

Myzostoma fuscomaculatum (Myzostomida), a new myzostome species from False Bay, South Africa

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Abstract A new myzostome species, described here as *Myzostoma fuscomaculatum* n. sp. was collected on *Tropiometra carinata* in False Bay (South Africa), during a survey of symbionts associated with comatulid crinoid species. *M. fuscomaculatum* n. sp. occurred only on *T. carinata* and not on the more common crinoid, *Comanthus wahlbergi*. It infested 61.7% of the 120 host specimens collected, of which 64.9% (48 specimens) hosted more than one individual (maximum of 32). *M. fuscomaculatum* n. sp. was always located on the host's arms and pinnules and was cryptically coloured, closely matching the colour pattern of the host. This is the first record of myzostomes from the cool temperate waters of South Africa's Atlantic coast. The new species is morphologically close to *M. gopalai* Subramaniam, 1938, collected on *T. encrinus* in Madras Harbour. *M. fuscomaculatum* n. sp. differs from *M. gopalai* in

lacking marginal cirri at the adult stage, the presence of three pairs of digestive diverticula, by the position of its lateral organs and by the shape of the manubrium. Molecular phylogenetic analyses based on 18S and 16S rDNA placed *M. fuscomaculatum* n. sp. into a clade including *Hypomyzostoma*, *Myzostoma* and *Mesomyzostoma* species.

Keywords Myzostomida · Annelida · Taxonomy · South Africa · Atlantic Ocean · Phylogenetic analysis

Introduction

Comatulid crinoids are a prominent component of the benthic fauna of False Bay in South Africa's Western Cape Province. *Comanthus wahlbergi* (Müller, 1843) is the most abundant species, often forming dense beds on shallow rocky reefs, while *Tropiometra carinata* (Lamarck, 1816) is commonly found singly on shallow rocky reefs, becoming more abundant on deeper reefs (Branch et al., 1994). During a survey of symbionts associated with these two common crinoid species, an undescribed species of Myzostomida was collected from *T. carinata*.

Myzostomida are a small group comprising over 170 described species that occur almost worldwide and from the intertidal zone to over 3,000 m depth (Grygier, 2000; Eeckhaut & Lanterbecq, 2005). They are obligate commensals or parasites of echinoderms,

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with about 78% of the described species being ectocommensals of crinoids. Myzostomida are commonly considered as constituting either a class of Annelida (Brusca & Brusca, 1990) or an aberrant family of Polychaeta (Ruppert & Barnes, 1994). During the last decade, several molecular phylogenetic analyses attempted to infer the relationships of Myzostomida. Eeckhaut et al. (2000) suggested that they were close to Platyhelminthes, a result that was later supported by Zrzavy et al. (2001) who placed myzostomes nested with Cyclophora, Rotifera, and Acanthocephala into Platyzoa. Passamanek & Halanych (2006) skeptically presented myzostomes within the Bryozoa, based on the analyses of LSU and SSU data. Recently, in their deep phylogeny analysis, Dunn et al. (2008) showed a myzostome coming out together with an acoele and a gnathostomulid. To date, only one molecular analysis (Bleidorn et al., 2007) obtained results in agreement with morphological characters (Rouse & Fauchald, 1997): analyses using data from nuclear genes (18S rDNA, 28S rDNA, myosin II, and elongation factor-1) and a nearly complete myzostome mitochondrial genome supports the position that myzostomes are part of the annelid radiation.

Many myzostomes are mobile, moving freely on their hosts and stealing food from them, while others are sessile, living near a 'convenient' feeding spot, and either inducing the formation of galls or cysts in their host integument, or living directly in the digestive system, coelom, or gonads of the host (see Grygier, 2000; Eeckhaut & Lanterbecq, 2005, for reviews). Probably due to their large variety of lifestyles, myzostomes exhibit a wide range of colours and body shapes. Flattened discoid body shapes are predominant in ectocommensals, but many species are elongate, dorsally ridged, and/or possess caudal extensions, and most tend to mimic the colours and structures of their host (calyx, pinnules, etc.) (Eeckhaut & Lanterbecq, 2005), as the present new species, *Myzostoma fuscomaculatum* n. sp., does (Fig. 1).

