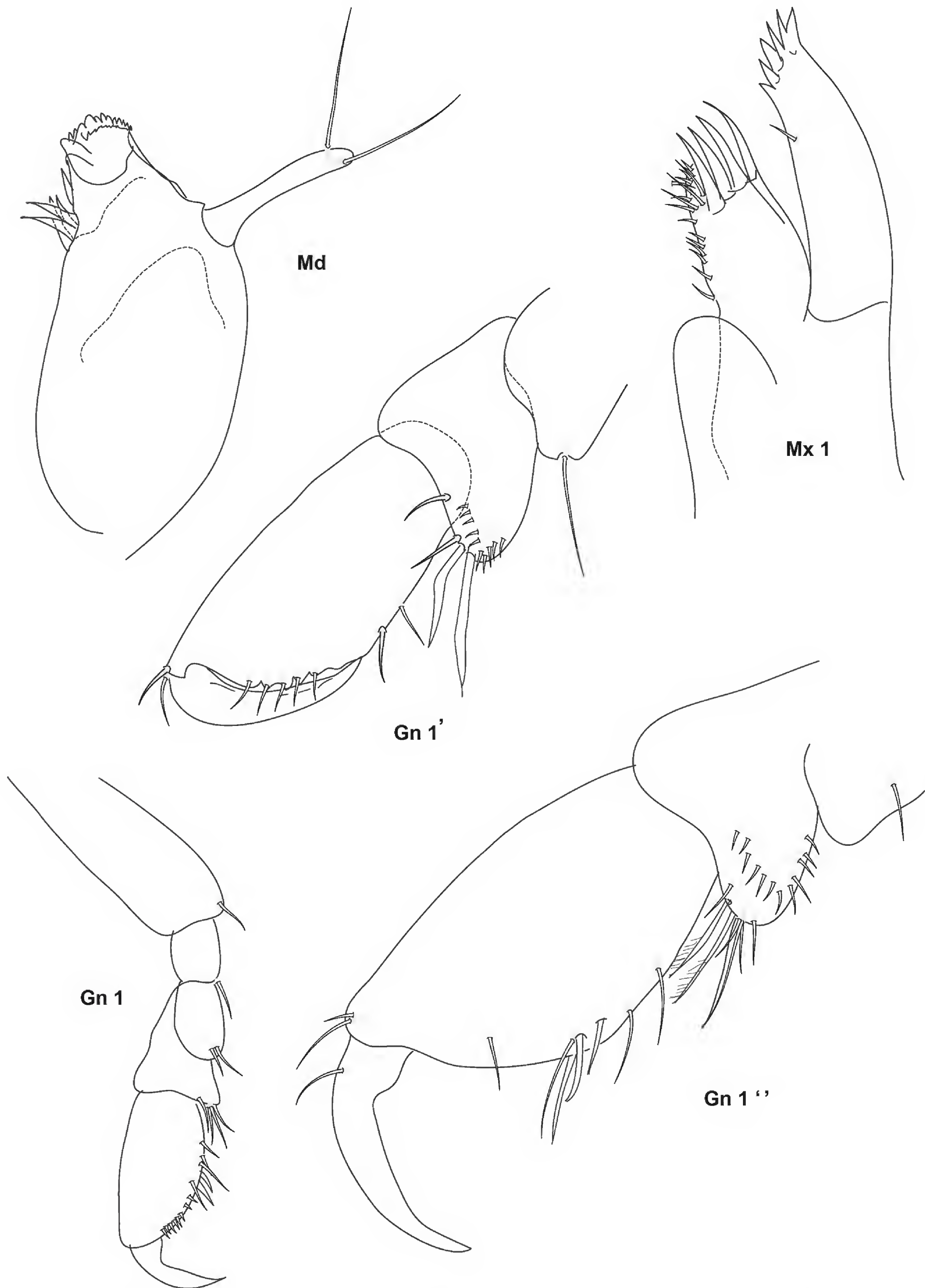**Figure 19.** *Stenula solsbergi* (Schneider, 1884): **Md** mandible; **Mx 1** maxilla 1; **Gn 1** gnathopod 1; **Gn 1', Gn 1 ''** gnathopod 1 right and left distally enlarged.
