

Morphological and molecular systematic review of *Marphysa* Quatrefages, 1865 (Annelida: Eunicidae) species from South Africa (#48929)

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Morphological and molecular systematic review of *Marphysa* Quatrefages, 1865 (Annelida: Eunicidae) species from South Africa

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A wide polychaete fauna has been hidden under species complexes, pseudo-cryptic or cosmopolitan species, which has triggered a dismissal of the species diversity present in different regions around the world. Among the eunicids, *Marphysa sanguinea* is a typical example, recorded in three oceans and various species placed under its synonym. In South Africa, the specimens previously misidentified as *M. sanguinea* are now known as *M. elityeni*. Also, of the six *Marphysa* species recorded in the same area, three have local distribution and the other three considered of wide distribution. We evaluated the presence of two species of the latter group in South Africa and the taxonomic status of the native *M. elityeni* through morphological and molecular reviews. The widely distributed *M. macintoshi* is now locally identified as *M. durbanensis*, which  previously considered as a junior synonymy; while *M. depressa* records in South Africa belong to a new overlooked species *M. sherlockae* **n. sp.**. Finally, we confirm that *M. sanguinea* is not found in South Africa. However, the local species should be named *M. haemasoma*, a valid species and senior synonym of *M. elityeni*. This study reveals that most South African *Marphysa* species (five out of six species) are only known from this coast and reiterates the importance of implementing an integrated framework in taxonomy to unravel species complexes that cover regional biodiversity.

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Abstract

A wide polychaete fauna has been hidden under species complexes, pseudo-cryptic or cosmopolitan species, which has triggered a dismissal of the species diversity present in different regions around the world. Among the eunicids, *Marphysa sanguinea* is a typical example, recorded in three oceans and various species placed under its synonym. In South Africa, the specimens previously misidentified as *M. sanguinea* are now known as *M. elityeni*. Also, of the six *Marphysa* species recorded in the same area, three have local distribution and the other three considered of wide distribution. We evaluated the presence of two species of the latter group in South Africa and the taxonomic status of the native *M. elityeni* through morphological and molecular reviews. The widely distributed *M. macintoshi* is now locally identified as *M. durbanensis*, which previously considered as a junior synonymy; while *M. depressa* records in South Africa belong to a new overlooked species *M. sherlockae* n. sp. Finally, we confirm that *M. sanguinea* is not found in South Africa. However, the local species should be named *M. haemasoma*, a valid species and senior synonym of *M. elityeni*. This study reveals that most South African *Marphysa* species (five out of six species) are only known from this coast and reiterates the importance of implementing an integrated framework in taxonomy to unravel species complexes that cover regional biodiversity.

Introduction

Studies implementing the use of molecular and morphological tools in an integrated framework have found that a large portion of polychaete diversity has been hidden among complexes of cryptic and pseudo-cryptic species (Knowlton, 1993; Nygren 2014; Hutchings & Kupriyanova 2018). Thus, unravelling these species complexes can help to uncover patterns of distribution, regional biodiversity and areas of endemism of previously overlooked polychaete species, which could have management and conservation implications (Bickford et al. 2007; Nygren 2014). Species belonging to *Marphysa* Quatrefages, 1865 (Quatrefages 1865a,b), which serve as important bait species around the world (Izuka 1912; Lewis & Karageorgopoulos 2008; Idris et al. 2014; Liu et al. 2017; Lavesque et al. 2017; Watson et al. 2017; Cole et al. 2018; Martin et al., 2020), are ideal candidates to investigate the incidence of complexes of pseudo-cryptic species. These complexes are frequently a consequence of very brief original descriptions of species, especially of *Marphysa sanguinea* (Montagu, 1813), the type species of the genus which resulted in several species around the world with similar morphology to be synonymized; the senior species (Hutchings & Karageorgopoulos 2003; Molina-Acevedo & Carrera-Parra 2015). As a consequence, its already broad distribution range was expanded and it was reported to occur in Spain (Parapar et al. 1993), South Africa (Day 1967), Australia (Day 1967), Mexican Caribbean (Salazar-Vallejo & Carrera-Parra 1998) and Japan (Miura 1977) among others. However, the detailed redescription of *M. sanguinea* and designation of the neotype (Hutchings & Karageorgopoulos 2003) resulted in the reinstatement of at least three junior synonyms, *M. acicularum* Webster, 1884, *M. nobilis* Treadwell, 1917, and *M. viridis* Treadwell, 1917 to valid species (e.g. Molina-Acevedo & Carrera-Parra 2015; Molina-Acevedo & Idris, in review).

Furthermore, several new indigenous species with restricted distributions were described (e.g., Hutchings & Karageorgopoulos 2003; Glasby & Hutchings 2010; Zanol et al. 2016; Zanol et al. 2017; Liu et al. 2017; Martin et al. 2020), some of which had been erroneously identified as *M. sanguinea* (e.g., Hutchings & Karageorgopoulos 2003; Lewis & Karageorgopoulos 2008; Lavesque et al. 2017; Wang et al. 2018). Detailed observations of specimen demonstrated the variability in diagnostic characters for *Marphysa* species that had previously been overlooked. This may apply to other species such as *M. teretiuscula* (Schmarda, 1861) and *M. macintoshi* Crossland, 1903, which also have suspiciously wide distribution ranges (Treadwell 1906, Read & Fauchald 2018).

Six valid species belonging to *Marphysa* are currently recognized as present in South Africa, three of which are endemic to the region, i.e., *Marphysa elityeni* Lewis & Karageorgopoulos, 2008; *Marphysa capensis* (Schmarda, 1861); and *Marphysa posteriobranchia* Day, 1962 (Day, 1967; Lewis & Karageorgopoulos 2008). *Marphysa elityeni*, commonly known as “wonderworm” by local fisherman, had been previously misidentified as *M. sanguinea* (Day 1967; Lewis & Karageorgopoulos 2008; Simon et al. 2019), making it part of the global *M. sanguinea* species complex. The remaining three *Marphysa* species recorded for the region namely *M. corallina* (Kinberg, 1865), *M. depressa* (Schmarda, 1861), and *M. macintoshi* Crossland, 1903 have type localities outside of South Africa and wide distributions (Day 1967). *Marphysa depressa* has a type locality in Auckland, New Zealand (Schmarda 1861) and has since been recorded in Hong Kong (Wang et al. 2018) and South African estuaries from Saldanha Bay to Durban Bay (Day 1953, 1967). *Marphysa macintoshi* was described from Zanzibar (Crossland 1903) and has since been recorded from several localities including Australia, South Africa, Caribbean Sea, Mozambique, Red Sea, Trinidad and Tobago and China (Read & Fauchald 2018). The South African distribution of *M. macintoshi* is from Cape St. Francis to Durban Bay (Day 1967). Thus, they could also be hiding indigenous species that were incorrectly identified or synonymized, such as *M. durbanensis* Day, 1934 described from KwaZulu-Natal in South Africa and currently considered a junior synonym of *Marphysa macintoshi* (Day 1967) and *M. haemasoma* Quatrefages, 1866 described from Table Bay in South Africa and currently considered a junior synonymized with *M. sanguinea*.

In this study, we evaluated the presence of *M. depressa* and *M. macintoshi* in South Africa and the validity of *M. haemasoma* using thorough taxonomic revisions and where possible molecular comparisons. We also provide detailed descriptions for *M. haemasoma*, *M. durbanensis* and a species new to science from South Africa, *M. sherlockae* n. sp.

Materials & Methods

Examined material

Fresh specimens of *Marphysa depressa* were collected from rock crevices in the fringing intertidal zones from Strand (n = 4) (Fig. 1). Fresh specimens of *M. elityeni* were collected from the fringing intertidal zone at low tide from burrows in gravely-sand type sediment under

boulders in Kommetjie (n = 5) (Fig. 1). Full collection data for both species can be found in the respective species accounts in the results section. Live specimens were brought back to the laboratory where they were anaesthetized with 7% MgCl₂, diluted in distilled water and photographed. Whole specimens from  and were fixed in 96% ethanol. Posterior ends of specimens from Kommetjie were fixed in 96% ethanol, whilst the anterior ends were fixed in a 4% seawater-formalin solution.

Type and non-type material of *M. depressa*, *M. macintoshi*, *M. durbanensis*, *M. haemasoma* and *M. elityeni* deposited at the Natural History Museum (BMNH), Museum National d'Histoire Naturelle, Paris (MNHN) and the Natural History Museum, Vienna, Austria (NHM) and the Iziko South African museum (SAM) were examined.

Morphological examination

Species descriptions were produced based on the type material, but a variation section with all specimens reviewed was also included.

The general structures such as the prostomium, peristomium, anterior region of the body, maxillary apparatus, branchiae, parapodia, chaetae and pygidium were included in the descriptions. A dorsal incision was made in the specimen to extract and describe the maxillary apparatus, after which it was returned to its original position. The maxillary formula (MF) and measures were taken according to Molina-Acevedo & Carrera-Parra (2015, 2017). Six parapodia (three from the anterior region, two from the median, and one from the posterior region) were dissected to describe in detail the morphology of the cirri and lobes, and simple and compound chaetae.

The start of the branchiae and subacicular hooks were indicated depending on the side where they began ('L' for Left, 'R' for Right) with the chaetiger number. In the region with the maximum number of branchial filaments, the long filaments are ≥ 4 times as long as dorsal cirri, whereas the short filaments are < 4 times as long as dorsal cirri. Terminology used for the descriptions of the pectinate chaetae are according to the classification proposed by Molina-Acevedo & Carrera-Parra (2015, 2017) and Zanol et al. (2016). Herein, thin and thick refers to the thickness of the pectinate shaft; wide and narrow refers to the width of the pectinate blade; and anodont and isodont refer to the relative length of external teeth in relation to each other and internal teeth, e.g. thin, wide isodont with long and slender teeth.

The length through chaetiger 10 (L10) and the width of chaetiger 10 excluding parapodia (W10) were measured in the specimens as standard measures when the specimens were collected incomplete. Likewise, the total length (TL) and variations of the total number of chaetigers (TChae) were recorded. All descriptions were illustrated with a series of photos taken with Canon EOS T6i. These were then stacked using Helicon Focus® 6 (Method A) software to improve the depth of field, and the final edition was performed in Adobe Photoshop® 2020. In order to understand patterns of intraspecific variation, linear regression analyses were conducted to evaluate the possible relationships between size (length of specimens using L10

measurement) and morphological features such as the chaetigers where branchiae or the subacicular hooks begin, the number of branchial filaments. The degree of predictability of the variation of the morphological features following size variation is given by R^2 (e.g., $R^2 = 0.63$, $p = 0.05$, $n = 34$, Fig. 4, 7).

Nomenclature

The electronic version of this article in Portable Document Format (PDF) will represent a published work according to the International Commission on Zoological Nomenclature (ICZN), and hence the new name contained in the electronic version is effectively published under that Code from the electronic edition alone. This published work and the nomenclatural act it contains has been registered in ZooBank, the online registration system for the ICZN. The ZooBank LSIDs (Life Science Identifiers) can be resolved and the associated information viewed through any standard web browser by appending the LSID to the prefix “<http://zoobank.org/>.” The LSID for this publication is: F420E96F-9B29-4288-B21B-8475F3DE4DDA. The online version of this work is archived and available from the following digital repositories: PeerJ, PubMed Central and CLOCKSS.