Most described myzostomes (more than 100 species) occur in warm waters of the Indo-Pacific, especially coral reef environments, where the diversity of crinoid hosts is high (see Grygier 1990, 2000). Records of myzostomes in cold waters are sparse, with only a few described species from European waters (see Grygier, 2000). The only record of myzostomes

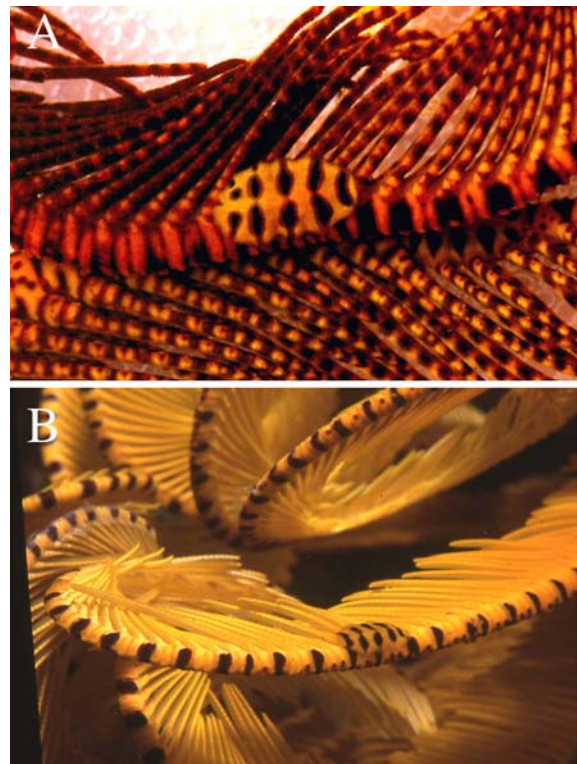


Fig. 1 *Myzostoma fuscomaculatum* n. sp. Specimens alive, showing the cryptic colouration compared to the colour pattern of the host

from South African waters to date is of the cyst-forming *M. willemoesii* Graff 1884 (Clark & Rowe, 1971; Grygier, 1990), associated with *T. carinata* (Clark & Rowe, 1971) and *Pachylometra sclateri* (Grygier's personal notes) off East London, on the south-east coast. In addition, an unopened twisted cyst of the *M. willemoesii* type, taken from a *Crotalometra magnicirra* from the Cape of Good Hope, has been found in the collections of the Natural History Museum in London, but this Atlantic record has never been published (Grygier's personal notes). The present description of the new species, *M. fuscomaculatum* n. sp., then represents the first published record to date of myzostomes in the cool temperate waters of the country's Atlantic coastline.

Grygier (1990, 1992) informally divided the genus *Myzostoma* into six recognizable groupings based on the external anatomic features (e.g. cirri and parapodial shape), lifestyle and distribution: the *gigas* group, the *platypus* group, the *ambiguum* group, the *costatum* group, the *crosslandi* group and the *wyvillethomsoni* group. The *wyvillethomsoni* group,

including *M. willemoesii* Graff 1884 cited above from South African waters, included species with long-oval bodies and prominent parapodial cirri (the first three and last two pairs are directed anteromedially and posteromedially, respectively) (Grygier, 1990). The *crosslandi* group included species with moderate to very elongate bodies with a dentate or scalloped margin instead of the usual 10 pairs of marginal cirri, and with lateral organs close to the margin (Grygier, 1990). This last group included (at the very least) *M. crosslandi*, *M. nanseni*, *M. sulcatum*, *M. elongatum*, *M. folium*, *M. taeniatum*, *M. membranaceum*, *M. fasciatum* and *M. maculatum* (plus a few other described species, not cited in Grygier 1990, and several undescribed ones). It is from that informal division of *Myzostoma* species (Grygier, 1990) that the generic name of *Hypomyzostoma* (originally proposed by Perrier, 1897) started to be used when Eeckhaut et al. (1998) applied it to the *crosslandi* species group, before being recognized as a generic name by Grygier (2000) in his review of Myzostomida. The *crosslandi* species group included *M. folium* Graff, 1877, that is now the type-species by monotypy of *Hypomyzostoma*. The genus *Hypomyzostoma* currently comprises at least 11 described species (including *H. crosslandi*, *H. nanseni*, *H. sulcatum*, *H. elongatum*, *H. folium*, *H. taeniatum*, *H. membranaceum*, *H. dodecaphalcis* and *H. longitergum*), but a number of other specimens are still undescribed, especially in Australia (Grygier, 2000).

