# Morphology, larval development, and distribution of *Bathypera feminalba* n. sp. (Ascidiacea: Pyuridae), a deep-water ascidian from the fjords and sounds of British Columbia

Craig M. Young and Elsa Vázquez

Harbor Branch Oceanographic Institution, Ft. Pierce, Florida 34946, USA

**Abstract.** A new species of the ascidian genus *Bathypera* (Ascidiacea: Pyuridae), *B. feminalba,* is described from deep-water habitats of Saanich Inlet and Barkley Sound, British Columbia, Canada, with data on the depth distribution, substratum use, reproduction, embryology, and larval development. The species is characterized by hourglass-shaped spicules topped with a single large spine and several smaller spines, a branchial sac with 7 folds per side, and irregular and curved stigmata. *B. feminalba* spawned in response to light following dark adaptation during the month of June. Development was similar to that of other pyurids. The tadpole larva, the first to be described in this genus, has the same sensory organs found in shallow-water relatives: an ocellus, a statocyst, and 3 large conical adhesive papillae. The ocellus is sensitive to monochromatic light between 475 and 600 nm with peak sensitivity in the blue region of the spectrum. This species occurs below 20–30 m depth, where it occupies vertical cliffs, the sides of boulders, and also smaller cobbles. It shares this habitat with a characteristic assemblage of ascidians, brachiopods, serpulid polychaetes, and sponges, few of which are found in shallow water. On a small scale, the distribution of *B. feminalba* is aggregated, with the pattern suggesting gregarious larval settlement. At the upper end of its depth range, this species appears to be excluded from surfaces with coralline algae; competition with algae or interactions with some organism associated with the coralline algal community may determine its upper limit.

The coast of the northeastern Pacific between southern Alaska and central California has a diverse ascidian fauna of more than 80 known species (Austin 1985). Many species occurring in the intertidal and shallow subtidal zones have been studied extensively; indeed, some species from this region are among the best-known ascidians anywhere. However, the biology of ascidians living beyond scuba depths in this region remains virtually unknown. Between 1978 and 1984 we examined the ascidians of Saanich Inlet and Barkley Sound fjords, using a manned submersible, dredges, and scuba. We found a relatively sharp faunal transition: below about 30 m depth, most but not all of the shallow-water ascidians are replaced by a distinct ascidian assemblage with several previously undescribed or recently described species. This assemblage includes, besides the pyurid *Bathypera feminalba* n. sp. described in this paper, the pyurid *Boltenia polyplacoderma* recently described from 91–117 m depth off central California (Lambert 1993); an ascidiid, *Rhopalaea cloneyi* n. sp. (Vázquez & Young, unpubl.); the corellid *Chelyosoma columbianum,* which lives in deep water throughout the north Pacific; and the ascidiid *Ascidia ceratodes,* which defies the pattern predicted by equatorial submergence by living in shallower water in California than in British Columbia. A few species of ascidians characteristic of shallow subtidal habitats, notably *Halocynthia igaboja, Styela coriacea,* and *Ascidia paratropa,* also extend into deep water in British Columbia. Ascidians share the deep fjordic habitats with a fauna dominated by brachiopods (Tunnicliffe & Wilson 1988), galatheid crabs (Burd & Brinkhurst 1984), hexactinellid sponges, serpulid polychaetes, and cup corals (reviewed by Levings et al. 1983). The physical and biological factors that impose the upper limit of these organisms remain unknown, though low oxygen levels may determine their lower depth limit in fjords (Tunnicliffe 1981; Burd & Brinkhurst 1984).

Ascidians of the genus *Bathypera* Michaelsen, 1904 are beautiful animals that resemble old-fashioned beaded purses. The "beads" are minute papillae arranged in a regular pattern and each containing a spiky calcareous spicule (Lowenstam 1989), often shaped like the acanthus-leaf decorations found atop Corinthian columns in ancient Greek architecture (Van Name 1945). The branchial sac has more than 4 plications on each side, and the stigmata are normally

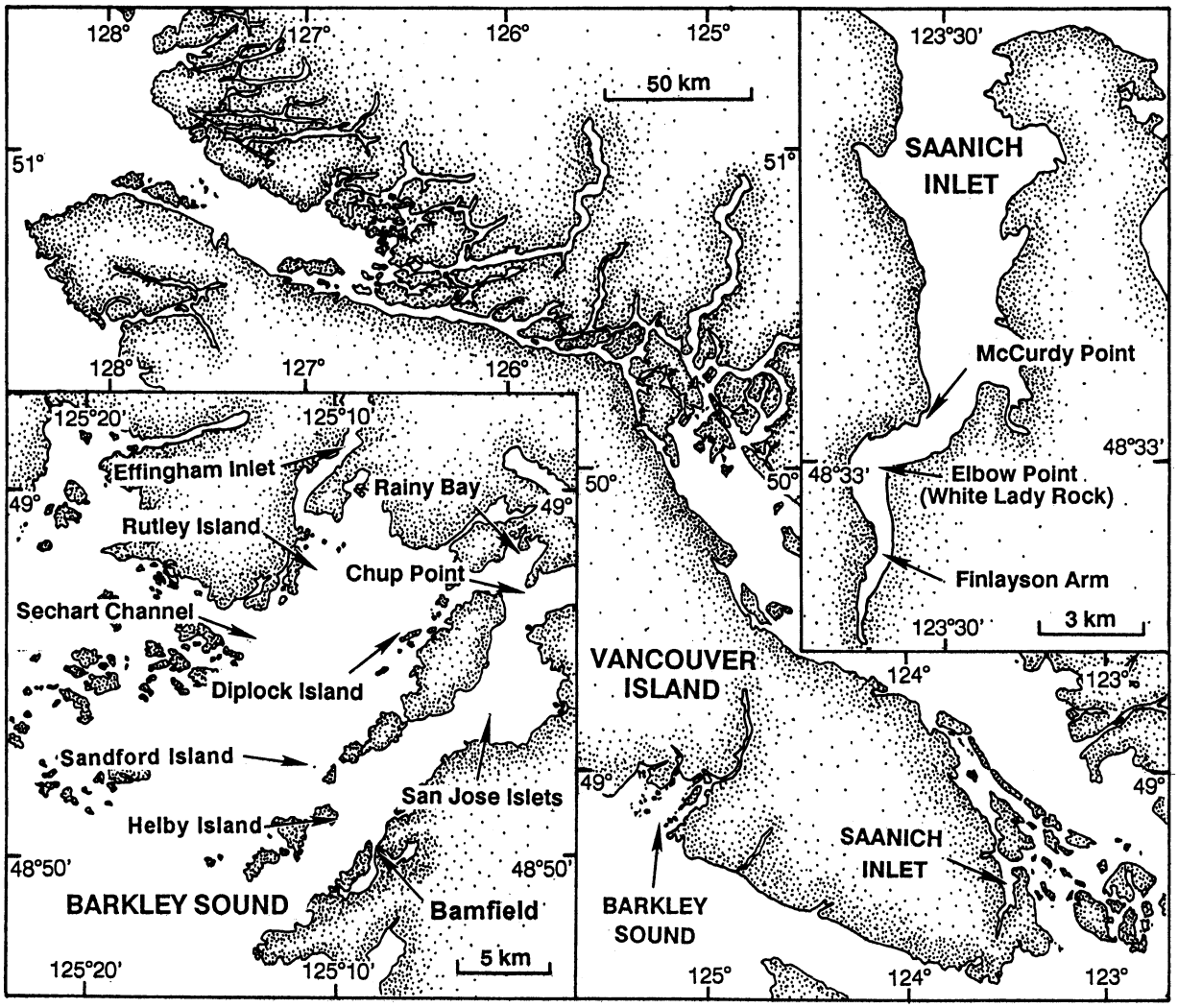



**Fig. 1.** Study sites in Barkley Sound and Saanich Inlet, Vancouver Island, Canada (see Table 1 for coordinates and depths).

much more regular than those of other pyurids (Monniot & Monniot 1972). The diagnostic characters of this genus have been reviewed by Monniot (1965).

Until now, only 3 valid species of *Bathypera* have been described. *B. splendens* (Michaelsen, 1904) occurs from 75 to 4636 m in antarctic waters (Van Name 1945; Vinogradova 1962; Kott 1969; Monniot & Monniot 1983) and off Tierra de Fuego (Kott 1971). Another antarctic species, *B. hastaefera* Vinogradova, 1962, has recently been synonymized with *B. splendens* (Monniot & Monniot 1983). *B. ovoida* (Ritter, 1907) is the only species known to occur in the Pacific Ocean. It has been collected from 3680 m off California (Van Name 1945; Fay & Vallee 1979), 184 m in Japanese waters (Nishikawa 1981), and 300 m in the Okhotsk Sea (Sanamyan 1992). *B. goreaui* is known only from a few specimens collected from a deep reef (53–90 m) near Discovery Bay, Jamaica (Millar & Goodbody 1974). These authors mention that Goodbody collected 10 specimens of a *Bathypera* from 100 m in Saanich Inlet, B. C., which "... appear to be of this species [*B. ovoida*]." Also, G. Lambert (pers. comm.) found a deep-water species of *Bathypera* off southern California with a similar spicule morphology, and Lowenstam (1989) described the spicules of an unidentified species dredged from 60–100 m off Santa Catalina Island, California. On the basis of distribution and spicule form, it seems likely that the specimens of Goodbody, Lambert, and Lowenstam belong to the new species described here.

In this paper, we describe *Bathypera feminalba* n. sp. and present information on the gross morphology of adults and larvae, embryology, larval behavior, reproduction, growth, recruitment, and the distribution of this species at several spatial scales—all that is now known about this unusual ascidian.

