CONTRIBUTIONS TO THE JAPANESE ASCIDIAN FAUNA XXXVI. A NEW SPECIES OF *ADAGNESIA* (FAMILY AGNESIIDAE) FROM THE JAPAN SEA, WITH A NOTE ON ITS STRANGE STRUCTURE, "EPIDERMAL VESICLE"

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With Text-figures 1-5

More than 60 specimens of a strange stalked simple ascidian were submitted to the author for identification by Mr. Tsutomu Shin'ya of the Noto Marine Biological Laboratory, Kanazawa University. These specimens were collected in 1981 by Mr. Shin'ya by dredging at a depth of 20 m in the northern part of Toyama Bay in the Japan Sea, far south off Noto-Ogi on the south coast of Noto Peninsula.

Closer examinations on these led the author to the conclusion that they belong to a new species of the genus *Adagnesia* Kott that is represented here for the first time from the western North Pacific. Further, a strange structure, "the epidermal vesicle", was detected at the base of the stalk on all the specimens examined. Seemingly, this structure might be related to the asexual reproduction by budding of a manner of so far unknown type.

Before going further, the author would like to express his cordial thanks to Mr. Shin'ya for this very interesting material and to Dr. Takasi Tokioka for his critical reading of the manuscript.

Adagnesia vesiculiphora n. sp.

(Figs. 1-5)

Type-series: Holotype (SMBL Type No. 330), 11.8 mm \times 8.1 mm body with 45 mm long stalk, chosen out of the sixteen specimens of the type-series picked up rather randomly and including the largest and the smallest bodies among the material. All of the specimens of the type-series matured; dredged at 37°20.6'N and 137°16.8'E, in the Japan Sea off Noto-Ogi, 20 m deep; April 24, 1981; collected by Mr. Tsutomu Shin'ya; preserved in 10% formalin and deposited at the Seto Marine Biological Laboratory (paratype specimens are designated as SMBL Type No. 331). Description: Body nearly oval, a little longer dorsoventrally than anteroposteriorly and in the preserved state compressed laterally, with fairly long and slender stalk projecting from the anteroventral corner. The largest body is 12.5 mm in long and

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Fig. 1 Adagnesia vesiculiphora n. sp. Holotype (SMBL Type No. 330). A: Entire animal.
B: Bud-like projections and fine filaments possibly of a protozoan on the stalk, enlarged. C: Mantle body, left side. D: Mantle body, right side; circular muscles on atrial siphon are omitted to show anus and oesophagus clearly.

8.0 mm in short axis, while the smallest 3.8 mm and 2.6 mm respectively. Stalk up to 57 mm long and 1 mm thick.

Test of body is as thin as paper, colourless and transparent, though rather tough; the surface is sometimes weakly wrinkled, but quite free from any foreign matters. On the largest specimen exclusively, several bud-like projections are found near the base of stalk. Test of stalk is tough, white and nearly opaque; the surface is almost wholly covered with fine filaments possibly of such a protozoan like *Zoothannium* (Fig. 1, B). Several root-like processes, carrying shell fragments, are born on the distal part of stalk; probably the animal is anchored on the sea floor by this distal portion of stalk, leaving the body and the rest part of stalk swinging in the water. In addition to the processes mentioned above, usually a few to many small bud-like projections just like those on the body surface near the base of stalk in the largest specimen are seen on the stalk (Fig. 1 B); these contain each usually an ampulla-like structure, apparently quite similar to the "ampullae" which are found inside the stalk and to be referred to later. Some projections are much elongated and may carry shell fragments along them (Fig. 4, A). Both apertures are nearly sessile; the atrial is terminal and clearly 6-lobed, while the branchial is situated with the dorsal base of the siphon around the middle of the body, indistinctly lobated and opened towards the ventral side.