In total, only 25% of the described species of Myzostomatidae, a family divided into three recognized nominal genera (*Myzostoma*, *Hypomyzostoma* and *Notopharyngoides*, see Grygier, 2000), fit into one of the five informal groups (Grygier, 1990, 1992) and this family obviously needs revisions (see Lanterbecq et al., 2006).

Here, we describe a new myzostome species, *M. fuscomaculatum* n. sp., the first species to be formally recorded from the cool temperate waters of South Africa.

Materials and methods

Crinoids and their associated symbionts were collected from shallow subtidal waters at Oatlands Point (34°12.48'S; 18°27.66'E) just south of Simon's Town in False Bay, South Africa, between 29 July and 8

September 2006. Each crinoid was gently lifted from the reef by a diver and enclosed in its own plastic bag filled with seawater, minimizing disturbance to the crinoids and their symbionts. The bags were carefully closed and then refrigerated for later processing in the laboratory. A total of 120 specimens of *T. carinata* were ultimately collected from depths of 0 to 20 m.

Each crinoid was placed in a solution of 20% ethanol in sea water and shaken vigorously. All symbionts were collected in a 265- μ m sieve, then counted and identified under a dissecting microscope. The symbionts of interest here, the ectocommensal myzostomes, were photographed and put into fixative for morphological study, or into ethanol for molecular analysis.

For histological observations, one paratype was fixed in Bouin's fluid for 24 h, and then dehydrated in graded concentrations of ethanol, embedded in paraffin, cut into 8 μ m thick sections with a HM 340E Microm microtome and stained with Masson's trichrome according to the procedure of Gabe (1968).

For SEM observations, myzostomes were fixed in Bouin's fluid for 24 h, and then dehydrated in graded concentrations of ethanol and critical point dried using CO₂ as the transition fluid. Specimens were then mounted on aluminium stubs, coated with gold in a sputter coater, and observed using a JEOL JSM 6100 scanning electron microscope.

Descriptions of the parapodial hook apparatus were made following digestion of the soft tissues in 25% bleach solution. Once most of the tissue was dissolved, the hooks and support rods were dried and mounted on slides. Microscopical observations were made using a Leica MZ8 zoom microscope.

Holotype and 10 paratypes of the new species have been deposited in the Institut Royal des Sciences Naturelles de Belgique (Bruxelles), under the IG number 31130/01–31130/11. Other specimens are conserved in the South African Museum (Cape Town).

The phylogenetic position of *M. fuscomaculatum* n. sp. was assigned based on the analyses of nuclear small ribosomal subunit (18S rDNA) and the mitochondrial large ribosomal subunit (16S rDNA) sequences, using 37 known myzostome sequences (Lanterbecq et al., 2006). Genomic DNA of *M. fuscomaculatum* n. sp. was extracted using the commercial Invitex Spin Tissue Mini kit (Invisorb). DNA fragments from the 18S rDNA (ca. 1,700 nucleotides) and 16S rDNA (ca. 360 nucleotides)

were amplified by PCR and sequenced following the procedure detailed by Lanterbecq et al., (2006). Sequences of the target gene fragments were successfully obtained for *M. fuscomaculatum* n. sp., edited with SEQPUP (Gilbert, 1996) and Se-AL v2.0a11 (Rambaut, 1996), and deposited in Genbank under accession number FJ346827 and FJ346828. The 18S and 16S rDNA alignment (where sites presenting a posterior probability of <90% were excluded in ProAlign; see Lanterbecq et al., 2006 for details) was used as a profile in ClustalX (Thompson et al., 1997), against which the new myzostome 18S and 16S rDNA sequences were aligned using default parameter settings. MP analyses were performed with PAUP*4.0b4a (Swofford, 1998) using a heuristic search (SeqAdd and TBR branch-swapping). Clade supports were estimated by bootstrapping (Felsenstein, 1985) (Simple SeqAdd and TBR branch-swapping; 1,000 replicates). Bayesian analyses were performed using MrBayes v3.0b4 (Ronquist & Huelsenbeck, 2003), using the model (GTR+I+G) selected by MrModelTest 1.0b (Nylander, 2002) based on Hierarchical Likelihood Ratio Tests (the same model was also chosen by the AIC test). Four Markov chains were run simultaneously for 5×10^5 generations, and trees were sampled every 100 cycles for a total of 5,000 trees. The first 1,000 trees with preasymptotic likelihood scores, i.e. the 100,000 first generations, were discarded as 'burn-in'. The remaining trees were used to compute Bayesian posterior probabilities (BPP) for each clade of the consensus tree.

