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New combinations of two *Pseudobranchiomma* species (Annelida: Sabellidae): The difficulty in establishing a boundary layer among genera within the Sabellinae

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Abstract

Due to the presence of companion chaetae on the thoracic notopodia, a feature unnoticed on the material of the original description, the species *Pseudobranchiomma tarantoensis* (Knight-Jones & Giangrande, 2003) and *P. marmarensis* (Çinar & Giangrande, 2018) are here transferred to different genera; the former to the genus *Sabella* Linnaeus, 1767 and the latter to *Bispira* Krøyer, 1856. A discussion on the boundary existing among the genera belonging to the clade Sabellinae is made, underlying the importance of the thoracic uncini shape, which shows consistency with species separation from a molecular point of view.

Keywords: Fan worms; West Mediterranean; new combination; companion chaetae; Bispira; Sabella.

Introduction

The genus Pseudobranchiomma Jones, 1962 is included in the Sabellinae clade sensu Tilic (2020), which seems to be a natural group well identifiable using morphological and molecular features (Capa, 2008). In the analysis by Tilic et al. (2020), the two groups are interconnected, the first one containing the genera Hypsicomus Grube, 1870 and Anamobaea Krøyer, 1856, and the second including the genera Pseudobranchiomma, Bispira Krøyer, 1856, Branchiomma Kølliker, 1858, Sabellastarte Krøyer, 1856, Sabella Linnaeus, 1767, and Stylomma Knight-Jones, 1997. This second clade is mainly characterized by the possession of interramal eyespots in the thorax and abdomen; having inferior thoracic chaetae arranged in longitudinal bundles; and with abdominal neuropodia as conical lobes and neurochaetae arranged in "tuft" fascicles (Fitzhugh, 1989; Rouse & Fitzhugh, 1994; Fitzhugh & Rouse, 1999; Capa, 2008). However, genera within this group still need to be better defined (Capa, 2008; Capa & Murray, 2016). The genus Pseudobranchiomma lacks companion chaetae in the thorax, which occurs in the genera Branchiomma and Sabellastarte. Among these three genera, Branchiomma seems to be well-defined by the presence of stylodes in the radioles, while Pseudobranchiomma can be determined by the presence of lateral flanges as small serrations on the radioles; however, these are not present in all the species, and by contrast, are present in *Bispira serrata* Capa, 2007 (Capa, 2008).

Bispira is one of the most problematic genera, including a heterogeneous species group not pooled by any synapomorphy. Therefore, its separation from Sabella is also difficult. In some previous analyses, Bispira was considered as the sister group of Sabella, based on the presence of companion chaetae with an asymmetrical distal membrane (Fitzhugh, 1989; Rouse & Fitzhugh, 1994; Fitzhugh, 2003), or presented as an unresolved polytomy (Fitzhugh & Rouse, 1999). In this respect, however, the shape of companion chaetae appears in different forms within the genus Bispira. A great heterogeneity is also reported in the genus Pseudobranchiomma (Nogueira & Knight-Jones, 2002; Knight-Jones & Giangrande, 2003; Tovar-Hernández & Dean, 2014; Capa & Murray, 2016), which species in the past were classified into three artificial groups, based on the absence or presence and relative length of the serrated radiolar flanges (Knight-Jones & Giangrande, 2003): Group A, with radiolar serrations evenly distributed along the entire length of the radioles, or at least for most of their length; Group B, with serrations restricted to the distal part of the radioles; and Group C with smooth radiolar flanges.

During the re-examination of material present in the collection of the Museum Parenzan (University of Salento, Lecce, Italy), the specimens of *Pseudobranchiomma tarantoensis*, which Knight-Jones & Giangrande (2003) described initially as from the Ionian Sea, and those of *P. marmarensis* which Çinar & Giangrande (2018) originally described as from the Sea of Marmara, were found to possess companion chaetae, and so were excluded from their present systematic position. However, the presence of companion chaetae was already detected in more recently collected specimens of *P. marmarensis* from a western Mediterranean site (North Corsica). This led to re-examining our material attributed to both *P. marmarensis* and *P. tarantoensis*, unveiling the necessity to transfer them into different genera from what they originally described. Both species were considered at the time of description as belonging to the *Pseudobranchiomma* group C sensu (Knight-Jones & Giangrande, 2003), increasing the variability already existing within this genus.