**Table 1.** Study sites in Barkley Sound and Saanich Inlet, B.C., showing coordinates, survey methods, range of depths surveyed, and range of depths where *B. feminalba* was observed or collected. Depths given in meters.

| Study site | Latitude and longitude | Survey methods | Depths surveyed | Depths of occurrence |
|---|---|---|---|---|
| Chup Point | 48°57.19′N 125°01.48′W | Submersible & Dredge | 135–250 | 180–239 |
| Diplock Island | 48°56.20′N 125°07.10′W | Dredge | 20–30 | 20–30 |
| Effingham Inlet | 49°01.10′N 125°09.40′W | Submersible | 9–120 | 22–93 |
| Elbow Point | 48°32.80′N 123°31.90′W | Submersible & Scuba | 0–200 | ~30–110 |
| Finlayson Arm | 48°31.50′N 123°32.30′W | Submersible | 16–216 | 91–118 |
| Helby Island | 48°50.10′N 125°09.70′W | Scuba | 0–40 | 25–40 |
| McCurdy Point | 48°33.60′N 123°31.50′W | Submersible & Scuba | 0–200 | ~30–100 |
| Rainy Bay | 48°58.50′N 125°02.40′W | Dredge | 50 | 50 |
| Rutley Island | 48°57.90′N 125°10.00′W | Submersible & Dredge | 34–98 | 40–45 |
| Sandford Island | 48°52.30′N 125°11.72′W | Submersible | 25–83 | 68–68 |
| San Jose Islets | 48°53.70′N 125°03.40′W | Submersible & Scuba | 0–193 | 19–165 |
| San Jose Islets | 48°53.87′N 125°02.85′W | Submersible | 34–199 | 36–123 |
| San Jose Islets | 48°53.88′N 125°02.86′W | Submersible | 22–200 | 30–190 |
| Sechart Channel | 48°55.70′N 125°11.80′W | Submersible | 10–96 | 60–74 |

## Methods

### Study sites and collection methods

Study sites were in Saanich Inlet, a narrow fjord at the southern end of Vancouver Island, and in Barkley Sound on the west coast of Vancouver Island (Fig. 1, Table 1). *Bathypera feminalba* was observed by scuba at 2 sites in Saanich Inlet (McCurdy Pt. and Elbow Pt.) and at 2 sites in Barkley Sound (Helby Island and San Jose Islets). Observations were made with the PISCES IV submersible at the same sites in Saanich Inlet and at a number of additional sites in Barkley Sound. Only 3 of the submersible dives were made by one of us (CMY); additional observations were by other submersible users. During the period in 1984 when PISCES IV was operating near the Bamfield Marine Station in Barkley Sound, we distributed drawings and descriptions of the common deep-water ascidians to all submersible observers and requested records of the depths and substratum distributions of these species.

Because most individuals of *B. feminalba* observed from the submersible were attached to large rocks or cliffs, we obtained few specimens by submersible. Larger collections, including the type specimens, were obtained by dredging shell rubble and cobbles from several sites in Barkley Sound using the MV *Alta* in 1984 and 1985. We dredged at comparable depths throughout the nearby San Juan Islands in Washington, USA, using the RV *Hydah* and RV *Nugget* from Friday Harbor Laboratories.

### Morphological studies

Animals for morphological studies were anesthetized in small jars of seawater by addition of menthol crystals, then fixed in buffered 5% seawater formalin. A few small individuals were fixed for scanning electron microscopy (SEM) using the double-fixation method of Cloney & Florey (1968). Spicules were removed for SEM by digesting away the tunic with sodium hypochlorite solution (5%), then rinsing with water and air drying. Specimens were sputter coated with

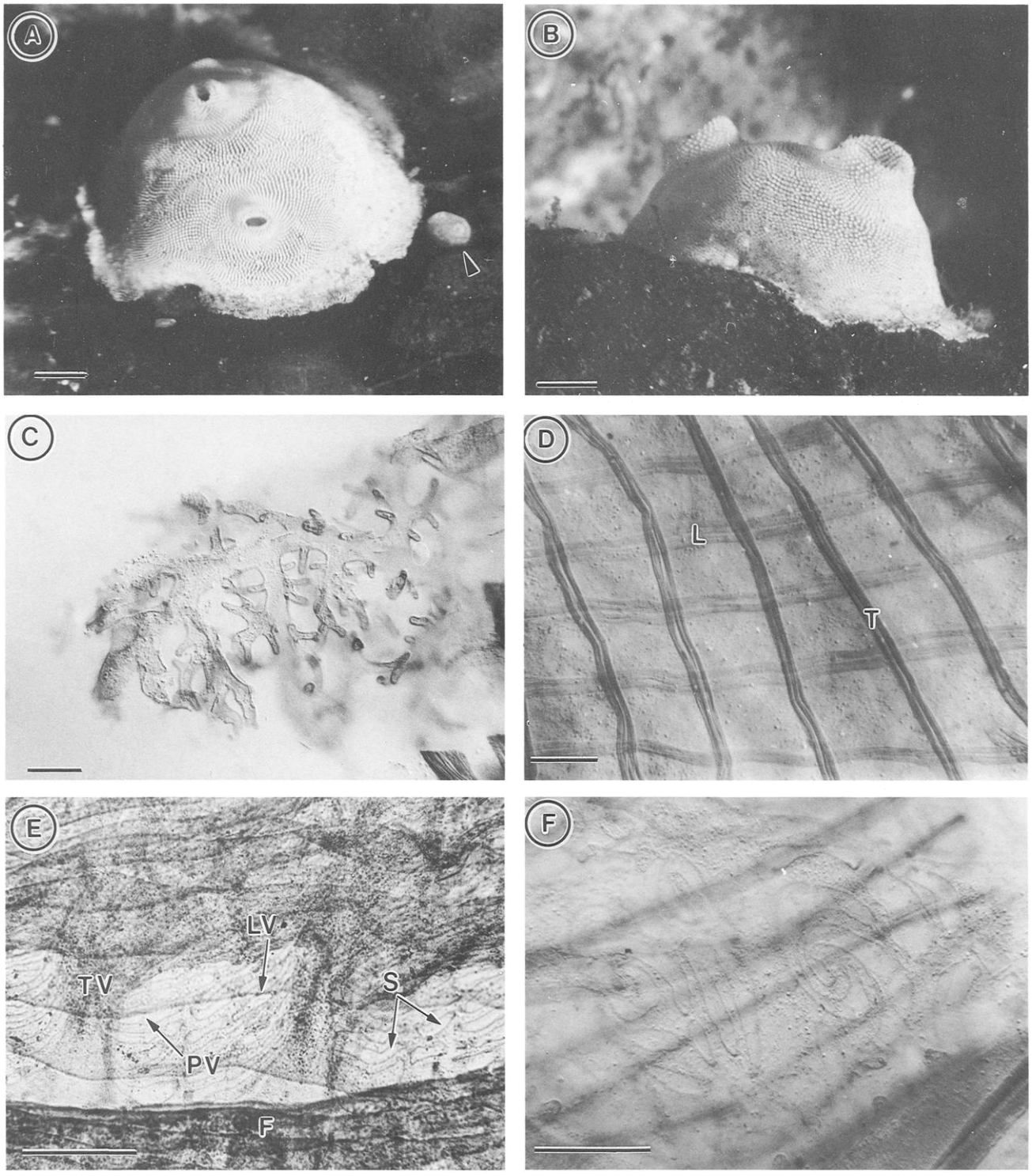



**Fig. 2.** External and internal anatomical features of *Bathypera feminalba.* **A.** Anterior view of an adult with partially contracted siphons, showing curved converging arrangement of spicule-bearing papillae and thin skirt of tunic attached to the substratum. A small juvenile (arrowhead) is attached near the adult. Scale bar 2 mm. **B.** Lateral view of adult with fully opened siphons. Scale bar 2 mm. **C.** A branchial tentacle of the first series photographed from a stained whole mount of a relaxed specimen. Scale bar 100 μm. **D.** Longitudinal (L) and transverse (T) muscle bands in the lateral body wall. Scale bar 100 μm. **E.** Detail of the branchial sac showing a fold (F), curved stigmata (S), longitudinal vessels (LV), transverse vessels (TV), and parastigmatic vessels (PV). Scale bar 300 μm. **F.** Stigmata at various stages of spiraling, showing that one end of each slit is wider than the other. Scale bar 50 μm.

gold and examined with a JEOL JSM-35 scanning electron microscope at Friday Harbor Laboratories or with a Cambridge Stereoscan-150 microscope at the University of Alberta.

Internal anatomical features were examined in fixed specimens using the technique described by Monniot & Monniot (1972). Dissected specimens were stained with Masson's Alum Hemalun, dehydrated in butyl alcohol, and mounted in Canadian balsam on microscope slides. Anatomical drawings were made with a camera lucida on a Wild M-5 dissecting microscope or a Zeiss compound microscope.

### Spatial distributions

Small-scale spatial distributions were described from 35-mm slides taken from the submersible or with a Nikonos V camera while scuba diving. The slides were projected on the rear of a small translucent screen and the position of each animal was traced on an acetate sheet. Onto a set of random points generated by computer (Sigmastat "Random" function), we superimposed the acetate map, measured the distances from each point to the nearest individual and from each individual to its nearest neighbor, and used these data to calculate the nearest neighbor statistic of Hopkins & Skellam (1954).

Densities of *Bathypera feminalba* were also obtained from the slides. Some slides taken by divers used a close-up framer of known area and some taken by submersible included a calibrated quadrat in the photograph. In the remaining slides, we estimated area by assuming that the largest visible individuals were approximately 2 cm long, an assumption that appeared to be warranted by analysis of size-frequency distributions.

We described the habitat of *B. feminalba* from submersible records by tabulating the substratum types on which this species was found. The distributions of animals by substratum type were compared with expected values based on the total number of records of all substratum types. We used a similar method to characterize the distributions of individuals on small cobbles collected by dredge.

### Reproduction and development

Living animals were transported to Bamfield Marine Station or Friday Harbor Laboratories, where they were held in flow-through seawater systems. Several animals with their original substrata were placed into the same dish. On 23 June 1981, four large individuals were kept in complete darkness from 1600 until 1000 the following morning, at which time they were exposed to diffuse sunlight entering through the laboratory window. Seawater temperature was 11° C. Eggs and sperm of different individuals were mixed, and the resulting embryos were maintained in finger bowls of filtered seawater and were examined and photographed periodically until hatching. Swimming larvae were transferred to polystyrene petri dishes, where they settled and metamorphosed. These juveniles were observed with a microscope in their dishes.

