BOTRYLLOPHILUS BAMFIELDENSIS, NEW SPECIES (COPEPODA: CYCLOPOIDA: ASCIDICOLIDAE), LIVING IN A COMPOUND ASCIDIAN FROM THE WEST COAST OF VANCOUVER ISLAND, CANADA

Shigeko Ooishi

Friday Harbor Laboratories, University of Washington, 620 University Road, Friday Harbor, Washington 98250–9299, U.S.A. (ooishi@fhl.washington.edu)

ABSTRACT

Botryllophilus bamfieldensis, new species, is described on the basis of specimens of both sexes living in a compound ascidian from the Bamfield area in Barkley Sound, west coast of Vancouver Island, Canada. The morphological details of the female fit the diagnostic features for morphotype A of the genus. The female can be distinguished from its eight congeners by the sharply pointed triangular apex of the cephalosome, the weakly segmented metasome, and the markedly elongated exopods of left legs 1–4. The male of the new species differs from the only known male of a species of morphotype A (*B. abbotti*) in that the endopod of leg 1 bears five variously shaped spines and five plumose steae. It is proposed that females of nine species of morphotype A be further divided into two subgroups based on two patterns of armature for legs 1–4.

Among 13 recognizable known species (based on Illg and Dudley, 1980; Ooishi, 1999) of the genus Botryllophilus, eight have been assigned to the female morphotype A in which the urosome consists of five segments (based on Ooishi and Illg, 1988; Ooishi, 1999) as follows: (1) B. brevipes Brément, 1909; (2) B. banyulensis Brément, 1909; (3) B. norvegicus Schellenberg, 1921; (4) B. bergensis Schellenberg, 1921; (5) B. brevipes Sars, 1921, which is a primary junior homonym of *B. brevipes* Brément, 1909; (6) B. inaequipes Hansen, 1923; (7) B. abbotti Ooishi and Illg, 1989; and (8) B. koreensis Seo and Lee, 1995. These species were described from various European seas and also from the Pacific Ocean: numbers 1 and 2 from the Mediterranean Sea (Port Vendres); numbers 3-5 from the Norwegian Sea (Trondheimsfjord, Bergen, and Espevaer, respectively); number 6 from the North Atlantic Ocean (Davis Strait); numbers 7 and 8 from the eastern and western Pacific Ocean (Monterey Peninsula and Chindo Island [Korea], respectively).

The copepod described in this paper is the third Pacific species of *Botryllophilus* and the second species of morphotype A in which the female and the male are described. Except for *B. norvegicus* and *B. koreensis*, which live in solitary ascidians (Styelidae), six others (one new, five described) are associated with various compound ascidians,

but the host of one (*B. inaequipes*) is not known.

The new Pacific species has many other diagnostic characters of type A: antennule 4-segmented; antenna with 7 armature elements; mandible including 2 sharp conical spines posteriorly; maxillulary precoxa with 6 setae; maxilla with 2 subordinate setae; leg 5 without conspicuous gap between 2 short distal setae; apparatus at gonopore including 2 conical elements and several sclerites; lateral caudal spine with rounded apical margin. The armature of the legs and other appendages includes variously modified elements. Some of them are illustrated by SEM in this paper. It is thought that these elements are also typical of certain other species of type A.

In a redescription of *B. ruber* Hesse, 1864, which is the type species and belongs to the female morphotype B (urosome consisting of 7–9 segments), it was stated that a combination of the leg morphology and its armature formula are species-specific for five members of type B (Ooishi, 1999). In the legs of these copepods, the arrangement of armature elements (spines, setae) was distinctly different even if the number of elements was the same.

In some species of type A, the armature elements are of the same number and of nearly the same arrangement. In order to explain the significance of the formula for leg armature in type A species, the formulas for the new species and for all its congeners will be compared and discussed in this paper, and type A species will be divided into two subgroups.

MATERIALS AND METHODS

Collecting copepods from the ascidian host was carried out at the Bamfield Marine Station, Vancouver Island, British Columbia, Canada, during October 1991. Purplish colonies (mostly 6-10 cm wide) of Eudistoma purpuropunctatum Lambert (Aplousobranchia: Polycitoridae) were obtained from the Bamfield area by dredging or diving arranged by the Station. A single female copepod was found in the branchial sac of one zooid, but all other specimens (females and males) were found in the common test. This ascidian also rarely harbors Haplostoma sp. (Ascidicolidae: Haplostomatinae). (In September 1993, attempts to collect the ascidian host by dredging were not successful.) The copepods were fixed in 95% ethanol and stored in 70% ethanol. Scanning electron micrographs (SEM) of the female were based on this preserved material. (They were taken at the Electron Microscopy Center, University of South Carolina, Columbia, in 1994.) The specimens were measured and dissected in lactic acid for drawings. Drawings were made with the aid of a camera lucida.

In the formula for the armature elements of legs 1–4, the total number of spines or spiniform elements (Roman numerals) is noted first and connected by a dash with the number of setae or setiform elements (Arabic numerals) in each segment. The total number (T) of these elements is given in parentheses for protopod, endopod, and exopod.