Results

Myzostoma fuscomaculatum n. sp. (Figs. 1A, B, 2A–I, 3).

Taxonomic account

Material examined

False Bay, South Africa: 11 specimens. Prevalence of infestation: 61.7% (74 specimens) of the 120 host specimens collected, of which 64.9% (48 specimens) hosted more than one myzostome (with a maximum of 32 per crinoid, and a mean of 7). Holotype (IG31130/01) preserved in 100% ethanol; 10 paratypes: two intact paratypes (IG31130/02 and

Fig. 2 *Myzostoma fuscomaculatum* n. sp. Dorsal (A) and ventral view (B) of one specimen alive, showing light beige colouration; introvert extruded but folded on itself at half length. Dorsal view (C) of another living specimen, showing orange colouration and the fully everted introvert. SEM ventral view of paratype IG31130/04 (D), showing the folded margin. SEM ventral view of paratype IG31130/05 (E), specimen unfolded. Detailed SEM view of the margin (F) showing most of three parapodia and three lateral organs. Detailed SEM view of the third parapodium (G), showing a lateral organ and the partially everted penis. Detailed SEM view of one parapodium (H) showing the parapodial cirrus at its base. Micrograph of a parapodium digested in bleach (paratype IG31130/10) showing the hook apparatus (I). The margin of all adult specimens lacks cirri. Scales: A, B, 1 mm; D, E, 1 mm; F, G, I, 100 μ m; H, 10 μ m. Abbreviations: a, ano-genital pore; ac, acicula; c, parapodial cirrus; ci, ciliature; h, hook; i, introvert; fm, folded margin; lo, lateral organ; m, margin of the trunk; ma, manubrium; p, parapodium; pc, parapodial cone; pe, penis; ph, parapodial hook; rh, replacement hook; vm, ventral midline; asterisks indicate the five pairs of parapodia

IG31130/03) in 100% ethanol and six paratypes (IG31130/04 to IG31130/09) used for SEM observations. One individual (paratype IG31130/10) dissolved in 25% bleach solution for observation of parapodial hook apparatus. One individual (paratype IG31130/11) used for histological sections. Other specimens retained for additional study in University of Cape Town (South Africa).

Etymology

The species name is a combination of the Latin adjective '*maculatum*' referring to the coloured patches that the specimens exhibit dorsally, and '*fusco*' referring to the brownish colour of these patches.

Diagnosis

Medium-sized myzostome with oval-elongate trunk ranging from 2.5 to 6 mm long. No marginal cirri observed, but some humps on margin of a few fixed specimens. Five pairs of robust, elongate and cirrate parapodia, arranged in two rows, located closer to midline than to body margin. Hook with moderately thick shaft, ending in large opened, rounded hook, tip curving 90°. Support rod (acicula) longer than hook; distal end of the acicula thinner than the hook; small axe-shaped manubrium, with translucent extremity.

Four pairs of lateral organs located between parapodia and body margin, closer to margin than

