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THE MORPHOLOGY OF *TRIPOLYDORA SPINOSA* WOODWICK (POLYCHAETA: SPIONIDAE): AN APPLICATION OF THE SCANNING ELECTRON MICROSCOPE TO POLYCHAETE SYSTEMATICS

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Abstract.—The morphology of *Tripolydora spinosa* Woodwick is reexamined using traditional light microscopy and the Scanning Electron Microscope (SEM). In addition to its already-known unique features, *T. spinosa* possesses modified spines on setiger 4, bristles on the major spines of setiger 5, unique "ribbed" notosetae on setigers 7–10, and neuropodial inferior sabre setae which accompany the hooded hooks. The setal arrangements, as revealed by SEM, permit a consideration of possible setal homologies of the modified spines of setigers 4 and 5. The position of *Tripolydora* among those genera having a modified fifth setiger is considered, relative to other genera of the subfamily Spioninae.

Tripolydora spinosa was first described from rocky intertidal habitats on Enewetak Atoll in the Marshall Islands by Woodwick (1964). Kohn and Lloyd (1964) subsequently reported the species from Easter Island where it occurred in rocky tidal pools with sand and algae.

In the original description, Woodwick (1964) found *T. spinosa* to possess an unusual combination of characteristics including a partially modified fifth setiger and branchiae on anterior setigers including the modified fifth. The species also has a characteristic unique for a member of the *Polydora*-complex, the presence of tridentate hooded hooks.

Because of this unusual combination of characteristics and the possible significance of *T. spinosa* in the evolution of the *Polydora*-complex (Blake, 1979), the species was selected to be the subject of a more complete morphological analysis. A detailed study of the morphology of *T. spinosa* using both the light and Scanning Electron Microscope (SEM) was initiated. For this work, it was possible to reexamine the Enewetak and Easter Island specimens, all of which are deposited in the National Museum of Natural History (USNM). In addition, new materials from Aitutaki in the Cook Islands, collected by the first author (JAB), Johnston Atoll, provided by the Naval Oceanographic Sorting Center (NOSC) in Hawaii, and from the Seychelles (Indian Ocean), deposited in the USNM were also examined.

The species is redescribed. A discussion considers the homologies of setal arrangements in forms having a modified fifth setiger and possible evolu-