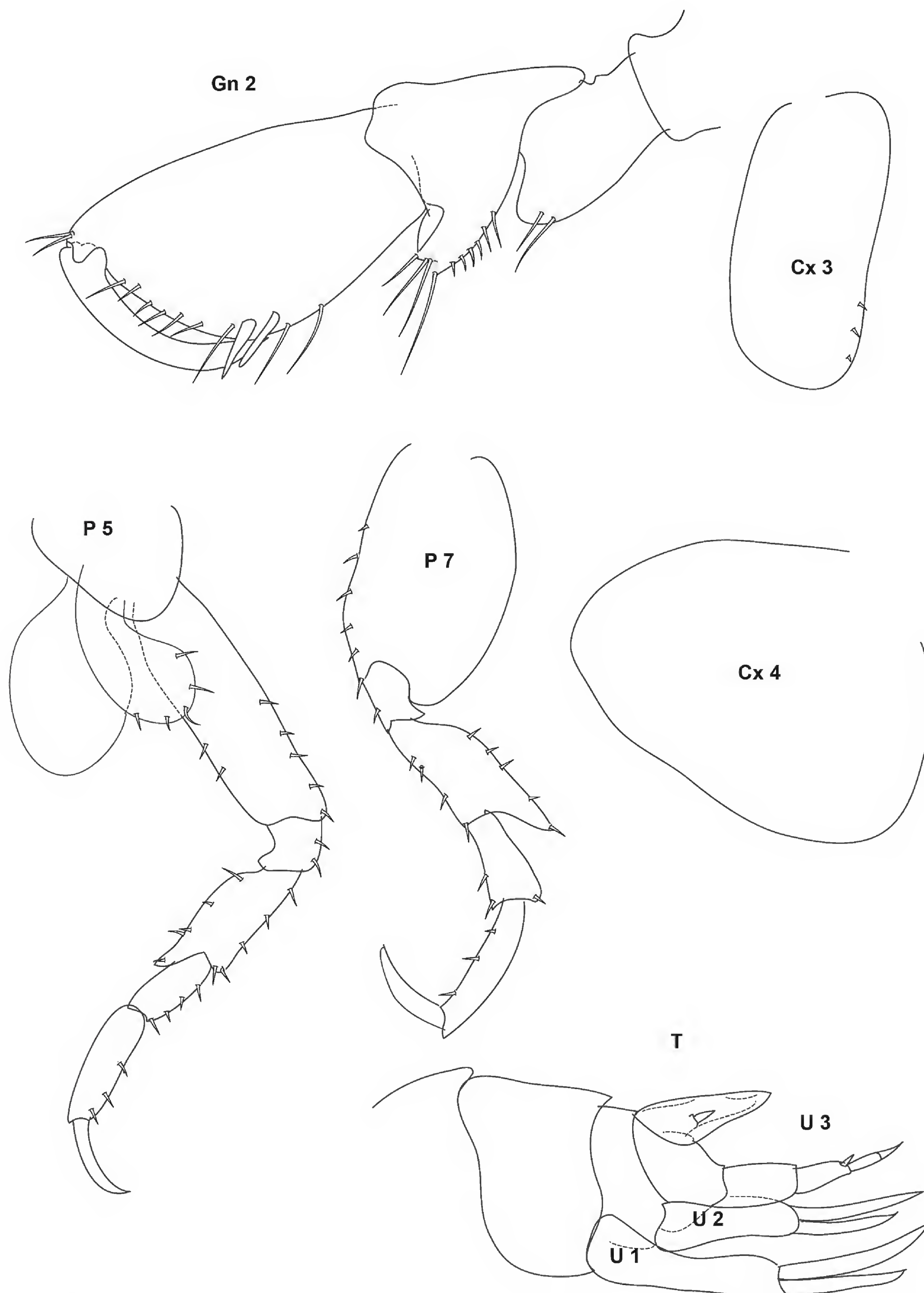
Figure 20. *Stenula solsbergi* (Schneider, 1884): **Gn 2** gnathopod 2; **Cx 3, 4** coxa 3, 4; **P 5, 7** peraeopod 5, 7; **U 1–3** uropod 1–3; **T** telson.