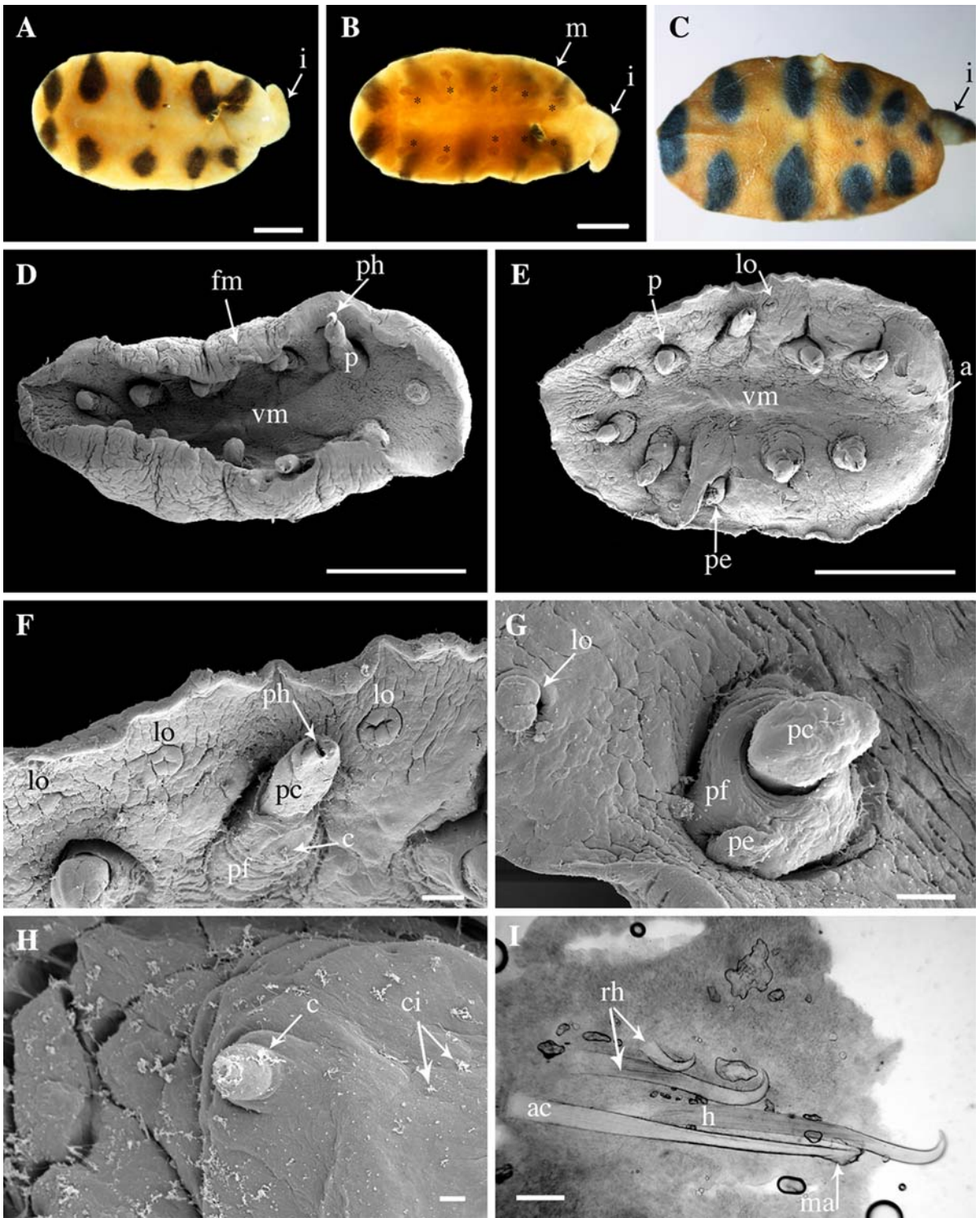
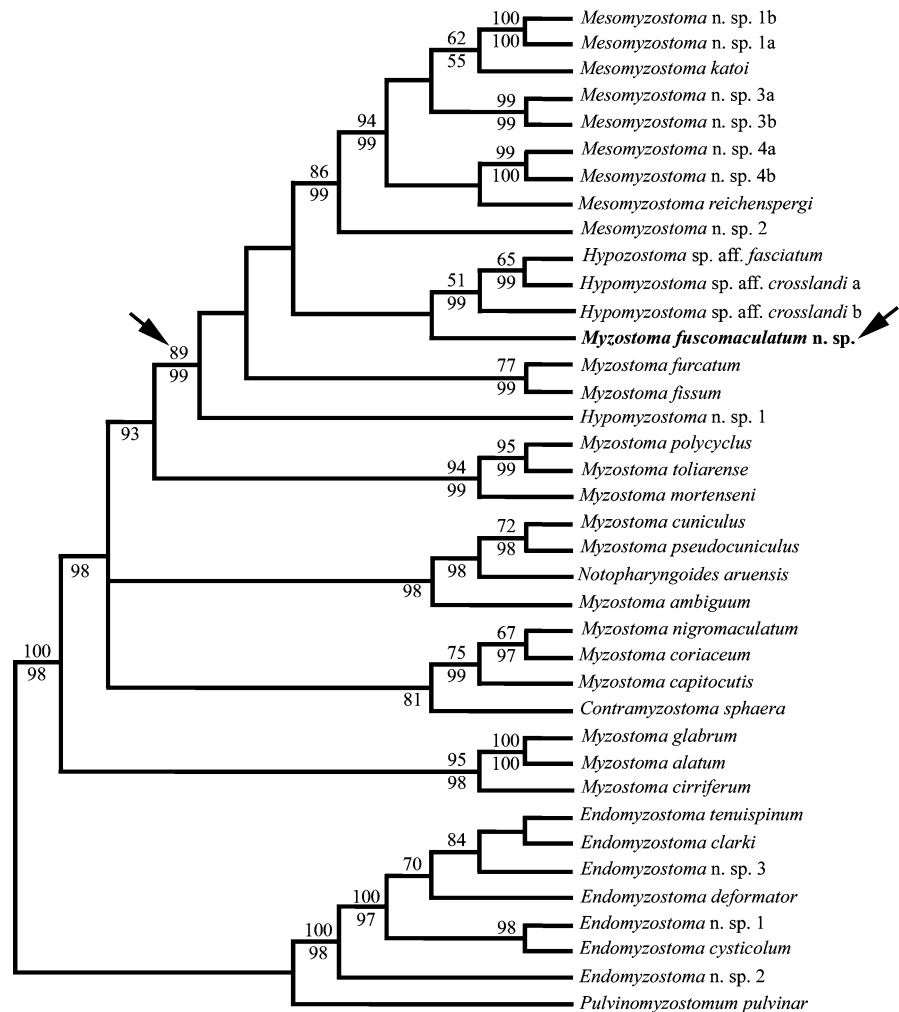