Molecular methods

DNA extraction, amplification and sequencing

DNA was extracted from tissue samples using the ZR Genomic DNA Tissue MiniPrep Kit according to standard manufacturer's protocol. The universal primer pair LCO1490 and HCO2198 (Folmer et al. 1994) was used to amplify a fragment of the mitochondrial gene cytochrome oxidase I (COI). PCR amplifications were carried out using 12.5 μ l of OneTaq Quick-Load Master Mix (New England BioLabs), 9.5 μ l of molecular biology grade water, 0.50 μ l of forward and reverse primer (10 μ M), 1 μ l of 1% bovine serum albumin (BSA) and 1 μ l of template DNA to make up a total reaction volume of 25 μ l. Thermal cycling conditions were as follows for *M. elityeni* and *M. sherlockae* n. sp.: initial denaturation at 95 °C for 3 minutes, followed by 35 cycles of 94 °C for 20 seconds, 45 °C for 30 seconds and 72 °C for 1 minute, followed by a final extension time at 72 °C for 5 minutes. Amplicons were Sanger sequenced at the Central Analytical Facility at Stellenbosch University using just the forward primer (LCO1490). Quality control was performed on sequences to check for any sequencing errors using BioEdit (v7.2.6) (Hall 1999).

Phylogenetic and species delimitation methods

The COI sequences were edited, trimmed and aligned with ClustalW (Thompson et al. 1994) using multiple alignment method in BioEdit (v7.2.6). Several species belonging to the *Marphysa*

genus were included in the analysis for comparison together with seven other species from different genera within the Eunicidae and one species from Onuphidae as outgroups to root the tree (see Table 1). DnaSP v5 (Librado & Rozas 2009) was used to generate a nexus file for subsequent analysis. PAUP (Swofford 2003) and MrModelTest v2.3 (Nylander 2004) were used to calculate the best fit model of evolution for the data set using Akaike Information Criterion (AIC). Bayesian inference (BI) was used to reconstruct phylogenetic relationships using the best fit model SYM+G in MrBayes 3.1.2 (Ronquist et al. 2012). The trees were calculated using 4 Markov Chains of 5 million generations sampled simultaneously with every 1000th tree sampled. A 50% majority rule consensus tree with posterior probability support was constructed by discarding the first 25% of trees as burn-in. Tracer v1.5 (Rambaut & Drummond 2009) was used to investigate the convergence of runs by analysing the average standard deviation of split frequencies (≤ 0.01) and the mixing quality of all parameters was verified by analysing the plot of likelihood versus the sampled trees and the effective sample sizes (ESS > 200), of which both criteria were satisfied. FigTree v1.4.4 (Rambaut 2013) was used to visualize trees.

A new formatted phylogenetic tree generated using FigTree v1.4.4 from the previous analysis was used as input for the Bayesian implementation of the Poisson tree process (bPTP) (Zhang et al. 2013) model for species delimitation using the online webserver <https://species.h-its.org/>. The tree was rooted and run for 500,000 MCMC generations, with thinning set to 100 and burn-in and seed set to 1 and 123, respectively. Convergence of MCMC chains was visually checked on the maximum likelihood plot generated by the online server. MEGA X (Kumar et al. 2018) was used to calculate the interspecific genetic distances between species using the Kimura 2-parameter (K2P) model with complete deletion of gaps and run for 500 bootstrap replicates.

Results

We could not confirm records of *M. macintoshi* and *M. depressa* in South Africa. Indigenous specimens of *M. durbanensis* have been identified as *M. macintoshi* after both species were synonymised. However, these species differ in the shape of the prostomium, of anterior post-chaetal lobes, of pectinate chaetae and in the shape and distribution of branchiae. Thus, we here consider *M. durbanensis* as a valid species.

South African specimens previously identified as *M. depressa* represent a species new to science, *M. sherlockae* n. sp. They differ from *M. depressa* type specimens in the shape and distribution of compound chaetae, shape of post-chaetal lobes, maximum number of branchial filaments. Sequences of *M. depressa* were not available from its type locality and therefore could not be compared with *M. sherlockae* n. sp. Nonetheless, *M. sherlockae* n. sp. forms an independent clade with high posterior probability (Fig. 8) and genetically differs from other *Marphysa* species by 18–25%, confirming that it is a separate species to all the other *Marphysa* spp. included in the phylogenetic analysis. Additionally, results from the bPTP analysis also supported *M. sherlockae* sp. nov. as a single independent species (BS>0.95) (S1, supplementary information). *M.*

sherlockae n. sp. is phylogenetically closest to *Marphysa californica* Moore, 1909 and *Marphysa brevitentaculata* but the clade is poorly supported. Nonetheless, all three species genetically differ from each other by 18–20%. *Marphysa haemasoma* is a valid species. The examination of type materials allowed us to confirm that *M. haemasoma* differs from *M. sanguinea* in the shape of postchaetal lobe in anterior chaetigers and of subacicular hooks, in the maximum number of branchial filaments and in the distribution of the swollen base of ventral cirri. Furthermore, types of *M. elityeni* only differ from those of *M. haemasoma* in size related features, such as the length of prostomial appendices, and branchiae and ventral cirri with swollen base start. For these reasons, and in view of the principle of priority (ICZN 1999, Arts. 23), we consider *Marphysa haemasoma* a senior synonym of *M. elityeni*. In molecular analyses, *M. haemasoma* forms a well-supported clade that is independent of the *M. sanguinea* clade (Fig. 8). The species differ from each other by 20%, with results from the bPTP analysis (S1 supplementary information) confirming their separation as independent species (BS>0.95). Thus, these species are not synonymous.

Systematics

Order EUNICIDA Dales, 1962

Family EUNICIDAE Berthold, 1827

Genus *Marphysa* Quatrefages, 1865

Marphysa durbanensis Day, 1934

Figure 2

Marphysa durbanensis Day, 1934:51–53, text-fig. 10.

Marphysa macintoshi Day 1967:378 (*non* Crossland, 1903); Day 1974:59; Branch et al. 2016:68–69, Pl. 26, Fig. 26.6.

Material examined. *Type material:* Lectotype designate here BNHM 1934.1.19.166, Durban, South Africa, 1933, coll. JH. Day. One paralectotype BNHM 1934.1.19.166 designated here, same information as lectotype.

Comparative material. *Marphysa macintoshi*, syntypes, three specimens, BNHM 1924.3.1.22–3, slide BNHM.1924.3.1.22A, Zanzibar, Africa, 1901–1902, by digging in sand between intertidal on both east and west coasts of Zanzibar (syntype 1 incomplete specimen with 262 chaetigers, L10: 8.1 mm, W10: 2.7 mm; syntype 2 incomplete specimen with 106 chaetigers, L10: 5.3 mm, W10: 3 mm; syntype 3 incomplete specimen with 160 chaetigers, L10: 7.8 mm, W10: 3 mm).

Description. Lectotype complete, ventrally dissected from peristomium until chaetiger 9, with 380 chaetigers, L10= 14 mm, W10= 3.6 mm, TL= 305 mm. Last 48 chaetigers regenerating.

Anterior region of body with convex dorsum and flat ventrum; body depressed from chaetiger 7, widest at chaetiger 24, tapering after chaetiger 37.

Prostomium bilobed, 1.7 mm long, 2.5 mm wide; lobes anteriorly rounded; median sulcus shallow dorsally (Fig. 2A), deep ventrally (Fig. 2B). Prostomial appendages in a semicircle, median antenna isolated by a gap. Palps reaching middle of first peristomial ring; lateral antennae reaching middle of second peristomial ring; median antenna broken, in paralectotype reaching middle of first chaetiger. Palpophores and ceratophores ring-shaped, short, thick; palpostyles and ceratostyles tapering, slender. Eyes not observed.

Peristomium (2.7 mm long, 3 mm wide) longer and wider than prostomium, first ring two and a half times longer than second ring; separation between rings distinct on all sides (Fig. 2A–C). Ventral anterior edge of peristomium longer than dorsal, remaining feature ventral distorted by the dissection (Fig. 2B–C).

Maxillary apparatus with MF= 1+1, 5+6, 6+0, 4+8, 1+1 (Fig. 2D). MI 3.1 times longer than maxillary carriers. MI forceps-like, MI 4.6 times longer than closing system (Fig. 2D–E); ligament between MI and MII sclerotized. MII wider than rest of maxillae, with triangular teeth; MII 3.6 times longer than cavity opening oval (Fig. 2D–E); ligament present between MII–MIII and right MII–MIV slightly sclerotized (Fig. 2E). MIII with triangular teeth; with rectangular attachment lamella, situated in the centre of ventral edge of maxilla, slightly sclerotized (Fig. 2D–E). Left MIV with two teeth bigger; attachment lamella semicircle, slender, better developed in central portion, situated 1/2 along anterior edge of maxilla. Right MIV with teeth of equal size; attachment lamella semicircle, slender, better developed in central portion, situated 2/3 along anterior edge of maxilla, sclerotized (Fig. 2D–E). MV square, with a short triangular tooth.

Mandibles dark; missing calcareous cutting plates; sclerotized cutting plates brown, with 20 growth rings (Fig. 2F).

Branchiae pectinate with up to 11 long filaments at around 64–80% of the body, present from chaetigers 28L–29R to 370 (Fig. 2J–K). First pair and last 10 with one filament; reach the maximum 10 or 11 filaments in chaetigers 241L–307L (Fig. 7). Branchial filaments longer than dorsal cirri except in first five and last seven branchiae.

First two parapodia smallest; best developed in chaetigers 6–26, following ones becoming gradually smaller. Notopodial cirri conical in anterior-median chaetigers, digitiform in posterior ones; longer than ventral cirri in anterior chaetigers, of similar length in posterior ones; best developed in chaetigers 3–30, following ones gradually smaller (Fig. 2G–K). Prechaetal lobes short, as transverse fold in all chaetigers (Fig. 2G–K). Chaetal lobes rounded in all chaetigers, shorter than postchaetal lobes in anterior region, longer than the other lobes in median-posterior region; with aciculae emerging dorsal to midline (Fig. 2G–K). Postchaetal lobes well developed in first 40 chaetigers; digitiform in first five chaetigers, rounded from chaetiger 6; progressively smaller from chaetiger 22; from chaetiger 41 inconspicuous (Fig. 2G–K). Ventral cirri bluntly conical in first five chaetigers; in chaetigers 6 to 355 with a short oval base and digitiform tip; conical from chaetiger 356, gradually reducing in size (Fig. 2G–K).

Aciculae blunt, reddish along most of  length, amber on the distal tip (Fig. 2G–K). First eight chaetigers with three aciculae; in chaetigers 9–18 with four aciculae; in chaetigers 19–44 with three or four aciculae; in chaetigers 45–124 with two aciculae; from chaetiger 125 with only one acicula.

Limbate chaetae of two lengths in same chaetiger, dorsalmost longer; reduced in number around chaetiger 30. Five types of pectinate chaetae, anterior chaetigers: thin, narrow isodont with long and slender teeth, 3–4 pectinate, with up to 14–15 teeth (Fig. 2L); median and posterior chaetigers: thin, wide isodont with short and slender teeth, 4–5 pectinate, with up to 23–24 teeth (Fig. 2M); thick, wide isodont with short and thick teeth, 1–2 pectinate, with up to 19 teeth (Fig. 2N); and thick wide anodont with short and slender teeth, 1–2 pectinate, with 19 teeth (Fig. 2O); posterior chaetigers: thick, wide anodont with long and thick teeth, 1–2 pectinate, with up to 17 teeth. Compound spinigers present in all chaetigers, in anterior-median chaetigers with blades of two lengths, shorter ones more abundant (Fig. 2P). Subacicular hooks unidentate, amber, present from chaetiger 46, one or two per chaetiger, with continuous distribution (Fig. 2Q).

Pygidium with dorsal pair of anal cirri as long as last eight chaetigers; ventral pair short, as long as last two chaetigers.

Variations. Material examined L10= 12–14 mm, W10= 3.6–4 mm, TChae= 322–380. Palps reaching middle of first or second peristomial ring; lateral antennae reaching middle of second peristomial ring or first chaetiger; median antenna reaching first chaetiger. The maxillary variations are MII 5–6+6–8, MIII 6, MIV 3–4+6–8. The proportion of maxillary apparatus varies as follows: MI are 3.1–3.2 times longer than maxillary carriers; MI are 4.6–5.3 times longer than closing system; MII are 3.5–3.6 times longer than length of cavity opening. Branchiae from chaetigers 28–32 to 10–13 chaetigers before pygidium. Maximum number of branchial filaments varied from 11 to 12. Postchaetal lobe well developed in the first 40 chaetigers. Ventral cirri with a swollen base from chaetigers 4–5 to 25 chaetigers before pygidium. Start of subacicular hooks in chaetigers 46–47.