1 Coxa 4 distal margin clearly sinuous; N. Norway, 4 mm...*M. sinuata* Sars, 1892

– Coxa 4 distal margin regularly convex or almost straight 2

2 Gn 1 propodus and carpus very long and slender, dactylus broad and heavily setose; Gn 2 rectipalmate 3

– Gnathopods not as above 4

3 Gn 1 propodus < carpus, palm concave, dactylus length to width > 2; N. Norway, 5 mm...
.... *M. palmata* Sars, 1892 (see note 1)

– Gn 1 propodus > carpus, palm convex, dactylus length to width = 3:2; Laptev Sea, 3.8 mm...
....*S. alexanderi* Tzvetkova & Golikov, 1990 (see note 2)

4 P 6 –7 basis and merus posteriorly serrated, hind margin of basis proximally with acute tooth; Kurile Islands, NE Pacific, 7 mm..*S. serripes* (Gurjanova, 1955)

– P 6 –7 basis not serrated nor with tooth 5

5 Gn 2 propodus in male with deep U-shaped excavation.. 6

– Gn 2 propodus in male not with deep U-shaped excavation.. 7

6 Gn 2 propodus palm in male semicircularly excavated near distal corner, rest of palm much longer than width of excavation; Japan Sea, 4 mm.. *S. ussuriensis* (Gurjanova, 1948)

– Gn 2 propodus palm in male deeply and irregularly excavated, rest of palm shorter than width of excavation. Japan Sea, 5 mm...*S. bassarginensis* (Gurjanova, 1948)

7 Gn 2 propodus male palm clearly irregular, defined by strong tooth... 8

– Gn 2 propodus in male smooth or slightly crenulate, convex or straight 9

8 Telson with 3 pairs of spines; Kamchatka, NE. Pacific, 3.5 mm....................................*S. beringiensis* (Gurjanova, 1948)

– Telson naked or with small setules; California, 2 mm...*S. incola* Barnard, 1969

9 Gn 2 propodus palmar corner prominent, shaped like a finger-tip; Chukchi Sea, 3 mm ....*S. pugilla* Krapp-Schickel & Vader, 2015

– Gn 2 propodus palmar corner blunt or with acute tooth 10

10 Gn 1 carpus unusually long, up to 3 × longer than wide; eyes very large.. 11

– Gn 1 carpus clearly not as long; eyes normal 12

11 Gn 1 dactylus not reaching half length of propodus; A1 clearly shorter than A2; P 6–7 basis with rounded hind margin; W. Greenland, 5 mm .... *S. nordmanni* (Stephensen, 1931) (= *S. arctica* (Gurjanova , 1951) (see note 3)

– Gn 1 dactylus reaching half length of propodus. A1 and 2 subequal; P 6–7 basis with straight hind margin; California, 2 mm...*S. modosa* Barnard, 1962

12 Telson with 2 pairs of spines; Bering Sea, 4 mm. ....*S. derjugini* (Gurjanova, 1948)

– Telson naked.. 13

13 Gn 1 carpus clearly shorter than propodus 14

– Gn 1 carpus subequal to propodus.. 15

14 P 7 very broad, posterior margin convex; mandiblepalp long, but unarticulated; Canada, 3–4 mm ....
.... *M. hearni* (Dunbar, 1954)

