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TO THE FUNNEL-LIKE STRUCTURE WITHIN
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**CONTRIBUTIONS TO THE JAPANESE ASCIDIAN FAUNA XXXV.
MOLGULA HOZAWAI OKA, 1932 AND M. VERRUCIFERA
RITTER AND FORSYTH, 1917, WITH SPECIAL REFERENCE
TO THE FUNNEL-LIKE STRUCTURE WITHIN THE SIPHON**

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With Text-figures 1-5 and Table 1

Molgula hozawai Oka, 1932 has been described only once on the two specimens obtained from near Asamushi, Mutsu Bay (Oka, 1932, 1935). Recently, a considerable number of specimens of a molgulid, collected from Tottori on the Japan Sea coast, Tan'nowa in Osaka Bay and Gobo on the west coast of Kii Peninsula, were examined, and all of these were proved to be referable to *M. hozawai*, though some variation was found in their structure of gonads and a remarkable structure seen inside the siphons of these specimens was left out of comparison, as this structure is not mentioned in the description of that species. In the majority of examined specimens, the branchial and atrial vela are developed inside the branchial and atrial siphons respectively into a funnel-like structure extended outwards, thus a duplicate internal siphon is formed within each siphon of the mantle body. To check these points, re-examination of the type specimens of *M. hozawai* was planned but in vain, as these were lost unfortunately. Then, it was tried to obtain some specimens referable to this species again from its type locality, Mutsu Bay, but this was not successful either.

On the other hand, general structure of the present specimens, and therefore of *M. hozawai*, resembles closely that of *Molgula verrucifera* Ritter and Forsyth, 1917 occurring on the coast of California, though nothing is described in this species on the internal siphons. Very fortunately, through the courtesy of Drs. H. S. Feinberg and E. Kirsteuer, four specimens of *M. verrucifera*, deposited at the American Museum of Natural History, were offered to me for closer observations. The existence of similar internal siphon was confirmed in these specimens and the relation between the present specimens and *M. verrucifera* has seemingly become much closer. The present paper is to give the full description of the present specimens, keeping in mind a possibility that these specimens, and then *M. hozawai*, might be identical with *M. verrucifera*, and to make some considerations on the structure of the internal siphon.

Before going further, I would like to express my hearty thanks to Mr. Tetsuo Kuwamura, Seto Marine Biological Laboratory and Dr. Mitsuaki Nakauchi, Kochi

University, who gave me a chance to examine a number of specimens, to Drs. Harold S. Feinberg and Ernst Kirsteuer of the American Museum of Natural History for their kindness in sparing for my study the specimens of *M. verrucifera* deposited at the museum, to Drs. Hiroshi Watanabe and Koichi Sekiguchi and Mr. Tetsuo Iwami, the University of Tsukuba, and to Dr. Makoto Tsuchiya, Tohoku University, for information about the type specimens of *M. hozawai*, and lastly to Dr. Takasi Tokioka for his precious advices and critical reading of the manuscript.

Molgula hozawai Oka, 1932

(Figs. 1–3 and 5)

Molgula hozawai—Oka, 1932, pp. 518–520, 2 figs.; 1935, pp. 430–432, 2 figs.

Material examined: Eighty-nine specimens shown in Table 1 were examined for the present study. Sixty-three specimens from Tottori were all found attached to the nearly vertical wall of moles, in the intertidal zone, built at Torigashima Islet about 500 m off Karo-cho near the estuary of River Sendai, Tottori Prefecture, and presented to me by Mr. Kuwamura and Dr. Nakauchi, while twenty-five specimens from Tan'nowa, Osaka Prefecture, were collected in the subtidal zone, about 2 m below the mean sea level, near the estuary of River Ban, and offered to me by Dr. Nakauchi. The last single one is the specimen obtained by myself on May 3, 1978 during the benthos survey, at a 3 m deep off Nada district of Gobo, Wakayama Prefecture and listed already in the ascidian fauna around Kii Peninsula (Nishikawa, 1980).