Material and Methods

Examined material comes from that already present in the collection of the Museum Parenzan Zoological Laboratory, University of Salento (Lecce, Italy) (Giangrande *et al.*, 2015) and from additional specimens more recently collected.

Two paratypes of *P. tarantoensis* were collected from the Gulf of Taranto (southern Italy), in July 1993, in a sand-muddy substrate, at a depth of 10 m, where the presence of the metallurgiccomplex "ILVA" has an important impact in term of pollution (Knight-Jones & Giangrande, 2003).

Specimens of *P. marmarensis* come from several areas: the Sea of Marmara (type locality of the species), where paratypes were collected between depths of 50 and 100 m, on sandy mud bottom with shell fragments, in June 2013 (Çinar & Giangrande, 2018); from Corsica, off the coast of Rogliano (the Tyrrhenian Sea, western Mediterranean Sea [coordinates: 43.019785°E; 9.428421°N]), specimens were collected during a survey conducted under the Water Framework Directive (WFD) of 2018, during the WFD campaign, financed by the Agence de l'Eau Rhône Méditerranée Corse (Delmas, 2018; Pelaprat *et al.*, 2018) 47 m depth on fine sediments in March 2018; from Elba Island (Tyrrhenian Sea, western Mediterranean Sea [coordinates: 10.2144420°E; 42.5029022°N]), at 68m depth on soft bottom, muddy sediment, May 2022.

Taxonomic remarks

Sabella tarantoensis comb. nov. (Knight-Jones & Giangrande, 2003) Figs. 1 and 2 Pseudobranchiomma tarantoensis Knight-Jones & Giangrande, 2003: 100–102, Fig. 3.

Material examined: Two paratypes (PCZL S. PBR. 1.1 and PCZL S. PBR. 1.2).

Description: Complete paratype with eight thoracic chaetigers and about 80 abdominal ones (Fig. 1A). Crown whitish, with numerous small darker spots, not clearly arranged in stripes (Fig. 1A, B).

Ten to twelve fully developed radioles, with pinnules of similar length and short, blunt radiolar tips. Radiolar flanges absent. Each radiole with three to six dark spots, without any clear photoreceptive structure (Fig. 1C). Radiolar skeleton not studied. Dorsal pinnular appendages



Fig. 1: Sabella tarantoensis comb. nov. paratypes from Taranto. A. Whole paratypes, B. Crown, C. Radiolar spots, D.Peristomium in ventral view (ss: squared shields at the base of the ventral lappets; fs: first thoracic shield M-shaped).



Fig. 2: Chaetae and uncini of *Sabella tarantoensis comb. nov.* A–C. Peristomial chaetae, A. Superior chaeta, B. Intermediate chaeta, C. Inferior chaeta; D–F. Thoracic chaetae, D. Superior chaeta, E. Intermediate chaeta, F. Inferior chaeta; G and H, Abdominal chaetae, G. Inferior chaeta, H. Superior chaeta in posterior abdomen; I. Thoracic uncinus, J. Companion chaeta, K. Scheme of disposition of teeth above the main fang of uncini, L. Abdominal uncinus. Scalebars: 20 μm.



Fig. 3: Thoracic uncini and companion chaetae (when present) from different genera belonging to the Sabellinae subfamily. A. *Pseudobranchiomma* cf. *orientalis* (McIntosh, 1885), B. *Pseudobranchiomma* cf. *emersoni* Jones, 1962, C. *Pseudobranchiomma pallida* Capa & Murray, 2016, D–F. *Bispira serrata* Capa, 2007, G and H. *Bispira volutacornis* Montagu, 1804, I and J. *Bispira riccardi* Giangrande, Putignano & Enrichetti, 2022, K and L. *Sabella spallanzanii* Gmelin, 1791, M and N. *Sabella pavonina* Savigny, 1822. Scalebars: A, D–F. 4 μm, B and C. 2 μm, G–N. 20 μm. A-C. redraws from Capa & Murray, 2016 (Fig. 3, elements A, G, and M); D–F. redraws from Capa 2008 (Fig.4, elements U, V, and H2).

absent, dorsal lips with long and tapered dorsal radiolar appendages. Ventral lips not seen, parallel lamellae present, ventral radiolar appendages absent. Rounded and prominent ventral sacs, placed outside radiolar crown, appearing under ventral collar as a fold (Fig. 1D).

