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Hawaiian Polyclad Flatworms: Prosthiostomids[1]

J. L. POULTER[2]

ALTHOUGH MANY MEMBERS of the family Prosthiostomidae Lang, 1884 (suborder Cotylea Lang, 1884), have been reported from the Indo-West-Pacific, no definitive report on Hawaiian polyclads has included prosthiostomids. However, Lang (1884: 606) suggested that *Peasia* (Gray) *irrorata* Pease, 1860, may belong to this group. The two species of *Prosthiostomum* described here probably do not comprise all the representatives of this genus in Hawaii.

The identification of *Enchiridium japonicum* Kato, 1943, is problematical and a comparison of the types from Taiwan with the Hawaiian material might have elucidated the status of the Hawaiian specimens. However, attempts to locate the type material were unsuccessful and it now appears that the specimens were destroyed during World War II. A comparison of Kato's description with the Hawaiian specimens, however, suggests conspecificity.

SYSTEMATICS OF THE GENUS *Prosthiostomum* QUATREFAGES, 1845

In describing *Prosthiostomum gabriellae* Marcus, 1949, Marcus suggested that binding of the accessory vesicles within a common muscle mantle could be of systematic value in distinguishing some of the many members of this genus where the copulatory apparatus demonstates a high degree of uniformity. Subsequently, Marcus and Marcus (1966, 1968) created the genus *Lurymare* to include species of *Prothiostomum* "with accessory vesicles bound in a common muscle sheath which may include the seminal vesicle." For a list of species, see Marcus and Marcus (1968).

Problems encountered in interpretation of the definition of *Lurymare* in relation to a species investigated here led to a modification of the systematics suggested by Marcus and Marcus (1966, 1968), with a subgeneric distinction being proposed in lieu of generic recognition and with the definitions of Marcus and Marcus being but slightly modified.

When the accessory vesicles are not closely bound, as in the type *Prosthiostomum* (*Prosthiostomum*) *siphunculus* (Chiaje, 1828) as redescribed by Lang (1884), the subgeneric name (*Prosthiostomum*) is maintained. This subgenus should include *P.* (*P.*) *drygalskii* Bock, 1931. The "binding" in *P.* (*P.*) *drygalskii* refers to a muscle and/or connective tissue development in the region of the seminal vesicle and accessory vesicles but not to a physical apposition of the accessory vesicles and consequently is more in accord with the condition found in *P.* (*P.*) *siphunculus* as redescribed by Lang (1884). An anatomical condition that may be similar to that of *P.* (*P.*) *drygalskii* may occur also in *P.* (*P.*) *singulare* Laidlaw, 1904.

In the subgenus *Lurymare*, the accessory vesicles lie in physical apposition, and the vesicles are bound within a common specialized tissue. Assigned to this subgenus are *Prosthiostomum* (*Lurymare*) *elegans* Laidlaw, 1902; *P.* (*L.*) *cooperi* Laidlaw, 1902; *P.* (*L.*) *purum* Kato, 1937; *P.* (*L.*) *delicatum* Palombi, 1940; *P.* (*L.*) *russoi* Palombi, 1940; *P.* (*L.*) *gabriellae* Marcus, 1949; *P.* (*L.*) *matarazzoi* Marcus, 1950; and *P.* (*L.*) *utarum* Marcus, 1952.

Prosthiostomum Quatrefages, 1845

Prosthiostomum (Prosthiostomum) montiporae, new species[3]

Figures 1–6, 18, 22

TYPE MATERIAL: Specimens collected 6 June 1971 and 14 October 1971 were provided for

[1] Work is from the laboratory of Dr. J. G. Forte. Manuscript accepted 7 January 1975.

[2] University of California at Berkeley, Department of Physiology-Anatomy, 2549 Life Sciences Building, Berkeley, California 94720.

[3] AUTHOR'S NOTE ADDED IN PROOF. Synonym: *Prosthiostomum* (*Prosthiostomum*) *montiporae* Jokiel & Townsley, 1974, p. 362, *nomen nudum*. The published specific name of *Prosthiostomum* (*Prosthiostomum*) *montiporae* Jokiel & Townsley, 1974, is invalid and unavailable according to the International Code of Zoological Nomenclature adopted by the xvth International Congress of Zoology, published for the International Commission on Zoological Nomenclature by the International Trust for Zoological Nomenclature (London, 1964), until a description of the species is published. The ensuing description validates the specific name and thus makes it available.

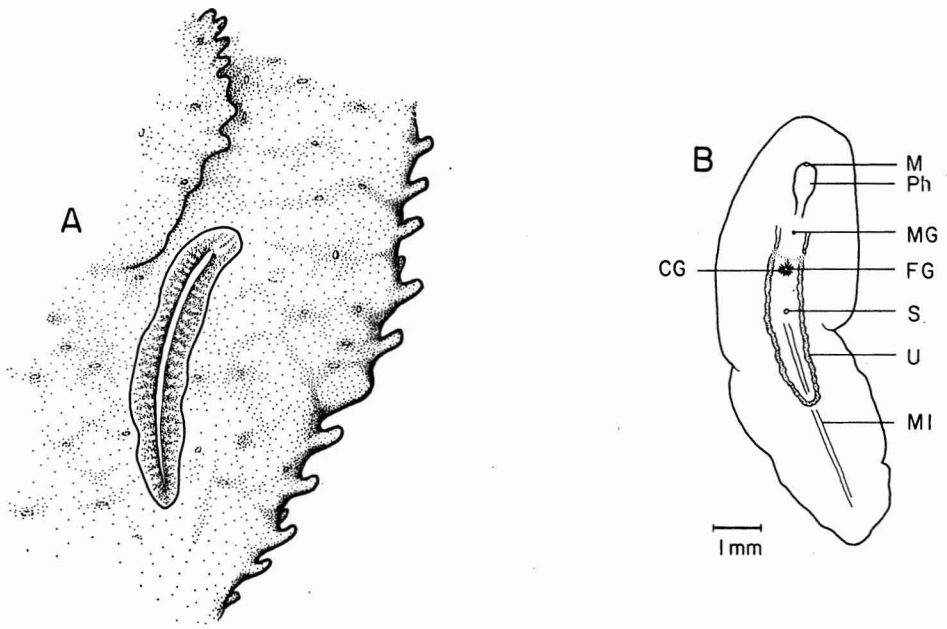

FIGURE 1. *Prosthiostomum* (*Prosthiostomum*) *montiporae*. *A*, living specimen on *Montipora verrucosa*, from a photograph by Paul Jokiel; *B*, anatomy of cleared holotype.
ABBREVIATIONS: CG, cement glands; FG, female gonopore; M, mouth; MG, male gonopore; MI, main intestine; Ph, pharynx; S, sucker; U, uterus.

identification by Dr. Sidney J. Townsley and Mr. Paul Jokiel of the University of Hawaii, Honolulu, Hawaii. These polyclads were originally discovered on the coral *Montipora verrucosa* (Lamarck) in experimental aquaria at the Hawaii Institute of Marine Biology on Mokuoloe Island (Coconut Island), Kaneohe Bay, Oahu[4], Hawaii. Subsequently, they were found associated with *Montipora verrucosa* in Kaneohe Bay, hereby designated the type locality (longitude 157°47′21″ W, latitude 21°26′10″ N).

The copulatory region of the Formalin-fixed holotype was sagittally sectioned (three slides), stained with periodic acid-Schiff (PAS) and Mayer hemalum (Pearse 1960), and the anterior and posterior portions mounted. The holotype is deposited in the United States National Museum (USNM 52695), where a paratype is also deposited. Paratypes are also deposited in the American Museum of Natural History and the British Museum (Natural History).

Other mercury-fixed paratypes were entirely sectioned: one sagittally, stained with PAS and Mayer hemalum; another frontally, stained with Mayer hemalum and phloxin. A juvenile paratype was cleared and whole mounted.

With mercury fixation specimens roll tightly, whereas with Formalin fixation specimens do not roll and less contraction appears to occur. Although the Formalin-fixed holotype is adequate for taxonomic purposes, Formalin fixation is histologically far inferior to mercury fixation and, consequently, some histological details are described from the mercury-fixed paratypes.

DESCRIPTION OF LIVING SPECIMENS: Form (Figure 1*A*) elongate, rounded anteriorly, bluntly pointed posteriorly, unusually thin dorsoventrally. Specimens may attain a length-to-breadth ratio of 7:1. Color pattern (from notes and a colored photograph accompanying specimens)—dorsal surface off-white; epidermal pigment lacking; density and color when present attributed to internal structures and mesenchyme. A rust-brown color delineates the gut. The gut exhibits the same rust-brown color as do the coral polyps on which the worms are found, the color being the result of zooxanthel-

[4] Geographic names from "Topographic map of the island of Oahu, 1954." United States Department of the Interior, Geological Survey, Washington, D.C.

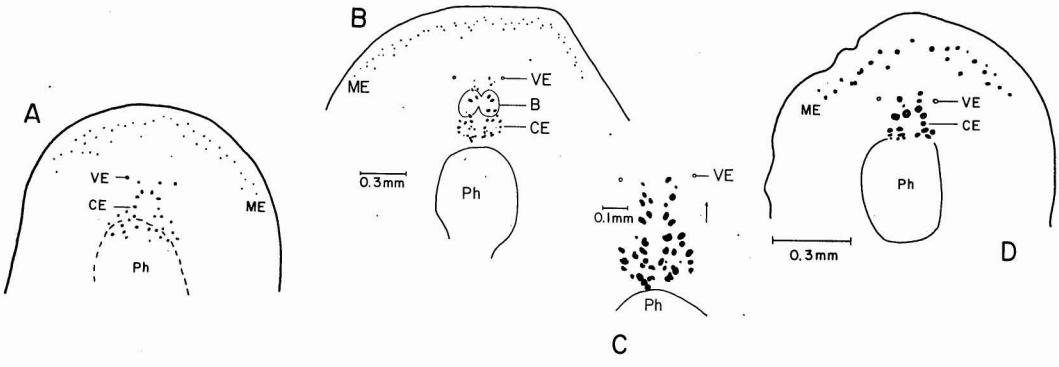

FIGURE 2. *Prosthiostomum* (*Prosthiostomum*) *montiporae*, eye arrangement. *A*, living specimen, from a photograph by Paul Jokiel; *B*, cleared holotype; *C*, cerebral eyes of cleared holotype; *D*, cleared juvenile paratype.
ABBREVIATIONS: B, brain; CE, cerebral eyes; ME, marginal eyes; Ph, pharynx; VE, ventral eye.

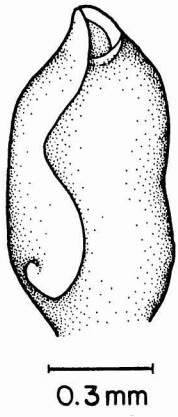

FIGURE 3. *Prosthiostomum* (*Prosthiostomum*) *montiporae*, pharynx morphology of a dissected paratype; pharynx viewed from ventral aspect, rotated slightly to right.

lae that are ingested with coral tissue. This distinctive coloration is absent in the midline except anterior to the cerebral eyes where a branch of the gut extends toward the anterior and from the peripheral region beyond the perimeter of the gut. Marginal and cerebral eye arrangement (Figure 2*A*) is similar to that of preserved specimens. The barrel-shaped pharynx occupies a length equal to 10 percent of the body length and lies 12 percent of the body length behind the anterior margin. The male copulatory apparatus is located close behind the pharynx, about 27 percent of the body length from the anterior margin. The cement glands of the female copulatory apparatus lie 37 percent of the body length from the anterior margin. The sucker lies just anterior to the body middle.