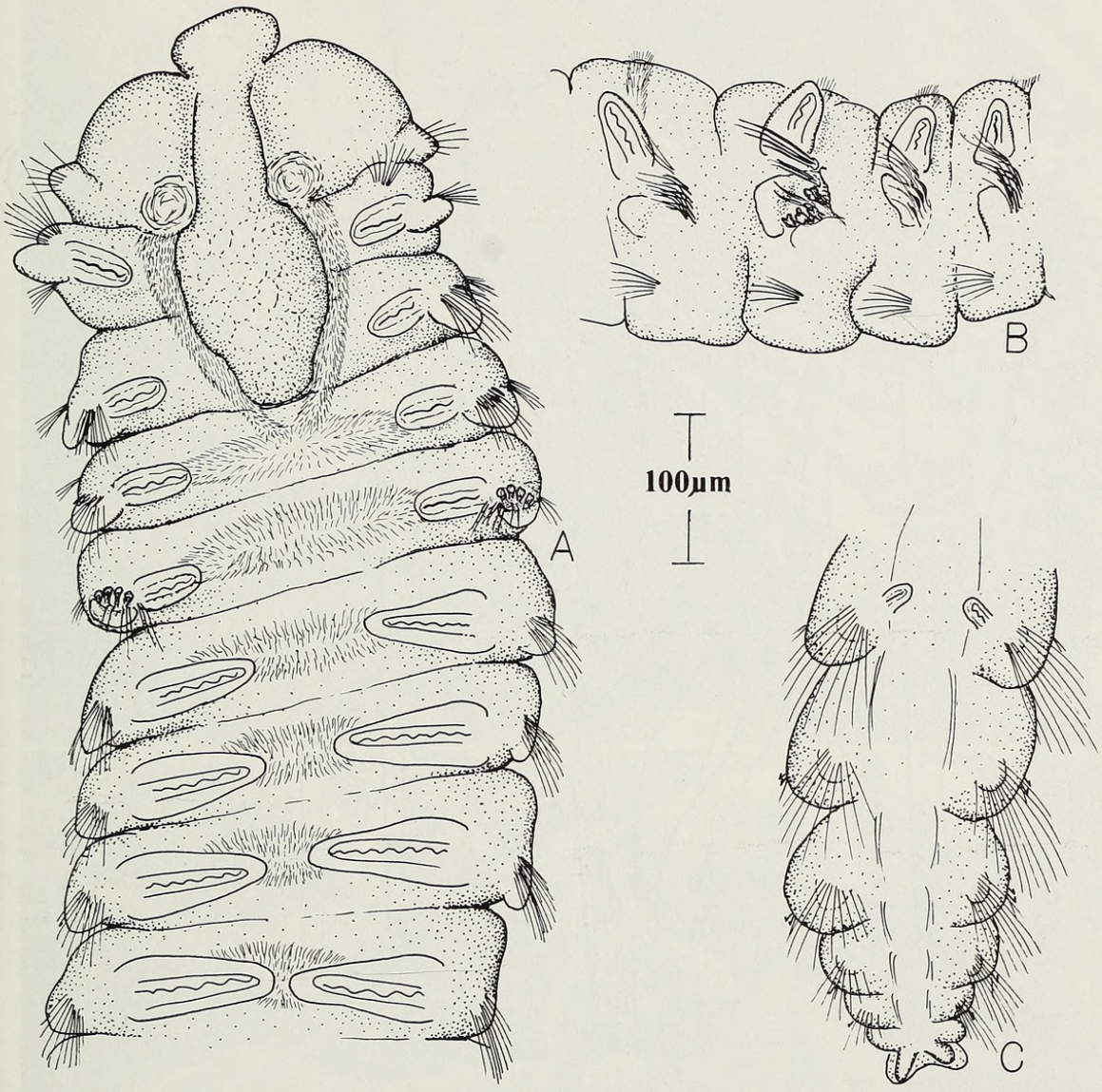

Fig. 1. *Tripolydora spinosa* (USNM 49530): A, Anterior end in dorsal view; B, Setigers 3–6 in right lateral view; C, Posterior end in dorsal view.

tionary significance of *T. spinosa* relative to its placement in the *Polydora*-complex of the subfamily Spioninae.

Methods

Nomarski Differential Interference Contrast (Zeiss RA) and Phase Contrast (Zeiss GFL) optics were used for light microscopy. SEM studies were made with a Zeiss Novascan 30 and JOEL JSM 35. SEM photographs were produced from negatives of Polaroid Type 55 film. The specimens were all from previously fixed museum specimens which are ordinarily fixed in formalin and preserved in 70% Ethanol. The specimens were dehydrated for SEM observation in a critical point drier and sputter coated with gold-palladium.