We tested for photosensitivity of the tadpoles at various wavelengths of monochromatic light generated by a Bausch and Lomb model 33-86-02 diffraction grating monochromator, using a 9.6-nm bandpass slit. Responsiveness was tested at 25-nm increments from 400 to 700 nm, maintaining a constant irradiance of $6.0 \times 10^{13}$ photons $cm^{-2}s^{-1}$ as measured with a Li-Cor quantum meter equipped with a cosine sensor. We focused on individual tadpoles under a dissecting microscope, then shaded them abruptly by passing a card over the monochromatic light source. The percentage of tadpoles (n = 20) exhibiting a photokinetic response (Svane & Young 1989) at each wavelength was recorded. We had a limited number of tadpoles, so the same test group was used for all wavelengths. An identical series of experiments was run with two species of ascidians also in the family Pyuridae, *Boltenia villosa* and *Pyura haustor.*

## Results

### Description of species

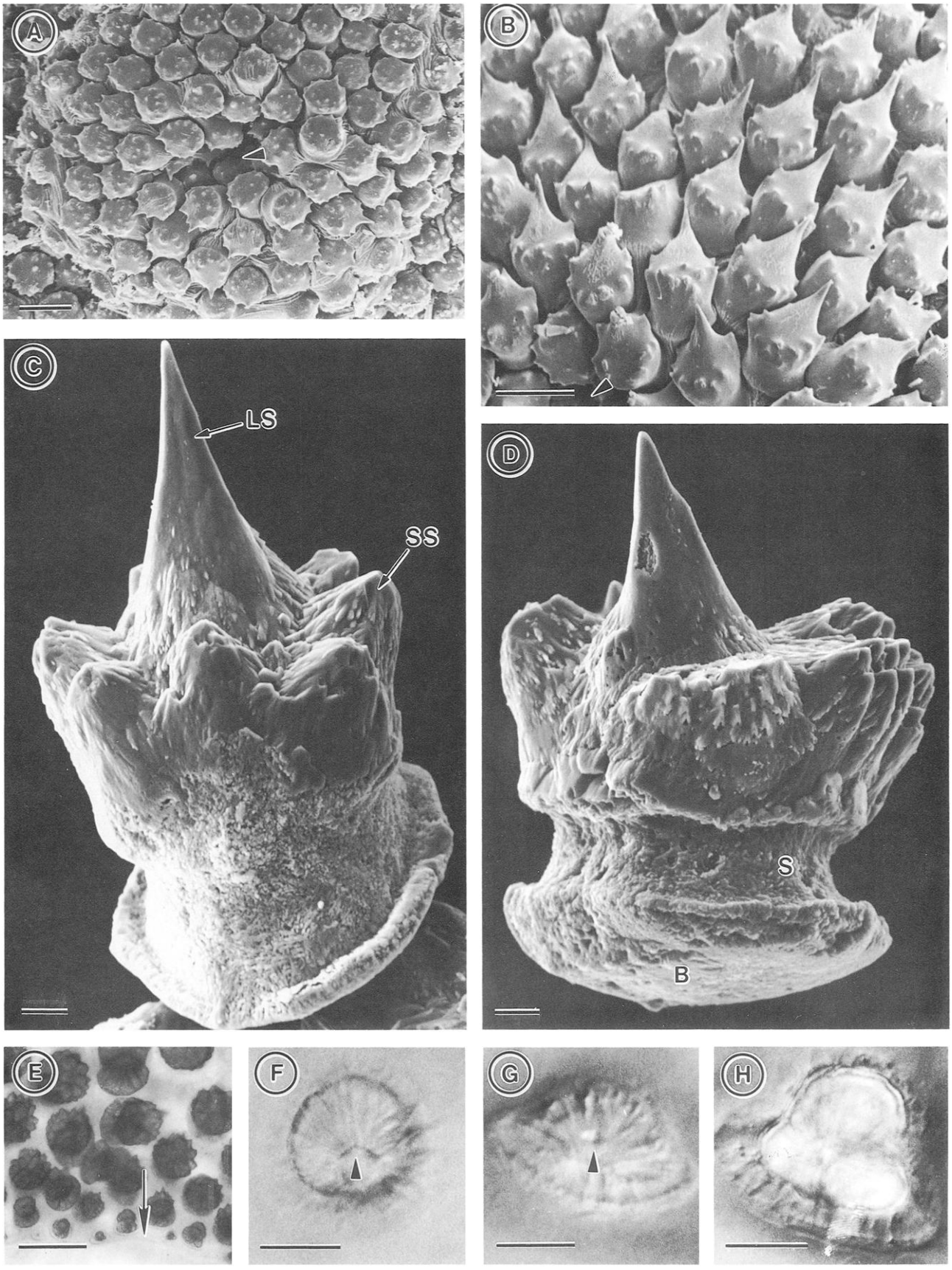
*Bathypera feminalba* n. sp.

**Types:** The holotype is deposited in the Smithsonian Institution, United States National Museum, Washington (catalog number USNM-20594), and paratypes are deposited in the Harbor Branch Oceanographic Institution (catalog number 096: 00130) and in the Museum National d'Histoire Naturelle in Paris (catalog number MNHN S2Bat.A7).

**Type locality:** Chup Point, Imperial Eagle Channel, 48°57.19′ N, 125°01.48′ W; 44–47 m depth. Collected 28 June 1985.

**Derivatio nominis:** The species name *feminalba* is derived from the Latin noun *femina* (woman or lady) and the Latin adjective *alba* (white). The species is named for White Lady Rock in Saanich Inlet, B.C., the site at which we first observed it from a manned submersible. The name is also descriptive of the animal's brilliant white color and delicate appearance.

**Description:** The body is dome-shaped to flattened, attached by a broad base (Fig. 2A,B). The largest animal fixed for morphological studies was 12 mm long, 5 mm high, and 10 mm wide, but specimens up to 20 mm long were collected and used as spawning stock


A
B
C
LS
SS
D
S
B
E
F
G
H

for embryological studies. The test is thin and fragile, and the surface is covered with minute papillae arranged in intersecting curved lines; these converge toward the two apertures and extend onto the part of the test that invaginates to line the interior surfaces of the siphons (Figs. 2A, 3A,B). These papillae contain pointed, calcareous spicules (Fig. 3A,B), which cause them to appear as white dots on the surface. Each spicule has one long and numerous (8–13) shorter spines (Fig. 3B,C,D). Near the siphons, the spicules are arranged with the long spines all pointing away from the siphon apertures (Fig. 3A,B). Spicules are added only at the basal margin of the tunic (Fig. 3E). In this region may be found a regular series of spicule sizes, with the smallest ones immediately adjacent to the margin and increasingly larger ones higher up (Fig. 3E). The smallest papillae contain circular sheaths in which the small spicules form (Fig. 3F). Viewed with cross-polarized light, these circular sheaths apparently contain no calcite crystals, though the site of eventual spicule formation is obvious. The circular sheaths elongate on two ends to become lemon-shaped and the site of crystal formation migrates from near the margin into the center of the sheath (Fig. 3G). At this stage, cross-polarized light reveals the initial mineralization of the small spicule (Fig. 3G). The sheath then elongates asymmetrically while additional calcite is added to form a bell-shaped spicule (Fig. 3H). As the spicule continues to grow, spines form on the actively growing apex, while the base widens.

Atrial and branchial siphons are almost sessile, without lobes, and directed anteriorly (Fig. 2A,B). Each siphon is encircled by 15 bands of transverse muscles (Fig. 4B); of these, 12 are very close together and the remaining 3 are slightly separated from the others. An additional 6 transverse muscles are located posterior to the tentacles. The transparent mantle has strong longitudinal muscle bands that extend ventrally as far as the gonad on the right side (Figs. 2D, 4B) and as far as the intestine on the left. Overall, the musculature forms a strong network.

Branchial tentacles are of 3 sizes, branched in 3 orders of magnitude, and alternating in size (Fig. 2C). The largest tentacles number 16; the intermediate and smaller tentacles are too numerous to count in contracted animals. The dorsal tubercle is small, oval, and transverse (Fig. 4B). The dorsal lamina consists of short languets on the border of a continuous membrane. The prepharyngeal groove is distant from the tentacular circlet.

The branchial sac has 7 high folds on each side. In the holotype, the number of longitudinal vessels is:

Dorsal lamina right: 1-**10**-1-**13**-1-**15**-1-**18**-2-**15**-2-**14**-2-**10**-0-E

Dorsal lamina left: 0-**15**-1-**13**-1-**14**-2-**15**-1-**16**-1-**12**-1-**4**-0-E

There are 6 transverse vessels with 2 infundibula between each two transverse vessels. Stigmata in the infundibula are regular and longitudinal, lightly curved at the ends but never spiral (Figs. 2E, 4A). Between folds, stigmata are small and irregularly distributed (Figs. 2F, 4A), usually short and elliptical, sometimes curved and with one end wider than the other. There are areas of unbroken membrane between folds.

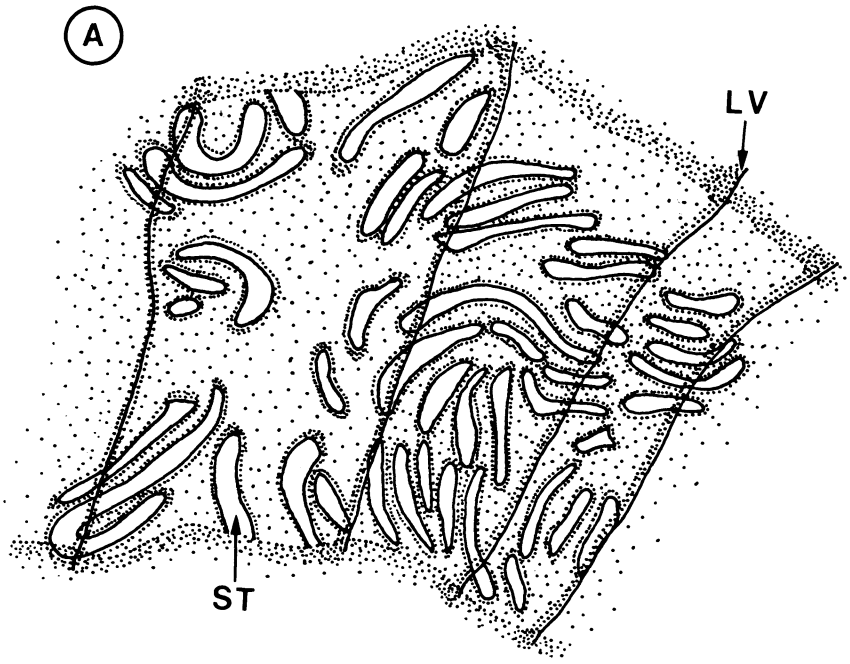
The gut loop is narrow and oriented horizontally in the posterior part of the body, enclosing the left gonad (Fig. 4B). The stomach has about 15 folds and the anus is bordered by lobes.

Endocarps form two lines, one dorsal to the gut loop on the left and the other dorsal to the gonad on the right (Fig. 4B).