DESCRIPTION OF PRESERVED SPECIMENS: The anatomy of the fixed cleared holotype (Figure 1*B*) corresponds well with living specimens. Length of holotype, 9.4 mm; breadth, 3.0 mm. Form elongate-oval, thin dorsoventrally: 0.44 mm thick in midline; 0.16 mm near margin. Color opaque off-white with a region of irregularly spaced minute rust-brown pigment granules over middorsal line; pigment dissipates about 15 percent of body length from each end. Zooxanthellae in gut represented by irregular dark blotches visible from both dorsal and ventral surfaces. Ground color of ventral surface off-white.

Marginal (Figure 2*B*) and cerebral (Figure 2*C*) eyes more densely aggregated than *in vivo*. Marginal eyes form a band, minimum distance from margin edge, 0.13 mm; maximum diameter of eyes, 20 μm. Larger eyes lacking in midline, smaller eyes continuous across midline. Band of marginal eyes extending posteriorly to level of anterior cerebral eyes. Paired cerebral eye clusters form roughly cuneiform groups; number per group in holotype, 22. Eye cluster located 0.46 mm (5 percent of body length) from anterior margin; length of cluster 0.46 mm (extending from 5 to 10 percent of body length); maximum eye diameter, 32 μm. A pair of ventral or "deep" eyes present lateral to most anterior cerebral eyes. Ventral eyes in sectioned material lie above musculature of ventral body wall as in *P.* (*P.*) *drygalskii* Bock, 1931. Eye arrangement in juvenile (length, 3.2 mm; breadth, 1.2 mm) similar to that of larger specimens but with fewer eyes (Figure 2*D*).

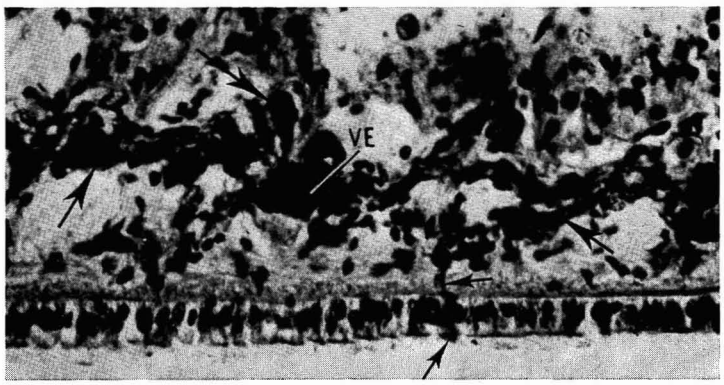

FIGURE 4. *Prosthiostomum* (*Prosthiostomum*) *montiporae*. PAS-positive secretion of mesenchyme anterior to pharynx (465 ×); sagittal section; stains, PAS and Mayer hemalum.
ABBREVIATION: VE, ventral eye.
SYMBOLS: large arrows, PAS-positive secretion of mesenchyme; small arrows, duct to ventral surface.

Mouth at ventroanterior extent of pharynx. Anterior limit of pharynx 1.0 mm (11 percent of body length) behind anterior margin. Pharynx barrel-shaped; length, 0.8 mm (9 percent of body length); maximum breadth, 0.56 mm. Pharynx remains *in situ* in fixed specimens. The morphology of the pharynx is unique in that the typical tubular structure of prosthiostomids has been modified by a deep longitudinal cleft (Figure 3), the overlapping edges rolled loosely, scroll-like; the base of the cleft terminates in a flat helical pattern. Once dissected free of surrounding tissue, the compact pharynx can be opened and unrolled revealing the interior surface. Pharynx base attached as in other prosthiostomids. Pharynx opens posteriorly into the main intestine, which passes posteriorly to near the posterior margin. Intestinal epithelium columnar; mean height, 24 μm; abundantly supplied with clavate basophilic gland cells. Three major trunks of the gut are anteriorly directed, one on each side of the pharynx, the third above. Gut branches do not anastomose.

Dorsal epithelium 20 μm thick, packed with rhabdites and numerous gland cells. Ventral epithelium 11 μm thick, with few rhabdite and gland cells. Muscle layers of both dorsal and ventral body walls poorly developed. A distinctive feature is a subepidermal PAS-positive secretion (Figure 4) elaborated in the densely packed mesenchyme and emptying by narrow ductules onto the ventral surface. The secretion occurs in isolated aggregations (generally around nuclei) along the entire ventral mesenchyme but is most abundant, forming a stratum, in the region anterior to the pharynx. The rust-brown pigment granules observed in fixed specimens were undetectable in sectioned material, and it is possible that the closely crowded rhabdites in the midline may cause the postmortem coloration.

Testes ventral. Spermiducal vesicles form rows, one on each side of the main intestine posterior to the male copulatory apparatus. Medially directed sperm ducts join each respective row of spermiducal vesicles, with a terminal spermiducal vesicle lying anterior to and on each side of the seminal vesicle. A sperm duct passes posteriorly from each terminal spermiducal vesicle, penetrates the muscle wall of the seminal vesicle (Figure 5), proceeds posteriorly just under the luminal epithelium, and opens independently into the posterior portion of the seminal vesicle. Seminal vesicle ovate; longest axis, 0.21 mm; shortest axis, 0.15 mm. The lumen is sperm filled; longest axis, 0.15 mm; shortest axis, 0.08 mm. The wide ejaculatory duct issues from the dorsoanterior extent of the seminal vesicle. It narrows anteriorly, detours one accessory vesicle, and enters the penis papilla. The two orbiculate accessory vesicles are stacked one atop the other; they are apposed but not bound. The ventral vesicle lies adjacent to the ventroanterior face of the seminal vesicle, apposed but not bound. The

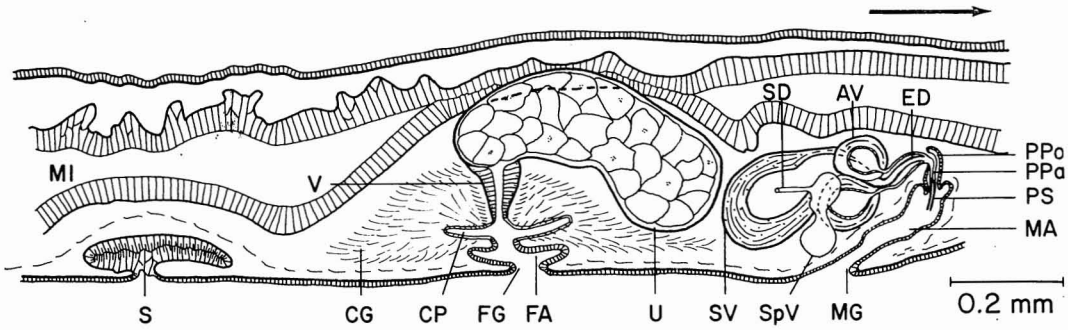

FIGURE 5. *Prosthiostomum* (*Prosthiostomum*) *montiporae*, copulatory region of holotype. Only one of a pair of sperm ducts is shown.

ABBREVIATIONS: AV, accessory vesicle; CG, cement glands; CP, cement pouch; ED, ejaculatory duct; FA, female antrum; FG, female gonopore; MA, male antrum; MG, male gonopore; MI, main intestine; PPa, penis papilla; PPo, penis pouch; PS, penis stylet; S, sucker; SD, sperm duct; SpV, spermiducal vesicles; SV, seminal vesicle; U, uterus; V, vagina.

accessory vesicles are chiefly composed of a relatively thin nonnucleated muscle and/or connective tissue hull; maximum thickness, 0.02 mm. The ventral accessory vesicle is 0.1 mm in diameter: the luminal long axis, 0.08 mm; short axis, 0.06 mm. The dorsal accessory vesicle is slightly smaller in all aspects. The luminal epithelium is thin with a few compressed nuclei. The lumina contain unidentifiable basophilic material. A duct from the lumen of each accessory vesicle passes anteriorly through the nonnucleated hull, roughly paralleling the ejaculatory duct, and becomes confluent with the ejaculatory duct in the penis base. The penis lies in the penis pouch and is armed with a stylet (90 μm in length) that protrudes into the male antrum. The usually prominent prostatic secretion in the mesenchyme surrounding and emptying into the penis pouch that has been reported for other prosthiostomids was undetectable in the dense mesenchyme of these worms but its presence may be indicated by a small quantity of secretion in the penis pouch. Penis pouch small, campanulate, opening into male antrum. Male antrum deep, passing obliquely posterior, opening on ventral surface at male gonopore, ventral to location of accessory vesicles. Terminal portion of male system located close behind posterior termination of pharynx. Male gonopore 2.4 mm (26 percent of body length) behind anterior margin. Length of terminal portion of male system including seminal vesicle to penis pouch, 0.3 mm. Male system miniaturized even though sperm were evident in smallest specimen sectioned.

Ovaries dorsal, but maturing ova may occupy the entire dorsoventral space between the musculature of the body wall. The uteri lie on each side of main intestine anterior and posterior to the female copulatory apparatus, joining posteriorly under the main intestine at 65 percent of body length from the anterior margin. This feature was also noted in *P.* (*L.*) *matarazzoi* Marcus, 1950, and *P.* (*L.*) *utarum* Marcus, 1952. An oviduct from each uterus converges anterior to the terminal portion of female copulatory apparatus, forming a large common ova fill-chamber that joins the vagina at its most ventroposterior extent. Vaginal epithelium tall, thicker in close proximity to the uterus, with extremely long cilia. Vagina opens into dorsoventrally compressed cement pouch. The cement pouch receives extensive cement gland secretion, opens by a narrow aperture into the female antrum (also with a pouch) which is broader and more compressed than usual. Female gonopore 60 μm in diameter, located 3.0 mm (32 percent of body length) behind the anterior margin equidistant between male gonopore and sucker.

Sucker 3.9 mm (42 percent of body length) behind anterior margin; diameter, 0.3 mm. Epithelium 30 μm in height. Muscular development of the sucker surpasses that of the adjacent body wall.

DIAGNOSIS: *Prosthiostomum* (*Prosthiostomum*) *montiporae* is most closely related to *P.* (*L.*) *purum* Kato, 1937, and *Amakusaplana ohshimai* Kato, 1938. *P.* (*P.*) *montiporae* is distinguished

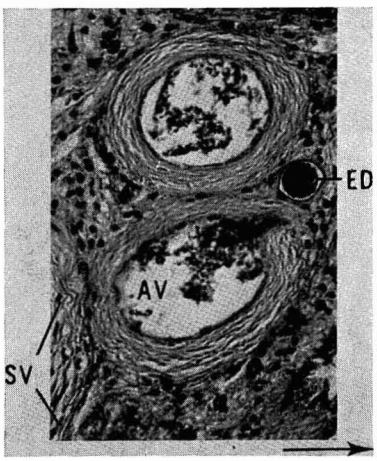

FIGURE 6. *Prosthiostomum* (*Prosthiostomum*) *montiporae*, accessory vesicles of holotype (265 ×); sagittal section; stains, PAS and Mayer hemalum.