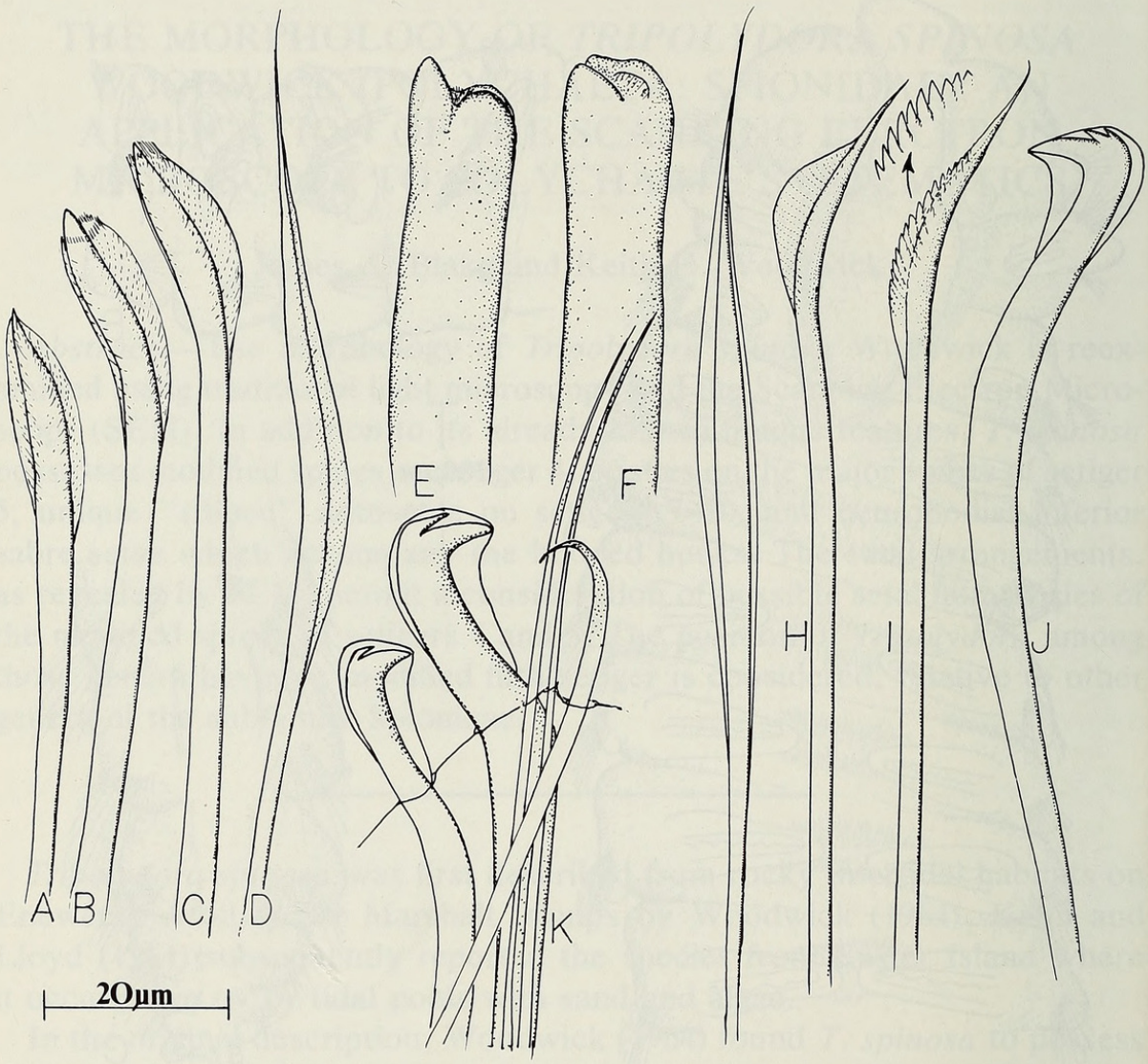

Fig. 2. *Tripolydora spinosa* (USNM 49530): A–C, Modified setae from setiger 4; D, Second tier capillary notoseta from setiger 3; E–F, Major spines from setiger 5; G, Companion bilimbate capillary setae from setiger 5; H, Anterior tier capillary notoseta from setiger 7; I, Posterior tier "ribbed" notoseta from setiger 7; J, Tridentate hooded hook; K, Fascicle of two bidentate hooded hooks, two capillary neurosetae and an inferior sabre seta.

Systematics

Tripolydora spinosa Woodwick, 1964
Figs. 1–5

Tripolydora spinosa Woodwick, 1964:155–156, fig. 4(6–9).—Reish, 1968:222.—Kohn and Lloyd, 1973:700.

Material examined.—MARSHALL ISLANDS: Enewetak Atoll, Bogambo, 2 July 1957, in beach rock, coll. D. J. Reish, holotype (USNM 32610).—EASTER ISLAND: between Hanga Roa and Hanga Piko, 15 Feb. 1969, in