Peristomial collar oblique, higher ventrally, with a deep cleft forming two rounded and not- overlapping ventral lappets (Fig. 1D).

Collar margins separated dorsally by a wide gap, with dorsal margins fused to first chaetiger, lateralcollar margins smooth, not reaching the junction between crown and thorax, leaving the anterior peristomial ring uncovered.

First ventral shield of complex shape: "M-shaped", divided almost completely into two halves by a median groove, slightly extending laterally in anterior half and with two circular depressions symmetrically disposed near the anterior edge; four smaller and polygonal shields placed symmetrically and anteriorly to it, at the start of the ventral lappets (two each side) (Fig. 1D).

Peristomial eyes absent; interramal eyespots and pygidial eyes present.

Peristomium with elongated, narrowly hooded superior chaetae and hooded inferior chaetae, disposed ventrally to superior ones (Fig. 2A-C); under superior chaetae, some chaetae of intermediate shape are present, being more elongated than proper inferior ones; they are referred to as "intermediate chaetae" (Fig. 2B). Thoracic superior chaetae elongated and narrowly hooded, intermediate chaetae present, inferior broadly hooded chaetae shorter, arranged in a tuft (Fig. 2D-F). Thoracic uncini with a well-developed and high breast, a really short handle, and numerous rows of teeth above the main fang, all similar in size; when seen frontally, they show a tessellated disposition of teeth (Fig. 2I-K). Companion chaetae, hard to see, were firmly embedded into the integument, present with an asymmetrical elongated blade developing from a bulbous head (Fig. 2J). Abdominal chaetae broadly hooded, superior abdominal chaetae thin, elongated, and extremely narrowly hooded; superior chaetae were only present in posterior abdominal segments (Fig. 2G-H). Abdominal uncini as the thoracic uncini, showing aprogressively higher crest towards posterior abdominal segments (Fig. 2L). Pygidium bilobed

Remarks. Although poor preservation of material did not allow a good observation of many features, most of them strongly suggested that this species, previously placed under the *Pseudobranchiomma* genus, indeed belongs to the genus *Sabella*. In particular, the presence of companion chaetae and the shape of thoracic uncini, the shape of the first thoracic shield, the absence of radiolar flanges and pinnular appendages, and the absence of radiolar eyes in *P. tarantoensis* are typical of the genus *Sabella*.

Knight-Jones & Perkins (1998) had revised most of the species belonging to *Sabella* by that time, transferring them to *Bispira*. The main differences between the two genera are the arrangement of abdominal chaetae, the presence of pinnular appendages, and the shape of the first ventral shield. In this way, only three species remained in *Sabella*: *S. pavonina* Savigny, 1822 (the type species), S. spallanzanii Gmelin, 1791, and S. discifera Grube, 1874. These species have thoracic uncini with numerous small teeth above the main fang, all similar in size and not clearly disposed in rows (Fig 3L, N). This shape of uncini differs from those present in the other genera of the Sabellinae subfamily(Fig. 3), especially in the Bispira species, which also represent different types within this groupe. The abdominal chaetal arrangement in our specimens was similar to that in Bispira (C-shaped). However, this feature was not discernable in the small-sized specimens, as also in Sabella; they show this shape during the formation of fascicles in the posterior segments to develop a spiral arrangement. The superior abdominal chaetae are absent in most abdominal fascicles and present only in the posterior and youngest segments. This is consistent with the genus Sabella, which loses its superior abdominal chaetae soon after the formation of fascicles in the posterior segments near the pygidium (Knight-Jones et al., 1991; Knight-Jones & Perkins, 1998). As already pointed out by Knight-Jones & Giangrande (2003), in S. tarantoensis the inferior abdominal chaetae are too few in number to form a spiral arrangement, as seen in the other species of the genus Sabella. The species seems to lack distinctive radiolar photoreceptive structures, as reported for the other Sabella species (except for S. pavonina). The absence of radiolar flanges and pinnularappendages are common features present in Sabella. However, in poorly preserved specimens, the radiolar flanges and skeleton cannot be easily spotted; the original description reported the absence of flanges but the presence of "longitudinal distal ridges." Lastly, the peristomial collar is consistent with the Sabella genus, showing well-developed and rounded ventral lappets, a wide dorsal gap, and fused to the posterior edge of the peristomium but lacking the mid-ventral cleft. In addition, the peristomial ventral shield appears different from the other species of the genus Sabella, being more complex and morphologically closer to *Bispira* (i.e., the presence of a smaller shield anterior to the main one and the presence of depressions in the latter).