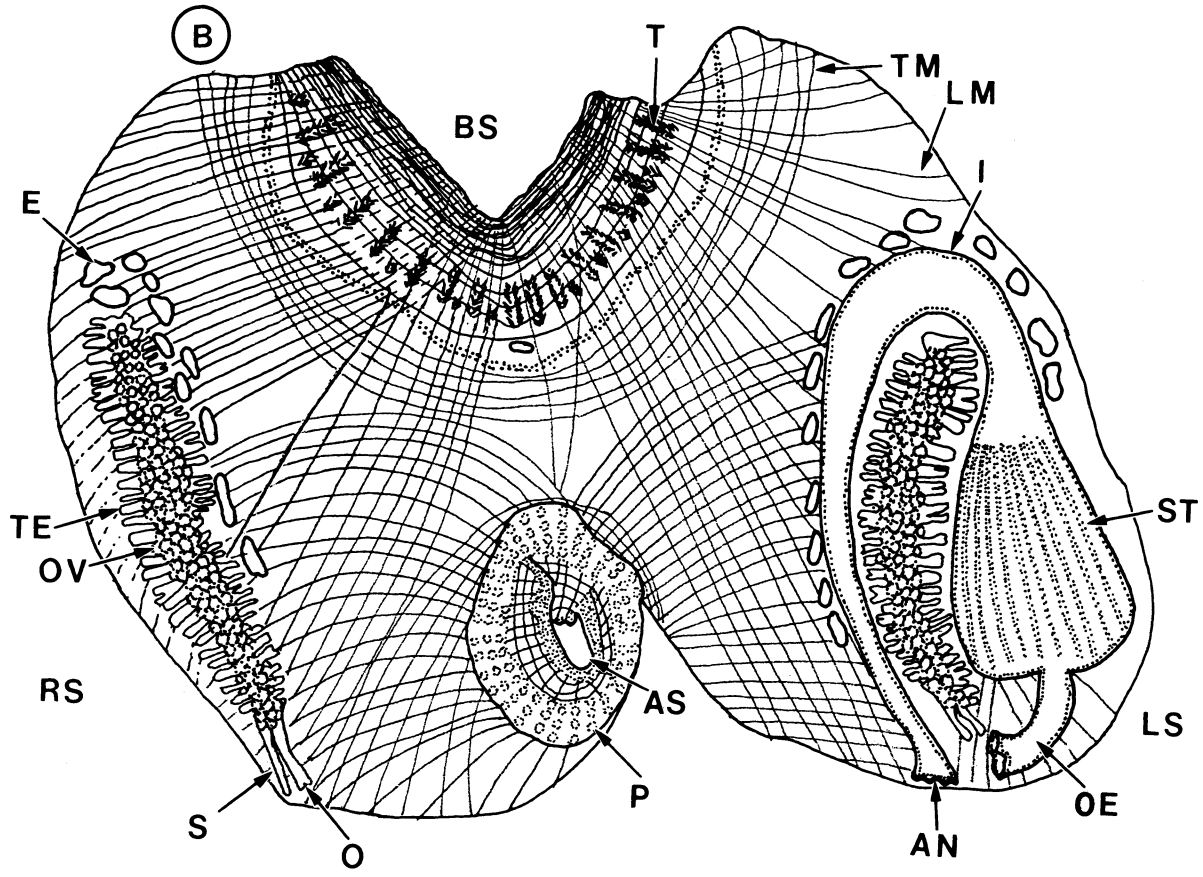
The body contains one pair of gonads; the left lies in the intestinal loop and the right is near the endostyle. In each, the elongate ovary ends in a very short oviduct and is bordered and overlapped by many-branched lobes of the testis (Fig. 4B). The ducts from individual testicular lobes join in a common sperm

←

**Fig. 3.** Spicules and spicule formation in *Bathypera feminalba.* **A–D,** SEM. **A.** Siphon, showing arrangement of tunic papillae with their enclosed spicules. Note that the tunic, papillae, and spicules extend into the siphonal opening. The long spine of each spicule points away from the siphon opening (arrowhead). Scale bar 50 μm. **B.** Close-up view of external siphon surface (the arrowhead indicates the siphon opening). Scale bar 50 μm. **C.** Oblique anterior view of a single, fully formed spicule removed from its papilla. The short spines (SS) are blunt and irregularly shaped; the long spine (LS) is smooth, regular, and sharply pointed. Scale bar 10 μm. **D.** Lateral view of a spicule, showing rounded base (B) and sulcus (S). **E–H,** photomicrographs. **E.** Spicules of various sizes forming at the basal margin of the tunic. Arrow indicates tunic margin and points in direction of tunic growth. Scale bar 100 μm. **F.** Early spicule sheath near the growing margin of the tunic. The arrowhead indicates where the initial spicule will form. Scale bar 10 μm. **G.** Lemon-shaped spicule sheath with tiny calcite crystal (arrowhead) that will eventually enlarge to form the spicule. Scale bar 10 μm. **H.** Bell-shaped early spicule within sheath. Note that the basal portion has already attained its final rounded form but the anterior spines have not yet been secreted. Scale bar 10 μm.




**5th left fold**



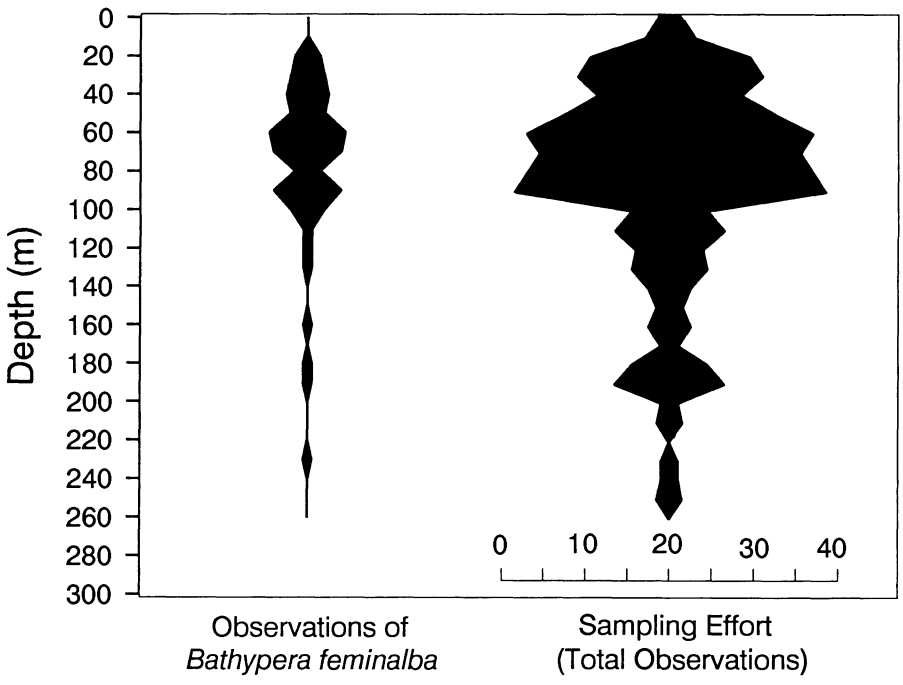



**Fig. 5.** Composite depth distribution of *Bathypera feminalba* at all sites surveyed by submersible in the Barkley Sound region. Left, the number of stations at each 10-m depth interval in which *B. feminalba* was observed from the submersible and noted in the dive log. Right, the total number of entries made in the log at each depth, as an indication of sampling effort.

duct, which runs alongside the oviduct and terminates at the same level.

### Habitat and distribution

Individuals of *Bathypera feminalba* were observed or collected at depths ranging from about 20 to 240 m in Barkley Sound (Fig. 5). Within this depth range, some areas were sampled heavily but yielded few or no observations of *B. feminalba* (e.g., 200–220 m). Few individuals were observed at depths shallower than 30 m; the shallowest record was 18 m. In Barkley Sound, we found a few specimens of *B. feminalba* as shallow as 19 m, but the populations did not become abundant until 30 m. In Saanich Inlet, we surveyed the bottom from the surface to 45 m using scuba at several sites. At sites where we collected by scuba, the characteristic fauna to which *B. feminalba* belongs generally began to appear between 30 and 45 m. We have found *B. feminalba* only in Canadian waters; many years of dredging and diving at appropriate depths in the San Juan Islands of Puget Sound, Washington, have failed to yield a single individual.

Individuals of *B. feminalba* occurred most commonly on vertical cliffs or the sides of large boulders (Fig. 6), though they were found on other hard surfaces, including rocky slopes and small cobbles. They were absent from habitats with sand or silt and mud, even on biogenic hard surfaces (e.g., crab carapaces, shells) in these areas. A goodness of fit test indicated that individuals occurred at approximately the frequency expected by chance on rock slopes, but more often than predicted on cliffs and cobbles (Fig. 6).

Nearest neighbor analysis of the small-scale distribution of individuals indicated highly significant aggregation. Of 14 slides examined, all but 2 produced nearest neighbor values $>1$, indicating aggregation (Fig. 7). Several points near the "random" line were from populations with low sample sizes. There was no apparent relationship between the degree of aggregation and the population density. Figure 8 shows the actual distributions of individuals from two representative slides taken at a depth of 30 m at San Jose Islets, Barkley Sound. Many animals occurred in discrete aggregations of 3 to 7 individuals while much open (and

←

**Fig. 4.** Camera lucida drawings. **A.** A portion of the fifth fold of the branchial sac on the left side of the body, showing stigmata (ST) and longitudinal vessels (LV). **B.** Interior view of the body wall (branchial sac removed) showing positions of the gut, gonads, branchial tentacles, and body wall musculature. Right side of the animal (RS); left side of the animal (LS). Anus (AN); atrial siphon (AS); branchial siphon (BS); endocarps (E); intestinal loop (I); longitudinal muscles (LM); oviduct (O); oesophagus (OE); ovary (OV); papillae with spicules (P); sperm duct (S); stomach (ST); tentacles (T); lobes of the testis (TE); transverse muscles (TM). Scale bar 2 mm.