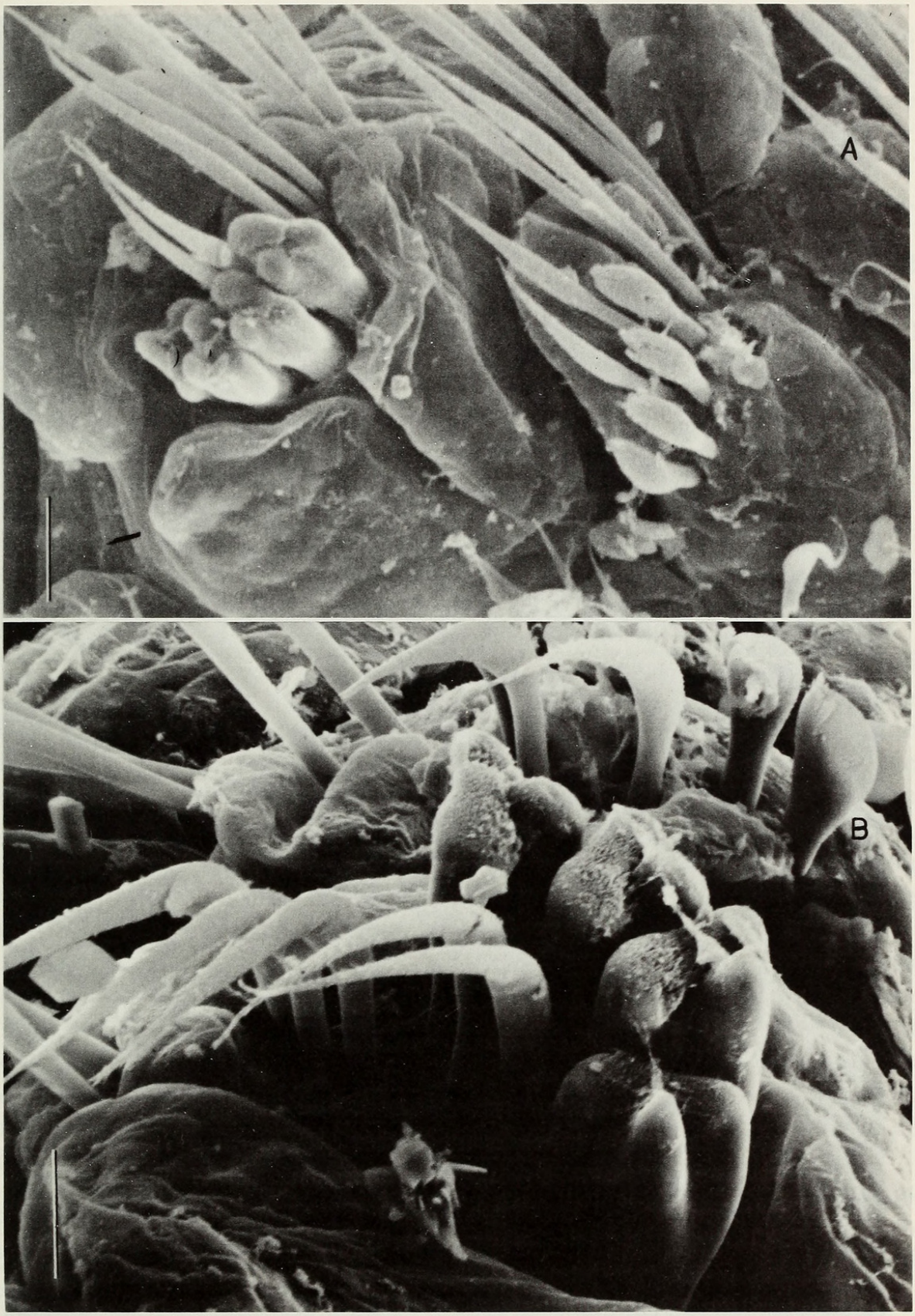

Fig. 3. *Tripolydora spinosa* (USNM 49530): A, SEM of notopodia of setigers 4 and 5 in right lateral location; B, SEM of same in posterior view of setiger 5. Scale: Fig. 3A = 20 μm; 3B = 10 μm.