ABBREVIATIONS: AV, accessory vesicle; ED, ejaculatory duct; SV, seminal vesicle.

from *P.* (*L.*) *purum* and other members of *Prosthiostomum* by the combination of eye arrangement, extremely short pharynx length, unusual morphology of the pharynx, and some details of the copulatory apparatus. *Amakusaplana ohshimai* and *P.* (*P.*) *montiporae* resemble each other with respect to pharynx length, but *P.* (*P.*) *montiporae* is distinguished from *A. ohshimai* on the basis of general shape, eye arrangement, morphology of the pharynx, and features of the copulatory apparatus, and the sucker apparently is lacking in *A. ohshimai*.

Prosthiostomum (*Prosthiostomum*) *montiporae* is easily distinguished from *Peasia irrorata* Pease, 1860, in which the pharynx length is probably more typical of that of *Prosthiostomum*.

DISCUSSION: Hyman (1959), on the basis of her studies of prosthiostomids, doubted that *Amakusaplana* could be maintained as a genus distinct from *Prosthiostomum*. The anatomical similarities between *Amakusaplana ohshimai* and *P.* (*P.*) *montiporae*, while interesting, offer no new evidence to erode further the generic separation of *Amakusaplana* as defined by Kato (1938) from *Prosthiostomum*. However, if the types or other specimens of *A. ohshimai* should be reinvestigated, the morphology of the pharynx should be of particular interest, and an unusual pharyngeal construction similar to that of *P.* (*P.*) *montiporae* is possible. Thus, the pharyngeal morphology makes it clear that the assignment of the species at hand to *Prosthiostomum* may be regarded at best as provisional. If *Amakusaplana* does have a cleft pharynx, then *A. ohshimai* and *P.* (*P.*) *montiporae* could be allied, and *Amakusaplana* would be clearly distinguished from *Prosthiostomum*. An alternative would be to create a new genus for *P.* (*P.*) *montiporae* based on pharyngeal morphology, but, in view of the unsettled questions, this could only be regarded presently as a dubious step. The possible phylogenetic significance of the cleft pharynx of *P.* (*P.*) *montiporae* remains problematical.

The morphology of the accessory vesicles of *P.* (*P.*) *montiporae* is relevant to its present systematic position within *Prosthiostomum*. Sectioned specimens apparently show an increasing apposition of these vesicles as correlated with specimen size. In the smallest sectioned paratype (length, 5.3 mm) some mesenchyme was found between the accessory vesicles, whereas in another sectioned paratype (length, 6.5 mm) little mesenchyme was found between these vesicles. In the holotype (length, 9.4 mm) the vesicles are apposed (Figure 6), and a few mesenchymal nuclei between the vesicles are the only residual evidence of mesenchymal tissue. There is no detectable muscle contribution from the seminal vesicle or specialized tissue involved in binding the accessory vesicles, nor is there nonnucleated muscle and/or connective tissue common to both vesicles. Where an accessory vesicle does not lie next to another anatomical structure, the nonnucleated hull lies adjacent to mesenchyme as described by Lang (1884:276) for *P.* (*P.*) *siphunculus* (Chiaje 1828). The latter condition rules out the possibility that a nucleated epithelial covering of the nonnucleated hull binds the vesicles. This aspect warrants consideration because the accessory vesicles of *P.* (*P.*) *drygalskii* Bock, 1931, are covered by such an epithelium but the vesicles are not apposed. Although the accessory vesicles were not deemed bound in this investigation of *P.* (*P.*) *montiporae*, the intimate approximation of the vesicles and the progressive diminution of interposed mesenchymal tissue does not permit one to conclude that the accessory vesicles could not be bound in larger specimens in the same manner as in *P.* (*L.*) *purum* Kato, 1937,

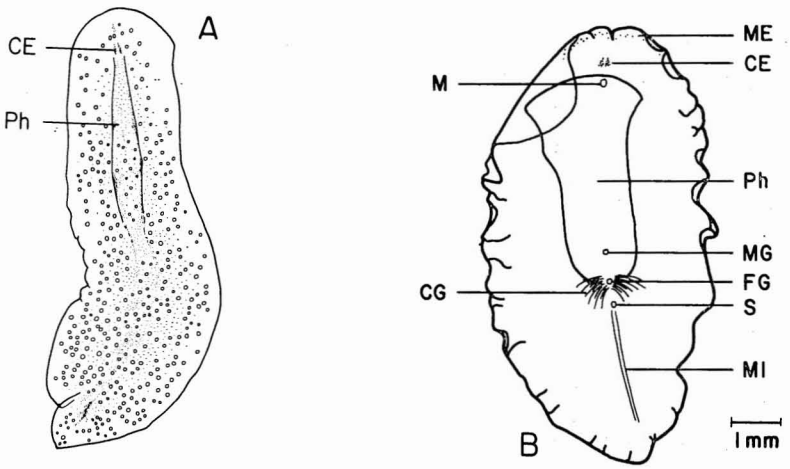

FIGURE 7. *Prosthiostomum* (*Lurymare*) *katoi*. *A*, living specimen from a photograph; specimen is beginning to crawl extended, anterior is expanding, posterior still contracted. Small circles indicate deep gold to orange-red pigment spherules; stippling indicates brown pigments. *B*., anatomy of cleared holotype.

ABBREVIATIONS: CE, cerebral eyes; CG, cement glands; FG, female gonopore; M, mouth; ME, marginal eyes; MG, male gonopore; MI, main intestine; Ph, pharynx; S, sucker.

where the accessory vesicles are "distinctly separated" in immature specimens but "a part of muscle fibers of each vesicle wall overlap" in a mature specimen.

Prosthiostomum (Lurymare) katoi, new species

Figures 7–11, 19

TYPE MATERIAL: Representatives of this species were usually collected among the shells of *Ostrea sandvichensis* Sowerby, 1871, on the reef at Ala Moana Park, Oahu, Hawaii, from 1 April 1961 through July 1962. The holotype (no. 9-E-1) was collected 17 March 1962. The type locality is here designated as the reef at Ala Moana Park, Oahu (longitude 157°51′ W, latitude 21°17′20″ N). Thirteen paratypes were collected from the same area. One specimen was collected by Dr. E. A. Kay near Chimney (old sugar mill) midway between Kuloa Point and Kaoio Point, Oahu.

The holotype was entirely sectioned sagittally (eight slides) and stained with PAS and Mayer hemalum (Pearse 1960). The copulatory region of a paratype was sectioned frontally and stained with PAS, Mayer hemalum, and fast green FCF; the anterior and posterior portions were mounted. The copulatory region of the smallest paratype was sectioned sagittally and stained with PAS, Mayer hemalum, and light green. A paratype was cleared and whole mounted. The copulatory region of the specimen from Chimney was sectioned sagittally and stained with PAS, Mayer hemalum, and fast green FCF. As reported for other members of the genus, specimens usually roll tightly on fixation and the pharynx is sometimes extruded.

The holotype (USNM 52698), the whole-mounted paratype, and two additional paratypes are deposited in the United States National Museum. Other paratypes are deposited in the American Museum of Natural History, the British Museum (Natural History), and the California Academy of Sciences.

DESCRIPTION OF LIVING SPECIMENS: Form (Figure 7*A*), when at rest, abbreviated elongate-oval; when crawling extended, elongate, rounded anteriorly, bluntly pointed posteriorly, margins usually smooth. Length of holotype, 17 mm; breadth, 5 mm. Size range and mean values of specimens are shown in Figure 8. Length-to-breadth ratio of resting animals 2:1; crawling animals may reach 6:1. Breadth is not appreciably altered by activity; breadth at rest approximates breadth when the worm is crawling extended. Dorsoventral thickness depends on activity: resting worms are thick, crawling extended worms are thin.

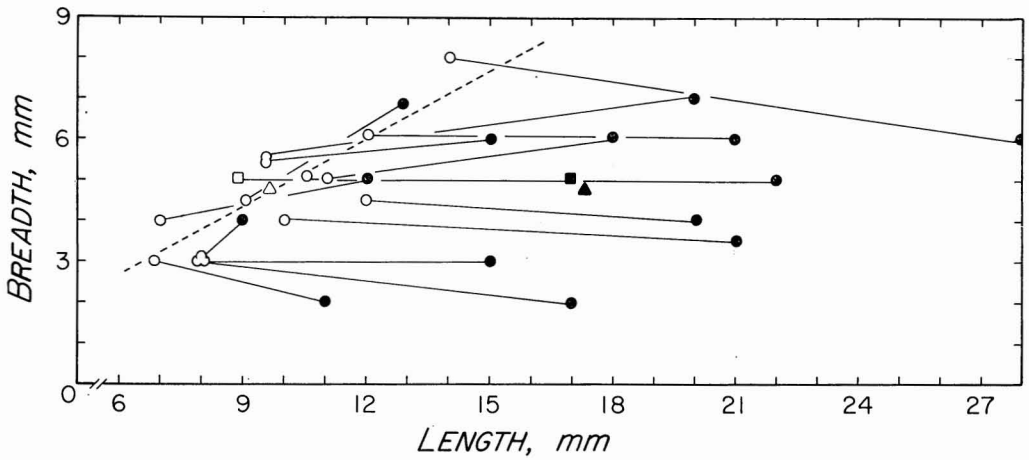

FIGURE 8. *Prosthiostomum* (*Lurymare*) *katoi*, graphic representation of length:breadth measurements of specimens collected.

SYMBOLS: black square, living holotype; black circles, living paratypes and other specimen; black triangle, mean value of living specimen; white square, fixed holotype; white circles, fixed paratype and other specimen; white triangle, mean value of fixed specimens; solid line joins measurement points of each living and fixed specimen; dashed line indicates trend line by method of least squares for fixed specimens.

Color pattern: ground color of dorsal surface off-white, indicating lack of pigment; mesenchyme and internal structures impart density and color. Perimeter of margin transparent and colorless for a distance of about 0.7 mm, indicating absence of gut and gonads. Pigment spherules present medial to margin, more abundant in middorsal region. Pigment spherules are uniform in color tone in each worm, but color varies among specimens from gold to orange-red. Minute brown pigment granules are scattered on the dorsal surface but are most prevalent on the middorsal line, especially over the pharynx, though they do not form a pronounced stripe. These pigments abruptly diminish anteriorly just posterior to the cerebral eyes and posteriorly in the area where the main intestine terminates. Density of both pigment types depends on state of expansion or contraction. Ventral surface off-white.

Marginal eyes form a band, minimum distance from margin edge, 0.3 mm. Although the marginal band usually extends posteriorly to the level of the cerebral eyes, shorter bands also have been observed. The cerebral eyes of the holotype lie 2.0 mm (12 percent of body length) posterior to the anterior margin and lie from 0.5 to 2.25 mm behind the anterior margin in the smallest and largest specimens examined, respectively. Eyes form a paired cluster, roughly triangulate in resting animals and elongate-oval in crawling animals.

Pharynx expands and contracts with body configuration. Copulatory region and sucker lie just back of the posterior termination of the pharynx, posterior to the middle of the body. Colored waste material often is evident in the gut posterior to the main intestine.

DESCRIPTION OF PRESERVED SPECIMENS: Form ovate (Figure 7*B*), rounded anteriorly, moderately pointed posteriorly, margins crenate. Holotype length in 70 percent alcohol, 8.8 mm; breadth, 4.9 mm; length after sagittal sectioning, 8.4 mm. Length and breadth measurements of fixed specimens are shown in Figure 8; length-to-breadth ratios range from 1.7:1 to 2.7:1. Body thickness, 1.0 mm in pharyngeal region; 0.45 mm on midline posterior to copulatory region.