Fig. 3 Phylogenetic position of *Myzostoma fuscomaculatum* n. sp. MP consensus tree (50% majority-rule consensus tree) using the PP90 18S-16S rDNA alignment (1866 bp, three MP trees obtained, tree length = 734, CI = 0.5940, RI = 0.8177). The Bayesian analysis produced a topology almost the same as this one. Numbers above branches indicate bootstrap values >50% (1,000 replicates). Numbers below branches indicate posterior probability obtained in the Bayesian analysis. Undescribed species (*Mesomyzostoma* n. sp. 1–4, *Endomyzostoma* n. sp. 1–3 and *Hypomyzostoma* n. sp. 1) are still under study (descriptions in preparation)



to parapodia. Introvert pouch opening located half-way between first parapodia and anterior body margin. Common pore of digestive tract and female genital duct closer to body margin than to last pair of parapodia. Penes arising just outside third parapodia. Dorsal colour pattern with 4–5 pairs of lateral patches, sometimes continuous across midline. Adults simultaneous hermaphrodites. Ectocommensals, living on crinoid arms.

Description

Body robust and oval-elongate, clearly longer than wide, often curved upwards laterally. Posterior trunk without any caudal processes. Holotype ca. 3.8 mm long and 2.7 mm wide. Paratypes (IG31130/04 to IG31130/09) 2.5–6 mm long and 1.5–3 mm wide.

Marginal cirri absent in holotype and paratypes. Only some humps or notches (<10 µm long or deep) observed on margin of some specimens (Fig. 2D, E).

Introvert thin and ca. 300 µm in diameter when protruding (Fig. 2A–C). Introvert pouch opening located between margin and first pair of parapodia (Fig. 2E). No buccal papillae observed.

Five pairs of cirrate parapodia arranged in two ventral parallel rows (deep within ventral concavity), closer to midline than to body margin (Fig. 2D, E). Parapodia ca. 750 µm long, with single emergent hook (Fig. 2F) and one basal parapodial cirrus, ca. 45 µm latter long (Fig. 2H). Hook shaft thick, ca. 725 µm long, with a thick, hook-like tip curving a little less than 90° (Fig. 2I). Support rod (i.e. acicule) longer than hook (760 µm long in 4 mm long digested individual), but thinner than hook only at

its distal end (proximal end of the acicula almost as thin as the hook); manubrium large (ca. 50 μm long) and axe-shaped on one side, and ending in four or five lobes on the other side (Fig. 2I). Each parapodium containing two replacement hooks, one long and one small (ca. 450 and 125 μm long, respectively).

Four pairs of round lateral organs between parapodia and body margin, alternating with parapodia (Fig. 2E, F), closer to body margin than parapodia (Fig. 2E–G). Some organs protruded as glabrous humps ca. 70 μm diameter (Fig. 2G), others retracted and represented by slits ca. 115 μm long (Fig. 2F).