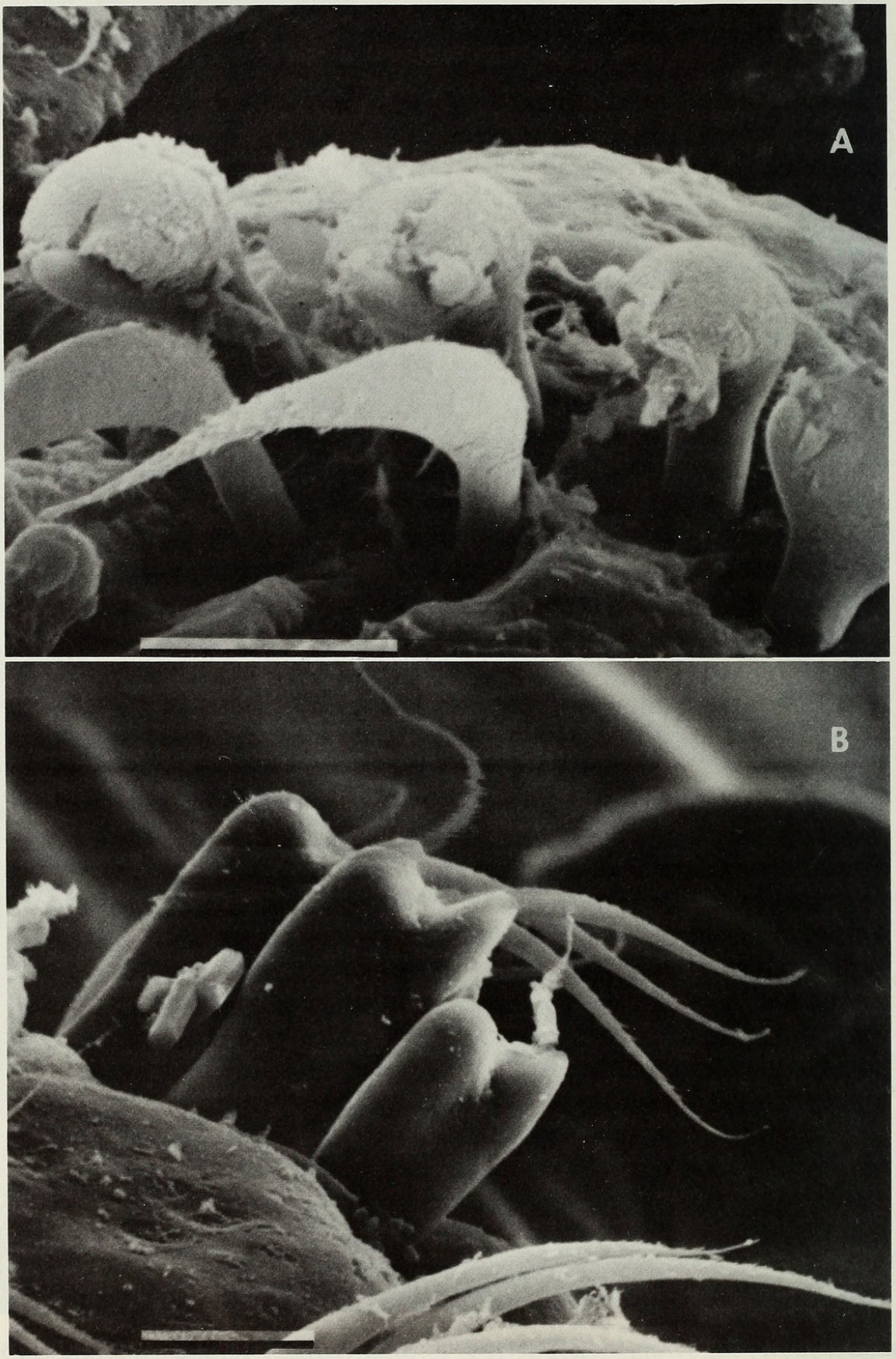

Fig. 4. *Tripolydora spinosa* (USNM 49530): A, SEM of modified notosetae (row I) and capillaries (row II) from setiger 4 in posterior view; B, Major spines of setiger 5 in lateral orientation. Scale: Fig. 4A = 10 μm; 4B = 10 μm.

rock in tide pool, coll. A. Kohn and M. Lloyd, 30 specimens (USNM 49537); same locality, in sand and algae, 125 specimens (USNM 49530).—COOK ISLANDS: Aitutaki, Akiami motu, south side, 1 Aug. 1978, boring into dead coral, coll. J. A. Blake, 3 specimens (JAB).—JOHNSTON ATOLL: Sta. 3, transect 6, 25 April 1976, coll. R. Brock, 11 specimens (NOSC 77003JI).—SEYCHELLES: Round Island, 19°45′S; 57°50′E, intertidal, from sponge, Dec. 1975, coll. K. Buchanan, 3 specimens (USNM 59926).

Description.—This is a small species, measuring up to 3.5 mm long and 0.4 mm wide for about 37 setigerous segments. In alcohol the color is opaque white to light tan with no body pigment. The prostomium is entire and broadly rounded on the anterior margin (Fig. 1A). The caruncle is broad and extends posteriorly over setigers 2 and 3 (Fig. 1A). There is no occipital tentacle and no eyes. Nuchal cilia occur in long tracts on both sides of the caruncle, merging posteriorly with the first dorsal sense organ on setiger 4. The peristomium is well developed and broad, but not inflated.

Setiger 1 is greatly reduced and lacks a notopodium and notosetae. The neuropodium is short, conical, and bears delicate capillary setae (Fig. 1A). Setigers 2–10 have elliptically shaped notopodial lamellae and indistinct neuropodial lamellae.