Description: Body roundish. Each siphon usually shrunk to only a low wart on the

Table 1. Characters in the specimens of *Molgula hozawai* Oka examined in the present study.

l: type with less developed testicular follicles. i: an intermediate type. w: type with well developed testicular follicles. +: present. —: absent. For grades I–III see the text (p. 344).

*1: Including the specimens partly matured. *2: Including a specimen with gonads much injured. *3: Including one or two specimens with the funnel inverted as seen in Fig.

5, B. *4: Including a single 12.0mm long specimen with spent(?) gonads.

Locality and date		Number of specimens examined	Body length (mm)	Number of immature specimens *1	Number of specimens with the gonad of type			Number of specimens with the funnel-like structure in the grade			Tailed larvae in the peribranchial cavity
					l	i	w	I	II	III	
Tottori	Oct. '77	4	1.2-7.0	4	0	0	0	1	2	1	—
	Dec. '77	7	3.0-9.0	4	2	1	0	1	0	6	—
	Feb. '78	13	4.5-10.9	7	0	0	6	0	0	13	+
	June '78	1	5.0	1	0	0	0	0	0	1	—
	Dec. '78	25 *2	4.8-9.7	6	11	7	0	3	5	17 *3	+
	Mar. '79	13	6.5-12.0	4 *4	0	0	9	0	0	13 *3	+
Tan'nowa Nov. '78		25	2.9-11.0	5	0	0	20	1	0	24 *3	+
Gobo May '78		1	6.0	0	1	0	0	0	0	1	—
Total		89		31	14	8	35	6	7	76	

dorsal surface; branchial siphon terminal, while the atrial sub-terminal or nearly in the middle of the body. Test surface wholly and densely coated, though not impregnated, with sand grains; numerous, sometimes rather small number of, short delicate test projections nearly all over the surface also gathering the grains. Test itself is very thin, tough, transparent and colorless.

Mantle musculature consists of fine muscle fibers, densely but randomly distributed all over the mantle body except both siphons, and thicker muscle bundles radiating from both siphons and usually confined to the siphonal area, but rarely extending to the middle on each lateral side. Margin of each aperture always cut into up to 20 lobules. Branchial and atrial vela well developed respectively to form in many specimens a funnel-like structure or an internal siphon mentioned in detail later. No atrial tentacles.

Fig. 1. *Molgula hozawai* Oka from Tan'nowa (9.3mm long specimen). Mantle body, right (A) and left (B) side.

Seven branchial folds on each side; the branchial formula are:

6.0 mm long specimen from Gobo

L. D. 0 (3) 0 (6) 0 (7) 0 (6) 0 (7) 0 (6) 0 (4) 0 V.

R. D. 0 (3) 0 (5) 0 (6) 0 (6) 0 (7) 0 (6) 0 (4) 0 V.

7.0 mm long one from Tottori (collected in Dec., '78)

L. D. 0 (3) 0 (5) 0 (6) 0 (6) 0 (5) 0 (5) 0 (4) 0 V.

R. D. 0 (3) 0 (5) 0 (6) 0 (6) 0 (6) 0 (5) 0 (3) 0 V.

10.3 mm long one from Tan'nowa

L. D. 0 (4) 0 (6) 0 (7) 0 (7) 0 (6) 0 (6) 0 (4) 0 V.

R. D. 0 (4) 0 (6) 0 (6) 0 (7) 0 (7) 0 (6) 0 (5) 0 V.

11.5 mm long one from Tottori (collected in Mar., '79)