The mantle body has a prominent conical projection, with the apical portion tapering, on the ventral side just ventral to the anterior half of the endostyle. Mantle very thin and transparent. The siphonal musculature consists almost of circular muscles and is more conspicuous on the atrial than on the branchial siphon. There are 8 to 12 fine longitudinal muscles on each side, radiating from the branchial siphon, branching and anastomosing in the posterior half on the left but around the middle of the body on the right side; the posterior fourth of the body holding the visceral mass, as well as the anteroventral conical projection, is quite free from any muscles (Fig. 1, C-D). Only some short and very fine muscle fibers are sometimes discernible radiating from the atrial siphon. The conical projection includes at it bottom an "epidermal vesicle", from which a tuft of tubules coated thinnly with transparent matter is issued to penetrate throughout the core of stalk to the distal end. The core leaves the conical projection at about the middle of its posterior margin. Atrial aperture is always clearly 6-lobed and opened dorsad; the branchial is also 6-lobed but very indistinctly in the holotype and some other specimens (Fig. 1, C-D), it is, however, more frequently surrounded by only two main plain lobes arranged dorsoventrally or anteroposteriorly and a few insignificant additional prominences and generally bent towards the ventral side. Neither branchial nor atrial velum. Tentacles simple, 20 to 24 in most specimens, though only 14 in the smallest one; usually



Fig. 2. Adagnesia vesiculiphora n. sp. A: 10.3 mm×6.0 mm (in body size) specimen.
B-C: Holotype. A: Ciliated groove. B: Anterodorsal part of branchial sac.
C: Middle part of the lateral wall of branchial sac.



Fig. 3. Adagnesia vesiculiphora n. sp. 10.3 mm×6.0 mm (in body size) specimen. A: Optical frontal section of the posterior part of mantle body, through stomach; ventral view. B: Optical frontal section of the posterior part of mantle body, through first intestinal loop; ventral view. b.c.: branchial cavity; h.: heart; i.: intestine; m. mantle; o.: ovary; o.d.: oviduct; p.c.: peribranchial cavity; r.g.: retropharyngeal groove; st.: stomach; t.: testicular follicles; v.d.: vas deferens.

larger and smaller ones alternating regularly, besides an additional minute papilla found generally in each interval. Prebranchial zone quite smooth. The peripharyngeal band figures a backward concavity on the right side of the ciliated groove which is an oval opening elongated transversely and situated just at the level of the band (Fig. 2, A-B). A conical prominence is formed along the left branch of the concavity of the band. The neural gland is situated on the right to and in lesser cases partially dorsal to the ganglion. Up to 11-12 (or very rarely 13) bifid branchial papillae born on five transverse vessels in each half; an additional transverse row of a few bifid papillae is found at the bottom of the branchial sac in the largest specimen, but this is not underlaid with any vessel. Row of five large dorsal languets displaced to the left of the dorsomedian line (Fig. 2, B). Stigmata C-shaped; typical arrangement may be represented by 4 transverse rows of stigmata between each pair of transverse vessels and 2 to 3 longitudinal rows in each interval between the longitudinal rows of branchial papillae (Fig. 2, C), but the arrangement is rather irregular at places (Fig. 2, B). Two stigmata adjoining each other anteroposteriorly or horizontally are generally paired with their open side face to face. In the smallest specimen, stigmata are usually rather rudimentary and only two transverse rows of them are seen between each pair of transverse vessels. Neither inner longitudinal nor parastigmatic vessels.

Stomach and the following half of the proximal branch of deep first intestinal loop are laid just under (posterior) the branchial sac, while the rest of the loop clearly on the left side of the rear portion of the sac (Fig. 3). Stomach seemingly globular and with smooth surface, though the actual structure is made obscure by testicular follicles wholly covering the stomach surface. The second intestinal loop very shallow and rectum situated on the left to rather short oesophagus. The anterior margin of the intestinal loop reaches to or slightly beyond the level of 5th transverse vessel. Anus bi-lobed. Heart is a simple sac situated on the right side of the posterior end of branchial sac and at the site not so far from the pyloric end of stomach. Ovary situated in the first intestinal loop, ovarian eggs up to 100μ in diameter; testicular follicles distributed over the whole surface of stomach and the proximal end of intestine. Apertures of vas deferens and oviduct open adjacently to each other behind the anus, and between oesophagus and rectum.

Stalk consists of the vacuolated tissue containing a tuft of tubules and a thick "vessel" running along a side of stalk and occupying about one third of cross section (Fig. 4, B). There are found in the "vessel" one to 13 "ampullae" in various parts along its length in respective specimens; some "ampullae" may be found attached to the vessel wall lining the side to the vacuolated tissue. In a few specimens, up to 7 "ampulla"-like structures are found between the mantle body and test firmly attached to the basal part of the stalk core. Further, several similar "ampullae" are also found liberated from the core and firmly attached to the inner surface of the test covering the conical projection of mantle at the anteroventral part of the body in some specimens. "Ampullae" are nearly cylindrical or sausage-shaped, up to 800 μ in length and nearly transparent except the nipple-like apical tip and a thin longitudinal band on the body wall; these parts, especially the former, are marked by cells aggregate so densely (Fig. 4, C). The basal end of ampullae is sometimes tapering into a long tubule, but nothing has yet been confirmed about the relation between this and the tuft of tubules running through the stalk core.