Setigers 2–10 bear notopodial fascicles of setae arranged in 2 rows (I and II), each running in a general dorso-ventral orientation, and a third group (III) of long superior capillaries located at a point near the uppermost setae of the second row. The setae vary in length and structure. The setae of the anterior row (I) are shortest, while those of the superior group (III) are the longest. The setae of setigers 2 and 10 are the least modified in the sequence, while those of setigers 4 and 5 are the most modified. There is a gradation of setal structure in setigers 3 and 4 leading to setiger 5 and a corresponding gradation in setigers 6–10 leading away from setiger 5.

The notosetae of setigers 2–3 and 6 are unilimbate capillaries (Fig. 2D) in which the setae of row I are shorter and have a broader sheath. In setigers 3 and 6 these setae are pennoned, with a long tapered tip. The enlarged portion of these setae is seen by SEM to be composed of free surface fibrils which are longest on one side of the seta. Varied lengths of these fibrils produce a tapered tip, which superficially resembles the acuminate setae of sabellids.

The notosetae of setiger 4 are arranged as on setigers 2 and 3 except that row I is modified and bears 3–4 setae having inflated sheaths, through which the tip of the shaft often protrudes (Figs. 2A–C; 3A–B; 4A). When observed with the SEM, the inflated sheath is seen to be a crest of bristles (Fig. 4A).

Setiger 5 appears only slightly modified superficially (Fig. 1A–B). The setae include a diagonal row of 3–4 major spines corresponding to row I and positioned anteroventral to 2 groups of 2–3 bilimbate companion capillaries (Fig. 2G) derived from row II, and a fascicle of dorsal capillaries (Fig. 3A–