Habitat. Day (1934) does not provide information about the specific substrate, although he did clarify that the collection was between the tidemarks in Durban Bay and Umkomaas.

Distribution. Day (1934) recorded this species from Durban Bay and Umkomaas in KwaZulu-Natal, South Africa.

Remarks. The original description of *Marphysa durbanensis* provides variation of the two specimens collected that matches with the specimens deposited in the BNHM. Day (1934) described almost colourless eyes, but they were not observed in this study. It is possible that the colour has totally faded due to the long-term preservation of the specimens. The best-preserved specimen is herein selected as lectotype in order to fix the species definition (ICZN 1999, Arts. 74.1, 74.7.3), whereas the other is considered as a paralectotype (ICZN 1999, Art. 74F).

Day (1934) considered *M. durbanensis* clearly different from morphologically similar species such as *M. simplex* Crossland, 1903 (= *M. teretiuscula*) and *M. acicularum* when he described the species. However, in his monograph of the polychaetes from South Africa, the same author considered *M. durbanensis* as a junior synonym of *M. macintoshi* without making any reference to this nomenclatural action (Day 1967, page. 378). Herein, clear differences were found between the species. *M. durbanensis* (L10: 14 mm) has a bilobed prostomium, the branchiae are pectinate and starts from chaetigers 28–32, the postchaetal lobe is digiform in first four chaetigers, and there are five types of pectinate chaetae; while in *M. macintoshi* (L10: 4.5 mm) the prostomium is unilobed with a shallow median sulcus at the anterior edge, the branchiae are palmate with a short button-shaped branchial stem and start from chaetiger 32–47, the postchaetal lobe is conical in the first four chaetigers, and there are only three types of pectinate chaetae. Due to these morphological differences, *M. durbanensis* is considered a valid species. *Marphysa durbanensis* resembles *M. haemasoma* (see below) by the presence of compound spinigers distributed in all chaetigers; however, *M. durbanensis* has more teeth in MII (5–6+6–8), digitiform postchaetal lobes in first four chaetigers, five types of pectinate chaetae, and the subacicular hook with a continuous distribution even in bigger specimens. However, *M. haemasoma* has 3 teeth in MII (4+4), the postchaetal lobe is ovoid in first four chaetigers, there are only four types of pectinate chaetae, and the subacicular hook has a discontinuous distribution in short specimens. *Marphysa durbanensis* resembles *M. bulla* Liu, Hutchings & Kupriyanova, 2018, *M. hongkongensa* Wang, Zhang & Qiu, 2018, *M. leidii* Quatrefages, 1866, *M. parishii* Baird, 1869 and *M. teretiuscula* by the presence of five types of pectinate chaetae; however, *M. durbanensis* has a digitiform postchaetal lobe in the first four chaetigers and the subacicular hook is amber, while *M. teretiuscula* has an ovoid postchaetal lobe in the first four chaetigers and the subacicular hook is reddish basally and translucent in the distal region. In addition, *M. leidii* has a conical postchaetal lobe in the first chaetigers. Otherwise, *M. durbanensis* has the long branchial filaments and the branchiae are pectinate; while for *M. hongkongensa* the branchial filaments are short and the branchiae are pectinate and palmate with a short button-shaped branchial stem in some regions of the body. On the other hand, in *M. durbanensis* (L10: 14 mm) the eyes are present, and the branchiae start in chaetigers 28–32; while *M. b.* (L10: 6.3–7.9 mm) lacks eyes, and the branchiae start in chaetiger 36. Finally, *M. durbanensis* has up to 11–12 branchial filaments whilst *M. leidii* (L10: 10.7–17 mm) and *M. parishii* (L10: 17.2 mm) only have 4 to 6 filaments.

***Marphysa haemasoma* Quatrefages, 1866**

Figure 3, 7–8

Marphysa haemasoma Quatrefages, 1866:334–335; Grube 1870:299.

Marphysa sanguinea von Marenzeller, 1888:11, Fauvel 1902:61; Day 1967:378 (*non* Montagu, 1813); Day 1974:59.

Marphysa sanguinea haemasoma Willey, 1904:263, Pl.13, Fig.15

Marphysa elityeni Lewis & Karageorgopoulos, 2008:279–281, Figs. 1–2, Table 1, 2,  Branch et al. 2016:68–69, Pl. 26, Fig. 26.5.

Material examined. *Type material:* Holotype *Marphysa haemasoma* MNHN type 613, Cape of Good Hope, South Africa. *Additional material:* Five incomplete specimens SAM-A090272, SAM-A090273, SAM-A090274, SAM-A090275, SAM-A090317, Kommetjie, South Africa from sand burrows under boulders at fringing intertidal zone, coll. A.N. du Toit, 10 Mar 2017, 18°19'40.7"E 34°09'33.0"S.

Comparative material:  Holotype *Marphysa elityeni* SAM-A21478, Cape of Good Hope, South Africa. Eight paratypes of *Marphysa elityeni* BNHM 2007.69, SAM-A21479, SAM-A21480, SAM-A21481, Buffels Bay in the Cape of Good Hope, South Africa, 15 Sep 2004, 18°29'27" E 34°21'6" S. Neotype *Marphysa sanguinea* BNHM 1867.1.7.24, Polperro, Cornwall, in mud and gravel at low water mark, coll. Laughrin, Redet. P. Hutchings (2 specimens from this lot), Desig. P. Hutchings (Neotype complete specimen with 286 chaetigers, L10: 16.7 mm, W10: 10 mm; topotype complete specimen with 239 chaetigers, L10: 20.4 mm, W10: 7.2 mm).

Description. Holotype complete, gravid female, with 322 chaetigers, L10= 12.3 mm, W10= 7 mm TL= 309 mm. Anterior region of the body with convex dorsum and flat ventrum; body depressed from chaetiger 5, widest at chaetiger 25, tapering after chaetiger 41. Prostomium bilobed, 2.8 mm long, 4 mm wide; lobes anteriorly rounded; median sulcus dorsally shallow (Fig. 3A), ventrally deep (Fig. 3B). Prostomial appendages in a semicircle, median antenna isolated by a gap. Palps reaching first chaetiger; lateral and median antennae reaching second chaetiger. Palpophores and ceratophores ring-shaped, short, thick; palpostyles and ceratostyles tapering, slender. Eyes colourless, as a scar between palps and lateral antennae. Peristomium (2.8 mm long, 6.3 mm wide) wider than prostomium; first ring three times longer than the  cond ring, separation between rings distinct only dorsally and ventrally (Fig. 3A–C). Ventral region of the first ring with a slight central depression in anterior edge (Fig. 3B). Maxillary apparatus with MF= 1+1, 4+4, 5+0, 3+7, 1+1 (Fig. 3D). MI 3 times longer than maxillary carriers. MI forceps-like, MI 4 times longer than closing system (Fig. 3D–E); ligament between MI and MII, sclerotized. MII with triangular teeth, right anterior teeth broken; MII 3.6 times longer than cavity opening (Fig. 3D–E); ligament present between MII–MIII and right MII–MIV slightly sclerotized (Fig. 3E). MIII with triangular teeth; with rectangular attachment lamella, situated only in the centre of right edge of maxilla, slightly sclerotized (Fig. 3D–E). Left MIV with all teeth of similar size; attachment lamella semicircle, wide, better developed in right portion, situated 2/3 of anterior edge of maxilla. Right MIV with lateral larger teeth; attachment lamella semicircle, wide, better developed in central portion, situated 2/3 of anterior edge of maxilla, sclerotized (Fig. 3D–E). MV square, with a short triangular tooth. Mandibles dark; with calcareous cutting plates present and sclerotized cutting plates brown, with nine growth rings (Fig. 3F).

Branchiae pectinate with up to six long filaments for around 20–54% of the body, present from chaetigers 26L–27R to 308L–311R (Fig. 3I–J). First two and last 13 pairs with one filament; with six filaments in chaetigers 79L to 173L (Fig. 7). Branchial filaments longer than dorsal cirri except in first two and last branchiae.

First two parapodia smallest; best developed in chaetigers 7–40, following ones gradually becoming smaller. Notopodial cirri conical in all chaetigers; of similar length as ventral cirri in anterior and posterior chaetigers, shorter than ventral cirri in median chaetigers; best developed in chaetigers 4–37, following ones gradually smaller (Fig. 3G–K). Prechaetal lobes short, as transverse folds in all chaetigers (Fig. 3G–K). Chaetal lobes in first 37 chaetigers rounded, shorter than postchaetal lobe in anterior region, with aciculae emerging dorsal to midline; from chaetiger 38 triangular, longer than other lobes in median-posterior chaetigers (Fig. 3G–K). Postchaetal lobes well developed in first 60 chaetigers; ovoid in first six chaetigers, rounded in chaetigers 7–9, auricular from chaetiger 10, progressively smaller from chaetiger 35; from chaetiger 61 inconspicuous (Fig. 3G–K). Ventral cirri digitiform in first three chaetigers; in chaetiger four to last chaetiger with a short oval base and digitiform tip (Fig. 3G–K).

Aciculae blunt, reddish along most of its length, amber on the distal tip (Fig. 3G–K). First 10 chaetigers with three aciculae; in chaetigers 11–77 with three or four; in chaetigers 78–161 with three; in chaetigers 162–322 with two or three.

Limbrate chaetae of two lengths in same chaetiger, dorsalmost longer, reduced in number around chaetiger 24. Four types of pectinate chaetae; in anterior chaetigers: thin, narrow isodont with long and slender teeth, with 2–3 pectinate, with up to 17 teeth (Fig. 3L); median-posterior chaetigers: thick, wide isodont with short and slender teeth, with 6–7 pectinate, with up to 17 teeth (Fig. 3M); posterior chaetigers: thick, wide anodont with short and slender teeth, with 6–7 pectinate, with up to 13–14 teeth (Fig. 3N), and thick, wide anodont with long and thick teeth, with 1–2 pectinate, with up to 10 teeth (Fig. 3O). Compound spinigers present in all chaetigers, with blades of two sizes in the same chaetiger (Fig. 3P), shorter slightly more abundant than longer blade. Subacicular hooks absent; in paratype of *M. elityeni* (L10= 9.3 mm) subacicular hook bidentate, translucent, present only in regenerating chaetigers, one per chaetiger; with triangular teeth, distal tooth smaller than proximal, directed upward; proximal tooth triangular, directed laterally (Fig. 3Q).

Pygidium with dorsal pair of anal cirri broken; ventral pair as long as last chaetiger.

Variations. Material examined L10= 9.3–20.1 mm, W10= 6.2–14.5 mm, TChae= 194–486.

Palps reaching second peristomial ring or first chaetiger; lateral antennae reaching first or second chaetiger; median antenna reaching first or middle of second chaetiger. The maxillary variations are MII 4+4, MIII 3–5, MIV 3–4+6–7. The proportion of maxillary apparatus varies as follows: MI are 2.6–3 times longer than maxillary carriers; MI are 4.1–4.6 times longer than closing system; MII are 4–4.3 times longer than cavity opening. Branchiae from chaetigers 26–37 to 10 chaetigers before pygidium. Maximum number of branchial filaments varied from six to 10.

Postchaetal lobe well developed in first 57–60 chaetigers. Ventral cirri with a swollen base from chaetigers 3–6 to last chaetigers.

DNA barcode. Kommetjie, Western Cape, South Africa (MB-A090272) (GenBank accession number: MN067877) (Simon et al. *unpublished data*); 577 bp fragment isolated with universal mitochondrial cytochrome oxidase subunit 1 gene, primer pair: LCO1490, HCO2198 (Folmer et al. 1994).