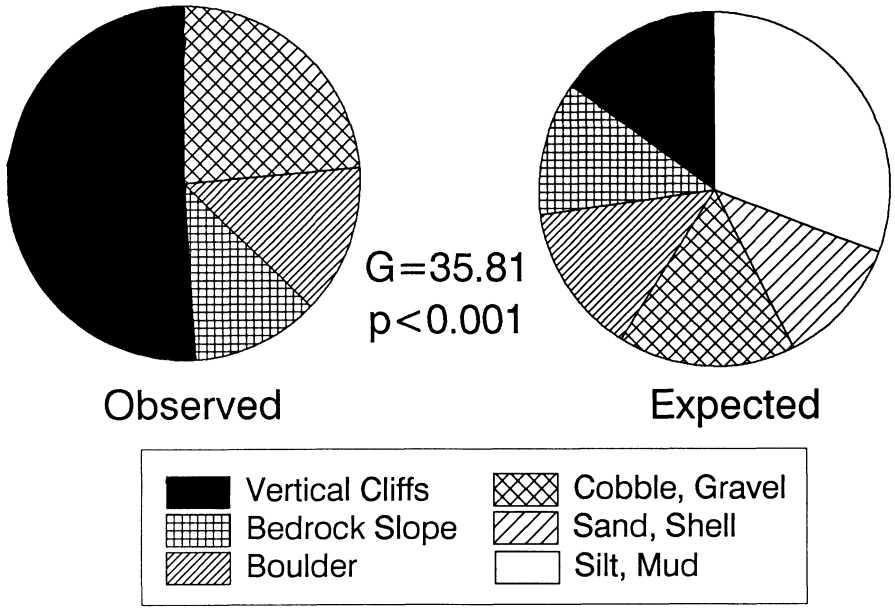



**Fig. 6.** Habitat use by *Bathypera feminalba* in Barkley Sound, as observed from the submersible and recorded in dive logs. Left, proportion of total observations of the species of various substrata. Right, proportion of total observations made on each substratum type. The difference between observed and expected distributions was highly significant ($p<0.001$).

apparently available) space contained few ascidians. Cobbles collected by dredging and examined carefully in the laboratory often had small individuals settled around the bases of adults (Fig. 2A). However, no young were ever observed attached directly to the tunic of an adult.

At San Jose Islets, there was very little bare rock exposed; virtually all surfaces not covered by sessile invertebrates were carpeted by either red coralline algae or an encrusting fleshy brown alga, perhaps of the genus *Ralfsia* (M. D. Hanisak, pers. comm.). Individuals of *Bathypera feminalba* often occurred on the brown algae, but they were seen on coralline algae only rarely, much less often than would be expected on the basis of available surface area (Fig. 8).

## Reproduction, recruitment, and growth

A single large adult dissected in early February 1981 was found to contain abundant ripe gametes; we attempted to fertilize the eggs *in vitro*. Although the sperm were active, no cleavage occurred, suggesting that there may be a complete block to self fertility, as in other pyurids (Svane & Young 1989). Two individuals spawned 15 minutes after they were exposed to light. Some sperm were released before any eggs ap-

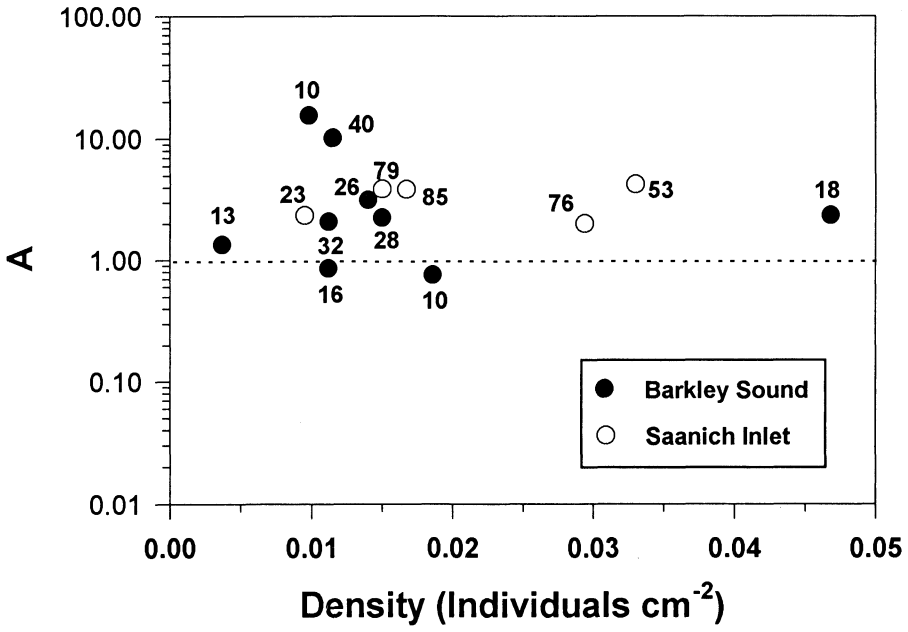



**Fig. 7.** The distribution of nearest neighbor values (the "A" statistic of Hopkins & Skellam 1954) as a function of population density in Barkley Sound and Saanich Inlet. Values >1 indicate aggregation. Numbers adjacent to the points are sample-sizes. Depth of all Barkley Sound stations was 30 m and depths in Saanich Inlet ranged from 55 to 75 m.

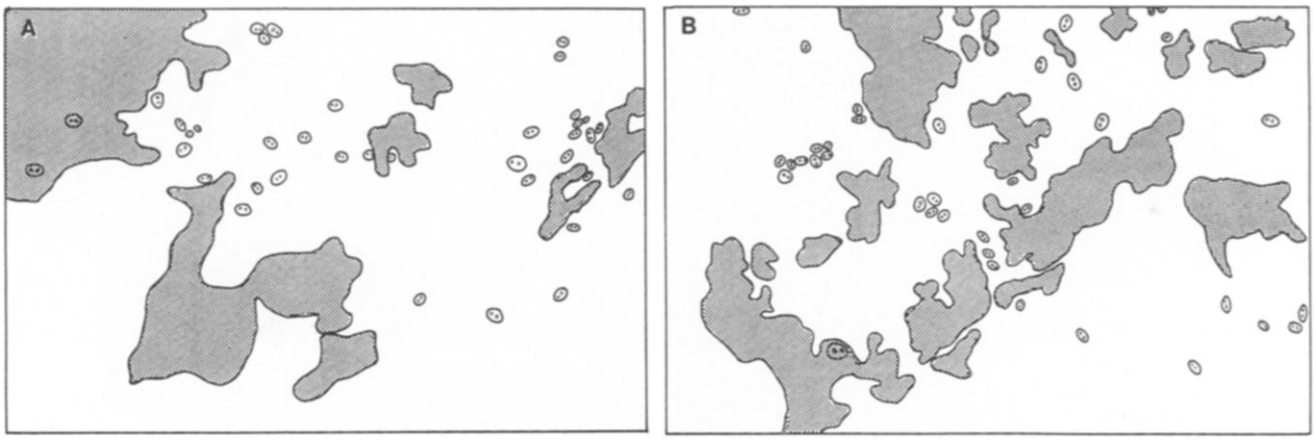



**Fig. 8.** Tracings of two representative transparencies taken by scuba divers at 30 m depth at San Jose Islets, showing the distribution of *Bathypera feminalba* in relation to cover of coralline algae (shaded regions). Ascidians of various sizes form discrete clumps; very few occur on coralline algae.

peared, but both eggs and sperm were later released simultaneously. Gametes from these two individuals were mixed and produced approximately 70 developing embryos.

The laboratory evidence that this species spawns in the very early spring is supported by a small amount of size-frequency data. Collections from Saanich Inlet at the same site and depth in February and June 1981 showed significant change in the size distribution (Fig. 9) that can be explained by growth and recruitment. In February, the population was virtually unimodal. In June, the peak of adult animals had shifted about 4 mm, indicating a linear growth rate of about 1 mm per month; an additional peak consisting of animals between 1.3 and 4.0 mm in length also appeared during this period, indicating a recent recruitment event.

## Embryogenesis, larval development, and larval behavior

The eggs were virtually identical to those of other pyurid ascidians (reviewed by Svane & Young 1989), being invested with a chorion (Fig. 10A), test cells, and follicle cells (Fig. 10B). The ova averaged 147.5 μm (S.D. = 3.53, n = 10) in diameter without the chorion and 193.5 μm (S.D. = 7.47) in diameter with the chorion. The follicle cells averaged 11.25 μm (S.D. = 2.12) in height (Fig. 10A), so the total diameter of

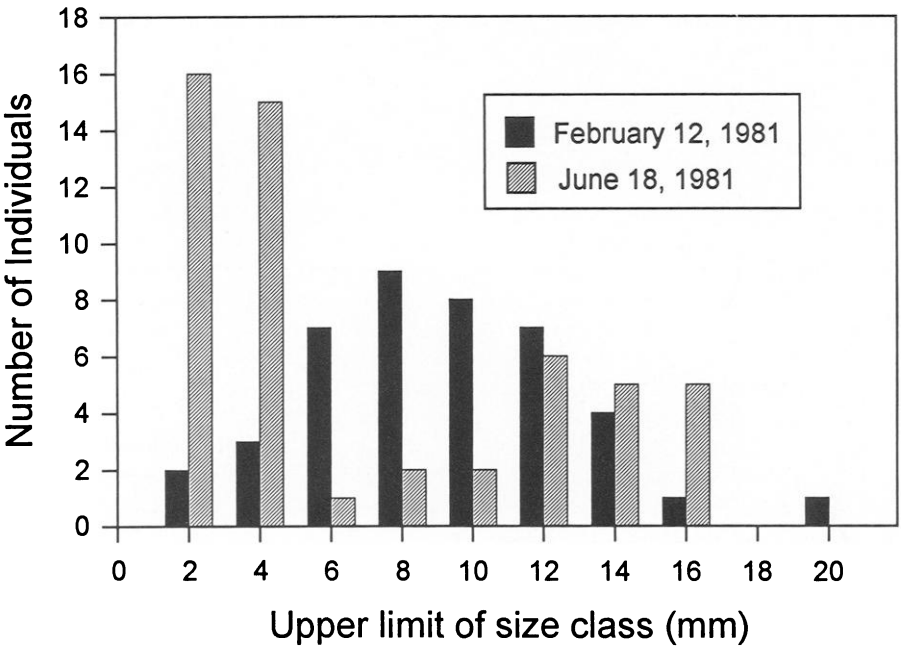



**Fig. 9.** Length-frequency distributions of *Bathypera feminalba* on two dates. These animals were collected on small cobbles between 30 and 40 m at Elbow Point, Saanich Inlet.