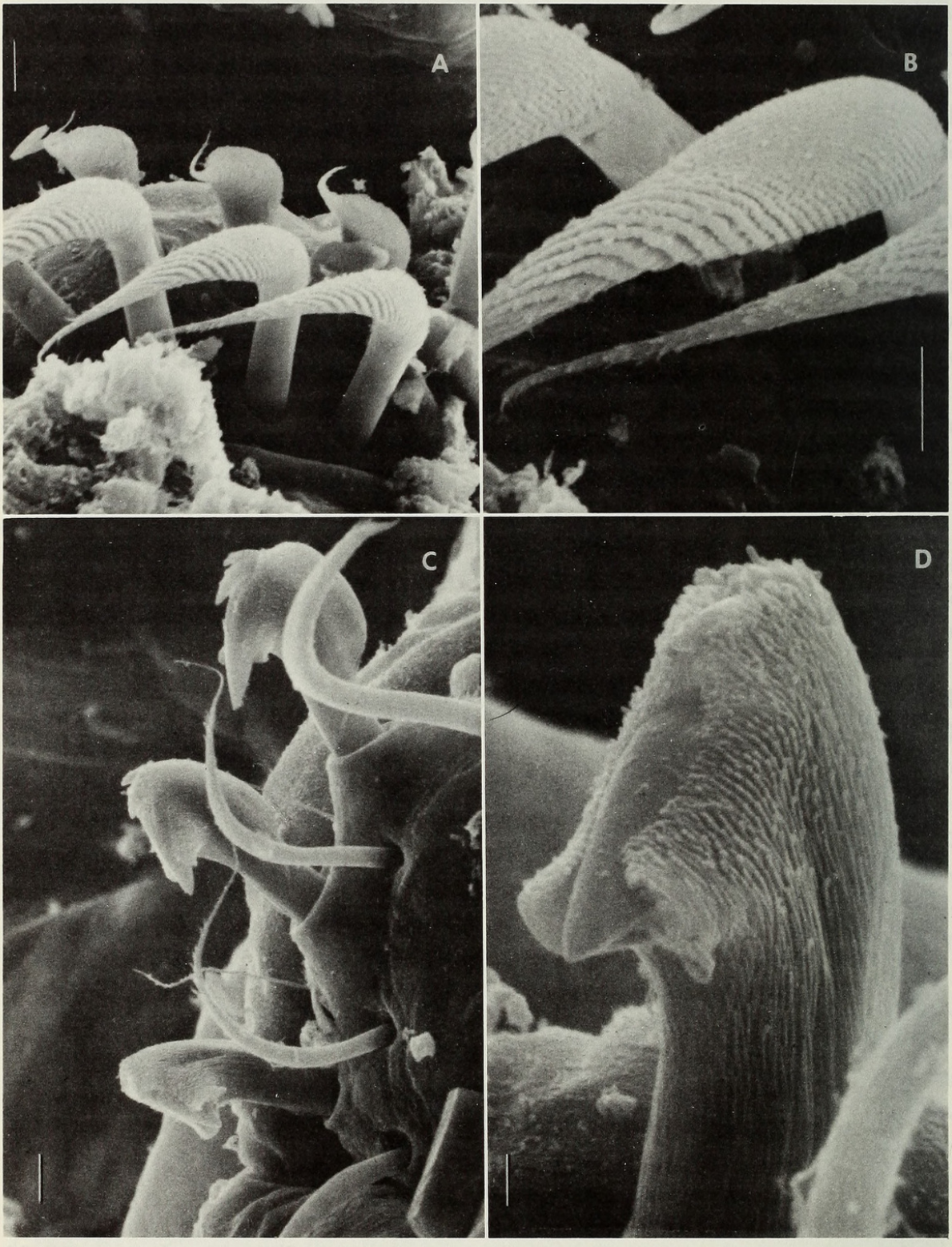

Fig. 5. *Tripolydora spinosa* (USNM 49530): A, SEM of "ribbed" row II capillary notosetae from setiger 7 in posterior view; B, Detail of same; C, SEM of tridentate hooded hooks in lateral view; D, Detail of individual hook in frontal orientation. Scale: Fig. 5A = 4 μm; 5B = 5 μm; 5D = 1 μm.

B) corresponding to row III. The major spines have a terminal tooth flanked on one side by 2 short knobs; the central depression between these prominences bears fine bristles (Fig. 2E; 3B; 4B). Worn spines have reduced knobs (Fig. 2F).

Setigers 7–10 have short unilimbate capillary notosetae in row I (Fig. 2H) similar to those of setigers 2–3 and 6 and long unilimbate capillaries with unusual riblike sculptured sheaths (Fig. 2I) in row II. The SEM reveals these ribs to consist of thickened whorls of minute bristle endings (Fig. 5A–B). The notosetae become longer and more slender in subsequent setigers, with those of the far posterior setigers being arranged as dense bundles of long needles which project posteriorly over the dorsum (Fig. 1C).

The neurosetae of setigers 2–8 consist of 4–5 unilimbate capillaries. Tridentate hooded hooks begin on setiger 9 and number 2–4 per neuropodium throughout the body. Initially, the hooks are accompanied by 4–5 unilimbate capillaries, but these are gradually replaced by 2–3 simple capillaries and a single large inferior sabre seta (Fig. 2K) bearing granulations near the distal end. The hooks have a large and a small apical tooth surmounting the main fang, to which the large apical tooth is closely applied (Fig. 2J). The small apical tooth is often worn and sometimes lacking, especially in posterior setigers (Fig. 2K). With the SEM, the hood is seen to have a fine microstructure, consisting of numerous minute bristle endings (Fig. 5C–D). In some cases, the hood is inflated (Fig. 5C). The hood opening is expanded near the tip of the main fang and flares somewhat laterally (Fig. 5D).

Branchiae occur from setiger 2, and continue without interruption to near the posterior end. The branchiae on setigers 2–5 are shorter than those of setiger 6 and subsequent setigers (Fig. 1A). Dorsal sense organs begin on setiger 4 and extend across the segments between the gills (Fig. 1A). The dorsal sense organs are difficult to see with light microscopy, but prominent with SEM. The pygidium is formed by 4 small lobes, the dorsal pair smaller (Fig. 1C).

Distribution.—Pacific Ocean: Marshall Islands, Johnston Atoll, Cook Islands, Easter Island.—Indian Ocean: Seychelles.

Discussion

The SEM reveals a significant number of details about setal structure and ciliary arrangement of *Tripolydora spinosa* that are not apparent with the light microscope. The SEM also demonstrates that it is possible to interpret setal homologies by considering setal arrangements over successive segments.

The arrangement of modified spines and capillaries on setigers 4 and 5 is clearly revealed in Fig. 3A. The modified spines of setiger 4 appear to be

homologous to the shorter and thicker unilimbate capillaries occurring in row I of setigers 2–3 and 6–10. The same homology holds for the major spines of setiger 5. Here the 3–4 spines are aligned in a more anteroventral position. The long, thin capillaries of row II are also shifted on setigers 4 and 5. On setiger 4, the 2–3 ventralmost and shortest capillaries lie just posterior to the modified setae. This same relationship occurs on setiger 5, with the ventralmost capillaries becoming the companion setae.