Habitat. Very common in the boulder fields at the lower intertidal zones of sheltered bays, and in rock pools. Worms can be found under rocks in sand burrows up to 1 m deep.

Distribution. Table Bay to Buffels Bay, Cape Point, Western Cape South Africa (Quatrefages 1866; Lewis & Karageorgopoulos 2008). Branch et al. (2016) recorded this species to occur from Namibia in southwest Africa to East London in South Africa. Simon et al. *unpublished*. recorded this species from Melkbosstrand to Knysna in the Western Cape and therefore falls within the currently accepted distribution range of this species according to Branch et al. (2016). However, the records from Namibia have not been verified and may also represent an indigenous overlooked species of that region and therefore should be revised.

Remarks. Specimens of *M. haemasoma* were previously redescribed by Grube (1870) and then identified as *M. sanguinea* after von Marenzeller (1888) synonymized *M. haemasoma* with *M. sanguinea* due to similarities in morphology and habitat observed in the specimens from the Cape of Good Hope. Later, Lewis & Karageorgopoulos (2008) realized that specimens from this region had been misidentified as *M. sanguinea*. This led to the description of *Marphysa elityeni* Lewis & Karageorgopoulos, 2008 in a study that seem to have overlooked *M. haemasoma*. *M. haemasoma* (L10: 9.3–18.5 mm) is considered a different species from *M. sanguinea* (L10: 11.5–20.4) because the former has up to 10 branchial filaments, and ovoid postchaetal lobe in anterior chaetigers; whereas the latter has 9–18 branchial filaments, and digitiform postchaetal lobes in anterior chaetigers. Moreover, in *M. haemasoma* the swollen base of the ventral cirri continues to the last chaetigers, and the subacicular hook is translucent; while in *M. sanguinea* the swollen base of the ventral cirri ends between 8–18 chaetigers before the pygidium, and the subacicular hook is reddish basally and translucent distally. *Marphysa haemasoma* resembles *M. aegypti* Elgetany, El-Ghobashy, Ghoneim & Struck, 2018, *M. fauchaldi* Glasby & Hutchings, 2010, *M. gravelyi* Southern, 1921, *M. nobilis*, *M. teretiuscula* and *M. tripectinata* Liu, Hutchings & Sun, 2017 by the presence of the ovoid postchaetal lobes; however, *M. haemasoma* has subacicular hooks that are completely translucent, while *M. nobilis*, *M. teretiuscula* and *M. tripectinata* have subacicular hooks that are reddish at the base and translucent in the distal region. Furthermore, *M. haemasoma* has four types of pectinate chaetae, while *M. fauchaldi* and *M. gravelyi* have only three types of pectinate chaetae. Additionally, when present in *M. haemasoma*, subacicular hooks (in regenerating chaetigers) are bidentate,

while *M. aegypti* bears unidentate subacicular hooks (Martin et al., 2020). Moreover, *M. haemasoma* has 3 teeth in MII and MIII (4+4, 4–5+0), while *M. gravelyi* have more teeth in the same plates (MI 8+7, MII 8+0). Finally, *M. haemasoma* has long branchial filaments, while in *M. fauchaldi* the branchial filaments are short. Type material of *M. elityeni* was collected from Buffels Bay, from the Cape Peninsula (Lewis & Karageorgopoulos 2008) which is ~58.4 km away from Table Bay where type material was collected for *M. haemasoma* (Fig. 1). Additionally, Kommetjie, where the fresh material examined in this study were collected, is near both Buffels Bay (~29.4km) and Table Bay (~43km). Thus, all these collections fall within the type region of the original collected material from Table Bay (Fig. 1).

Marphysa sherlockae n. sp.

Figure 4–8

Marphysa depressa Day 1953:434, text-figs. 5 n, p; 1967:395–396, Figs. 17.5 n–t (*non* Schmarda, 1861); Day 1974:59; Branch et al. 2016:68–69, Pl. 26, Fig. 26.8.

Material examined. *Type material* Holotype BNHM 1963.1.84, Langebaan Lagoon, South Africa, coll. J.H. Day. Paratype, one specimen BNHM 1952.5.10.7. Paratype, two specimens SAMC-A089089 and SAMC-A089090), Strand, False Bay, South Africa, coll. J. Kara, 20 March 2019, det. J. Kara. *Additional material*: two specimens BNHM 0000, same data as holotype. One incomplete specimen SAMC-A20578, Langebaan lagoon, South Africa, coll. UCT ecological survey, 24 April 1949, det. J.H. Day. one complete specimen SAMC-A60425, Langebaan Lagoon, South Africa, coll. UCT ecological survey, 24 April 1949, det. D. Clarke. Two complete specimens, SAMC- A089091 and SAMC- A089092), Strand, False Bay, South Africa, coll. J. Kara, 20 March 2019, det. J. Kara.

Comparative material. Syntypes, two specimens, *Marphysa depressa* NHM 0000, New Zealand, Port of Auckland, coll. Schmarda (syntype 1 complete specimen with 328 chaetigers, L10: 9.5, W10: 4 mm; syntype 2 complete specimen with 132 chaetigers, L10: 9.5 mm, W10: 4.8 mm).

Description. Holotype complete, with 208 chaetigers, L10= 6.6 mm, W10= 1.7 mm, TL= 67 mm. Anterior region of body with convex dorsum and flat ventrum, body depressed from chaetiger 6, widest at chaetiger 38, tapering after chaetiger 112. Prostomium bilobed, 1 mm long, 1.1 mm wide; lobes frontally oval; with median sulcus dorsally shallow (Fig. 4A), ventrally sulcus deep (Fig. 4B). Prostomial appendages in a semicircle, median antenna isolated by a gap. Palps reaching first chaetiger; lateral antennae reaching second chaetiger; median antenna reaching middle of second chaetiger. Palpophores and ceratophores ring-shaped, short, thick; palpostyles and ceratostyles tapering, slender. Eyes as a brown line, between palps and lateral antennae.

560 Peristomium (1.1 mm long, 3.2 mm wide) wider than prostomium, first ring  times longer
561 than the second ring, separation between rings distinct on all sides (Fig. 4A–C). Ventral region of
562 the first ring with a slight central depression in anterior edge (Fig. 4B).
563 Maxillary apparatus with MF= 1+1, 3+5, 5+0, 4+8, 1+1 (Fig. 4D). MI 2.3 times longer than
564 maxillary carriers. MI forceps-like, MI 4.3 times longer than the closing system; ligament
565 between MI and MII, slightly sclerotized (Fig. 4D–E). MII with recurved teeth; MII 
566 longer than cavity opening oval (Fig. 4D–E); ligament present between MII and MIII and right
567 MIV slightly sclerotized (Fig. 4E). MIII with blunt teeth; with rectangular attachment lamella,
568 situated in the anterior of right edge of maxilla, slightly sclerotized (Fig. 4D–E). Left MIV with
569 le ost tooth larger; attachment lamella semicircle, wide, better developed in right portion,
570 situated along anterior edge of maxilla (Fig. 4D–E). Right MIV with rig ost tooth larger;
571 attachment lamella semicircle, wide, better developed in central portion, situated along anterior
572 edge of maxilla (Fig. 4D–E). MV square, with a short-rounded tooth. Mandibles dark; missing
573 calcareous cutting plates, sclerotized cutting plates brown, with 10 growth rings (Fig. 4F).
574 Branchiae palmate with a short button-shaped branchial stem, with up to two long filaments,
575 present from chaetigers 28R–37L to 195L–196R (Fig. 4J–K). One filament in chaetigers 28L and
576 31L–45L; without filament in chaetigers 29L–30L; two filaments in chaetigers 46L–170L; one
577 filament in chaetigers 171L–196L (Fig. 7). Branchial filaments longer than dorsal cirri.
578 First two parapodia smallest; best developed in chaetigers 6–42, following ones becoming
579 gradually smaller. Notopodial cirri conical in all chaetigers; longer than ventral cirri in anterior
580 chaetigers, shorter in median chaetigers, of similar size in posterior ones; best developed in
581 chaetigers 3–41, following ones gradually decreasing in size (Fig. 4G–K). Prechaetal lobes short.
582 Chaetal lobes in first 29 chaetigers rounded, shorter than postchaetal lobe, with aciculae
583 emerging dorsal to midline; from chaetiger 30 triangular, longer than other lobes (Fig. 4G–K).
584 Postchaetal lobes slightly developed in first 24 chaetigers; triangular first 5 chaetigers, auricular
585 following ones, progressively smaller from chaetiger eight; from chaetiger 25 inconspicuous
586 (Fig. 4G–K). Ventral cirri conical in first six chaetigers; from chaetigers 7 to 138 with a short
587 oval base and digitiform tip; conical from chaetiger 139, gradually smaller (Fig. 4G–K).
588 Aciculae blunt, reddish from base to most of its length, translucent on the distal tip (Fig. 4G–K).
589 First five chaetigers with 2 aciculae; in chaetiger 6–10 with three aciculae; in chaetigers 11–73
590 with two aciculae; from chaetiger 74 with only one acicula.
591 Limbate chaetae of two lengths in same chaetiger, dorsalmost longer, reduced in number around
592 chaetiger 13. Two types of pectinate chaetae; in anterior chaetigers: thin, narrow isodont with
593 long and slender teeth, 1–2 per parapodium and up to 10–11 teeth (F  4L, 5A–C); in median-
594 posterior chaetigers, thick, wide isodont with long and thick teeth, 4–5 per parapodium and up to
595 14 teeth (F  M, 5D–E); anodont pectinate not observed. Compound spiniger chaetae present in
596 all chaetigers, with blades of similar size in the same chaetiger (Fig. 4N), longer blades in
597 median-posterior chaetigers. Compound falciger chaetae in anterior-median chaetigers, more
598 abundant than compound spiniger in first 26 chaetigers; in anterior region blades of similar
599 length (56 µm, Fig. 4O), with triangular teeth, both of similar size, proximal tooth directed

laterally, distal directed upward; in median chaetigers with blades shorter (38.5 μm) with teeth of similar shape. Subacicular hooks bidentate, reddish from base to most of its length, with translucent end tip, starting from chaetigers 41R–42L, one per chaetiger, with continuous distribution; with blunt teeth, distal tooth smaller than proximal, both teeth directed upward (Fig. 4P); some chaetigers with subacicular hook unidentate with hoods. Pygidium with dorsal pair of anal cirri as long as last seven chaetigers; ventral pair short, as long as the last chaetiger.

Variations. Material examined varied in the following features: L10= 3–6.6 mm, W10= 1.3–2.1 mm. Palps reaching second peristomial ring or first chaetiger; lateral antennae reaching middle of first or second chaetiger; median antenna reaching third-fourth chaetiger. Maxillary formula varies as follows: MII 3–4+4–5, MIII 5–6, MIV 3–4+7–8. The proportion of maxillary apparatus varies as follows: MI are 2.4–2.7 times longer than maxillary carriers; MI are 4.3–5 times longer than closing system; MII are 3–3.3 times longer than cavity opening. Branchiae from chaetigers 25–34. The maximum number of branchial filaments 2. Postchaetal lobe well developed in first 17–91 chaetigers. Ventral cirri with a swollen base from chaetigers 3–7 to 70 chaetigers before of pygidium. Falcigers present up to last chaetiger (L10= 3–6 mm) or median region (L10= 6.1–66 mm). Start of subacicular hooks in chaetigers 28–43.

Regression analyses indicated that in this species, there is no correlation between the start of the branchiae ($R^2 = 0.0702$, $p = 0.26$, $n = 11$, Fig. 6), the maximum number of branchial filaments ($R^2 = 0.000$, $p = 0.00$, $n = 11$, Fig. 6) or the start of the subacicular hooks ($R^2 = 0.1307$, $p = 0.35$, $n = 11$, Fig. 6) with the length to chaetiger 10. The chaetiger where the branchiae start does not follow a pattern with respect to their growth, but its start occurs between the chaetigers 20 and 30 (Fig. 6, blue points). This same situation is repeated with the start of the subacicular hook, only they start chaetigers 30 and 40 (Fig. 6, points red). However, the number of filaments (2 filaments) seems to be fixed regardless of the size of the organism, a contrasting pattern with several species in which the number of filaments appears to increase with length of the specimen.