– P 7 basis slender, posterior margin rather straight; mandible-palp shor, Point Barrow, Alaska, 3 mm. ....
.... *S. angusta* (Shoemaker, 1955)

15 P 6–7 posterior tip on merus reaches halfway of less along carpus 16

– P 6–7 posterior lobe on merus reaches clearly further than halfways along carpus 17

16 Gn 2 propodus length:width = 3. P 7 basis broad, length = 2 width.; N. Norway, 4 mm..............*S. invalida* (Sars, 1892)

– Gn 2 propodus l:w = 2; P7 basis less wide, l< 2w, N. Norway, 7 mm.......................... *S. solsbergi* (Sp. Schneider, 1884)

17 We have been unable to find reliable differences between *Stenula latipes* (Chevreux & Fage, 1925), a species associated with hermit crabs in W. Europe, and *S. ratmanovi* (Gurjanova, 1948), an only partly described species from Kamchatka in the northern Pacific. Moreover, the illustrations of this latter species and those of *Stenothoe peltata* (Smith, 1874) from Georges Banks, NW Atlantic, are, as far as they go, practically identical.

Notes

1) This is the type species of Norman's (1902) short-lived genus *Sthenometopa*.

2) Probably not a *Stenula*, to be transferred to *Metopa* or *Sthenometopa*.

3) In transferring *Metopa nordmanni* to *Stenula*, Just (1980) apparently overlooked the fact that this species is clearly identical to *Stenula arctica* (Gurjanova, 1951).

Table 1. Associations of Stenothoidae with larger marine invertebrates.

Amphipods	Hosts	Reference
on Sea anemones		
Parametopella antholobae Krapp-Schickel & Vader, 2009	*Antholoba achates* (Drayton, 1846)	Krapp-Schickel and Vader 2009
Stenothoe barrowensis Shoemaker, 1955 Point Barrow, Alaska	unidentified	Shoemaker 1955; Vader 1983
Stenothoe bartholomea Krapp-Schickel & Vader, 2015 Florida Keys	*Bartholomea annulata* (Lesueur, 1817)	this paper, Vader 1983 (as *Stenothoe* sp. n.)
Stenothoe boloceropsis Krapp-Schickel, Häussermann & Vader, 2015 Chiloe Island, Chile	*Boloceropsis platei* McMurrich, 1904	Krapp-Schickel et al. 2015
Stenothoe brevicornis G. O. Sars, 1883 N. Norway, Newfoundland, Canada Stellwagen Bank, Canada	*Actinostola callosa* (Verrill, 1882) *Liponema multicornis* (Verrill, 1880)	Vader and Krapp-Schickel 1996 Fenwick and Steele 1983 Auster et al. 2011
Stenula pugilla Krapp-Schickel & Vader, 2015 Chukchi Sea	*Haliactis arctica* Carlgren, 1921	this paper (see also Vader 1983, as *Stenothoe* sp.)
Stenula solsbergi (Sp. Schneider, 1884) W.Scotland Newfoundland, Canada	*Metridium senile* (L., 1767)	(as *Metopa solsbergi*) Elmhirst 1925 Fenwick and Steele 1983
Other large coelenterates		
Metopa bruzelii (Goes, 1866) Newfoundland, Canada	*Primnoa resedaeformis* (Gunnerus, 1763)	Buhl-Mortensen and Mortensen 2004, 2005
Proboloides calcarata G. O. Sars, 1883 Newfoundland, Canada	*Primnoa resedaeformis*	Buhl-Mortensen and Mortensen 2004, 2005
Stenothoe minuta Holmes, 1905 N. Carolina, USA	*Astrangia danae* Milne-Edwards & Haime, 1849	Pearse 1947
Stenothoe valida Dana, 1853 Barbados, W.Indies	*Millepora complanata* Lamarck, 1816	Lewis 1992
Stenula nordmanni (Stephensen, 1931) Newfoundland, Canada	*Gersemia* sp.	Fenwick and Steele 1983 (as *S. arctica*)
Torometopa sp. Antarctic	*Primnoella* sp.	De Broyer et al. 2003?
Hydromedusae		
Metopa alderi (Sp. Bate, 1857) E. Scotland Norfolk, England Bohuslän, Sweden N Norway	*Tima bairdii* (Johnston, 1933)	Evans and Ashworth 1909 Hamond 1967 Dahl 1946; Hansson 1971 Vader 1972
Metopa borealis G. O. Sars, 1883 W. Scotland	*Phialidium* sp.	Elmhirst 1925
Ascidians		
Malvinometopa porcellana (K. H. Barnard, 1932) Falkland islands	'pharynx of large ascidian'	K.H.Barnard 1932 (as *Metopoides p.*)
Metopa groenlandica Hansen, 1888 Maine, USA W. Greenland	*Pyura ovifera* (Linnaeus, 1767) *Boltenia* sp., various ascidians	Blake 1929 (as *M. hirsutimana*) Stephensen and Thorson 1936 Tandberg and Vader 2009
Stenothoe eduardi Krapp-Schickel, 1976 Napoli, Mediterranean	on and in ascidians (*Microcosmos*)	Krapp-Schickel 1976
Stenothoe marina Sp. Bate, 1857 Norway	"inside ascidians"	G. O. Sars 1892, Vader 1984
Stenothoe minuta Holmes, 1905 N. Carolina W. France	*Styela plicata* (Lesueur, 1823)	Pearse 1947 Pirlot 1933 Toulmond and Truchot 1964) (as *Microstenothoe ascidiae* Pirlot)
Stenothoe valida Dana, 1853 E. Greenland	*Boltenia* sp.?	Stephensen and Thorson 1936
Torometopa paralellocheir (Stebbing, 1888) Falkland Islands	'branchial sac of simple ascidian'	Stebbing 1920 (as *Metopoides p.*)
Bivalves		
Metopa alderi (Sp. Bate, 1857) Svalbard	*Musculus discors* (Linnaeus, 1767), *M. niger* (JE Gray, 1824)	Tandberg et al. 2010