Ecology. The species was first found on muddy substrata in Taranto harbor, characterized byenriched organic content and high level of pollution.

> Bispira marmarensis comb. nov. (Çinar & Giangrande, 2018) Figs. 4 and 5

Pseudobranchiomma marmarensis Çinar & Giangrande, 2018: 1564–1568, Figs. 2–5.

Material examined. Additional material from Elba Island (seven specimens) (PCZL S.B. 10.1) and Corsica Island (20 specimens).

Description: All specimens with 8 thoracic chaetigers and about 40 abdominal chaetigers (Fig. 4A).Crown whitish in preserved specimens, with darker spots, clearly organized in only one basal stripe (Fig. 4A–B).

Twelve to fourteen fully developed radioles, with pinnules of similar length and extremely elongated, tapering radiolar tips. Radiolar flanges present without serrations.



Fig. 4: Bispira marmarensis comb. nov. specimen from Elba Island. A. Whole specimen, B. Crown and anterior chaetigers, C. Dorsal radiolar appendage, D. Radiolar compound eyes and radiolar flanges, E. Ventral peristomium with ventral lappets and ventral sacs (photographs of one specimen from Elba's Island).



Fig. 5: Chaetae and uncini of *Bispira marmarensis comb. nov.* A-C. Peristomial chaetae; A. Superior chaeta, B. Intermediate chaeta, C. Inferior chaeta; D–F Thoracic chaetae, D. Superior chaeta, E. Intermediate chaeta, F. Inferior chaeta; G–I, Abdominal chaetae; G and H. Inferior chaetae, I. Posterior superior chaeta; J. Thoracic uncinus, K. Companion chaeta, L. Abdominal uncinus; M–O, Radiolar skeleton, M. Section at the base of the radiole, N. Section medial to the radiole, O. Section with radiolar compound eyes. Scalebars: 20 μm (A–L) and 0.05 mm (M–O).

Each radioles with 2 to 4 pairs of purple compound eyes (Fig. 4D). Radiolar skeleton composed of 4 vacuolated cells, disposed in a square (Fig. 5 M–O). Dorsal lips pointed, with one pair of long, tapering dorsal radiolar appendages (Fig. 4C). Ventral lips rounded and parallel lamellae present, with one pair of ventral radiolar appendages. Highly developed and pointed prominent ventral sacs, placed outside the radiolar crown, appearing under the ventral collar as a protuberance (Fig. 4E).

Peristomial collar oblique, higher ventrally with a deep cleft forming two triangular ventrals, pointed and not-overlapping lappets (Fig. 4E). Collar margins separated dorsally by a wide gap, with dorsal margins fused to the first chaetiger. Lateral collar margins smooth, not reaching the junction of the crown and thorax, leaving the anterior peristomial ring uncovered. First ventral shield "W-shaped", with two smaller, circular shields placed anteriorly to it.

Peristomial eyes present, interramal eyespots and pygidial eyes present.

Peristomium with elongated, narrowly hooded superior chaetae and two rows of (broader) hooded inferior chaetae, disposed ventrally to the superior ones (Fig. 5A-C); the middle row show chaetae of intermediate shape, being more elongated than inferior ones; they are referred to as "intermediate chaetae" (Fig. 5B, E). Thoracic superior chaetae elongated and narrowly hooded chaetae, the ventral-most ones with a longer hood, similar to peristomial intermediate chaetae; inferior broadly hooded chaetae shorter, arranged in a tuft (Fig. 5D-F). Thoracic uncini with a rounded breast, short handle, and three rows of teeth above the main fang, with a marked increase in the size of teeth from apical row to basal one; when seen frontally, the teeth were disposed in ordinated rows (Fig. 5J). Companion chaetae, hardto see, firmly embedded into the integument, present with a symmetrical elongated hood with a long, tapering mucro arising on its right side (Fig. 5K). Abdominal broadly hooded chaetae with thin, elongated, and extremely narrowly hooded superior chaetae appearing only in the posterior abdominal segments (Fig. 5G-I). Abdominal uncini as the thoracic, showing a progressively higher crest moving toward posterior abdominal segments (Fig. 5L).