On the other hand, *M. sherlockae* n. sp. has similar condition as other species of *Marphysa* where the presence of compound chaetae is size dependent (Aiyar 1931; Pillai 1958; Salazar-Vallejo & Carrera-Parra 1998; Molina-Acevedo & Carrera-Parra 2017; Molina-Acevedo 2018). *Marphysa sherlockae* n. sp. specimens with $L10 \leq 6\text{mm}$ presented compound falcigers to last chaetiger. In this group of individuals, we observed that the number of falcigers per chaetiger decreased from median to posterior region, and this decrease was more noticeable in specimens with L10 close to 6 mm. Additionally, the specimens with $L10 > 6\text{ mm}$ do not present falcigers in the posterior region. This probably indicates that in the largest specimens of *M. sherlockae* n. sp. falcigers will be lost and only compound spinigers will be observed, as demonstrated in *M. gravellyi* Southern, 1921, *M. borradailei* Pillai, 1958 and *M. brevitentaculata* Treadwell, 1921.

Etymology: The species is named after Emma Sherlock, in recognition of her valuable work on the polychaete collections of BHNM.

DNA barcode: Strand, False Bay, Western Cape, South Africa (MB-XXXXXX) (Genbank accession number: XXXXXX). 577 bp fragment isolated with universal mitochondrial cytochrome oxidase subunit 1 gene, primer pair: LCO1490, HCO2198 (Folmer et al. 1994).

Habitat. Found in the fringing rocky zones at low tide in sheltered bays. Worms can be found in rock crevices.

Type locality. Langebaan Lagoon, South Africa.

Distribution. Day (1953; 1967) and Branch et al. (2016) recorded this species to occur in rocky coasts and estuaries from Saldanha Bay in the Western Cape to Durban in KwaZulu-Natal, South Africa.

Remarks. Day (1953) studied the material collected by himself and other members of the Zoology Department at the University of Cape Town during the course of ecological surveys of the rocky coasts and estuaries in South Africa. The author identified some specimens as *Marphysa depressa* collected from localities such as East London, Bushman's Estuary, Still Bay, Cape Agulhas and Langebaan Lagoon due to the presence of compound spinigers and falcigers in the same chaetiger which overlapped with the New Zealand species. As a result, this was the first record of the species in South Africa. Additionally, Day compared his material with a specimen collected from New Zealand by Ehlers (1904), most likely to confirm his identification. However, thorough taxonomic revisions revealed strong differences between the material from South African and New Zealand and led us to conclude that the South African specimens belong to a new species named herein as *Marphysa sherlockae* n. sp. *Marphysa sherlockae* n. sp. differs from *M. depressa* in the chaetal distribution, for example, the former has compound spinigers in all chaetigers, and compound falcigers restricted to median and posterior chaetigers; whereas in *M. depressa* the compound falciger is present in all chaetigers, and the spinigers are only in the anterior region. Also, *M. sherlockae* n. sp. has a triangular postchaetal lobe; while *M. depressa* has digitiform postchaetal lobe. Furthermore, *M. sherlockae* n. sp. (L10: 5.7–6.6 mm) has only two branchial filaments, while *M. depressa* (L10: 9.5 mm) has up to four filaments.

Marphysa sherlockae n. sp. resembles *M. durbanensis* and *M. haemasoma* by having compound spinigers. However, *M. sherlockae* n. sp. (L10: 5.7–6.6 mm) has two branchial filaments, triangular postchaetal lobe in anterior chaetigers, and ventral cirri with a swollen base ending 70 chaetigers before pygidium; whereas *M. durbanensis* (holotype, L10: 14 mm) has 11–12 branchial filaments, digitiform postchaetal lobes, and ventral cirri with a swollen base ending 25 chaetigers before pygidium. Further, *M. haemasoma* (L10: 9.3–18.5 mm) has 6–10 branchial filaments, ovoid postchaetal lobe, and ventral cirri with a swollen base until the last chaetiger.

Marphysa sherlockae **n. sp.** resembles that of *M. angelensis* Fauchald, 1970, *M. brevitentaculata*, *M. digitibranchia* Hoagland, 1920, *M. emiliae* Molina-Acevedo & Carrera-Parra, 2017, *M. formosa* Steiner & Amaral, 2000, *M. mangeri* Augener, 1918, *M. orensanzii* Carrera-Parra & Salazar-Vallejo, 1998 and *M. sebastiana* only having compound falcigers and spinigers present; however, *M. brevitentaculata*, *M. digitibranchia* and *M. mangeri* have limbate capillaries in the subacicular position from middle to posterior region of the body, while in *M. sherlockae* **n. sp.** these simple chaetae are absent. Furthermore, *M. angelensis* and *M. emiliae* have digitiform postchaetal lobe in first four chaetigers, while in *M. sherlockae* **n. sp.** the postchaetal lobe is triangular at the same first chaetigers. Also, in *M. emiliae* (L10: 3.5–5.4 mm) branchiae begin in chaetigers 8–12; while in *M. sherlockae* **n. sp.** (L10: 3–6.6 mm) branchiae begin from 25–34. On the other hand, *M. formosa* have pectinate branchiae, while *M. sherlockae* **n. sp.** have palmate branchiae with a short button-shaped branchial stem. Furthermore, *M. formosa* (TL: 55 mm), *M. orensanzii* (TL: 12 mm) and *M. sebastiana* (LT: 120 mm) have up to 4–6 branchial filaments whilst *M. sherlockae* **n. sp.** (TL: 67 mm) only have 2 filaments. Finally, *M. sebastiana* and *M. angelensis* has short branchial filaments, while the filaments in *M. sherlockae* **n. sp.** are long.

Discussion

This study revealed that *M. macintoshii* and *M. depressa* recorded for the region actually represent (1) an incorrectly synonymised indigenous species, i.e., *M. durbanensis* that was reinstated herein, and (2) a new indigenous species that was previously overlooked and herein described, i.e., *M. sherlockae* **n. sp.**, respectively. We also confirm Lewis & Karageorgopoulos (2008) in that *M. sanguinea* is not present along the South African coast; however, the local species should be named *M. haemasia* not *M. elityeni*, since the latter is a junior synonym of the former.

Marphysa depressa and *M. macintoshii* were first recorded on the South African coast (Day (1953, 1967) with summary descriptions and general illustrations. The recurrent identification of *M. macintoshii* and *M. depressa* along the South African coast (e.g., Branch et al. 2016) reflects the overlooking of detailed characteristics and the use of traditional and conspicuous diagnostic features considered enough to define *Marphysa* species; such as, the colour and shape of the subacicular hook, distribution of compound chaetae throughout the body, the shape and distribution of branchiae, and the number of branchial filaments (Quatrefages 1866, Grube 1878, McIntosh 1910, Hartman 1944, Fauchald 1970, among others). The sole use of distinctive conspicuous features in the identification may lead to spurious records of cosmopolitanism in species (Hutchings & Kupriyanova 2018), and also to the propagation of misleading species records and synonymizations.

The detailed study of the traditional conspicuous features, the finding of novel characters as well as the examination of type specimens, as carried out here, has improved the morphological delimitation of *Marphysa* species, and the understanding of the diversity within the genus (e.g., Glasby & Hutchings 2010; Molina-Acevedo & Carrera-Parra 2015, 2017). Therefore, recent

studies on *Marphysa* have focused on the detection of novel characters or in the re-assessment of those forgotten features, such as the shapes of dorsal cirri, postchaetal lobes, and pectinate chaetae, and the first appearance of the ventral cirrus with swollen base. For instance, Miura (1986) and Molina-Acevedo & Carrera-Parra (2015) have shown that the distribution of the number of filaments and the region where the maximum number is reached can be informative in species delimitation. Here, the distribution of branchial filaments is different in each analysed species (Fig. 7). Thus, whenever possible, it should be incorporated in future descriptions of *Marphysa* species. A challenge of using “new” features in the taxonomy is the lack of this information in older descriptions preventing comparison. Thus, the examination of type material is an essential step towards improving the taxonomy and recognizing new or misled synonyms as in the case of *M. haemasoma*.

Molecular data bring an additional source of information that improves our knowledge on species boundaries and aiding in the recognition of intraspecific variation (e.g., Lewis & Karageorgopoulos 2008; Zanol et al. 2016, 2017, Lavesque et al. 2017, Elgetany et al. 2018, Lavesque et al. 2019, Glasby et al. 2019, Abe et al. 2019, Martin et al. 2020). The phylogenetic tree revealed two distinct South African monophyletic clades, one of them belonging to the new species *M. sherlockae* n. sp., and the other to *M. haemasoma*. The molecular analyses reinforced the establishment of *M. haemasoma* and its distinction from *M. sanguinea*, which agrees with previous findings from the region (Lewis & Karageorgopoulos 2008) and for the first time provided COI sequences of this species from South Africa.

Considering the present study, a total of nine *Marphysa* species have been newly proposed or redescribed under a taxonomic integrative framework since 2003 (e.g., Zanol et al. 2016; Zanol et al. 2017; Lavesque et al. 2017; Elgetany et al. 2018; Lavesque et al. 2019; Glasby et al. 2019; Abe et al. 2019; Martin et al. 2020), thus, increasing the number of publicly available sequences of *Marphysa* species globally, which in turn provides a starting point from which other studies can address more complex hypotheses such as resolving the phylogenetic placements of species within the genus.

This study has indicated that the indigenous diversity of *Marphysa* in South Africa was indeed previously underestimated and thus increases the number of described indigenous species from three to five (Day 1967; Lewis & Karageorgopoulos 2008) and reduces the number of putative cosmopolitan species to one (i.e., *Marphysa corallina*). Similar to studies by Lewis & Karageorgopoulos (2008); Clarke et al. (2010); Kara et al. (2018) and Simon et al. (2019), the present study provides additional evidence that many cosmopolitan species reported in the Day (1967) polychaete monograph for this region are actually incorrect assignments. Undoubtedly, the polychaete monograph authored by John Day is an invaluable resource for polychaete descriptions and distributions. However, it is widely used by researchers from many disciplines including those working outside of the region (Hutchings & Kupriyanova 2018). Thus, biologists locally and internationally should take cognisance of this fact and use the monograph with caution, especially with regard to species that are considered “cosmopolitan”.

Using information from Day (1967), Awad et al. (2002) determined that only 20% of polychaete species in South Africa are endemic to the region. Thus, if only half the remaining 80% prove to be misidentifications of indigenous species, our understanding of diversity, biogeography and endemism of polychaete worms in South Africa have been severely underestimated and priority conservation areas may need to be reviewed. Furthermore, the resolution of taxonomically confusing species, such as those belonging to *Marphysa*, and consequently more realistic diversity estimates will be improved if voucher specimens are deposited in museums for taxonomy and molecular investigations.

Conclusions

Marphysa in South Africa is now represented by six species namely, *M. capensis*, *M. corallina*, *M. durbanensis*, *M. haemasoma*, *M. posteriobranchia* and *M. sherlockae* n. sp. Although the number of species is similar with previous identifications, the resurrection of *M. haemasoma*, downgrading of *M. elityeni*, reinstatement of *M. durbanensis* from *M. macintoshi* and redescription of *M. sherlockae* n. sp. from *M. depressa* has changed the composition of endemic and cosmopolitan species. As such, gaining a better understanding of our true local biodiversity may help us to understand the extent of biodiversity loss in the face of climate change and also help to make better decisions regarding the designation of marine protected areas.

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Table 1 (on next page)

Sequence data of *Marphysa*

Sequence data of *Marphysa* species used in the phylogenetic analysis

Table 1. Sequence data of *Marphysa* species used in the phylogenetic analysis. 