The color pattern is altered with fixation and preservation, and the gold to orange-red pigment spherules are undetectable. However, the minute brown pigments are retained in the same general pattern as in living animals but appear to be more pronounced because of contraction. Ground color of dorsal and ventral surfaces off-white.

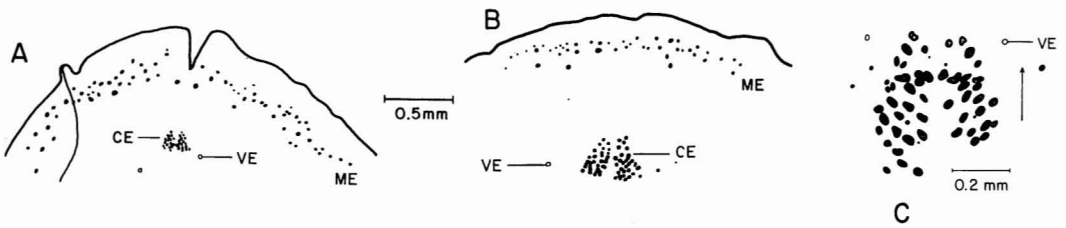

FIGURE 9. *Prosthiostomum* (*Lurymare*) *katoi*, eye arrangement. *A*, holotype; *B*, paratype with shorter band along margin; *C*, cerebral eyes of paratype.
ABBREVIATIONS: CE, cerebral eyes; ME, marginal eyes; VE, ventral eye.

Marginal eyes (Figure 9*A*, *B*) form an uninterrupted band 0.2 mm from margin edge. Maximum eye diameter in holotype, 40 μm. The wide variation in length of the bands with a concomitant variation in eye number noted in living worms was verified in cleared specimens. Cerebral eyes in paired, roughly triangulate to ovate groups, 0.34 mm in length, 0.7 mm behind anterior margin (8 percent of body length in holotype). A precise determination of the cerebral eye arrangement could not be made on the holotype because of distortion by the pharynx at fixation, and, consequently, the cerebral arrangement is described from paratypes (Figure 9*C*). Each group contains from 25 to 33 eyes; maximum diameter of eyes, 50 μm; most eyes are of relatively uniform diameter. A pair of ventral or "deep" eyes present. Their location in relation to the cerebral eyes is associated with distortion incurred at fixation, the pharynx forcing the cerebral eyes anteriorly. When the pharynx is retained within the body, the ventral eyes are usually posterior to the cerebral eyes; when the pharynx is extruded, the ventral eyes usually occur on a level with the most anterior cerebral eyes. In one paratype there is only one ventral eye. In the sectioned holotype, the ventral eyes lie in the mesenchyme above the musculature of the ventral body wall as described for *P.* (*P.*) *drygalskii* Bock, 1931.

Mouth 1.4 mm (17 percent of body length) posterior to anterior margin, subterminal relative to pharynx. Pharynx large, campanulate, 4.0 mm in length (48 percent of body length), anterior extent 1.0 mm (12 percent of body length) behind anterior margin. Anterior portion of main intestine badly distorted by pharynx at fixation. Main intestine posteriorly directed, giving rise to major trunks of gut branches, terminating 7.0 mm (83 percent of body length) posterior to anterior margin. Intestinal epithelium undulating, cell boundaries indistinct, cytoplasm granular, deficient in individual gland cells. Gut branches do not anastomose.

Dorsal epithelium 35 μm thick, about equally supplied with moderate numbers of rhabdite and granular gland cells; rhabdites one-half the height of the epithelium; gland cells equal the height of the epithelium. Musculature of dorsal body wall not well organized. Brown pigment of color pattern interspersed among dorsal musculature, prevalent medially but sparse peripherally. Ventral epithelium 4 μm thick, cuboidal, with a few tiny rhabdite and gland cells, ciliated; cilia length twice the thickness of epithelial cells. Ventral muscle layers distinctly developed, 54 μm thick. Mesenchyme well packed but not densely so. No evidence of the PAS-positive material observed in the mesenchyme of *P.* (*P.*) *montiporae* was detected.

Testes ventral. Spermiducal vesicles forming two rows, one on each side of midline. Medially directed sperm ducts, one from each row, enlarge irregularly and form additional spermiducal vesicles near the seminal vesicle. Terminal spermiducal vesicles lie almost under anterior portion of seminal vesicle. Terminal sperm ducts independently and symmetrically penetrate the muscle hull of the anterior region of the seminal vesicle, discharging into the seminal vesicle lumen at less than one-half of its length. Seminal vesicle (Figure 10) long axis 0.43 mm, short axis 0.29 mm, luminal epithelium thin, lumen diameter roughly 0.1 mm; sperm and a PAS-positive secretion from epithelium of male tract present. Ejaculatory duct issues from most

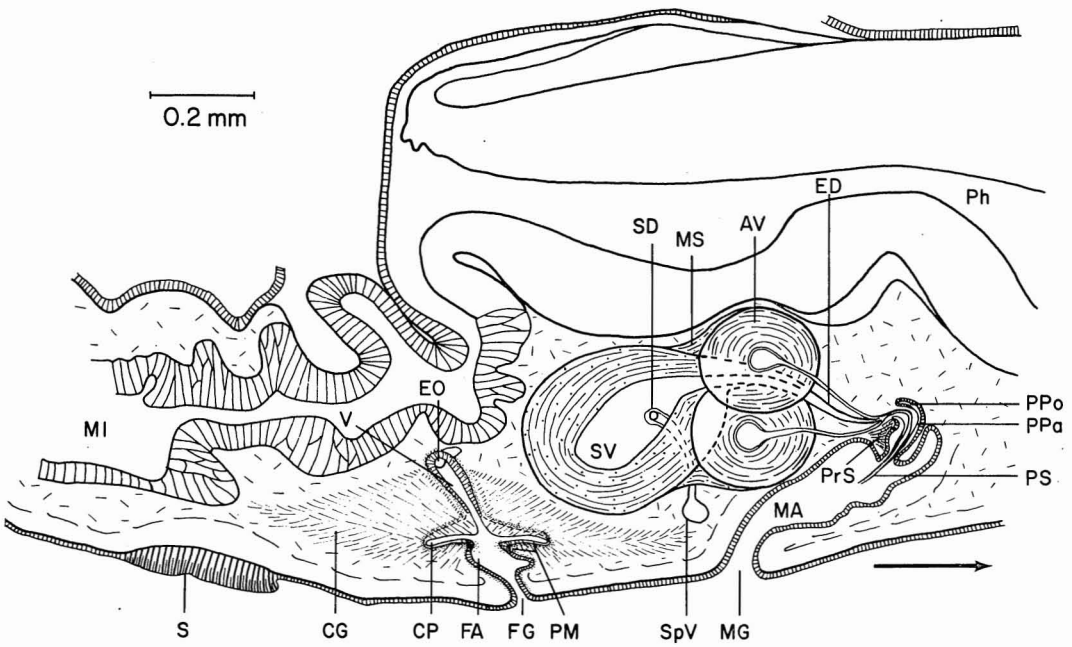

FIGURE 10. *Prosthiostomum* (*Lurymare*) *katoi*, copulatory region of holotype. Only one of a pair of sperm ducts is shown.

ABBREVIATIONS: AV, accessory vesicle; CG, cement glands; CP, cement pouch; ED, ejaculatory duct; EO, entrance of oviduct; FA, female antrum; FG, female gonopore; MA, male antrum; MG, male gonopore; MI, main intestine; MS, muscle involvement of seminal vesicle with accessory vesicle; Ph, pharynx; PM, PAS-positive material along terminal portion of female tract; PPa, penis papilla; PPo, penis pouch; PrS, prostatic secretion; PS, penis stylet; S, sucker; SD, sperm duct; SpV, spermiducal vesicle; SV, seminal vesicle; V, vagina.

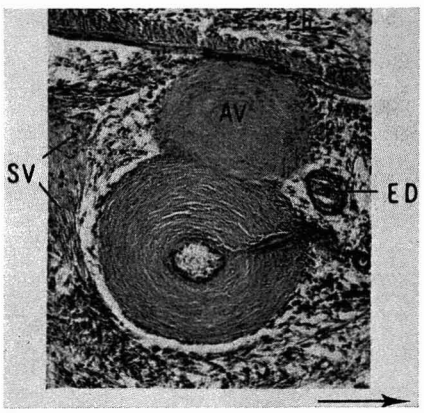

FIGURE 11. *Prosthiostomum* (*Lurymare*) *katoi*, accessory vesicles of holotype demonstrating the muscle involvement of seminal vesicle with accessory vesicles (115 ×); sagittal section; stains, PAS and Mayer hemalum.

ABBREVIATIONS: AV, accessory vesicle; ED, ejaculatory duct; SV, seminal vesicle.

dorsoanterior extent of seminal vesicle, passes anteriorly along cleavage of accessory vesicles, then to penis papilla. The two accessory vesicles apposed, one atop the other, the dorsal vesicle pressing against the pharynx. The vesicles are bound by a muscle contribution from the seminal vesicle (Figure 11), as described for *P.* (*L.*) *utarum* Marcus, 1952. Accessory vesicles are composed primarily of a relatively thick (90 μm) nonnucleated muscle and/or connective tissue hull. Ventral accessory vesicle 0.22 mm in diameter; lumen central, 54 μm in diameter. Luminal epithelium thin; lumen with stringy basophilic material. Dorsal accessory vesicle almost identical with respect to dimensions and histology. Duct from each accessory vesicle narrow, lacking nuclei until free of nonnucleated hull of vesicles, following a course nearly parallel to that of the ejaculatory duct and becoming confluent with the ejaculatory duct at the base of the penis papilla. Penis papilla in

penis pouch, curves sharply downward, armed with a long stylet (0.2 mm). Penis pouch receives "prostatic secretion" primarily from posterior wall. Male antrum 0.48 mm in depth, with epithelium 4 μm thick, passes obliquely posterior. Male gonopore 43 μm in diameter, located 4.2 mm (50 percent of body length) behind anterior margin. Length of terminal portion of male system (including seminal vesicle to penis pouch) 0.85 mm. The terminal apparatus lies entirely under the most posterior portion of the pharynx.

In paratypes where the pharynx is extruded, the orientation of the accessory vesicles is altered and the vesicles lie in an anterior–posterior configuration, with the comparable dorsal vesicle lying anteriorly. The smallest paratype clearly demonstrates the muscular contribution of the seminal vesicle in binding the accessory vesicles, and in this specimen the male copulatory apparatus lies posterior to the pharynx.

The ovaries of the holotype are not in the typical dorsal location; instead, they lie above the testes between the gut branches about midway between the ventral and dorsal surfaces. In more mature paratypes the ovaries lie in the more typical position, closer to the dorsal surface. The uteri, one on each side of the main intestine, extend anterior and posterior to the female gonopore. The uteri may join posteriorly some distance behind the sucker as in *P.* (*P.*) *montiporae* and a few other prosthiostomids. A medially directed oviduct from each uterus passes above the cement gland region, uniting at the vagina. Vaginal epithelium is cuboidal, with long cilia. Vagina turns downward and opens into an inverted funnelform cement pouch, 0.2 mm in length. The boundaries of the cement pouch are outlined by a PAS-positive basement membrane and distended by the secretion of the cement glands. Near the ventral constriction where the cement pouch joins the female antrum, the histology alters and PAS-positive material is predominantly secreted. The female antrum enlarges into a wide chamber, deeply undulated on the dorsoventral axis, the cuboidal epithelium densely ciliated and PAS-positive. Female gonopore 30 μm in diameter, 0.6 mm posterior to male gonopore, 4.85 mm (58 percent of body length) behind anterior margin.