L. D. 0 (4) 0 (6) 0 (6) 0 (7) 0 (6) 0 (5) 0 (5) 0 V.

R. D. 0 (4) 0 (6) 0 (6) 0 (7) 0 (6) 0 (5) 0 (5) 0 V.

Basically there are 5 thick transverse vessels and 6 transverse rows of infundibula;

under each fold only a single infundibulum is inserted between each pair of transverse vessels in smaller specimens or in the dorsal part of branchial sac in larger ones, but two infundibula are inserted there in larger specimens or sometimes in the ventral part of branchial sac in smaller ones; the apex of each infundibulum usually divided into two summits. Stigmata nearly straight in the interspace. Tentacles ca. 15 in smaller specimens, but 20 to 30 in larger ones exclusive of several minute simple papillae; branched in two orders; generally larger and smaller ones alternating almost regularly. Ciliated groove a longitudinal slit or an oval laterally compressed. Dorsal lamina with smooth margin. First intestinal loop very narrow, and the second loop very wide and shallow.

Fig. 2. *Molgula hozawai* Oka. Gonads. A-D: Immature gonads, attachment surface (A, B) and free surface (C, D). E-I: Matured gonads, free surface (above) and attachment surface (below); E-F: type with less developed testicular follicles; G-H: intermediate type; I: type with well developed testicular follicles. J: Optical longitudinal section of I. A: Specimen from Tottori (Oct., '77), 6.5mm long. B: Tottori (June, '78), 5.0mm. C: Tottori (Dec., '78), 4.8mm. D: Tottori (Dec., '77), 9.0mm. E: Gobo, 6.0mm. F: Tottori (Dec., '78), 7.0mm. G: Tottori (Dec., '78), 8.5mm. H: Tottori (Dec., '77), 6.0mm. I and J: Tottori (Feb., '78), 10.3mm.

Gonads are matured even in specimens ca. 5 mm long. Left gonad in the second intestinal loop, while the right one anterior to the renal sac (Fig. 1). Each gonad consists of an oval ovary and some to numerous testicular follicles gathered at the posterior end of ovary (Fig. 2). Male genital aperture is situated very close to the aperture of short oviduct, vas deferens straight and running along the median line on the free surface of ovary (Fig. 2 C, D, E). Ovarian eggs up to 200μ in diameter. Tailed larvae with trunk 250 to 275μ in length (Fig. 3 A, B); very rarely even the larvae in metamorphosis and with several ampullae may be found in the peribranchial cavity (Fig. 3, C-E).

Remarks: In the present material, the number of testicular follicles and then their arrangement are markedly variable seen in Fig. 2, but the variation is quite continuous and seemingly no correlation is found between the developmental grade of testicular follicles and body size. For convenience' sake, the developmental degree

Fig. 3. *Molgula hozawai* Oka. Larvae, usual one (A), unusual (B) and those in metamorphosis (C-E). A: From 10.0mm long specimen from Tottori (Mar., '79). B: From 7.0mm specimen, Tottori (Dec., '78). C and D: From 4.8mm specimen, Tottori (Dec., '78). E: From 11.0mm specimen, Tan'nowa.

of testicular follicles may be divided into the following three grades: Type w (well developed) representing the grade with a number of follicles radially arranged (Fig. 2 I), Type l (less developed) showing the state with only a limited number of follicles arranged transversely (Fig. 2 E, F) and Type i (intermediate) showing intermediate grades between these two (Fig. 2 G, H). As seen in Table 1, there are seemingly some trends in the relative frequency of respective types according to seasons and localities, though less clearly in the latter. Therefore, the variation itself may be safely regarded at present as intraspecific, but it might reflect some significant factors in the environment or with the aid of future more crucial studies might suggest further subdivision of the taxon.

Molgula hozawai described by Oka is the only species from the Japanese waters, provided with the gonads of the same structure as the Type w in the present specimens. All other morphological characters are shared by *M. hozawai* and the present specimens, except that the former is furnished with very long "hair-like processes of the test" carrying sand grains (Oka, 1935, p. 430) and provided with the ciliated groove "horse-shoe shaped" and open posteriorly (ibid., p. 431). These differences are, however, probably of minor taxonomic significance. For these reasons, the present specimens may safely be identified with *M. hozawai*, though the structure of internal siphon is put aside from checking.