Species	Genbank accession number	Location	Reference
<i>Marphysa haemasoma</i>	MN067877	South Africa	Simon et al. unpublished data.
<i>Marphysa sherlockae</i> n. sp.	GBxxxxxxx	South Africa	This study
<i>Marphysa aegypti</i>	MF196971, MF196969, MF196970, MF196968	Egypt	Elgetany et al. 2018
<i>Marphysa chirigota</i>	MN816441, MN816442, MN816443	Iberian Peninsula	Martin et al. 2020
<i>Marphysa bifurcata</i>	KX172177, KX172178	Australia	Zanol et al. 2016
<i>Marphysa brevitentaculata</i>	GQ497548	Mexico	Zanol et al. 2010
<i>Marphysa californica</i>	GQ497552	California	Zanol et al. 2010
<i>Marphysa corallina</i>	KT823271, KT823300, KT823306, KT823343, KT823371, KT823389, KT823410	South Africa	Kara et al. unpublished
<i>Marphysa fauchaldi</i>	KX172165	Australia	Zanol et al. 2016
<i>Marphysa gaditana</i>	MN816444, KR916870, AY040708, KR916871, KR916872, KR91687, KP254503, KP254537, KP254643, KP254743, KP254802	Iberian peninsula, Portugal, France, Virginia (USA)	Martin et al. 2020, Lobo et al. 2016, Siddal et al. 2001, Leray et al. 2015
<i>Marphysa honkongensa</i>	MH598526	China	Wang et al. 2018
<i>Marphysa iloiloensis</i>	MN133418, MN106279, MN106280, MN106281	Phillipines	Glasby et al. 2019
<i>Marphysa kristiani</i>	KX172141, KX172142, KX172143, KX172144, KX172145, KX172146,	Australia	Zanol et al. 2016

Species	Genbank accession number	Location	Reference
	KX172147, KX172148, KX172149, KX172150, KX172151, KX172155, KX172152, KX172153, KX172154, KX172156, KX172157, KX172158, KX172159, KX172160, KX172161, KX172162, KX172163		
<i>Marphysa mossambica</i>	JX559751, KX172164	Philippines, Australia	Zanol et al. 2010, Zanol et al. 2016
<i>Marphysa mullawa</i>	KX172166, KX172167, KX172168, KX172169, KX172170, KX172171, KX172172, KX172173, KX172174, KX172175, KX172176	Australia	Zanol et al. 2016
<i>Marphysa pseudosessiloides</i>	KY605405, KY605406	Australia	Zanol et al. 2017
<i>Marphysa regalis</i>	GQ497562	Brazil	Zanol et al. 2016
<i>Marphysa victori</i>	MG384996, MG384999, MG384997, MG384998	France	Lavesque et al. 2017
<i>Marphysa viridis</i>	GQ497553	Brazil	Zanol et al. 2010
<i>Marphysa sanguinea</i>	GQ497547, MK541904, MK950851, MK950852, MK950853, MK967470, MN106282, MN106283, MN106284	Cornwall (UK), France	Zanol et al. 2010, Lavesque et al. 2019, Glasby et al. 2019
<i>Marphysa tripectinata</i>	MN106271, MN10622, MN1062723, MN106274, MN106275, MN106276, MN106277, MN106278	China	Liu et al. 2017
<i>Marphysa</i> sp.	KP255196, KP254890, KP254644, KP254223, NC023124, KF733802	Florida (USA), China	Leray et al. 2015, Li et al. 2016
<i>Paucibranchia</i>	KT307661	Spain	Aylagas et al. 2016

Species	Genbank accession number	Location	Reference
<i>bellii</i>			
<i>Paucibranchia disjuncta</i>	GQ497549	California, USA	Zanol et al. 2010
<i>Paucibranchia</i> sp.	JX559753	Phillipines	Zanol et al. 2014
<i>Palola viridis</i>	GQ497556	Micronesia	Zanol et al. 2010
<i>Eunice cf. violaceomaculata</i>	GQ497542	Belize	Zanol et al. 2010
<i>Leodice rubra</i>	GQ497528	Brazil	Zanol et al. 2010
<i>Nicidion angeli</i>	GQ497550	Brazil	Zanol et al. 2010
<i>Hyalinoecia</i> sp.	GQ497524	Massachusetts, USA	Zanol et al. 2010

3
4

Figure 1

Sampling localities

M. depressa (Strand), *M. macintoshi* (Durban Bay), *M. haemasoma* (Table Bay) and *M. elityeni* (Buffels Bay and Kommetjie) from South Africa.

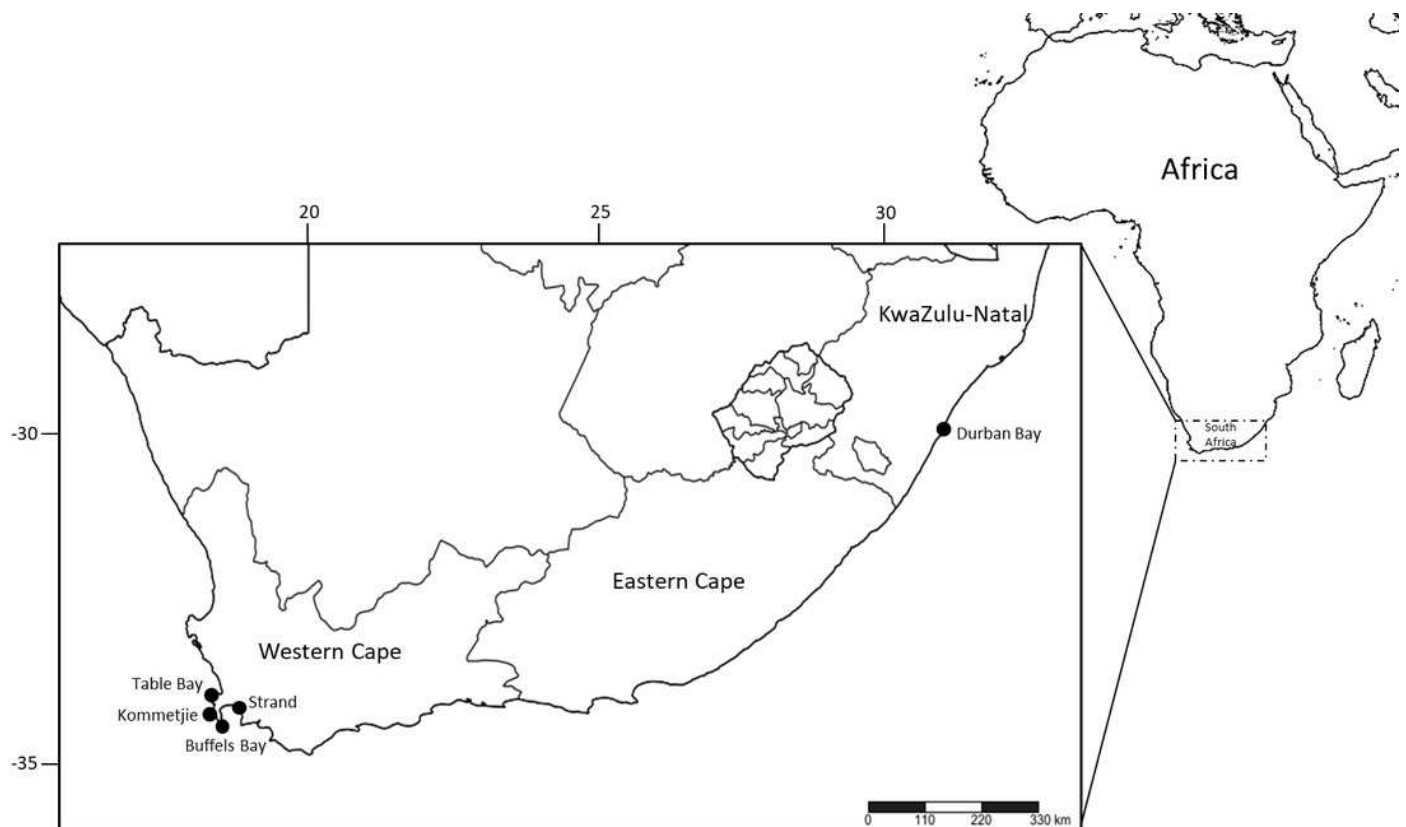


Figure 2

Marphysa durbanensis Day, 1934

A. Anterior end, dorsal view; B. Anterior end, ventral view; C. Anterior view, lateral view; D. Maxillary apparatus, dorsal view; E. Left MI-II-III-IV-V, lateral view; F. Mandible; G. Parapodium 3; H. Parapodium 8; I. Parapodium 12; J. Parapodium 69; K. Parapodium 217; L. Thin narrow isodont pectinate with long and slender teeth, chaetiger 3; M. Thin wide isodont with short and slender teeth, chaetiger 69; N. Thick wide isodont pectinate with short and thick teeth, chaetiger 140; O. Thick wide anodont with short and slender teeth, chaetiger 140; P. Compound spinigers, chaetiger 3; Q. Subacicular hook, chaetiger 278. A-C, G-P from Lectotype BNHM 0000; D-F, Q from paralectotype BNHM 0000. All chaetigers in anterior view; LMI-II: Ligament between MI and MII; LMII-III: Ligament between MII and MIII. Scale bars: A-C, 3.5 mm; D-E, 0.9 mm; F, 0.8 mm; G-K, 0.2 mm; L-O, Q 30 μ m; P, 0.1 mm