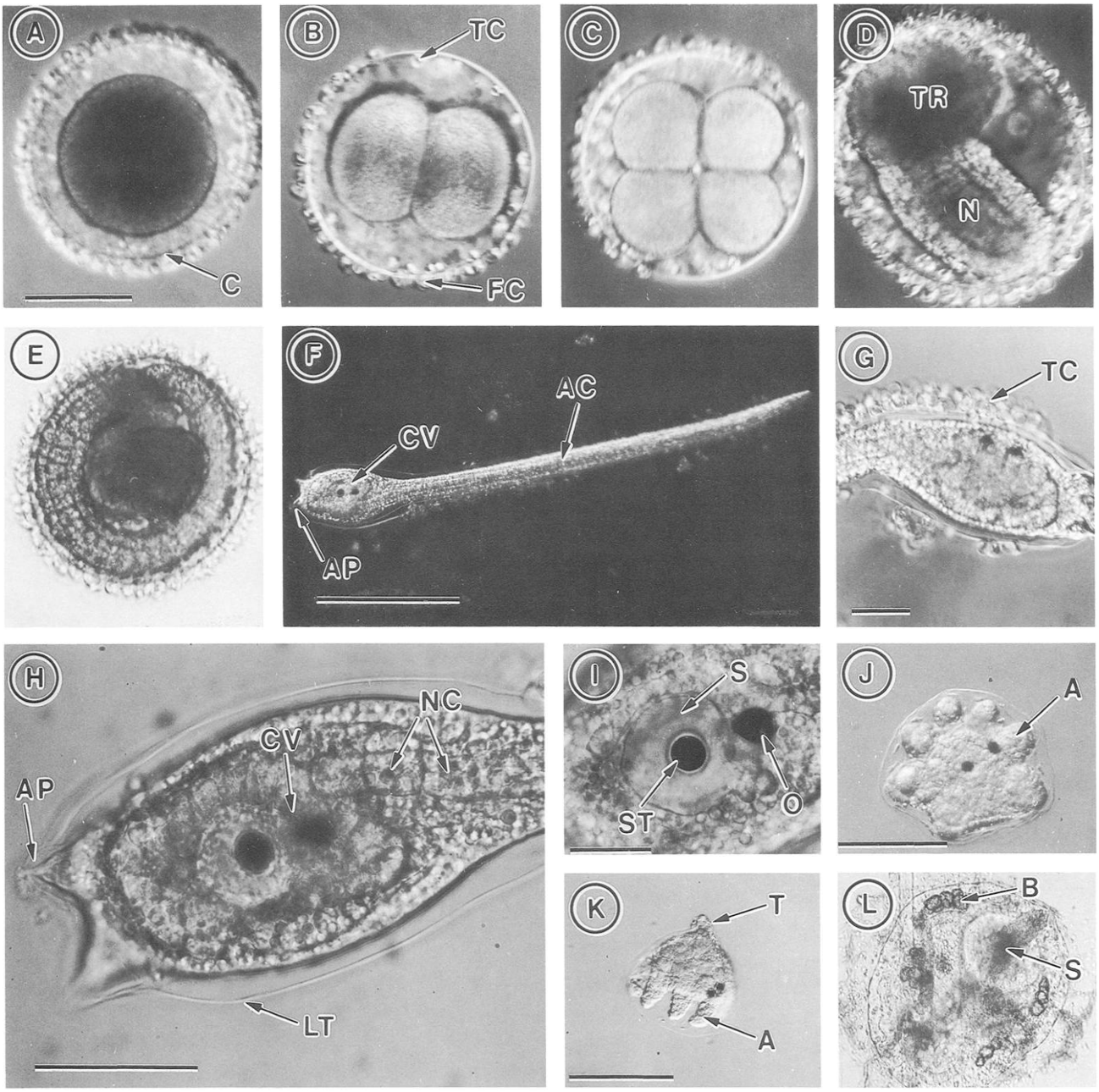



**Fig. 10.** Embryogenesis and larval development of *Bathypera feminalba.* **A.** Newly spawned egg showing chorion (C). Scale bar A–E, J, L 100 μm. **B.** Two-cell embryo with extraembryonic cell layers in focus. Test cell (TC); follicle cell (FC). **C.** four-cell embryo. **D.** Early tail-bud stage showing differentiation between trunk region (TR) and tail with notochord (N). **E.** Embryo just before hatching. Tail is coiled completely around trunk. **F.** Tadpole larva. Adhesive papillae (AP); cerebral vesicle (CV); axial complex of the tail (AC). Scale bar 250 μm. **G.** Trunk of a newly hatched tadpole showing adherent test cells (TC). Scale bar 50 μm. **H.** Trunk and anterior end of the axial complex. Adhesive and sensory papilla (AP); outer cuticular layer of larval tunic (LT); cerebral vesicle (CV); notochordal cells (NC). Scale bar 50 μm. **I.** Cerebral vesicle. Statocyst vesicle (S); statolith (ST); pigmented spot of the ocellus (O). Scale bar 25 μm. **J.** Anterior view of recently metamorphosed individual showing 6 primary ampullae (A) surrounded by tunic. Scale bar 100 μm. **K.** Lateral view of juvenile ascidian showing ampullae (A) and remainder of the resorbed tail (T). Scale bar 50 μm. **L.** Juvenile ascidian 33 days after settlement with open siphons (S) and a complete circulatory system with blood cells (B).

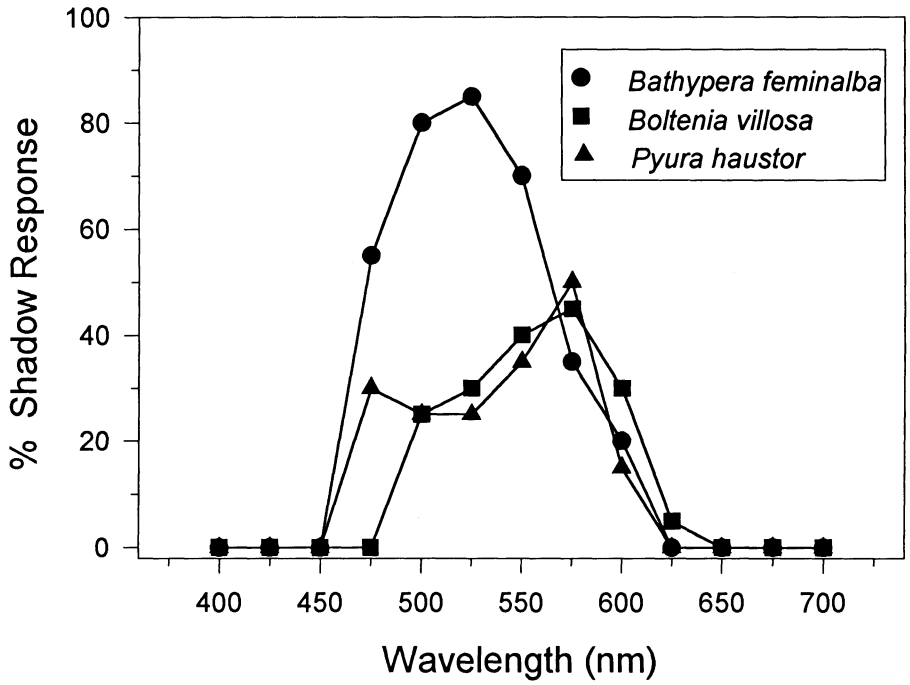



**Fig. 11.** The percentage of tadpoles of *Bathypera feminalba, Boltenia villosa,* and *Pyura haustor* responding to shadows at various wavelengths of monochromatic light. See text for experimental details.

the egg and associated structures was 216 μm (S.D. = 6.99). Sperm resembled those of other ascidians, with a distinct asymmetry due to a large eccentric mitochondrion at the side of the head.

First cleavage (Fig. 10B) began only 1 hour and 45 minutes after fertilization. The embryos originating from the eggs of one individual were white and quite transparent, whereas those originating from the eggs of the second animal were either light yellow or dark yellow. Gastrulation occurred about 9 hours after fertilization and by about 15 hours, elongate embryos with distinct tails and axial complexes had formed (Fig. 10D). Embryos appeared fully formed 24 hours after fertilization (Fig. 10E) and about half of the tadpoles had hatched by 31 hours after fertilization (Fig. 10F).

The tadpoles resembled those of other pyurids and had an overall length of 900 to 970 μm (mean = 945, S.D. = 17.32, n = 20), not including the tail tunic. The average length of the tail tunic was 203 μm (S.D. = 31.97), making the total tadpole length an average of 1148 μm (S.D. = 42.25, range 1100–1210 μm). Tadpoles were transparent. At hatching, they were covered with adherent test cells (Fig. 10G), but these fell off within a few hours. The anterior end of the larval trunk supported 3 large simple conical papillae, the tips of which appeared to extend beyond the outer layer of tunic (Fig. 10H). The trunk also contained a well-formed spherical statolith in a large statocyst and darkly pigmented ocellus (Fig. 10I).

The larvae of *B. feminalba* were generally lethargic in culture. Short bouts of upward swimming alternated with longer periods of resting on the bottom of the culture vessel. The larvae exhibited a well-developed shadow response in white light and responded to monochromatic light at wavelengths between 475 and 600 nm (Fig. 11), with a peak sensitivity in the blue region of the spectrum, at 500 to 525 nm. Two shallow-water pyurids from the Puget Sound region, *Boltenia villosa* and *Pyura haustor,* displayed similar ranges of sensitivity but their peaks were in the green region of the spectrum at 575 nm (Fig. 11). At the single light intensity tested, a larger percentage of tadpoles of *B. feminalba* responded than either of the other species, possibly indicating that the deep-water species is more sensitive to low light levels.

The first settlement was observed 48 hours after fertilization and approximately 19 hours after hatching. Following tail resorption, the juveniles produced 6 primary ampullae, which spread the tunic out over the substratum (Fig. 10J). The darkly pigmented structures of the cerebral vesicle remained visible for several days after settlement, but other internal structures were not discernable with light microscopy. Some tadpoles remained active and demonstrated strong shadow responses for at least 21 hours longer than the first individual that settled. A single juvenile that survived in culture for 33 days after settlement (Fig. 10L) had open siphons, a clearly visible circular blood vessel with large blood cells, and a few tiny spicules. At this time, an array of regularly spaced spicules was growing around the incurrent siphon, but only a few irreg-

ularly spaced ones were present around the excurrent siphon.

## Discussion

The fauna of the slope depths and deep fjordic habitats of the Northeast Pacific remains relatively poorly known. Whereas finding a new ascidian species in the shallow waters of this region is relatively unusual, at least 5 new species from deeper waters have been described in the past 2 years (Lambert 1993; this paper; Vázquez & Young, unpubl.). From the standpoint of biological processes such as recruitment, reproduction, and physiology, the deep-water fauna of the North Pacific remains almost completely unknown.