Amphipods	Hosts	Reference
Metopa glacialis (Krøyer, 1842) N. Brunswick, Canada Bear Island Svalbard Korea W. Greenland	*Musculus discors* (Linnaeus, 1767) *M. koreanus* Ockelmann, 1980 *M. laevigatus* (JE Gray, 1824)	Shoemaker 1955 Vader and Beehler 1983 Tandberg, Vader and Berge 2010 Ockelmann 1980 Just 1983
Metopa groenlandica Hansen, 1888, E. Greenland	*Pandora glacialis* Leach, 1819	Stephensen and Thorson 1936
Hermit crabs		
Metopa rubrovittata G. O. Sars, 1883 N.W. Europe	*Pagurus bernhardus* Linnaeus, 1758	many authors
Metopelloides micropalpa Shoemaker, 1930 St Laurent estuary, Canada	*Pagurus pubescens* Krøyer, 1838	Besner 1976
M. paguri Marin & Sinelnikov, 2012 Russian coast Japan Sea	*Pagurus pectinatus* (Stimpson, 1858) & *Elassochirus cavimanus* (Miers, 1879)	Marin and Sinelnikov 2012
Stenula latipes (Chevreux & Fage, 1925) W. France Ireland England	*Pagurus bernhardus* (L., 1758)	Chevreux and Fage 1925, many later authors, ?Giard 1908 (as *Metopa rubrovittata*) McGrath 1978 Lincoln 1979
Spider crabs		
Stenothoe symbiotica Shoemaker, 1956 Florida, USA Florida, USA	'large spider crab' *Stenocionops spinimana* (Rathbun, 1892)	Shoemaker 1956 Thomas and Cairns 1984

A short survey of associations between stenothoids and larger marine invertebrates (Table 1).