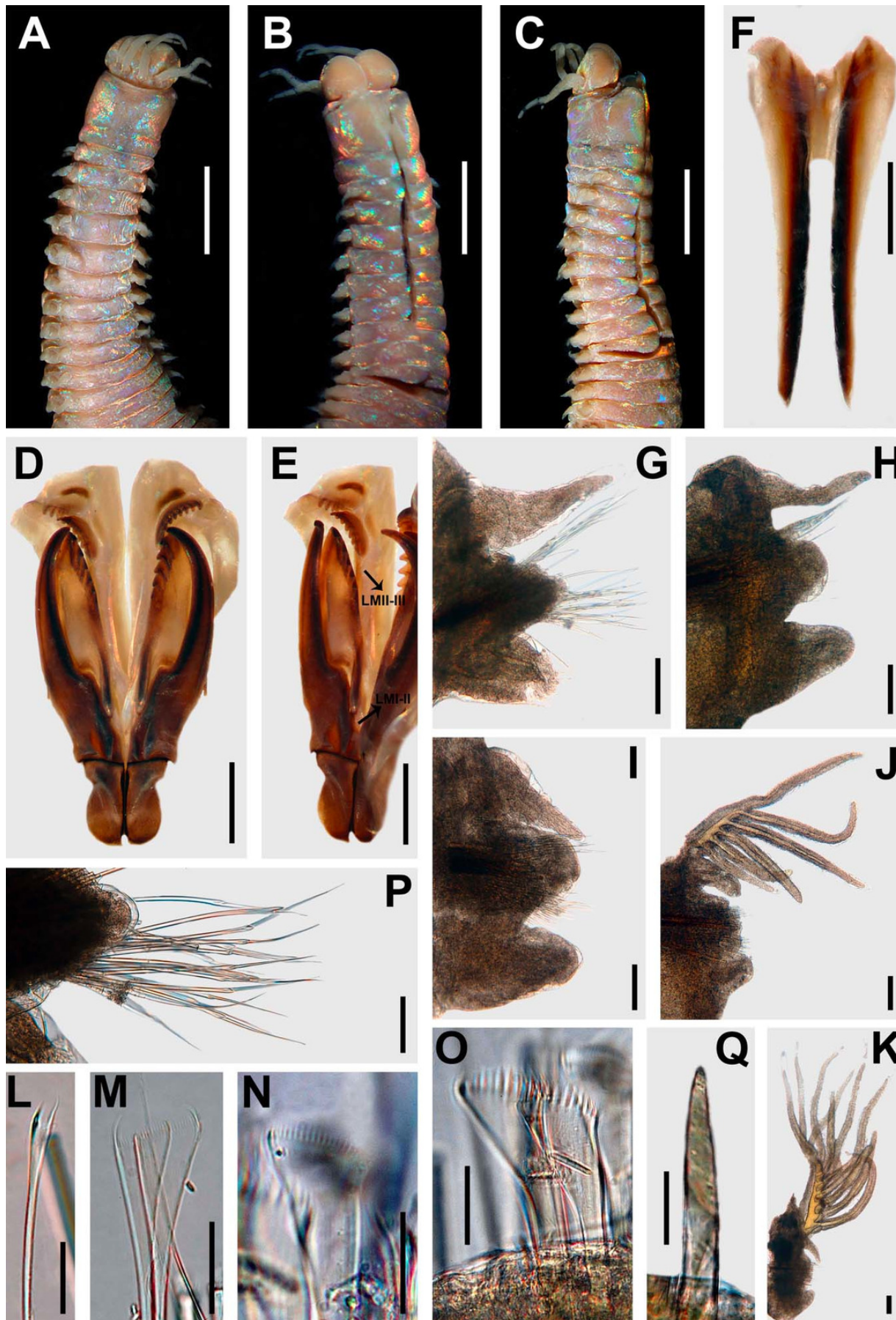


Figure 3

Marphysa haemasoma Quatrefages, 1866

A. Anterior end, dorsal view; B. Anterior end, ventral view; C. Anterior view, lateral view; D. Maxillary apparatus, dorsal view; E. Left MI-II-III-IV-V, lateral view; F. Mandible; G. Parapodium 3; H. Parapodium 12; I. Parapodium 30; J. Parapodium 154; K. Parapodium 307; L. Thin narrow isodont with long and slender teeth, chaetiger 3; M. Thick wide isodont with short and slender teeth, chaetiger 251; N. Thick wide anodont with short and slender teeth, chaetiger 307; O. Thick wide anodont with long and thick teeth, chaetiger 251; P. Compound spinigers, chaetiger 3; Q. Subacicular hook, chaetiger 209. A-B, D-E, G-L, N, P from Holotype *M. haemasoma* MNHN type 613; F, M, O, Q from Paratype *M. elityeni* BNHM 2007.69. All chaetigers in anterior view; al-MIII: attachment lamella MIII; al-MIV: attachment lamella MIV; LMI-II: Ligament between MI and MII; LMII-III: Ligament between MII and MIII. Scale bars: A-B, 3.1 mm; C, 3.8mm; D-E, 1.2 mm; F, 1.7 mm; G-K, 0.2 mm; L-O, Q, 30 µm; P, 0.1 mm.

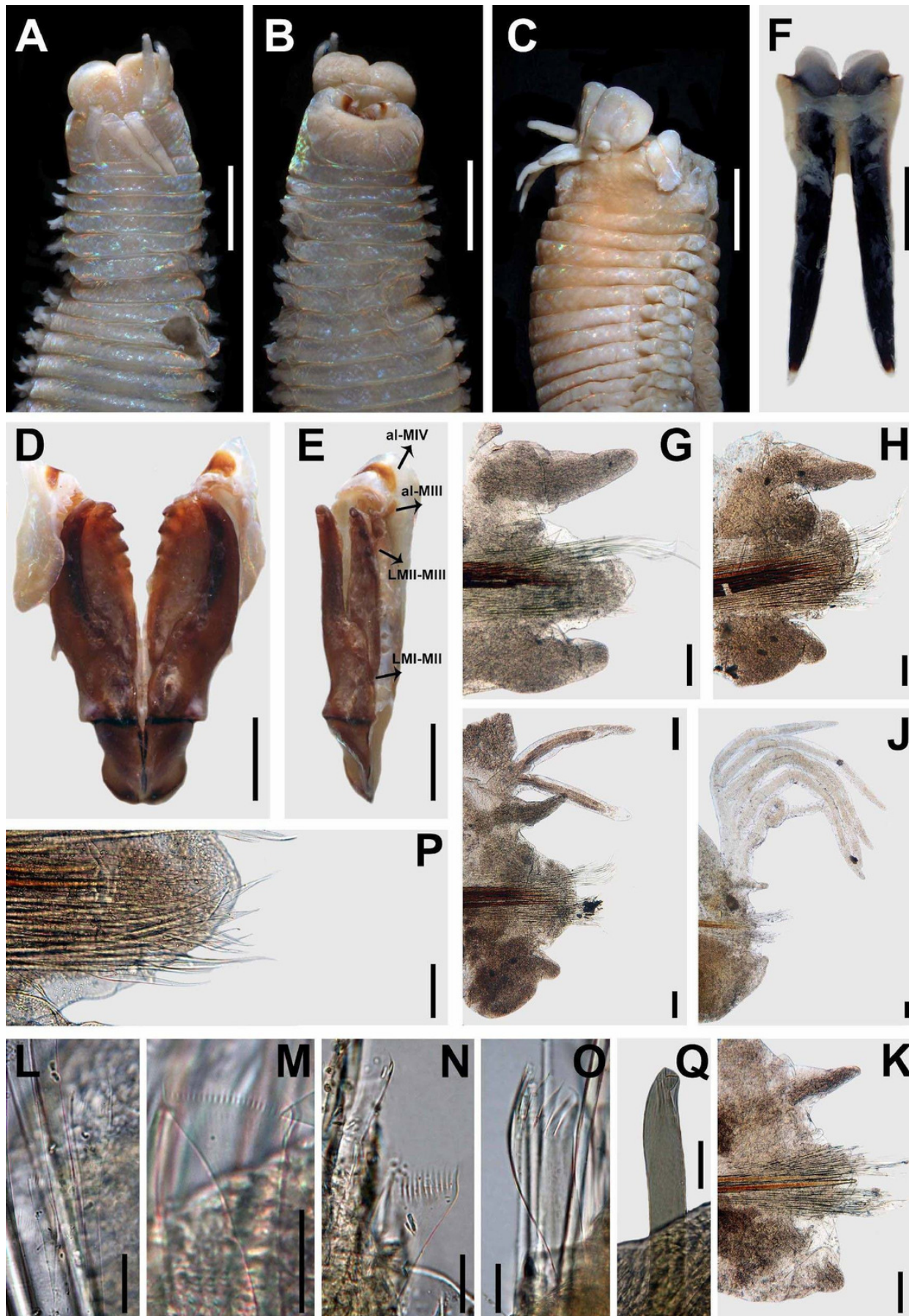


Figure 4

Marphysa sherlockae n. sp. Holotype BNHM 1963.1.84.

A. Anterior end, dorsal view; B. Anterior end, ventral view; C. Anterior end, ventral view; D. Maxillary apparatus, dorsal view; E. Left MI-II-III-IV-V, lateral view; F. Mandible; G. Parapodium 3; H. Parapodium 6; I. Parapodium 14; J. Parapodium 114; K. Parapodium 185 L. Thin narrow isodont with long and slender teeth, chaetiger 3; M. Thick wide isodont with long and thick teeth, chaetiger 185; N. Compound spinigers, chaetiger 3; O. Compound falcigers, chaetiger 3; P. Subacicular hook, chaetiger 49. All chaetigers in anterior view; al-MIII: attachment lamella MIII; al-MIV: attachment lamella MIV; LMI-II: Ligament between MI and MII; LMII-III: Ligament between MII and MIII. Scale bars: A-C, 1.7 mm; D-E, 0.6 mm; F, 0.4 mm; G-K, 0.1 mm; N-P, 30 μ m.

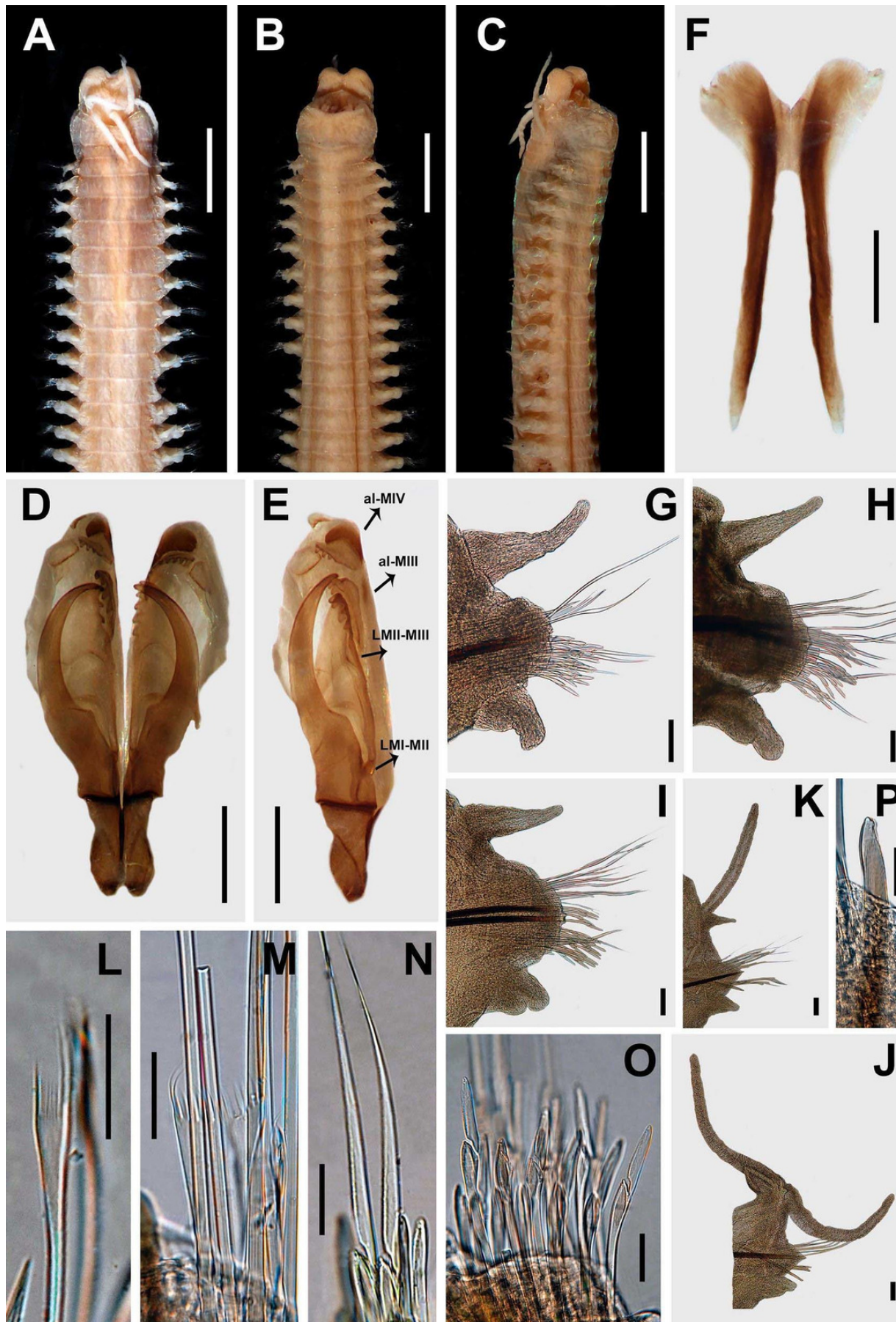


Figure 5

Marphysa sherlockae n. sp.

A. Thin narrow isodont pectinate chaetae with long and slender teeth, anterior chaetiger; B. Thin narrow isodont pectinate chaetae with long and slender teeth, anterior chaetiger; C. Thick narrow isodont pectinate chaetae with long and slender teeth, chaetiger 32; D. Thick wide isodont pectinate chaetae wide with long and thick teeth, posterior chaetiger; E. Thick wide isodont pectinate chaetae with long and thick teeth, posterior chaetiger. A, B, C from SAMC-A20578; D, E SAMC-A089089 Scale bars: A-E, 0.05 mm.