Sucker lies 0.6 mm posterior to female gonopore, 5.4 mm (64 percent of body length) behind anterior margin; it was everted in all specimens sectioned. Sucker diameter, 0.15 mm; epithelium, 54 μm thick; musculature more developed than that of adjacent body wall.

DIAGNOSIS: The type of binding of the accessory vesicles in *Prosthiostomum* (*Lurymare*) *katoi* indicates that this species is most closely related to *P.* (*L.*) *utarum* Marcus, 1952. However, *P.* (*L.*) *katoi* is distinguished from *P.* (*L.*) *utarum* and other presently designated members of (*Lurymare*) by the combination of color pattern, eye arrangement, and details of the copulatory apparatus.

Peasia irrorata Pease, 1860, described from the Hawaiian Islands, little resembles *P.* (*L.*) *katoi* with respect to color pattern.

DISCUSSION: The binding of the accessory vesicles in the three Indo-West-Pacific members of *Prosthiostomum* assigned to (*Lurymare*)—*P.* (*L.*) *elegans* Laidlaw, 1902; *P.* (*L.*) *cooperi* Laidlaw, 1902; and *P.* (*L.*) *purum* Kato, 1937—appears to be morphologically distinct from that of *P.* (*L.*) *katoi*, and none seem to include the muscle contribution of the seminal vesicle.

The wide variation in marginal eye band length concomitant with wide variations in eye number cannot be correlated with specimen size (Table 1) and exceeds the range of variability that one expects to find in *Prosthiostomum* where eye arrangement is a major diagnostic character. Worms with both longer and shorter bands were sectioned and no significant internal differences were found. Among the specimens collected, longer bands are more typical than are shorter bands.

The two distinct secretions of the cement pouch of *P.* (*L.*) *katoi* are perhaps reminiscent of *P.* (*P.*) *lineatum* Meixner, 1907. However, the extent and staining characteristics of the ventral cement pouch of *P.* (*P.*) *lineatum* apparently do not correspond with the highly restricted PAS-positive secretion of *P.* (*L.*) *katoi*.

It is a pleasure to name thiss pecies for Dr. Kojiro Kato, who has contributed so extensively to our knowledge of Pacific polyclads.

TABLE 1

MARGINAL BAND EXTENT AND EYE NUMBER IN *Prosthiostomum* (*Lurymare*) *katoi*

SPECIMEN LENGTH (MM)	MARGINAL EYES (NUMBER)	POSTERIOR EXTENT OF MARGINAL BAND
6.8 (paratype)	54	on level with midregion of cerebral eyes
8.8 (holotype)	73	on level with posterior cerebral eyes (Fig. 9*A*)
9.6 (paratype)	35	limited to anterior margin
11.9 (other specimen)	80	on level with midregion of cerebral eyes
14.3 (paratype)	45	limited to anterior margin (Fig. 9*B*)

NOTE: data are given for four sectioned specimens and one whole-mounted specimen of *P.* (*L.*) *katoi* and indicate a wide variation in eye number that is unrelated to specimen length.

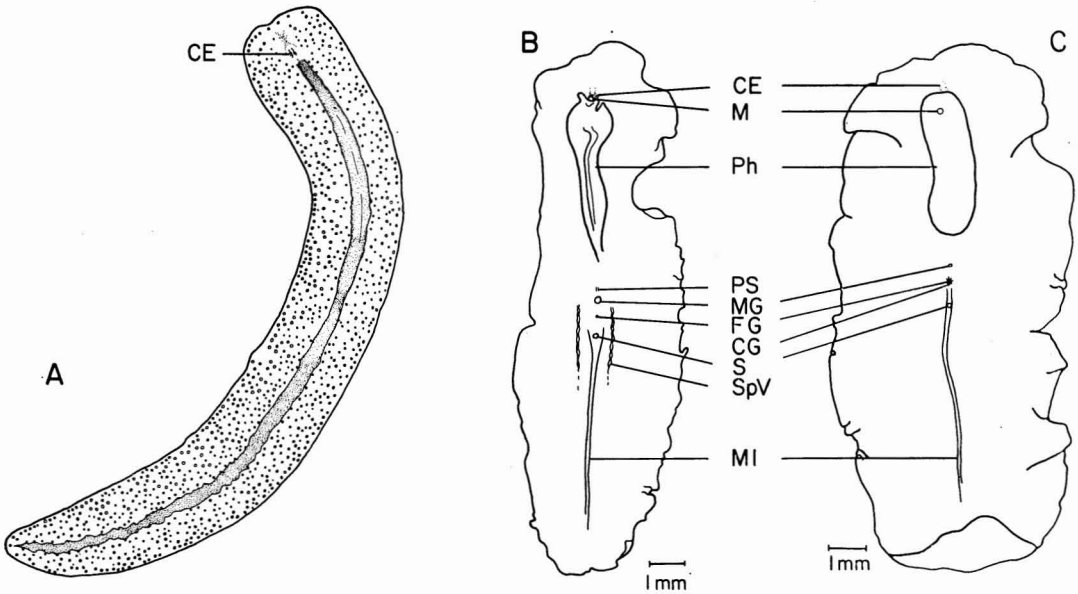

FIGURE 12. *Enchiridium japonicum* Kato, 1943. *A*, living specimen from a photograph; specimen is beginning to crawl extended. Small circles represent brown maculae; stippled band is brown. *B*, *C*, anatomy of cleared specimens. ABBREVIATIONS: CE, cerebral eyes; CG, cement glands; FG, female gonopore; M, mouth; MG, male gonopore; MI, main intestine; Ph, pharynx; PS, penis stylet; S, sucker; SpV, spermiducal vesicle.

Enchiridium Bock, 1913

Enchiridium japonicum Kato, 1943

Figures 12–17, 20

In many respects the Hawaiian specimens conform with the original description. However, because of the difficulties encountered in obtaining the description, the brevity of the description, and anatomical differences discovered in the investigation of the Hawaiian specimens when compared with the original description, a more than cursory report is warranted.

MATERIAL: Four specimens total; one collected 14 April 1961, one 11 June 1961, and two 4 April 1962, at Kupikipikio Point (Black Point) (longitude 157°47′38″ W, latitude 21°18′30″ N), Oahu, Hawaii.

The copulatory region of two specimens was sagittally sectioned and stained with PAS and Mayer hemalum (Pearse 1960). The anterior and posterior portions of one sectioned specimen

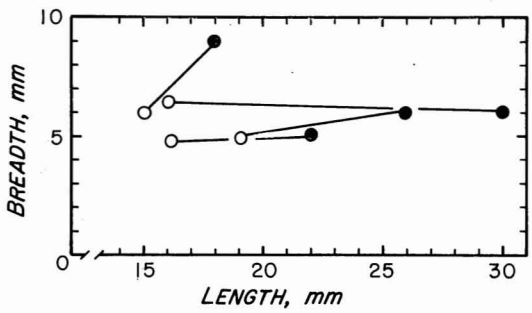

FIGURE 13. *Enchiridium japonicum* Kato, 1943, graphic representation of length: breadth measurements of specimens collected.

SYMBOLS: black circles, living specimens; white circles, fixed specimens.

were mounted. Specimens roll tightly with mercury fixation and the pharynx is sometimes extruded.

DESCRIPTION OF LIVING SPECIMENS: Form elongate, with smooth margins, anteriorly truncate, sometimes with a slight indentation on the anterior midline, posteriorly bluntly cuspate. Length-to-breadth measurements are given in Figure 13. The worms are capable of great elongation with a maximum length-to-breadth ratio recorded photographically of 10:1 although the specimen was not fully extended. Breadth is not appreciably increased in the contracted state but the worms do appear thicker on the dorsoventral axis. Ground color off-white resulting from a lack of pigment and represents color of internal tissues; margin translucent. Cocoa-brown maculae are dispersed over the dorsum, absent on the margin, prevalent and generally larger medially where they coalesce forming a cocoa-brown middorsal band (1.5 to 2.0 mm in width). Anteriorly the band narrows, becoming abruptly discontinuous in the cerebral region; anterior to the cerebral region component maculae become more distinct and terminate short of the anterior margin. The band is posteriorly cuspate and does not reach the posterior margin. Golden yellow pigment spherules punctuate the dorsum, appearing more prevalent laterally but may be obscured medially by brown maculae. Ventral surface off-white. Marginal eyes form a broad band along the anterior margin. Cerebral eyes form paired elliptical aggregations, 1.5 to 2.3 mm (7 to 8 percent of body length) behind the anterior margin. Pharynx long and expands and contracts with body configuration. Copulatory apparatus close behind posterior termination of pharynx.

DESCRIPTION OF PRESERVED SPECIMENS: Form elongate (Figures 12*B*, *C*). Length-to-breadth measurements are given in Figure 13, ratio range 2.5:1 to 3.8:1. Body thick. Color pattern altered following fixation: brown maculae appear less numerous, especially laterally; middorsal band (maximum width, 1.2 mm) clearly retained; golden yellow spherules undetectable. Ground color of dorsal and ventral surfaces light gray.

Margin perimeter eye free up to 80 μm. Marginal eyes (Figure 14*A*) abundant along anterior margin, maximum eye diameter 20 μm. Marginal band uninterrupted across midline, anteriorly forming a broad band (0.8 mm wide). Marginal band width and eye number rapidly diminish posteriorly, with eyes rarely posterior to female gonopore. The marginal band does not completely encircle the margins. Cerebral eyes form paired, roughly triangulate aggregations (Figure 14*B*), 0.45 mm in length, lying 1.0 mm (5 to 6 percent of body length) behind the anterior margin. Each cerebral group comprises about 30 eyes, maximum eye diameter 32 μm, most eyes are of relatively uniform size. A pair of ventral or "deep" eyes is present, located near the more anterior cerebral eyes.

Mouth near anterior extreme of pharynx, 1.6 mm (7 to 8 percent of body length) behind the anterior margin. Pharynx broad, sometimes bulbous anteriorly, tapering posteriorly, length in intact specimens ranges from 23 to 37 percent of body length. Pharynx unites posteriorly with main intestine. Main intestine extends posteriorly, terminating short of margin. Main intestine epithelium columnar, 40 to 55 μm thick, with clavate basophilic gland cells. Gut branches arising from main intestine do not anastomose.