Sectioned adult of 5 mm long with digestive system consisting of a straight tube of 4,500 μm long and three pairs of caeca. Tube consisting of a pharynx included within a protrusible introvert, stomach and intestine, each consisting of one-third of the whole length of the tube. Simultaneous hermaphrodites. Common exit of digestive tract and female genital duct located closer to body margin than last pair of parapodia. One pair of protruding penises, 140 μm long, arising immediately from outer basal region of third parapodia (Fig. 2E, G). Female genital system located dorsally with a diffused ovary and genital ducts. Oogonia and oocytes observed in ovary. Female genital ducts below the ovary, following the digestive tract, made of a sagittal uterus and three pairs of genital diverticula that dichotomize above digestive diverticula. Male genital system located ventrally, below the digestive system, made of two diffuse testes and ducts. Testes made of cyst cells including developing spermatozoa. Male ducts starting from the two testes and converging towards two small seminal vesicles connected to penises. Forming spermatophores observed into seminal vesicles. Integument including bundles of collagen fibres, i.e. the 'cutis' of Graff (1877), in the dorsal part of the trunk.

Dorsal and ventral body surfaces with sparse tufts of cilia (Fig. 2H).

Colour

Colour pattern similar to that of host (Fig. 1). Light in life, with dorsal side lightly beige (Fig. 2A) to orange (Fig. 2C) with usually five pairs of dark brown to black patches (Fig. 2A–C), located along the same pair of axes as the ventral lateral organs (Fig. 2A, C). Ventral side beige to orange with darker areas corresponding to dorsal dark patches (Fig. 2B).

Host

Ectocommensal on arms and pinnules of *T. carinata*.

Phylogenetic account

Myzostoma fuscomaculatum n. sp. clusters within a clade including *Hypomyzostoma* sp. aff. *crosslandi* a and b and *Hypomyzostoma* sp. aff. *fasciatum* in the MP analysis (see Fig. 3). This clade is the sister group of the *Mesomyzostoma* (nine species) (Fig. 3). Yet, another *Hypomyzostoma*, *Hypomyzostoma* n. sp. 1, is more basal and form a monophyletic grouping with all the last cited species, plus *M. furcatum* and *M. fissum* (with 99% of Bayesian posterior probability and 89% of MP bootstrap support, Fig. 3). *Mesomyzostoma* is thus monophyletic (with 99% of Bayesian posterior probability and 86% of MP bootstrap support), but the *Hypomyzostoma* and *Myzostoma* groupings are not.

Discussion

The external anatomy of *M. fuscomaculatum* n. sp. is close to that of *M. gopalai* Subramaniam, 1938 (q.v.) collected on *T. encrinus* A.H. Clark, 1911 in Madras Harbour. Both species live at the surface of crinoids of the family Tropiometridae, and present an elongate, thick trunk and a yellowish-orange colouration with darker dorsal patches. *M. fuscomaculatum* n. sp. differs from *M. gopalai* in totally lacking marginal cirri at the adult stage: no cirrus was observed on any of the 12 examined specimens (in life and fixed in alcohol or after SEM preservation procedures), whereas *M. gopalai* has a minimum of 20 (in a 2.26 mm long specimen) and a maximum of 136 (in a 7.74 mm long specimen) well-developed cirri (Subramaniam, 1938). The two myzostome species were collected in two different sampling sites separated by almost 8,400 km, and in two different oceans (Atlantic and Indian Ocean, respectively). Yet, despite extensive researches, type specimens of *M. gopalai* or other specimens have not been located. Comparisons of these two species are thus only possible through the Subramaniam (1938) and Rao & Sowbhagyavathi (1972) descriptions. In the first article, the description and the drawing of parapodial chaetae in *M. gopalai* are not sufficiently described to

conclude if South African specimens should be included in *M. gopalai* or not. The only morphological difference between the two species is thus the presence, in *M. gopalai*, and the absence, in *M. fuscomaculatum*, of cirri in adults. Concerning the cirri, Subramaniam (1938) states the following: "...Magnesium chloride and menthol do not induce myzostomes to expand, and later preservation of animals so treated, in alcohol or formalin does not show the cirri properly. The body in such cases becomes rolled up...it appears to me that the best method is to kill the animal with Bouin...". Yet, Subramaniam (1938) gave a table indicating the length, the width and the number of cirri of the 10 examined specimens. The number of cirri was variable at 20–136. Length also varied between 8 and 215 μm . Nine of the specimens had cirri of at least 100 μm , which is huge (it is the length of the cirri of myzostomes such as the European *M. cirriferum*). At present, there is no recorded species of myzostomes where cirri are lost when individuals become adults, which suggests that South African specimens are not young individuals of *M. gopalai*. Moreover, the presence of three pairs of digestive diverticula (instead of two in *M. gopalai*), the presence of lateral organs very near the margin and the three large teeth on the support rod manubrium in *M. gopalai* (if drawn accurately in Subramaniam, 1938) are additional characters differentiating the two species. Rao & Sowbhagyavathi (1972) made some additional observations on *M. gopalai* collected on *T. encrinus* at Waltair Coast. They made sections in specimens of this species and of *M. striata*, the later collected on *Lamprometra palmata*. They observed in the dorsal part of both species bundles of collagens known as the cutis described by Graff (1877), and also observed in *M. fuscomaculatum* (present study).