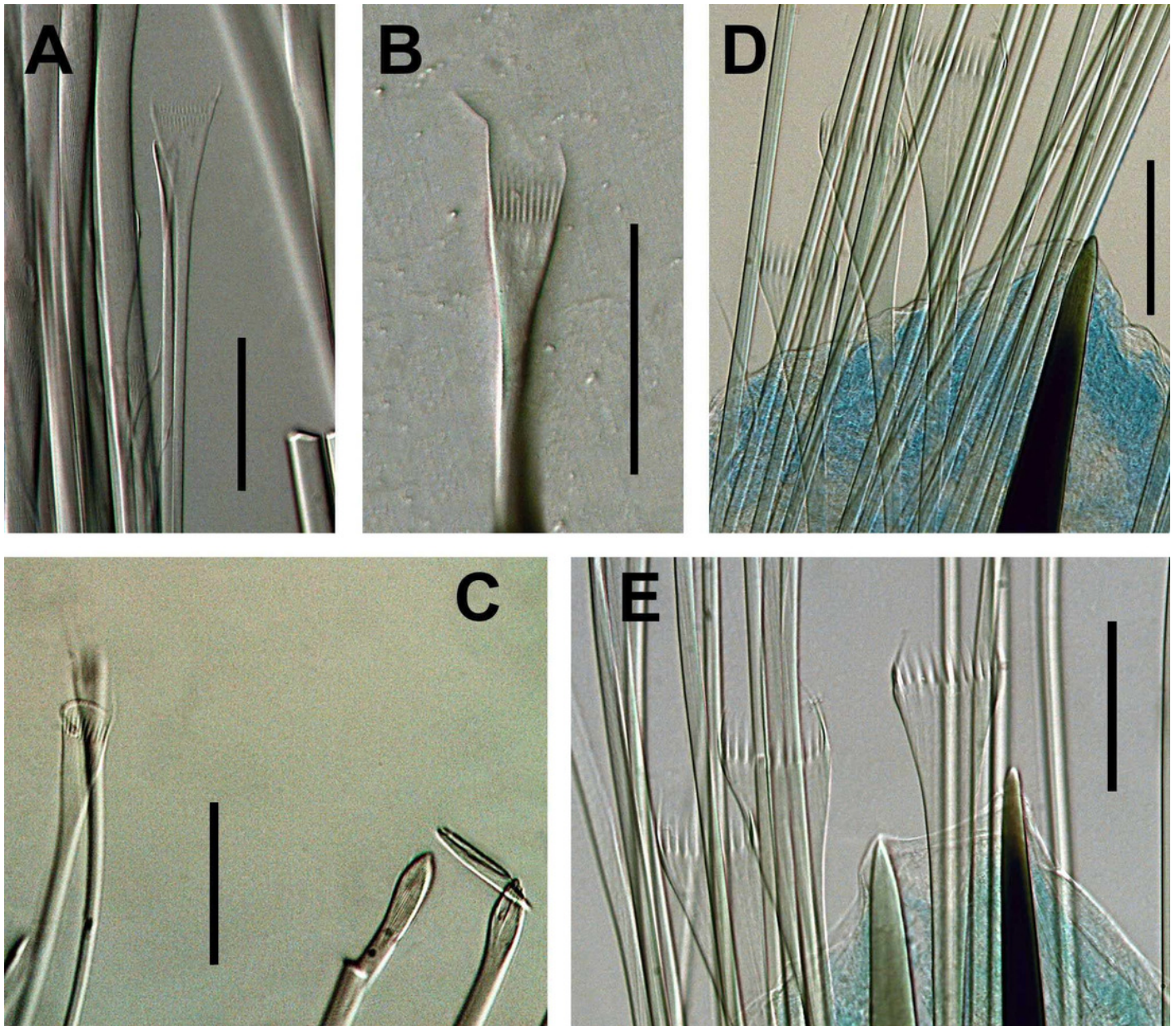


Figure 6

Length-dependent variation of some morphological features in *Marphysa sherlockae* n. sp.

Red point: Chaetiger where subacicular hook start ($p=0.35$, $n=11$). Blue points: First chaetiger with branchia ($p=0.26$, $n=11$); Green points: Maximum number of branchial filaments ($p=0.00$, $n=11$).

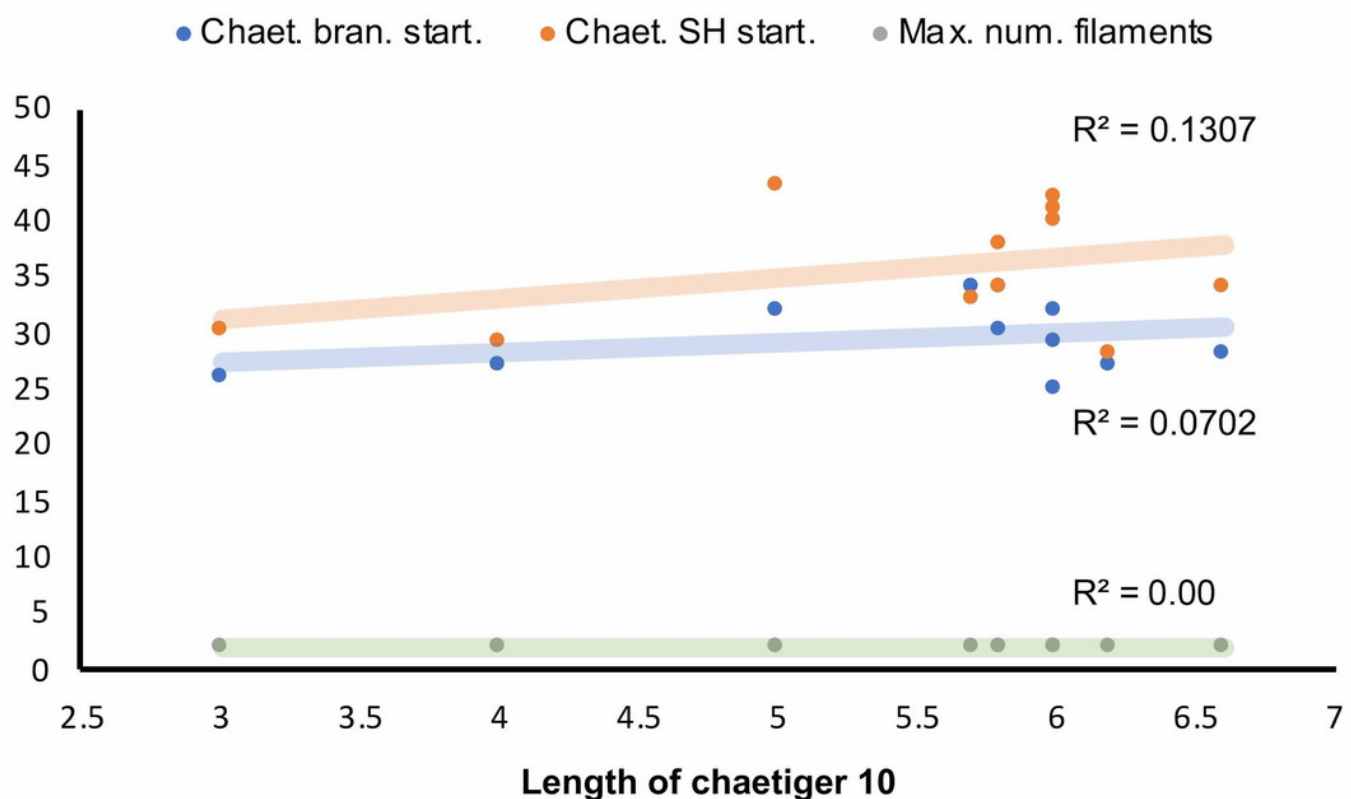


Figure 7

Distribution of branchial filaments throughout the body.

Marphysa durbanensis Day, 1934, *Marphysa haemasoma* Quatrefages, 1865 and *M. sherlockae* n. sp.

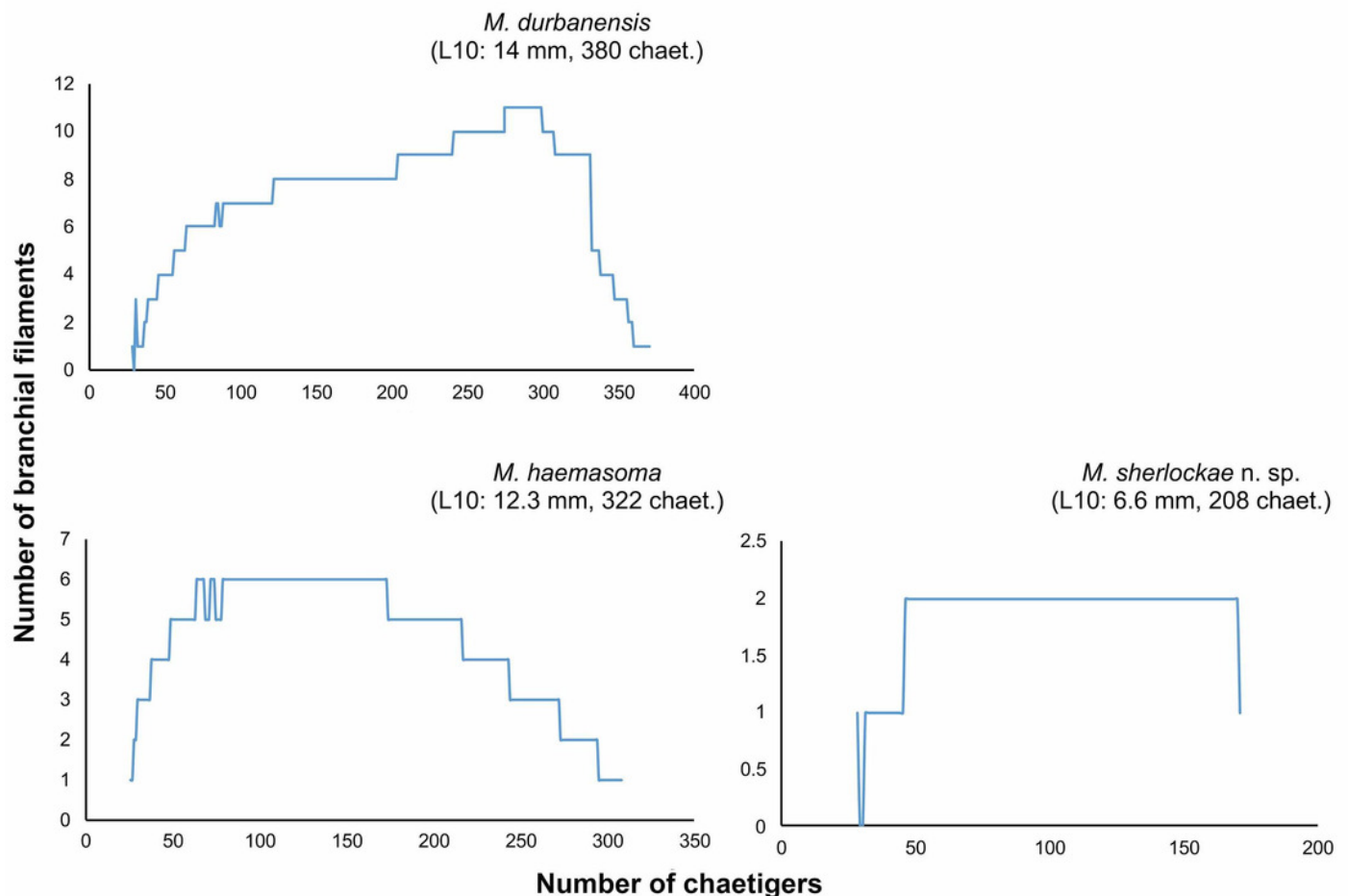


Figure 8



Phylogenetic tree based on the mitochondrial cytochrome c oxidase subunit 1 alignment of *Marphysa* spp. globally.

Bayesian probabilities >95% are represented by an * at each node. The two South African species described in text are: Purple clade - the reinstated *M. haemasoma* and blue clade - newly described *M. sherlockae* **n. sp.**