*Bathypera feminalba* n. sp. is most clearly distinguished from other species of the genus by the presence of 7 longitudinal folds in the branchial sac. *B. splendens* and *B. goreaui* have 6 branchial folds and *B. ovoida,* the other species occurring in the North Pacific, has 9 folds. The shape of fully formed spicules is also diagnostic; only in *B. feminalba* do the spicules always bear a single long spine at the apex. The spicules of *B. ovoida* have several long spines of equal length surrounding the apex, and those of both *B. goreaui* and *B. splendens* have a large number of small spines covering the entire apex. Monniot & Monniot (1983) synonymized the two previously described antarctic species, *B. splendens* and *B. hastaefera,* eliminating the latter species. However, according to the original description of *B. hastaefera* by Vinogradova (1962), the spicules of this species have one spine that is much larger than the others (see figure in Kott 1969). In this respect, it appears that at least some individuals of *B. splendens* must have spicules resembling those of *B. feminalba.* The new species described here is clearly distinguished from *B. splendens* not only by a different number of branchial folds, but also by the relative lengths of the gonoducts. In *B. feminalba,* the oviduct and sperm duct are both very short, whereas in *B. splendens,* the oviduct is much longer than the sperm duct.

The characteristic tunic spines of *Bathypera* spp. were studied in some detail by Lowenstam (1989), who included in his study one unidentified species dredged from 60–100 m depths off Santa Catalina Island, California. Scanning electron micrographs of spines from this species are identical to those of *Bathypera feminalba,* so we can assume that Lowenstam's conclusions about biomineralization apply equally to the latter. Thus, the spicules are made of magnesium calcite with 8.4% of the weight being composed of $MgCO_3$ (Lowenstam 1989). Lowenstam described the fully formed spicules, but did not discuss the ontogenetic changes in spicular form that occur as the tunic margin grows. Our examination of the basal edge of juveniles of *B. feminalba* indicates that the spicule begins as a tiny round crystal, becomes lens-shaped, and then elongates to form the castellate and spined form of the adult spicule. The early stages were found only on the margin of the tunic, indicating that only this region of the tunic grows.

The genus *Bathypera* has a disjunct biogeographical distribution with one species being found at bathyal depths in the Antarctic, *B. splendens*; one at slope depths in the tropical Caribbean, *B. goreaui*; and one at abyssal depths off California and in shallower water in the Northwest Pacific, *B. ovoida.* With the addition of a fourth species, *B. feminalba,* from the same general area of the North Pacific as *Bathypera ovoida,* we see for the first time two species of *Bathypera* occurring within the same hemisphere and ocean. *B. feminalba,* like all other known ascidians, has relatively short-lived larvae with little ability to disperse long distances, so this highly disjunct distribution could possibly be an example of vicariance. As the larvae of *Bathypera* species swim for no more than a few days, vast soft-bottom regions separating suitable hard-bottom habitats in the deep sea could easily provide barriers to dispersal that would promote allopatric speciation. However, this interpretation must be made cautiously. Because of limited sampling in deep-sea hard-bottom areas, the actual ranges of the species of *Bathypera* are not well documented. The bathyal hard-bottom regions of South and Central America remain virtually unstudied, so there remains a good chance that one or more species of *Bathypera* span the broad geographical gap between *B. ovoida* in California and *B. splendens* in the Antarctic. Millar & Goodbody (1974) speculated that *B. goreaui* may be widespread in the Caribbean, as the deep reef habitats in this part of the world have seldom been examined carefully.

Kott (1969) hypothesized that the genus *Bathypera* originated in the Antarctic and that *B. ovoida,* which at that time was known only from a depth of 3680 m (where it must live at near-antarctic temperatures), is an example of tropical submergence. Several recent pieces of evidence cast doubt on this scenario. Nishikawa (1981) found *B. ovoida* at a depth of only 184 m in Sagami Bay and additional species have now been described from both the temperate North Pacific and the tropical Atlantic. Because the Atlantic species lives in quite shallow waters and at relatively warm temperatures, it is more likely to have originated from a Pacific stock isolated by the Isthmus of Panama than from equatorial submergence of an antarctic form. Moreover, with the synonymy of *B. hastaefera* and *B. splendens* (Monniot & Monniot 1983), this small ge-

nus now appears more diverse in the northern hemisphere than in antarctic waters. Until we have adequately explored the appropriate depths and habitats along both coasts of Central and South America, the biogeographical origin and spread of this genus must remain enigmatic.

*Bathypera* is characteristically a deep-water genus, with at least 2 of the 4 species occurring at depths greater than 3000 m, and with all but one known species (*B. goreaui*) extending into bathyal or abyssal depths. The species that have been found as shallow as 30–50 m all have eurybathal distributions that range from several hundred to several thousand meters, with the possible exception of *B. goreaui,* which has been collected over only a 40–m depth range (Millar & Goodbody 1974). However, the narrow depth distribution of this latter species probably reflects a limited collecting effort at depths beyond those that can be worked by scuba divers.

Many abyssal and bathyal ascidians are adapted to life in deep water and soft sediments by having ramified attachment surfaces and modified branchial sacs (reviewed by Monniot & Monniot 1978). Members of the genus *Bathypera* demonstrate none of these characteristic deep-sea adaptations; indeed, they more closely resemble typical shallow-water ascidians by having a wide base for attachment to firm substrata and normal, functional stigmata. Indeed, *B. feminalba* was never found in soft-bottom areas except on firm surfaces such as shells, cobbles, or larger ascidians. The absence of *B. feminalba* at 200–220 m was probably due to the local absence of suitable hard surfaces. This is also the likely reason that the species was absent below 240 m; all of our observations at these depths were of very soft bottoms. It occurred much more commonly on the vertical faces of cliffs and the vertical or downward facing surfaces of large rocks and boulders. As a rule, juveniles of hard-bottom ascidians are very susceptible to smothering by silt (Young & Chia 1984). *Bathypera feminalba* is probably no exception, as individuals were never found on silt-covered slopes or platforms. Likewise, every specimen of *B. goreaui* collected from the deep reef face off Jamaica was found on the underside of a horizontal slab of coral rock (Millar & Goodbody 1974). Other species of *Bathypera* from the North Pacific and Antarctic have all been collected by dredge, so we know little about the microhabitats in which they live.

Many ascidians in the family Pyuridae occur in shallow water; indeed, a number of them are dominants in the lower intertidal zone (Millar 1971). We must ask, therefore, why species in the genus *Bathypera,* which normally have very wide depth ranges, virtually always have an upper depth limit somewhere between 25 and 50 m. Larval, juvenile, or adult stages may be involved in the control of this distribution.

The larvae of *B. feminalba,* the first to be described in this genus, are similar to the larvae of other pyurids except for their unusually large adhesive/sensory papillae. The tadpoles are photosensitive, display a characteristic ascidian shadow response (low photokinesis), and have a gravity receptor. Although the behavior has not been studied in detail, the larvae are apparently well equipped for making adjustments in their vertical position, using both light and gravity as cues (reviewed by Svane & Young 1989). Perhaps swimming behaviors facilitate retention in deep water. Even with good depth regulation capabilities, it seems almost inconceivable that some individuals would not be carried into shallow water before settlement to be affected by selective mortality occurring during the juvenile and/or adult stage.

The wavelength sensitivity of *B. feminalba* peaks in the blue region of the spectrum, in contrast to shallow-water confamilials from the same geographic region, which respond more frequently to green than to blue light (Fig. 11). A shift from green to blue sensitivity might be expected if we were comparing coastal animals with oceanic species (reviewed for fishes by Blaxter 1970). Coastal species should have greater sensitivity to green light because phytoplankton, particulate matter, and humic acids filter out much of the light at those shorter wavelengths that penetrate deepest in clear oceanic waters. It is difficult to understand why a deep-water species living in coastal waters should have greater blue sensitivity than shallow-water animals in the same coastal regions. One possibility is that the coastal populations of *B. feminalba* represent only a small portion of the total population and that visual pigments are actually adapted to the light regimes in offshore habitats. At the single light intensity tested, a larger percentage of tadpoles of *B. feminalba* responded than either of the other species, possibly indicating that the deep-water species is more sensitive to low light levels than the two species from shallow water.

Predation by shallow-water fishes or other predators could set the upper depth limits of *B. feminalba,* but we have no evidence for or against this. All members of the genus *Bathypera* have a delicate, thin tunic compared to the leathery tunic of most other pyurids—one exception is *Herdmania momus,* which also produces spicules (Lambert 1992)—but they are also generously endowed with sharp spicules that could make penetration by gastropods and other small predators difficult. Lowenstam (1989) speculated without evidence that the spines of *B. feminalba* function as a defense mechanism. Hard non-mineralized tunic spines in larger

pyurids have been shown experimentally to deter predation by gastropods (Young 1986).

Analysis of small-scale distributional patterns near the upper depth limit of *B. feminalba* indicates that density is negatively correlated with the percent cover of red coralline algae. Moreover, the lower distributional limit of these algal crusts is very near the upper limit of the ascidians. Perhaps larvae avoid settling on the algae. Alternatively, juveniles may be dislodged by algal herbivores (e.g., the chiton *Tonicella lineata* and the limpet *Acmaea mitra* specialize on coralline algae) or by exfoliation of the algal cells. Coralline algae are known to stimulate metamorphosis of various motile invertebrates including abalones (Morse & Morse 1984), chitons (Barnes & Gonor 1973), and echinoderms (Barker 1977; Rowley 1989), all of which probably use the algal cells or associated organisms as food. Some sessile invertebrates such as corals (Sebens 1983; Morse et al. 1988) and spirorbid polychaetes (Gee 1965) also settle preferentially on corallines, but these species have hard skeletal structures from shortly after metamorphosis, unlike small ascidians, which tend to be soft and easily removed from a surface. Although there has been much work on inhibition of settlement in recent years (reviewed by Davis et al. 1989; Pawlik 1992), inhibition of settlement by coralline algae has not, to our knowledge, been reported for any invertebrate. The observed negative correlation between coralline algae and *B. feminalba* deserves attention. Whether the pattern involves larvae or settlement at all, or results from differential post-settlement survival, remains to be discovered.

On a scale of centimeters, the distribution of *B. feminalba* is highly aggregated even deeper than coralline algae occur. Aggregation has been attributed to gregarious settling in other ascidian species (Young & Braithwaite 1980; Young 1988; Havenhand & Svane 1989). In ascidians, several different kinds of gregariousness are known. Larvae may settle directly on or adjacent to established adults (Young 1988), or cohorts of larvae may settle together, responding either to other larvae or to newly established juveniles. Similar spatial patterns can sometimes be attributed to hydrodynamic effects (Havenhand & Svane 1991) or philopatric dispersal of anural species with adhesive eggs (Young et al. 1988). As *B. feminalba* often lives where there is little or no current and has active tadpoles well endowed with sensory organs, it seems most likely that the small-scale aggregation in this species results from larval behavior. We never found small individuals of *B. feminalba* attached to larger ones, but we commonly found small recruits around the base of an adult (Fig. 2A). This suggests that the larvae respond to the presence of adults, but do not prefer the adult tunic as a settlement site as in *Corella inflata* (Lambert 1968). Perhaps spicules render the tunic of *B. feminalba* inappropriate for settlement. The surface of this species, unlike that of many pyurids, is always completely free of fouling organisms.