Table 1 lists the associations between stenothoid amphipods and other marine invertebrates known to us, with the exception of those reported from sponges, hydroids or bryozoans. These latter are excluded because in most cases it is unclear what the exact niche of the amphipods is: usually the labels say only ‚among hydroids and bryozoans‘ or ‚found together with sponges‘. Among the others the associates of various large coelenterates and also those found in ascidians generally do not seem to be obligate symbionts. Although now and then found in large numbers (e.g. *Metopa bruzelii* and *Proboloides calcarata* on gorgonians), the same species are also regularly found apparently free-living.

The situation is different for the associates of mollusks (all *Metopa* species) and those on sea anemones (mostly *Stenothoe* and *Stenula* species). Practically all these species appear to be obligate associates of only a single or in some cases a few hosts, and they have never been found free-living (for a possible exception see Blain and Gagnon 2014, who claim to have found numbers of *Stenothoe brevicornis* on the alga *Desmarestia viridis*). The amphipod associates of sea anemones always live on the column of the host or among the tentacles (Elmhirst 1925, Krapp-Schickel and Vader 2009, Krapp-Schickel et al. 2015; Vader 1983, Vader and Krapp-Schickel 1996), with the possible exception of *Stenula pugilla*, found according to the label ‚in the coelenteron of *Haliactis arctica*‘ (this paper). At least *Stenothoe brevicornis*, somewhat surprisingly, feeds to a large extent on host tissue (Moore et al. 1994), contrary to earlier assumptions (Vader 1983). Large numbers of amphipods are usually found on a single host, and ovigerous females are commonly present. Interestingly, sexual dimorphism is in most cases much

less developed in the associates of sea anemones than in related free living stenothoids (see also Vader 1983).

In contradistinction to the case with sea anemones, all the stenothoid associates of bivalve mollusks are *Metopa* species. Once more the associations seem to be obligate ones, the amphipods are rarely found free-living (and never leave their hosts in laboratory observations) and they are confined to a single host or, in the case of *Metopa glacialis*, to a series of closely related host species. A partial exception is *Metopa alderi*, usually an associate of large hydroids and hydromedusae, that recently was found in *Musculus* spp in N. Spitsbergen (Tandberg et al. 2010b). The data on mollusk-associated stenothoids have recently been reviewed by Tandberg et al. (2010a): the amphipods live inside the host and feed on that part of the ingested material that the host does not consume itself. In addition, the stenothoid symbionts of bivalves seem to exhibit territoriality as well as extended parental care: invariably only a single pair of adults is present within a single host, often together with several cohorts of juveniles.

In the case of the single, quite aberrant *Stenothoe* species that lives on a spider crab, *S. symbiotica* Shoemaker, 1956, its biology is as yet completely unknown, but also this association appears to be an obligate and probably species-specific one; the species has never been collected elsewhere and it has clearly prehensile peraeopods. Also the amphipod associates of hermit crabs and their tenanted mollusk shells are of unknown biology. *Metopelloides paguri* Marin & Sinelnikov, 2012 and *M. micropalpa* (Shoemaker, 1930) have slightly but clearly prehensile posterior peraeopods, and may therefore well be direct associates of their host hermit crabs (Vader 1983b). But the somewhat mysterious pair of *Metopa rubrovittata* Sars and *Stenula latipes* (Chevreux & Fage) do not have prehensile peraeopods (even though the posterior legs

carry maybe more spines than is usual in *Metopa* species?) and many authors have associated these species primarily with the *Hydractinia*-cover of the tenanted gastropod shells rather than with the hermit crabs themselves, although without any proof. These two species occupy the same niche, and slightly different, but possibly overlapping distributions, and have the exactly identical, quite special coloration pattern, but according to present classifications they have to be placed in different genera. Also the species associated with hermit crabs seem to be largely obligate symbionts, although possibly occurring on a larger range of hosts.

Acknowledgements

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