Pygidium bilobed.

Remarks. All morphological features of the new material from Corsica and Elba, Bispira marmarensis comb. nov., are similar to those of the material from the type locality, except for the presence of a lesser number of compound eyes in each radiole (two to four pairs, usually three pairs). A peculiarity of B. marmarensis comb. nov. is the presence of pointed ventral sacs, a unique feature among sabellids. The presence and the shape of companion chaetae on the thoracic notopodia (as well as the presence of pinnular appendages), the shape of the peristomial ventral shields ("W-shaped"), and the presence of paired compound eyes on radioles are consistent with the *Bispira* genus, leading us to transfer the species from Pseudobranchiomma to Bispira. In addition, the shape of the latero-posterior peristomial collar and the shape of the companion chaetae are consistent with those of *Bispira* viola (Grube, 1863). Bispira marmarensis comb. nov. differs from most of the other species of Bispira, especially in the shape of their thoracic and abdominal uncini, which are short-handled and have 2–3 rows of big teeth above the main fang (Fig. 5J, L), instead of having medium-sized handles and small, numerous rows of teeth over the main fang (Fig. 3H, J) This feature is more similar to some *Pseudobranchiomma* species (Fig. 3C) than *Bispira*. The only *Bispira* species with a similar uncinal shape is *B. serrata* (Fig. 3E, F), which differs from *B. marmarensis comb. nov.* in having serrated radiolar flanges (Capa, 2008). Interestingly, the serrated flanges and the uncinal shape make *B. serrata* very close to *Pseudobranchiomma*.

This seems to be consistent with results from molecular analysis, where B. serrata, as well as B. manicata and B. porifera are always separated from the other examined Bispira (see Enrichetti et al., 2022); there are some features common to both Sabella and Bispira, especially with regard to the shape of the thoracic uncini, which are typically composed of numerous rows of teeth, all similar in size. The need for a deep revision of the whole Sabellinae subfamily is firmly shared by taxonomists worldwide, as well as the necessity to find informative morphological characters for itstaxonomy. We noted that features of hard structures better follow the separation from themolecular point of view. Within the Bispira genus, for example, the shape of companion chaetae appears highly variable among the species, as well as the shape of thoracic uncini, and their distribution is in accordance with the separation of some taxa from molecular results.

Ecology. Twenty specimens were collected in coarse sand with shell fragments at a depth of 47 m. with a low percentage of clay particles (1%). The sediment type corresponds to what is generally indicated as coastal detritus (PNUE-PAM-CAR/ASP, 2007). This habitat, widely distributed around Corsica, is also present on an important part of the eastern facade of the French coast of the Mediterranean Sea (Pelaprat *et al.*, 2018) and is dominated by species with a wide ecological distribution, and other annelidspecies with a preference for coarse or fine sands such as *Aponuphis bilineata* Baird 1870; *Lumbrineris latreilli* Audouin and Milne Edwards 1833; *Galathowenia* sp., *Syllis gerlachi* Hartmann-Schröder 1960; *Aricidea* (*Acmira*) *cerrutii* Laubier, 1966; and *Gyptis propinqua* Marion & Bobretzky 1875 (Pelaprat *et al.*, 2018).

Final Remarks

The transfer of *P. tarantoensis* and *P. marmarensis* to the *Sabella* and *Bispira* genera, respectively, is based on characters that univocally claim their unsuitability for placement in the *Pseudobranchiomma* genus. However, as previously stated, some common characters suggest a more complex situation inside the Sabellinae subfamily, challenging the definition of the accepted genera. Although a deep morphological analysis is required for correct taxonomical evaluation, a complete and coupled approach between molecular and morphological analysis

is needed to re-arrange the taxonomical structure within the Sabellinae clade to establish a boundary layer among genera.

Lastly, we wish to give some recommendations. Rougher dissection likely makes it difficult to see companion chaetae, and it is possible that what we experienced has also happened during the examination of other taxa. Therefore, we call for paying close attention to examining this character; in this case, the use of scanning electron microscopy analysis should be paramount.

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