Structure of the "epidermal vesicle" and possible function of "ampullae": The "vesicle" seems to represent a structure invaginated from the posterior surface of the conical projection into the projection cavity, and the cavity itself might be originally a part of the peribranchial cavity (atrium) and then completely separated from the atrium. Therefore, the vesicle wall is considered to be built of the mantle epidermis and the epithelium of the atrium (Fig. 5, B). The bottom of the "vesicle", facing the endostyle but never adhering to it, is oval in outline, longer anteroposteriorly and up to



Fig. 4. Adagnesia vesiculiphora n. sp. A: 3.8 mm×2.6 mm (in body size) specimen. B: 7.4 mm×6.0 mm specimen. C: 6.0 mm×4.2 mm specimen. A: Elongated buds issued from stalk; filamentous structures are removed. B: Optical cross section of stalk, asterisk showing the "vessel". C: An "ampulla" found in stalk, lateral view.



Fig. 5. Adagnesia vesiculiphora n. sp. A: "Epidermal vesicle" in a 8.6 mm × 5.8 mm (in body size) specimen, left side. B: Schematic representation of a transverse section of the ventralmost part of mantle body through the middle of "vesicle". b.p.: basal papilla; e.: endostyle; tu.: tubule; for other abbreviations see fig. 3.

1.5 mm. The bottom thicker than the general epidermis, slightly pinkish and nearly opaque; its inner surface carries 4 to 10 conical or cylindrical, rather stout "basal papillae", usually less than 300μ long. There are tubules, up to 20μ in diameter and lined with only a single layer of cells, issued from the vesicle bottom and gathered in a tuft. The lumen of these tubules is seemingly connected to the cavity of the conical projection surrounding the "vesicle". The remaining space of the vesicle is filled with a mass of tough, white and translucent matrix which enters the stalk core, coating the tuft of tubules and then joins to the test in a short distance from the mantle body. Some basal papillae may rarely be elongated up to 1 mm along the tuft of tubules firmly attached to it, their wall becomes translucent, except the apical tip and a thin longitudinal band marked by densely aggregate cells just like the "ampullae" mentioned previously, and they are connected to the bottom wall of the vesicle through a very thin neck (Fig. 5, A).

Resting on the observations given above, it may safely be supposed that the "basal papillae" will leave the "vesicle", move into the stalk and travel through the vessel along the tuft of tubules, till they reach respectively into the bud-like projections from the stalk and there become to be the "ampulla" at the tip, and later separated from the stalk. The tubules in the vesicle and the stalk might be nothing but only the stolon-like structures respectively connecting the ampulla-hollow to the cavity of the conical projection and thus homologous to the thin neck of elongated basal papillae in the vesicle. If these "ampullae" grow up to be individuals, it follows that this animal can reproduce asexually in a quite unique manner of budding, that is, however, not essentially different from "peribranchial" or "pallial" budding, where buds consist at least of both epidermis and atrial epithelium, but surprisingly adapted to produce by budding the juveniles anchored on the soft substratum by stalk. Actual observations of living specimens are urged to confirm the abovementioned supposition.

Remarks: In the shape of the entire animal, the present specimens may be reminiscent

of Corellopsis pedunculata Hartmeyer, 1903 from the Arctic waters (Hartmeyer, 1903, pp. 273–278); indeed these two are identical with each other in the presence of bifid branchial papillae, more or less curved stigmata and of dorsal languets. In the former, however, the visceral mass is situated on the left side of, though partially posterior to, the branchial sac, instead of the right side seen in the latter arctic species that is assigned to the family Corellidae; thus the former should be assigned to Agnesiidae.