Extending our comparison to other myzostomes infesting species of the same host genus, *Tropiometra*, some other myzostomes collected on *T. afra*, in Australia and Asia, are morphologically related to the present new species. Of these, the one that is the most similar to *M. gopalai* and *M. fuscomaculatum* n. sp. is *M. nasonovi* (Fedotov, 1938). The latter, originally described by Fedotov (1938) based on specimens from Japan, and redescribed by Grygier (1992) based on specimens from Hong Kong, is elongate-oval, with a convex dorsal side and a concave ventral side, transverse markings (going from light bands to dark-

bordered lozenges), numerous tiny marginal cirri (not always clearly evident on all investigated specimens), parapodia in two parallel rows (deep within ventral concavity), and parapodial cirri. Grygier (1992) already compared *M. nasonovi* to *M. gopalai* and even suggested a possible synonymy of the two. However, the position of the lateral organs very near the margin and the three large teeth present on the support rod manubrium of *M. gopalai* (characters that may also differentiate *M. gopalai* from *M. fuscomaculatum* n. sp.) are different from *M. nasonovi*. The absence of marginal cirri and the presence of two replacement hooks in *M. fuscomaculatum* parapodia are features that are not observed in *M. nasonovi*, which presents numerous tiny cirri and parapodia with only one replacement hook. Another species, *M. bocki* (Jägersten, 1937), is a very prevalent myzostome on *T. afra*, and similar to *M. nasonovi* (synonymy investigated by Grygier, 1992). However, *M. bocki* does not have dorsal transverse markings and possesses 10 pairs of marginal cirri (Grygier, 1992).

Six subgroups (*gigas*, *crosslandi*, *platypus*, *costatum*, *ambiguum* and *wyvillethomsoni*) have been informally described (Grygier, 1990, 1992) and the new species, *M. fuscomaculatum* n. sp., clearly does not fall perfectly within any of them. *M. fuscomaculatum* n. sp. exhibits the characteristics of the *crosslandi* subgroup, as follows. They live extended lengthwise along the arms and pinnules of crinoids, have an oval-elongate trunk with a convex dorsal side and a concave ventral side, the parapodia lie at equal intervals in two parallel rows (the first pair being closer to the anterior margin than the fifth is to the posterior margin), lack proper cirri (some *crosslandi* subgroup species have rounded projections or scallops on the margin), have transverse dorsal colour bands (some *crosslandi* subgroup species also have stripes or ridges), and possess basal parapodial cirri. *M. fuscomaculatum* n. sp. does not have, as it is the case in the *crosslandi* subgroup, the villose pad and the longitudinal groove on the parapodia and the first pair of parapodia are not closer to the anterior end than the fifth is to the rear end.

M. fuscomaculatum n. sp. clusters with nine species of *Mesomyzostoma*, four species of *Hypomyzostoma* and two species of *Myzostoma*. The *Mesomyzostoma* genus is monophyletic. *M. fuscomaculatum* n. sp. clusters with three of the four *Hypomyzostoma*, the *Hypomyzostoma* genus being paraphyletic due to the position of *Hypomyzostoma* n. sp. 1. The two

Myzostoma species (*M. fissum* and *M. furcatum*) included in the clade belong to the *costatum* group (Grygier, 1990) that includes species with dorsal ridges and, often, caudal appendages. The phylogenetic analyses support the view that species of the *costatum* group, whose caudal appendages mimic pinnules of the hosts, and *Mesomyzostoma* species, that parasite arms and pinnules, develop from *Hypomyzostoma* species, where colour patterns mimic the host's arms and pinnules. If a phylogenetic systematics of the Myzostomida is considered in the future, authors would have to take into consideration that *Hypomyzostoma* may not be a valid taxon, but actually lies within the clade including all the *Mesomyzostoma* and some *Myzostoma*.

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