The modified notosetae of setiger 4 are unusual. In some respects they resemble the major spines of setiger 5 found among some species of *Polydora* and *Boccardia* where the curved shaft is surmounted by a crest of bristles. In *T. spinosa* this crest is transparent when viewed with the light microscope, but is a solid feature in *Polydora caulleryi* Mesnil (see Blake, 1971). It is probable that the modified notosetae on setiger 4 of *T. spinosa* are transitional in degree of modification between unilimbate capillaries and the heavy crested spines of *P. caulleryi*.

The major spines of setiger 5 on *T. spinosa* are revealed by SEM to have a terminal tooth and 2 shorter knobs between which are located fine bristles. The terminal structure of this seta is very difficult to observe with light microscopy and the bristles are rarely seen. This emphasizes the difficulties of making accurate portrayals of setal structure using only the light microscope. The bristles had not been previously reported on the major spines of *T. spinosa*.

The ribbed nature of the capillary notosetae of setigers 7–10 of *T. spinosa* has not been previously described. With light microscopy these setae have a unilimbate appearance in which the ribs appear to be surface sculptures of the sheath. With SEM, it can be seen that each "rib" is part of a whorl of thickened fibril endings which completely cover the ends of these setae.

The fine details of the hoods of the hooded hooks are different in *T. spinosa* than we have observed using SEM in related species of *Polydora* and *Boccardia*. These differences manifest themselves in the degree of bristling apparent upon the hood, position and shape of the hood opening, and the number of teeth which project through the hood opening. These differences suggest that microarchitecture of the hooded hooks may prove to be useful in systematic studies of spionid polychaetes. Readers should be aware, however, that hood form will differ depending upon the drying technique employed during specimen preparation. If the specimens are air dried prior to coating and observation, the hoods tend to collapse, tear and more closely adhere to the shaft and teeth. The bristles also tend to be distorted with air drying. With critical point drying the hoods remain inflated, are seldom torn, and the bristles remain undistorted.

Among the genera of the *Polydora*-complex, *Tripolydora spinosa* is unique in many respects. It is the only species to have tridentate hooded

hooks, prominent granulated inferior sabre setae, and unique "ribbed" notosetae on setigers 7–10. Moreover, with the exception of *Boccardiella limnicola* (Blake and Woodwick, 1976), it is the only species with gills from setiger 2 continuing uniformly to near the end of the body. The species has modified notosetae on setiger 4 *in addition to* the major spines on setiger 5. Modifications to notosetae on setiger 4 are known for some species of *Pseudopolydora* (see Woodwick, 1964), but are not known for other genera of the *Polydora*-complex. *Tripolydora spinosa* also resembles *Pseudopolydora* in the lack of major parapodial alteration to setiger 5 and in the reduced angle between the main fang and apical tooth (teeth) of the hooded hooks. The modified notosetae of setiger 4, the "ribbed" notosetae of setigers 7–10 and the inferior sabre setae were not reported by Woodwick (1964) because some of these setae were sheared off the holotype.

Blake (1979) proposed that *Tripolydora* exhibits a close relationship to the non-polydorid genus *Microspio*. This conclusion was originally based on similarities in branchial distribution, hooded hook structure, and the presence of inferior sabre setae in both genera. Additional support for this conclusion is the recent discovery that *Microspio granulata* Blake and Kudenov from Australia, the Hawaiian Islands, and Ifaluk Atoll in the Caroline Islands, and an undescribed species of *Microspio* from Ecuador have enlarged capillary notosetae on rows I and II over setigers 4–6 (Blake, unpublished). In addition, the setae of these 2 rows come together ventrally, close and form a loop. This arrangement is reminiscent of some species of *Polydora* where the modified spines of setiger 5 are positioned into a J-shape (see Blake, 1971). This trend in *Microspio* towards modification of setae in the corresponding modified region of polydorids occurs in no other member of the Spionidae. Blake (1979) emphasized that larval characters are also useful in interpreting relationships of the *Polydora*-complex; however, the larval morphology of *Tripolydora spinosa* has not been described.

From the information available, it would appear that *Microspio* is an appropriate genus to consider as having given rise to the *Polydora*-complex. In order to pursue this line of research SEM studies are being undertaken on selected species of *Microspio, Pseudopolydora,* and other genera of the *Polydora*-complex. Following completion of those studies it should be possible to describe more fully the interrelationships of the genera of the subfamily Spioninae.

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