Dorsal epithelium columnar, 35 μm thick, well supplied with rhabdites but with few gland cells. Brown maculae of color pattern represented by dense brown pigment granules lying immediately above basement membrane of epithelium. In areas of dense pigmentation (e.g., the midline), strings of tiny pigment granules

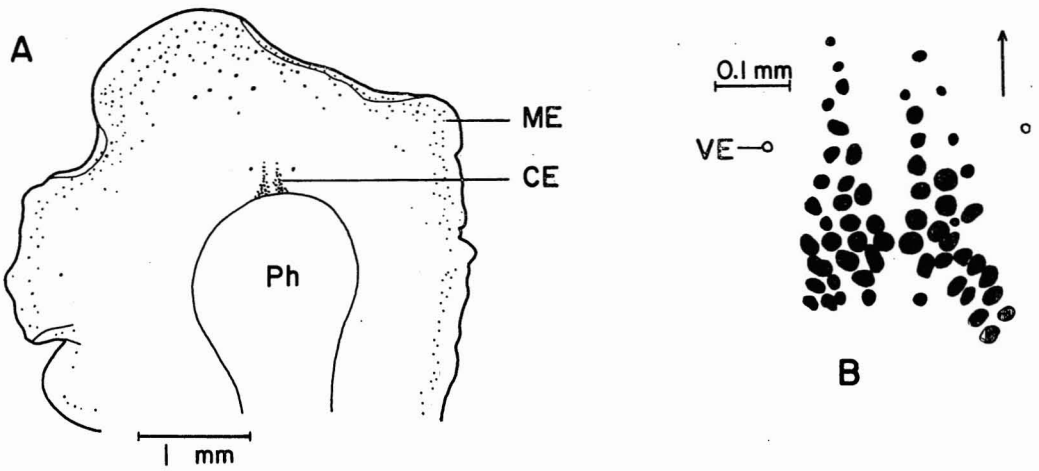

FIGURE 14. *Enchiridium japonicum* Kato, 1943, eye arrangement. *A*, anterior region; *B*, cerebral eyes. ABBREVIATIONS: CE, cerebral eyes; ME, marginal eyes; Ph, pharynx; VE, ventral eye.

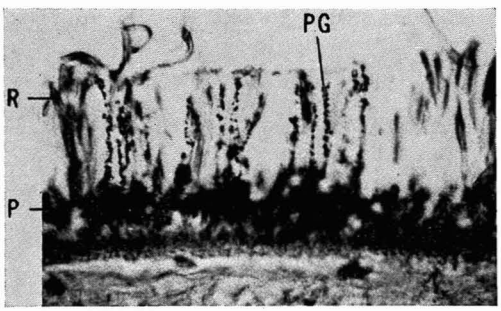

FIGURE 15. *Enchiridium japonicum* Kato, 1943, brown pigmentation of dorsal epithelium and rhabdites (570 ×); sagittal section; stains, PAS and Mayer hemalum. ABBREVIATIONS: P, pigment; PG, pigment granule; R, rhabdites.

are found from the basement membrane to the apices of epithelial cells (Figure 15). Dorsal muscle layers 40 μm thick. Ventral epithelium cuboidal, 11 μm thick; densely ciliated, the cilia one-third the height of epithelial cells. Rhabdites relatively numerous, gland cells few. Ventral muscle layers 75 μm thick.

Testes small, numerous, ventrally situated. Spermiducal vesicles form long chains one on each side of the median line, extending both anterior and posterior to the copulatory region; epithelium of vesicles thickened ventrally, secreting PAS-positive material. A sperm duct from each row of spermiducal vesicles passes dorsally and medially, penetrates the muscle wall of the seminal vesicle near its anterior extent, and discharges independently into the lumen. Seminal vesicle (Figure 16) is 0.58 mm on its long axis, 0.40 mm on its short axis; interior epithelium is thin, and the lumen contains sperm and PAS-positive material. Seminal vesicle narrows anteriorly, forming the ejaculatory duct which almost immediately penetrates the accessory vesicle's hull (Figure 17), and passes within the nonnucleated hull at a uniform distance from the surface until it reaches the ventroanterior region where it emerges and immediately enters the penis papilla. Accessory vesicles paired, orbate, horizontally oriented, one beside the other. The vesicles are 0.33 mm in diameter, composed primarily of nonnucleated muscle and/or connective tissue, 0.11 mm thick, and with nonnucleated fibers binding the vesicles. A thin nucleated epithelium envelops the nonnucleated hull. Lumina of accessory vesicle are 75 μm in diameter, the interior epithelium is thin, nucleated, and ciliated. Similar ciliation has been reported for all other members of the genus except *E. punctatum* Hyman, 1953. Accessory vesicle ducts pass in a ventroanterior direction through the nonnucleated hull and, once emerging, follow a course roughly paralleling the ejaculatory duct, becoming confluent with the ejaculatory duct at the base of the penis papilla. Penis papilla long, curving, lying within the penis pouch, and armed with penis stylet 0.2 mm in length. Penis pouch enlarges distally and receives prostatic secretion. Prostatic

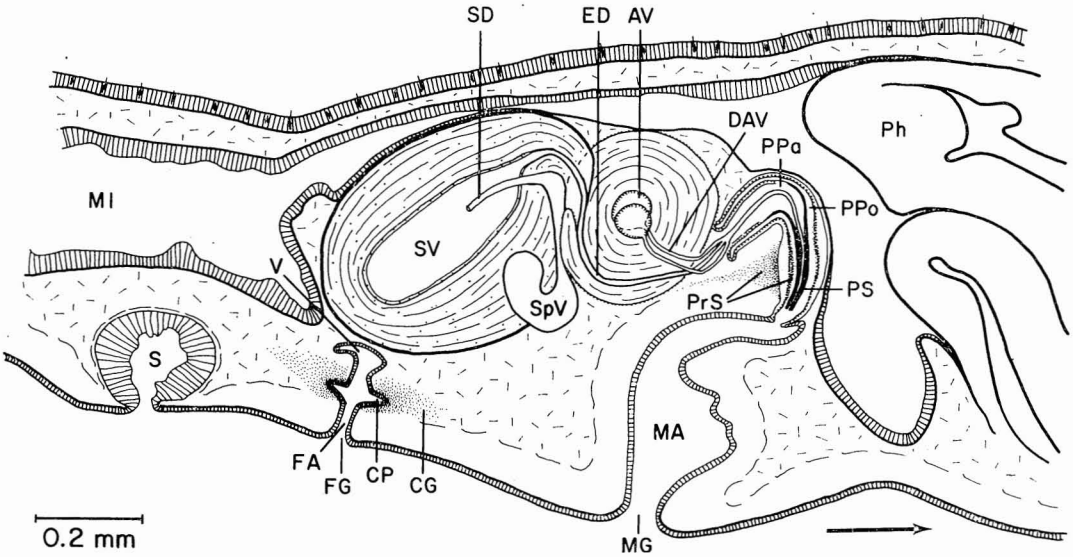

FIGURE 16. *Enchiridium japonicum* Kato, 1943, copulatory region. Only one of a pair of sperm ducts is shown.
ABBREVIATIONS: AV, accessory vesicle; CG, cement glands; CP, cement pouch; DAV, duct of accessory vesicle; ED, ejaculatory duct; FA, female antrum; FG, female gonopore; MA, male antrum; MG, male gonopore; MI, main intestine; Ph, pharynx; PPa, penis papilla; PPo, penis pouch; PrS, prostatic secretion; PS, penis stylet; S, sucker; SD, sperm duct; SpV, spermiducal vesicle; SV, seminal vesicle; V, vagina.

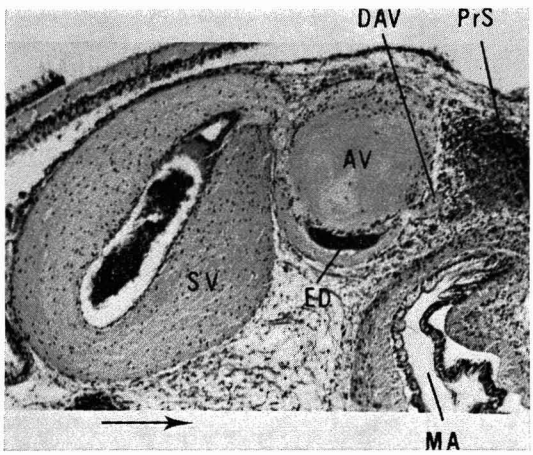

FIGURE 17. *Enchiridium japonicum* Kato, 1943, male copulatory apparatus showing relationship of ejaculatory duct with accessory vesicles (80 ×); sagittal section; stains, PAS and Mayer hemalum.
ABBREVIATIONS: AV, accessory vesicle; DAV, duct of accessory vesicle; ED, ejaculatory duct; MA, male antrum; PrS, prostatic secretion; SV, seminal vesicle.

secretion was found in the mesenchyme surrounding and in the epithelium of the penis pouch but was most abundant in the posterior region. Penis pouch opens into deep male antrum. Antrum heavily muscularized, epithelium cuboidal, 11 μm thick, secretes PAS-positive material. Male gonopore 70 μm in diameter, located 5.4 to 7.4 mm (33 to 46 percent of body length) posterior to anterior margin. The terminal portion of the male system including the seminal vesicle to penis pouch is 0.9 mm in length and lies under the main intestine, with the anterior extent in close proximity to the pharynx.

Only the terminal portion of the female system commencing with the proximal oviducts was clearly evident in both sectioned specimens. Oviducts originate anteriolaterally and discharge at the vagina. Vagina curves ventrally, opening into cement pouch, 76 μm in diameter, which in turn opens into female antrum. Epithelium of female antrum 7 μm thick, secretes PAS-positive material. Female gonopore small, 11 μm in diameter, lies 6.0 to 7.9 mm (37 to 50 percent of body length) behind anterior margin. Terminal portion of female system lies directly under large seminal vesicle.

Sucker 0.27 mm in diameter. Epithelium columnar, 60 μm in height. Underlying musculature appears less well developed than that of

adjacent body wall. Sucker lies 6.7 to 8.6 mm (41 to 54 percent of body length) behind anterior margin.

DISCUSSION: As implied in the description, pharynx length was deceptive in the specimens examined and a wide variation in pharynx length with respect to body length is indicated. At fixation, one specimen (length, 19 mm) had entirely extruded its pharynx and this was folded upon itself in a J-shape, resulting in an apparent length of 25 percent of body length but an actual length of 37 percent relative to body length. The specimen represented in Fig. 12*C* (length, 16.2 mm) was sectioned and it also showed a similar deep J-shape of the posterior, narrow, more inconspicuous, ventrally located portion of the pharynx. This configuration suggests that the pharyngeal pouch usually contracts far more than does the pharynx, which results either in a fold or in expulsion of the pharynx. The only exception was the specimen represented in Figure 12*B* (length, 15.8 mm), and here the posterior termination of the pharynx is slightly everted as depicted in Figure 16. That the degree of folding of the pharynx is of consequence in relation to structures lying posteriorly is graphically illustrated by a comparison of Figure 12*B* with 12*C*. The findings here may be relevant to the differences in pharynx length (expressed as percent of body length) reported for *Enchiridium periommatum* Bock, 1913 (13.3 percent), and *Enchiridium evelinae* Marcus, 1949 (26 to 27 percent). Marcus and Marcus (1968) regarded pharynx length relative to body length as a major diagnostic character distinguishing *E. evelinae* from *E. periommatum*. Other systematic differences between these two species concern cerebral eye number and details of the copulatory apparatus with special emphasis on the morphology of the musculature common to both accessory vesicles.

By definition, Prosthiostomidae assigned to *Enchiridium* have eyes completely encircling the margin, a feature not shared by the specimens at hand. Although juvenile specimens of *E. evelinae* Marcus, 1949, were reported to lack eyes completely encircling the margin (Marcus 1949), it is difficult to reconcile the very small specimens commented on by Marcus with the rather large specimens investigated here, albeit these are smaller than those described by Kato (1943). However, it cannot be presumed that larger specimens will not demonstrate the characteristic feature of eyes completely encircling the margin. In addition, within the Prosthiostomidae, the intimate involvement of the ejaculatory duct with the accessory vesicles that is demonstrated by the specimens investigated here seems to be peculiar to *Enchiridium* and perhaps should be considered in the definition of the genus.