Considerations

Four specimens of *Molgula verrucifera*, deposited at the American Museum of Natural History were examined closely. This species was first described on the several specimens collected "in the littoral zone, La Jolla, California" (Ritter and

Forsyth, 1917, p. 447). The four specimens treated here were collected from "among the roots of sea weeds on surf-beaten rocks on the ocean side at Corona Del Mar, California" (No. 1574, 6.3 mm and ca. 9 mm long specimens, July 3, 1939; No. 1575, 9.3 mm long specimen, July 3, 1939) or found "as well as attached to other ascidians dredged a short distance off shore in that locality" (No. 1576, ca. 7.5 mm long one, 1939), and were already described by Van Name (1945, pp. 414-415). Some additional features of these specimens are given below:

Fig. 4. *Molgula verrucifera* Ritter and Forsyth. A-C: Ciliated grooves. D-F: Gonads, free surface (above) and attachment surface (below). A and E: 6.3mm long specimen (No. 1574). B and F: 9.3mm (No. 1575). C: ca. 7.5mm (No. 1576). D: ca. 9.0mm (No. 1574).

The structure of internal siphons is more or less distinct in these four specimens. Ciliated groove a longitudinal slit or an oval laterally compressed (Fig. 4 A-C). Gonads matured already in all of them (Fig. 4 D-F). Ovarian eggs up to 150μ in diameter, a little larger than $130-140\mu$ given by Berrill in the same species (1931, 1937). The sperm duct opens very close to the aperture of oviduct. Many tailed larvae with the trunk $200-210\mu$ long were found in the peribranchial cavity, except in No. 1576 specimen ca. 7.5 mm in length; but no larvae with ampullae as shown by Berrill in this species (1931, p. 298) were not detected in any of the four specimens. Evidently, this species is related very closely to *M. hozawai*, especially to the specimens of Type 1, with the gonad furnished with much fewer testicular follicles, except that the ovarian eggs and then the trunk of larvae are smaller in the former. Probably these two might be identical with each other and belong to the single, same and amphi-Pacific species. However, I am hesitating at present to take this thought conclusively, as the variation in the size of ovarian eggs is not yet studied fully in Japanese specimens and further the difference in egg size might be followed by some differences of taxonomic significance in the course of metamorphosis.

There are four other species that are similar to *M. hozawai*, of the Type w with well developed testicular follicles, in the structure and position of gonads, the course of alimentary canal and the structure of branchial sac: *M. amokurae* Bovin, 1922 from

New Zealand (Bovin, 1922, pp. 34–36), *M. enodis* Sluiter, 1912 from the Antarctic (Sluiter, 1912, pp. 452–453; 1914, pp. 4–6), *M. sabulosa* (Quoy and Gaimard, 1834) from East Australia (Kott, 1976a, pp. 86–87; Millar, 1975, pp. 320–321) and *M. tumulus* (Quoy and Gaimard, 1834) from Southeastern Australia (?) (Pizon, 1898, pp. 366–367). All these species come from the southern hemisphere and thus the locality is so far apart between these species, and *M. hozawai* and *M. verrucifera*. However, a symmetrical distribution of some boreal form in the antiboreal waters might be not impossible, especially in the case when such a form was first introduced there by vessels. At least, the taxonomic re-examination of these species may be urged by the present studies on the two boreal forms.

Notes on the Funnel-like Structure within the Siphon

In many of the examined specimens of *M. hozawai*, the funnel-like structure or the internal siphon is formed with its base along the anterior border of the tentacular ring within the branchial siphon and generally directed outwards (Fig. 5 A), though very rarely inwards (Fig. 5 B). From this relative position, the funnel may safely be regarded as nothing but the velum itself markedly developed inside the branchial or atrial siphon. Actually, in some specimens the branchial and atrial vela are found remaining in a state well developed but not so much to form a funnel inside the siphon. As seen in Fig. 5 C showing a section of this structure illustrated rather schematically on the series of sections, the funnel consists only of circular muscles; the longitudinal muscle-like images seen under the stereomicroscope are probably, judging from the Nomarskii-images, attributable to partial thickening of the epithelial layer.