*Bathypera feminalba* probably reproduces and recruits seasonally rather than continuously near the shallow end of its depth range, and examination of individuals from below the euphotic zone suggests that these portions of the population are in synchrony with the shallow portions. The factors entraining reproductive cycles are poorly studied in ascidians. It would be especially interesting to know how cycles are synchronized in species such as this one, which spans a vast vertical range and must experience very different environmental conditions at the extremes of the range. Based on a sequential pair of samples from a single population (Fig. 9), it appears that early stages of *B. feminalba* grow about one mm per month. Assuming that changes in diameter are a linear function of age, an animal should attain reproductive size within one year. However, without any data on survival of marked populations, we do not know if the entire life cycle is completed in one year or if individuals persist for multiple years. Other pyurids that have been studied tend to be perennial species reproducing iteroparously. There remain many questions concerning the life-history, population biology, and ecology of this and other deep-water ascidians in the fjords of western Canada. We hope that this preliminary information will stimulate more complete physiological and ecological studies of the deep fjordic fauna and of other deep-water ascidians worldwide.

**Acknowledgments.** Opportunities to observe *Bathypera* from the Pisces V submersible were provided by Verena Tunnicliffe (University of Victoria) and Ron Shimek (Bamfield Marine Station). Ron Shimek provided welcome encouragement and academic and logistical support throughout this study and served as dive buddy on every deep scuba dive. Chuck Anderson arranged a dive trip to Saanich Inlet on his personal boat, *Foxy Lady.* Dredging opportunities and scuba support were made available by Bamfield Marine Station (R. Foreman, director) and Friday Harbor Laboratories (D. Willows, director). Early field work was supported by a graduate research fellowship to C.M.Y. from the University of Alberta. Later work was assisted by members of the 1985 ascidian biology class at Bamfield Marine Station. E.V. was supported by a postdoctoral fellowship from the Conselleria de Educacion e Ordenacion Universitaria, Xunta de Galicia (Spain). We thank Bill Austin, Anne Bergey, Dave Denning, Kerry Irons, Sabina Leader, Claudia Mills, Steve Rumrill, Ron Shimek, and Verena Tunnicliffe for making observations of ascidians during their submersible dives. This is Harbor Branch contribution No. 1063.

## References


Austin WC 1985. An annotated Checklist of Marine Invertebrates in the Cold Temperate Northeastern Pacific. Vol. 1. Khoyatan Marine Laboratory, Cowichan Bay, B.C.

Barker MF 1977. Observations on the settlement of brachiolaria larvae of *Stichaster australis* (Verrill) and *Coscinasterias calamaria* (Gray) (Echinodermata: Asteroidea) in the laboratory and the shore. J. Exp. Mar. Biol. Ecol. 30: 95–108.

Barnes JR & Gonor JJ 1973. The larval settling response of the lined chiton, *Tonicella lineata.* Mar. Biol. 20: 259–264.

Blaxter JHS 1970. Light-Animals-Fishes. In: Marine Ecology, Vol. I. Environmental Factors, Part 1. Kinne, O ed., pp. 213–320. Wiley-Interscience, London.

Burd BJ & Brinkhurst RO 1984. The distribution of the galatheid crab *Munida quadrispina* (Benedict, 1902) in relation to oxygen concentrations in British Columbia fjords. J. Exp. Mar. Biol. Ecol. 81: 1–20.

Cloney RA & Florey E 1968. Ultrastructure of cephalopod chromatophore organs. Z. Zellforsch. 89: 200–280.

Davis AR, Targett NM, MacConnell OJ, & Young CM 1989. Epibiosis of marine algae and benthic invertebrates: natural products chemistry and other mechanisms inhibiting settlement and overgrowth. Bio. Mar. Chem. 3: 85–114.

Fay RC & Vallee JA 1979. A survey of the littoral and sublittoral ascidians of southern California, USA, including the Channel Islands. Bull. S. Calif. Acad. Sci. 78: 122–135.

Gee JM 1965. Chemical stimulation of settlement in larvae of *Spirorbis rupestris* (Serpulidae). Anim. Behav. 13: 181–186.

Havenhand JM & Svane I 1989. Larval behaviour, recruitment, and the rôle of adult attraction in *Ascidia mentula* O.F. Müller. In: Reproduction, Genetics, and Distributions of Marine Organisms. Ryland JS & Tyler PA, eds., pp. 127–132. Olsen & Olsen, Fredensborg, Denmark.

——— 1991. Roles of hydrodynamics and larval behaviour in determining spatial aggregation in the tunicate *Ciona intestinalis.* Mar. Ecol. Prog. Ser. 68: 271–276.

Hopkins B & Skellam JG 1954. A new method for determining the type of distribution of plant individuals. Ann. Bot. Lond. N.S. 18: 213–227.

Kott P 1969. Antarctic Ascidiacea. American Geophysical Union Antarctic Res. Ser. 13: 1–239.

——— 1971. Antarctic Ascidiacea II. American Geophysical Union Antarctic Res. Ser. 17: 11–82.

Lambert G 1968. The general ecology and growth of a solitary ascidian, *Corella willmeriana.* Biol. Bull. 135: 296–307.

——— 1992. Ultrastructural aspects of the spicule formation in the solitary ascidian *Herdmania momus* (Urochordata, Ascidiacea). Acta Zoologica 73: 237–245.

——— 1993. Three new species of stolidobranch ascidians (Chordata: Ascidiacea) from the California continental shelf. Proc. Calif. Acad. Sci. 48(4): 109–118.

Levings CD, Foreman RE, & Tunnicliffe VL 1983. Review of the benthos of the Strait of Georgia and contiguous fjords. Can. J. Fish. Aquat. Sci. 40: 1120–1141.

Lowenstam HA 1989. Spicular morphology and mineralogy in some Pyuridae (Ascidiacea). Bull. Mar. Sci. 45: 243–252.

Millar RH 1971. The biology of ascidians. Adv. Mar. Biol. 9: 1–100.

Millar RH & Goodbody I 1974. New species of ascidians from the West Indies. Stud. Fauna Curacao 45: 142–161.

Monniot C 1965. Etude systématique et evolutive de la famille des Pyuridae (Ascidiacea). Mém. Mus. natl. Hist. nat., Paris, Sér.A, Zool. 36: 1–203.

Monniot C & Monniot F 1972. Clé mondiale des genres d'Ascidies. Arch. Zool. esp. gén. 113: 311–367.

——— 1978. Recent work on the deep-sea tunicates. Oceanogr. Mar. Biol. Annu. Rev. 16: 181–228.

——— 1983. Ascidies antarctiques et subantarctiques: morphologie et biogeographie. Mem. Mus. natl. Hist. nat., Paris, ser. A, Zool. 125: 1–168.

Morse ANC & Morse DE 1984. Recruitment and metamorphosis of *Haliotis* larvae induced by molecules uniquely available at the surfaces of crustose red algae. J. Exp. Mar. Biol. Ecol. 75: 191–215.

Morse DE, Hooker N, Morse ANC, & Jensen RA 1988. Control of larval metamorphosis and recruitment in sympatric agariciid corals. J. Exp. Mar. Biol. Ecol. 116: 193–217.

Nishikawa T 1981. Contributions to the Japanese ascidian fauna XXXIV. Record of *Bathypera ovoida* (Ritter, 1907) from Sagami Bay. Publ. Seto Mar. Biol. Lab. 26: 187–190.

Pawlik JR 1992. Chemical ecology of the settlement of benthic marine invertebrates. Oceanogr. Mar. Biol. Annu. Rev. 30: 273–335.

Rowley RJ 1989. Settlement and recruitment of sea urchins (*Strongylocentrotus* spp.) in a sea-urchin barren ground and a kelp bed: are populations regulated by settlement processes? Mar. Biol. 100: 485–494.

Sanamyan K 1992. Ascidians from the Sea of Okhotsk collected by R.V. *Novoulyanousk.* Ophelia 36(3): 187–194.

Sebens KP 1983. The larval and juvenile ecology of the temperate octocoral *Alcyonium siderium* Verrill. I. Substratum selection by benthic larvae. J. Exp. Mar. Biol. Ecol. 71: 73–89.

Svane I & Young CM 1989. The ecology and behaviour of ascidian larvae. Oceanogr. Mar. Biol. Annu. Rev. 27: 45–90.

Tunnicliffe V 1981. High species diversity and abundance of the epibenthic community in an oxygen-deficient basin. Nature 294: 354–356.

Tunnicliffe V & Wilson K 1988. Brachiopod populations: distribution in fjords of British Columbia (Canada) and tolerance of low oxygen concentrations. Mar. Ecol. Prog. Ser. 47: 117–128.

Van Name WG 1945. The North and the South American ascidians. Bull. Amer. Mus. Nat. Hist. 84: 1–479.

Vinogradova NG 1962. Ascidiae simplices of the Indian part of the Antarctic. Biol. Results Soviet Antarctic Exped.

(1955–1958), 1. Explorations of the fauna of the seas. Acad. Sci. USSR, Zoological Institute 1(9): 195–215.

Young CM 1986. Defenses and refuges: alternative mechanism of coexistence between a predatory gastropod and its ascidian prey. Mar. Biol. 91: 513–552.

——— 1988. Ascidian cannibalism correlates with larval behavior and adult distribution. J. Exp. Mar. Biol. Ecol. 117: 9–26.

Young CM & Braithwaite LF 1980. Larval behavior and post-settling morphology in the ascidian *Chelyosoma productum* Stimpson. J. Exp. Mar. Biol. Ecol. 42: 157–169.

Young CM & Chia FS 1984. Microhabitat associated variability in survival and growth of subtidal solitary ascidians during the first 21 days after settlement. Mar. Biol. 81: 61–68.

Young CM, Gowen RF, Dalby, J Jr, Pennachetti CA, & Gagliardi D 1988. Distributional consequences of adhesive eggs and anural development in the ascidian *Molgula pacifica* (Huntsman, 1912), Biol. Bull. 174: 39–46.