The assertion that the Hawaiian material is conspecific with *E. japonicum* Kato, 1943, requires critical consideration. The following differences were noted. (1) The marginal eyes completely encircle the margin but are "very sparse at the posterior" (Kato 1943); the marginal eyes do not completely encircle the margin in the Hawaiian material. (2) The distance between the cerebral eye groups and the anterior margin equals the length of the cerebral eye groups as photographically and diagrammatically indicated by Kato (1943) and commented on by Marcus and Marcus (1968); the length of the cerebral eye groups is less than one-half this distance in the Hawaiian material. (3) The distance between the pharynx and male gonopore is "widely separated" (Kato 1943); the male system and pharynx are closely associated in the Hawaiian material. (4) The length of the male copulatory apparatus calculated from Kato's (1943) figure (1.7 mm) is nearly double the length found in the Hawaiian material (0.9 mm). Kato's (1943) comment that the male copulatory apparatus lies "directly behind the main intestine" is regarded as a *lapsus calami*. The differences considered may be attributed almost entirely to state of fixation and maturity.

The two forms are similar in general color pattern, arrangement of the anterior marginal and cerebral eyes, and the distinctive morphology and orientation of the copulatory apparatus. These similarities substantially support the contention that the Hawaiian specimens are conspecific with *E. japonicum*.

Enchiridium japonicum is the only known representative of the genus from the Indo-West-Pacific, where it is restricted in its distribution to the tropical west and central Pacific. *E. punctatum* Hyman, 1953*a*, has been reported from southern California and the Gulf of

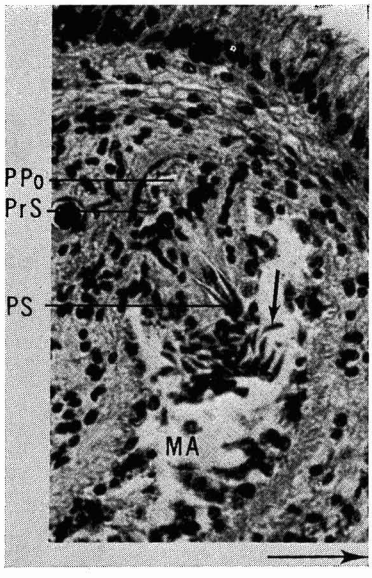

FIGURE 18. *Prosthiostomum* (*Prosthiostomum*) *montiporae*, penis pouch of holotype; sagittal section; stains, PAS and Mayer hemalum. Small arrow indicates sperm at tip of penis stylet (392 ×).

ABBREVIATIONS: MA, male antrum; PPo, penis pouch; PrS, prostatic secretion; PS, penis stylet.

California. Two additional members of the genus occur in the tropical west Atlantic.

DISTRIBUTION: Suô (Su-ao), Taiwan; Oahu, Hawaii.

SPECIMENS: Specimen no. 1-E-2 with the copulatory region sagittally sectioned and the anterior and posterior portions mounted is deposited in the United States National Museum (USNM 52702) together with another entire specimen.

GENERAL DISCUSSION

The yellow to orange-red pigment spherules found *in vivo* in both *Prosthiostomum* (*Lurymare*) *katoi* and *Enchiridium japonicum* are of similar size and have the bright transparent appearance of an oil droplet. The latter characteristic may have led Marcus (1949) to suggest that similar pigments in *Enchiridium evelinae* are composed of a lipoid material "provàvelmente constituído por lipóides" and are subject to alcohol extraction. Hyman (1955) observed similar pigment in the living specimens she identified as *Enchiridium periommatum* Bock, 1913. In the species considered above, the similar pigment spherules do not withstand fixation and alcohol preservation.

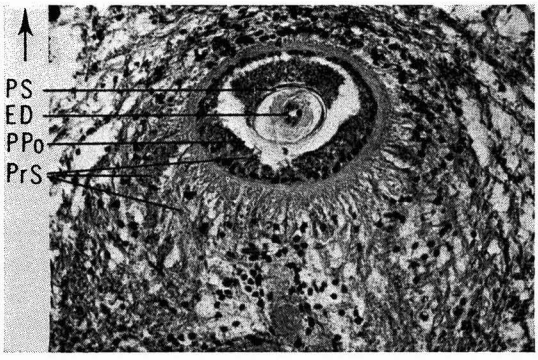

FIGURE 19. *Prosthiostomum* (*Lurymare*) *katoi*, penis pouch of paratype (200 ×); frontal section; stains, PAS, Mayer hemalum, and fast green FCF.

ABBREVIATIONS: ED, ejaculatory duct; PPo, penis pouch; PrS, prostatic secretion; PS, penis stylet.

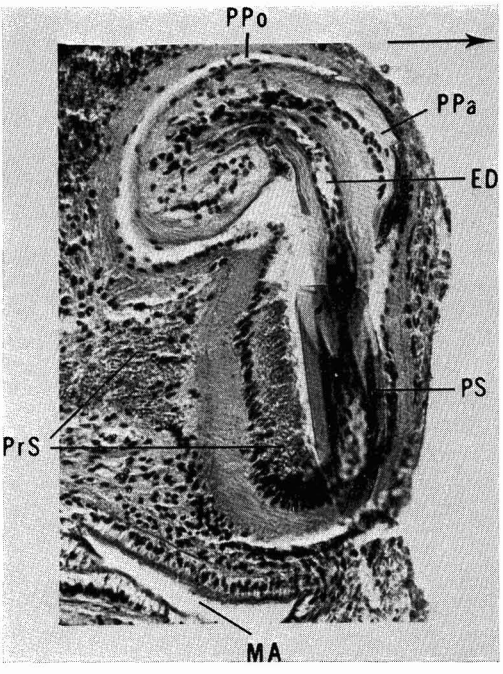

FIGURE 20. *Enchiridium japonicum* Kato, 1943, penis pouch (225 ×); sagittal section; stains, PAS and Mayer hemalum.

ABBREVIATIONS: ED, ejaculatory duct; MA, male antrum; PPa, penis papilla; PPo, penis pouch; PrS, prostatic secretion; PS, penis stylet.

In both *P.* (*L.*) *katoi* and *E. japonicum*, the consideration of a lipoidal composition can apparently be dismissed, since these spherules become undetectable at fixation prior to possible alcohol extraction. The possibility that these pigment spherules represent ovaries is discounted by the lack of these structures in specimens of *E. japonicum* investigated here.

In *Prosthiostomum* (*Prosthiostomum*) *montiporae* the equivocal nature of the prostatic secretion, generally an outstanding feature among prosthiostomids, is perhaps compatible with the miniaturization of the male system in general. A small quantity of possible prostatic secretion may be found in the tiny penis pouch (Figure 18), but the secretion in the dense mesenchyme is indistinct in comparison with the other two prosthiostomids investigated here (Figures 19, 20). The histology of the penis pouch and surrounding mesenchymal tissue is similar among the species investigated here. The luminal boundaries of the epithelium are indistinguishable from the free prostatic secretion of the lumen, primarily due to the deep basophilic staining of the prostatic secretion. Closely placed nuclei define the base of the epithelium. A pellucid region underlies the epithelium (indicated as a muscularis by Lang, 1884, plate 29, figure 6), across which the prostatic secretion passes to discharge into the penis pouch. The pellucid region is smooth on the epithelial (luminal) side but coronated on the mesenchymal side (see Figure 19), with long projections (septa) penetrating the prostatic secretory mesenchyme.

In both *P.* (*P.*) *montiporae* and *P.* (*L.*) *katoi*, the sperm are fusiform, whereas in *E. japonicum* they are decidedly filiform as described by Lang (1884: 221) for *P.* (*P.*) *siphunculus*. The staining characteristics of the sperm of *P.* (*P.*) *montiporae*, *P.* (*L.*) *katoi*, and *E. japonicum* are identical. A central core of chromatin is encapsulated with PAS-positive material. The thickness of this layer of PAS-positive material varies: thick in *P.* (*P.*) *montiporae*; intermediate in *P.* (*L.*) *katoi*; thin in *E. japonicum*. Where hematoxylin and a counterstain are used, the strands of chromatin do not appear to be closely packed as is typical of many other polyclads. The PAS-positive material is accumulated by the sperm in the testes follicle during spermiogenesis, and this process is easily followed in *P.* (*P.*) *montiporae* where the sperm are larger (see Figure 18) than those of *P.* (*L.*) *katoi*, or of *E. japonicum* where they are smallest.

BIOLOGY AND ECOLOGY OF SPECIES INVESTIGATED AND A NOTE ON *Stylochoplana inquilina* HYMAN, 1950

Observations on prosthiostomids in the field and laboratory are here restricted to *Prosthiostomum* (*Lurymare*) *katoi* and *Enchiridium japonicum*. However, the association of *Prosthiostomum* (*Prosthiostomum*) *montiporae* with the coral *Montipora verrucosa* is of interest with respect to the microtomized worms and a tissue preparation of *M. verrucosa* fixed and processed for histological examination in the same manner as were specimens of *P.* (*P.*) *montiporae*.

Prosthiostomum (*Prosthiostomum*) *montiporae*

A number of polyclads reportedly are associated with coelenterates, but predation has been inferred for only a few by the presence of nematocysts in the epithelium and/or gut of these polyclads. These predators include *Stylochoplana tarda* (Graff, 1878); *Anonymus virilis* Lang, 1884; *Chromoplana bella* Bock, 1922; and *Amyella lineata* Bock, 1922. Bock (1922) suggested that hydroids are the source of the nematocysts found in *C. bella* and *A. lineata*. Recently, Karling (1966) reinvestigated and reported on Bock's material of *C. bella*, which is included in a general review of nematocysts found in turbellarians.

Stylochoplana inquilina Hyman, 1950, which participates in a hermit carb–anemone complex, is also suspected of preying on a Zoantharia. I discovered spirocysts (mean length, 25 μm; mean breadth, 3.3 μm) in the gut and syncytial gut epithelium when examining Hyman's sectioned paratype (Figure 21). There is a distinct possibility that *Calliactis armillatas* Verrill, 1928, the anenome in the complex, is the source of these spirocysts. The worm may ingest them accidently along with food stolen from *C. armillatas* or may prey directly on the anemone. A more comprehensive study of the relationships between *S. inquilina* and *C. armillatas* could be illuminating.