Fig. 5. *Molgula hozawai* Oka. Funnel-like structure within the siphon. A: Optical longitudinal section of the branchial siphon, the structure directed outwards. B: Do., directed inwards. C: Schematic representation of the structure showing the muscle arrangement. br. s.: branchial siphon. circ. m.: circular muscles. f.: funnel-like structure. long. m.: longitudinal muscles. t.: tentacle.

The following three grades may be defined in the formation of this structure:

I: Branchial and atrial vela well developed, but not so much to form a funnel.

II: Either branchial or atrial velum developed to a funnel.

III: Both vela developed respectively to a funnel.

Formation of this structure seems to be general and stable regardless of seasons and localities (see Table 1). Further, no correlation is seen between the development of this structure and the body size.

So far as I am aware, the development of the branchial and/or the atrial velum to a distinctly funnel-like structure within the siphon has not been reported yet, though the strong development of the branchial and/or the atrial velum is noted in many species such as *Polycitorella mariae* Michaelsen (Michaelsen, 1924, p. 282 and fig. 6), *Pyura ganglion* (Savigny) (Michaelsen, 1919, p. 27 and fig. 6), *P. pantex* (Savigny) (Michaelsen, 1919, p. 23), *P. vittata* (Stimpson) (Monniot, 1972, fig. 8, B), *Molgula amesophleba* (Codreanu and Mack-Fira) (Monniot, 1969, p. 243 and fig. 35, D), *M. bancalis* Monniot (Monniot, 1970, p. 353 and fig. 4, C; Monniot, 1978, fig. 14, B, C), *M. bleizi* (Lacaze-Duthiers) f. *typica* (Monniot, 1969, p. 223 and fig. 24, C), *M. complanata* Alder and Hancock (Monniot, 1969, p. 237 and fig. 32, C), *M. diversa* Kott (Kott, 1972, p. 252), *M. exigua* Kott (Kott, 1972, p. 249), *M. herdmanni* Bjerkan (Monniot and Monniot, 1979, fig. 3, A), *M. manhattensis* (De Kay) (Kott, 1976b, p. 451), *M. roulei* Monniot (Monniot, 1969, p. 249), *M. setigera* Ärnäck-Christie-Linde (Monniot, 1978, p. 211 and fig. 15, F) and *M. tubifera* Örstedt (Monniot, 1969, p. 203 and fig. 13, C).

The funnel-like structure may be reminiscent of the specialized apertures on the mantle body of *Stomozoa murrayi* Kott (Kott, 1957, pp. 131–134, figs. 2–8; Millar, 1977, pp. 169–174, figs. 1–2), *Clavelina roseola* Millar (Millar, 1955, pp. 183–185, fig. 13; Millar, 1962, pp. 139–140) and *Clavelina dentatosiphonis* Millar (Millar, 1975, pp. 211 and 213–214, fig. 7). In each of these three species, however, it is suggestible that the original siphon is surrounded externally by some mantle projections that will be further extended and fuse one another to form a circular external siphon. This process may be seen in the following two molgulids. Such mantle projections are not so well developed in both *Molgula janis* Kott (Kott, 1952, p. 295) and *Molguloides immunda* (Hartmeyer) (Kott, 1969, p. 158 and fig. 220), but several mantle projections are extended respectively into the hollow test extensions around respective apertures. If this is true, the resemblance in the structure of siphons between *M. hozawai* and *M. verrucifera*, and the three species mentioned above (*S. murrayi*, *C. roseola* and *C. dentatosiphonis*) is to be regarded as not homology but analogy. Further closer examinations of the structure under consideration in respective species referred to here are urged. Validity of the genus *Stomozoa* and the subfamily Stomozoinae of the family Clavelinidae, defined by Kott (1957), should be discussed again when these examinations are over (see Millar, 1962, p. 140).

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