The specimens from the Japan Sea are quite unique among the members of Agnesiidae in having a slender stalk, C-shaped stigmata, testicular follicles nearly exclusively covering the whole stomach surface and the "epidermal vesicle"; the last two characters may be very strange even in Ascidiacea. Nevertheless, the present specimens may safely be assigned to the genus Adagnesia Kott, 1963 defined on the existence of bifid branchial papillae and dorsal languets (Kott, 1969 a, p. 99; Monniot, C. and Monniot, F., 1973 a, p. 345). In this genus, the following 6 species have been so far recorded: A. opaca Kott, 1963 from sublittoral to 140 m deep in the eastern coast of Australia (Kott, 1963; 1969b, pp. 454-455; 1972, p. 238; 1973, p. 250), A. antarctica Kott, 1969 from 86 to 101 m deep of the subantarctic waters (Kott, 1969a, p. 99; 1969b, pp. 453-454), A. bifida Millar, 1970 from 3517-5841 m deep off the Pacific coasts of Central America and Peru and 4820 m deep in the Indian Ocean off East Africa (Millar, 1970, pp. 118-120), A. charcoti Monniot, C. and Monniot, F., 1973 from 500 to 4758 m deep in the northern and tropical Atlantic waters (Monniot, C. and Monniot, F., 1973b, pp. 424-428; 1974, p. 744; 1976, p. 666; Monniot, F. and Monniot, C., 1976, p. 634: Millar, 1978, pp. 104-105), A. rimosa Monniot, C. and Monniot, F., 1974 from 4159 to 4829 m deep in the Bay of Biscay and off the southwestern coast of Africa (Monniot, C. and Monniot, F., 1974, pp. 744-747) and A. fissa Monniot, F. and Monniot, C., 1976 from 5223 to 5208 m deep off Argentina (Monniot, F. and Monniot, C., 1976, pp. 633-634). The specimens from the Japan Sea are clearly distinguishable from all these hitherto known species by, in addition to the above-mentioned 4 characters, the poorly developed mantle musculature, expressed by the absence of any transverse muscle fibers around the body proper exclusive of siphons and of any remarkable muscle bundles radiating from the atrial siphon, the simple structure of the branchial sac, shown by the shape of stigmata never coiling but only curved as C, instead of coiling 1.5 to 3 times or more as seen in the known species excepting A. bifida without obvious spiral arrangement of stigmata, and by the existence of only 5 transverse vessels instead of 6 (in antarctica by Kott, 1969b, though 5 is suggested by Kott, 1969a) to about 30 (in opaca by Kott, 1969b). Thus, there is no doubt that the present specimens represent a new species, to which the specific name vesiculiphora is proposed after the existence of prominent "epidermal vesicle". The formation of a conical prominence of the peripharyngeal band on the right side of the ciliated groove in the present new species seems peculiar, too, but this feature has been described already not only in Adagnesia antarctica (Kott, 1969a, p. 99 and fig. 133) but also in other agnesiids such as Agnesia glaciata Michaelsen, 1898 (Kott, 1969b, p. 452), A. himeboja Oka, 1915 (Oka, 1915, p. 5 and fig. 2 on pl.

3; Nishikawa, unpublished) and A. bergica Ritter, 1913 (=septentorionalis Huntsman, 1912) (Ritter, 1913, p. 493 and figs. 39 and 40).

Of other solitary ascidians, *Rhizomolgula globularis* (Pallas, 1776) from the Arctic waters and *Hemirhizomolgula utidai* Oka, 1926 from North Sakhalin are noted in having the "peduncular gland" (name after Oka, 1926) on the inner surface of the mantle along each side of the posterior end of the endostyle. This structure is named differently according to the researchers, "rundliche Körper" by Hartmeyer (1903), "Fußdrüse" by Redikorzev (1907, 1908) or "gland" by Huntsman (1912) and Ärnbäck-Christie-Linde (1928), and is explained by Hartmeyer (1903) as consisting of a pair of branched hollow or solid capsular glands covered with the peribranchial epithelium, the duct from respective branch capsules are united into a common duct on each side and thus a pair of common ducts penetrate throughout the rooty processes of test to each distal end. Mentioning this, Hartmeyer (1903, p. 170) suggested that the "peduncular gland" might be comparable in function to the byssus gland of some lamellibranchs. Anyhow, the "peduncular gland" referred to in the two molgulids mentioned above are of the structure and nature evidently different from those of the "epidermal vesicle" found in the present new species.

Lastly, closer reexamination of *Corellopsis pedunculata*, especially of its peduncle and related structures, is very desirable to check again the relation between this and the present new species.

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