Four Indo-West-Pacific prosthiostomids have

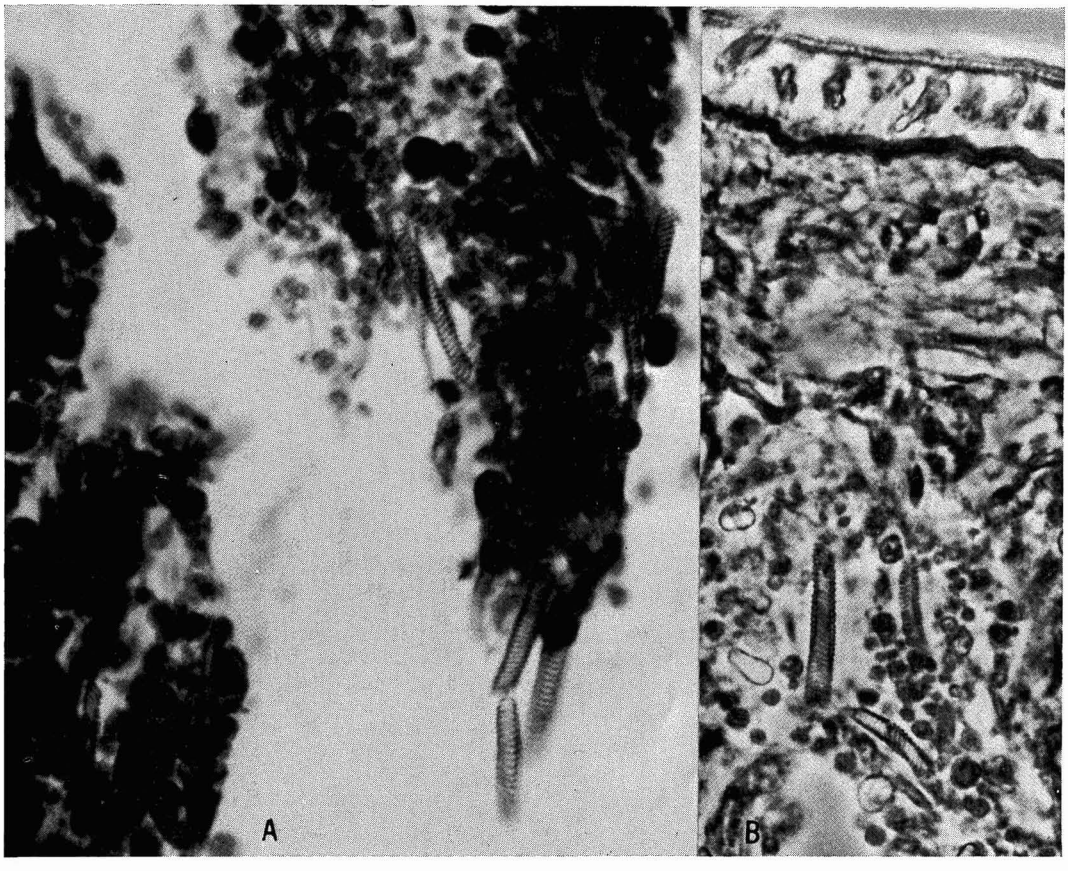

FIGURE 21. *Stylochoplana inquilina* Hyman, 1950, spirocysts in gut of paratype. *A*, 1800 ×; *B*, phase contrast (920 ×).

been reported in association with Zoantharia but the associations are decidedly nebulous. Three specimens of *P.* (*P.*) *exiguum* Hyman, 1959, were taken from "coral rock," presumably not living coral. A single specimen of *P.* (*P.*) *sonorum* Kato, 1938, was dredged from a depth of 10 fathoms "along with some corals" but the condition of the coral was unreported. More specific associations with Madreporaria or true stony corals are found with the single specimen of *P.* (*P.*) *lineatum* Meixner, 1907, which was found on a *Porites*, and the two specimens of *Amakusaplana ohshimai* Kato, 1938, were taken on madreporarians.

Evidence of predation by *P.* (*P.*) *montiporae* on the madreporarian *Montipora verrucosa* appears substantial. (1) Tissue comparable with that of the sectioned preparation of *M. verrucosa* was found within the lumen of the main intestine of one sectioned paratype of *P.* (*P.*) *montiporae*. (2) In the gut of *P.* (*P.*) *montiporae*, brownish zooxanthellae (Figure 22) were found in profusion either in the gut lumen or within the syncytial gut epithelium. Both normal appearing and empty cell walls (presumably digested cells) of zooxanthellae were found. The zooxanthellae of both the coral and polyclad are identical with respect to mean size (8 μm) and distinctive staining characteristics, including the nucleus and a PAS-positive structure. The presence of zooxanthellae is the probable result of ingesting coral polyps. (3) The PAS-positive secretion elaborated in the ventral mesenchyme and discharging onto the ventral surface of *P.* (*P.*) *montiporae* (see Figure 4) probably represents a viscous mucous glycoprotein. This secretion may offer protection from nematocysts and is one of several possible defense

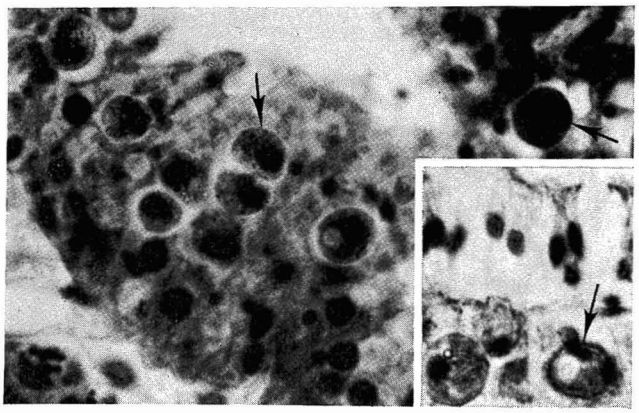

FIGURE 22. *Prosthiostomum* (*Prosthiostomum*) *montiporae*, zooxanthellae in gut epithelium (950 ×). Tissue of *Montipora verrucosa* inset in lower right (950 ×). Arrows indicate a few of the zooxanthellae. Stains, PAS and Mayer hemalum.

mechanisms suggested by Salvini-Plawen (1972) for species that ingest cnidarians. (4) It is possible that the cleft pharynx of *P.* (*P.*) *montiporae* relates to a specialized feeding habit and may be employed as a typical tubular pharynx or, once protruded, may be opened along the deep cleft and spread over a broad or uneven area for feeding. Histological evidence regarding predation on *M. verrucosa* by *P.* (*P.*) *montiporae* and various other aspects of life history of *P.* (*P*). *montiporae* have been investigated and reported on by Jokiel and Townsley (1974).

The corresponding *in vivo* coloration of the gut (exclusive of pharynx, main intestine, or the proximal portion of the gut branch above the pharynx) of *P.* (*P.*) *montiporae* with the *in vivo* coloration of polyps of *M. verrucosa* is attributed to zooxanthellae, a symbiont of corals in general. While symbiotic associations of zooxanthellae with turbellarians are not uncommon (Hyman 1951), this type of association is unknown among polyclads. The evidence of possible digestion of zooxanthellae by *P.* (*P.*) *montiporae* and the restriction of zooxanthellae to epithelial gut tissue and their conspicuous absence from mesenchymal tissue indicate that a symbiotic association probably does not obtain in this polyclad.

Nematocysts were sought in the histological preparations of both *P.* (*P.*) *montiporae* and *M. verrucosa*. Although hematoxylin staining, phase optics, and the Feulgen nucleal reaction (Pearse 1960) were employed to differentiate between basophilic nuclei and basophilic nematocysts, nematocysts were not clearly identified either in the coral or the worms. This may have been the result of harsh treatment with hot corrosive sublimate at fixation.

Prosthiostomum (*Lurymare*) *katoi*

Specimens are commonly found in the spring and summer, and many more were observed than collected. These worms are found under rocks either of inorganic or organic origin on a broken rock substratum at depths ranging from 14 to 100 cm at low tide. The rocks are located on the reef platform well removed from the reef front in areas of mild wave action but subject to tidal interchange. These rocks usually harbor the rock oyster *Ostrea sandvichensis* Sowerby, 1871, on their sides and undersides. The distribution of these worms is not entirely limited to the range of *O. sandvichensis* and specimens are found occasionally under rocks where the rock oysters are absent. These polyclads are solitary and are found among the shells or hidden in the empty shells of *O. sandvichensis*. It is not an obvious predator of the oysters and did not effectively invade living oysters under laboratory conditions. Other prosthiostomids have been reported to be associated with oysters, and these include *P.* (*P.*) *ostreae* Kato, 1937, found with "cultivated oysters" (Kato 1937) and *P.* (*P.*) *lobatum* Pearse, 1938 (Pearse and Wharton 1938).

These worms do not swim, and locomotion is restricted to gliding which may be rapidly executed. Starting from a resting or contracted state, the anterior portion begins to expand on the anterior-posterior axis while the posterior region remains stationary. Expansion continues until the posterior region is affected and begins to move. On a number of occasions these worms were observed to pursue *Thysanozoon tentaculatum* (Pease, 1860) and another species of *Thysanozoon*, apparently by following the mucous path of the thysanozoons. Since the thysanozoons are usually larger (maximum length, 35 mm; breadth, 15 mm) than are *P.* (*L.*) *katoi* (maximum length, 28 mm; breadth, 6 mm), this appears unusual. When this prosthiostomid contacts a *Thysanozoon*, its pharynx is quickly protruded and a portion of the margin of the *Thysanozoon* is ripped off with a backward motion and ingested. The attack was never lethal to the thysanozoons, which may swim to avoid further attacks. Histological evidence of these encounters is found in the sectioned prosthiostomids where rhabdites that are longer and stouter than those of *P.* (*L.*) *katoi* and are comparable to those found in the dorsal epithelia of thysanozoons preyed upon are found in the syncytial gut epithelium. A species of *Pseudoceros* is commonly found in the same area during winter months when *P.* (*L.*) *katoi* is rarely seen. It cannot be assumed that predation by *P.* (*L.*) *katoi* is limited to Pseudoceridae and other small invertebrates, and invertebrate egg masses upon which specimens of *P.* (*L.*) *katoi* often were found "resting" were probably consumed. A species of *Stylochus* also common where *P.* (*L.*) *katoi* and the rock oysters are abundant was never noted to be attacked by *P.* (*L.*) *katoi*, this due perhaps to the firm consistency of the stylochid's body.

In an unexpected laboratory observation, I saw a specimen of *P.* (*L.*) *katoi* consumed by a small (length, 50 mm preserved) spiny puffer fish, *Diodon hystrix* Linnaeus. The polyclad was dropped into a bucket with the puffer and, before the worm had reached the bottom, the fish had ingested it and similarly consumed two more worms in the next 10 minutes. The puffer was kept for a week and demonstrated no detectable ill effects. This incident may not reflect "usual" predation by the spiny puffer but does indicate that not all fish regard all polyclads as distasteful (see Hyman 1951: 194).

Although internal fertilization has been reported by Kato (1940) for *P.* (*P.*) *auratum* Kato, 1937, and was inferred with regard to *P.* (*P.*) *ostreae* Kato, 1937, in *P.* (*L.*) *katoi*, the evidence does indicate hypodermic impregnation; sites of recent hypodermic impregnation have been detected in sectioned material and sperm have been found free in the mesenchyme. Haswell (1907) reported evidence of hypodermic impregnation in *P.* (*P.*) *maculatum* Haswell, 1907. In neither *P.* (*P.*) *montiporae* nor *E. japonicum* was evidence of hypodermic impregnation discovered.

Enchiridium japonicum Kato, 1943

These worms were found under relatively "clean" basalt rocks at and below a depth of 60 cm at low tide. The rocks lie at a cliff base in line with the reef front in the breaker zone. These rocks form part of a large, tightly wedged basalt rubble. The area can be collected only under extremely reduced wave conditions. *Pericelis hymanae* Poulter, 1974, is found in the same area at Kupikipikio Point. The mollusk *Isognomon* is represented by a few scattered specimens, and patches of *Sargassum* are found among the rocks. Unlike *Pericelis hymanae*, *E. japonicum* does not obviously use the undersides of shells of *Isognomon* as a refuge.

Locomotion in *E. japonicum* is similar to that in *P.* (*L.*) *katoi*. Although one of the sectioned specimens had recently fed, the tissue in the pharynx and digestive system yielded no distinctive morphological structures. Digestion as in *P.* (*P.*) *montiporae* and *P.* (*L.*) *katoi* is intracellular and seems to occur in the syncytial gut epithelium.

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