The abbreviations used are: A1 = antennule, A2 = antenna, MD = mandible, MX1 = maxillule, MX2 = maxilla, MXP = maxilliped, PG = paragnath, R = rostrum, I = first armature element of antenna or leg 1 and also first major seta of maxilla, I_1 = first major seta on first segment of antennule, 1 = 1 independent seta of antennule, and +1 = 1 subordinate seta associated with a major seta of antennule or maxilla.

Systematics

Family Ascidicolidae Thorell, 1859 Subfamily Botryllophilinae Sars, 1921 Genus *Botryllophilus* Hesse, 1864 *Botryllophilus bamfieldensis*, new species Figs. 1–10

Material Examined.—51 $\[mathcar{e}\]$ and 4 $\[mathcar{e}\]$ living in common test of *Eudistoma purpuropunctatum* Lambert, 5 m depth, Ohiat Island, Bamfield area (125°08′W, 48°50′N). Holotype $\[mathcar{e}\]$ (USNM 298311) and allotype $\[mathcar{e}\]$ (USNM 298312), collected on 4 October 1991, and paratypes 40 $\[mathcar{e}\]$ and 1 $\[mathcar{e}\]$ (USNM 298313), collected on 4 and 5 October 1991, deposited in the National Museum of Natural History, Smithsonian Institution, Washington, D.C. Remaining specimens (intact, dissected, or sputter-coated with gold) in the collections of the author.

Female.—Body (Fig. 1a-c) almost straight, without distinct curvature ventrally in pro-

some and urosome, although anal segment and caudal rami directed somewhat ventrally. Length (excluding caudal spines) 1.63 mm; greatest width and thickness 0.40 and 0.45 mm, respectively, in fourth metasomal segment. Average length 1.66 mm, based on 5 females. Proportional lengths about 1:2.27:2.65 for cephalosome, metasome, and urosome. Ratio of length of prosome to that of urosome 1.2:1. Single globular egg sac (Fig. 1d) dorsal between fifth legs, usually containing 15 eggs (embryos). Sparsely scattered hairlike sensilla (Fig. 6h) found on most of body surface.

Cephalosome triangular, slightly longer than wide, bearing appendages through maxillipeds (Fig. 1e, f); antennae asymmetrical. Rostral area sclerotized, not distinctly protruded ventrally (in lactic acid), but its area (shown by SEM) recognizable as somewhat expanded portion incorporated distally into sharply pointed triangular apex of cephalosome (Fig. 6a). Two small sensilla (right, left) located on ventral sclerotized rostral area and 2 pairs (anterior, posterior) of larger sensilla on pointed apex of cephalosome (Figs. 1e–g, 6a); each larger sensillum protruding from mammiform or tubular projection.

Metasome (Fig. 1b) subequally and weakly divided into 5 segments by integumental indentations; in contracted specimens (in life or fixed material, not in lactic acid), segmentation distinct. Width and thickness gradually increasing from first to fourth segments. Legs (Fig. 1c) biramous, asymmetrical in structure and arrangement. Right and left legs of first pair clearly separated from each other, but right legs coming gradually closer to left legs in second to fourth pairs; intercoxal sclerite absent. Thus, legs 1–4 obliquely placed from right side to left side. Fifth segment, with uniramous fifth legs protruded from posterolateral sides, abruptly narrowed toward urosome (Fig. 1b).

Urosome (Fig. 1a, c) cylindrical, 5-segmented (1 genital, 3 abdominal, and 1 anal). Anterior margin of genital segment recognizable behind metasome; 2 gonopores dorsolateral, and transverse slit for genital atrium midventral. Anal segment with terminal caudal rami; anus opening posterodorsally.

In living specimens, body white, opaque, with small red eye (depicted by dotted lines,



Fig. 1. *Botryllophilus bamfieldensis,* new species, female. a, body form, dorsal; b, same specimen, lateral; c, same specimen, ventral; d, egg sac between right and left fifth legs, dorsal; e, cephalosome, ventral; f, cephalosome, left, lateral; g, arrangement of sensilla on rostral area (1 pair on sclerotized portion) and on apex of cephalosome (2 pairs), anteroventral.

Fig. 1a) internally near anterior distal margin of cephalosome; gut similarly red. Eggs in body and eggs (embryos) in egg sac green.

Antennule (Figs. 2a, b, 6b) divided into 4 segments, with total of 28 simple setae including 1 aesthete-like seta (see arrow) on fourth segment. First segment with 8 setae (I_1+2 , II_1 , III_1+1 , IV_1 , V_1); second segment with 5 setae (1 [indicated by black dot], I_2 , II_2+2); third segment with 3 setae (I_3+2); and fourth segment with 12 setae (I_4 , 11).

Antenna 4-segmented and asymmetrical with respect to arrangement of armature elements. In right antenna (Fig. 2c), proportional lengths about 1:2.7:0.5:2.2 for segments 1-4, measured along anterior margins. Fourth segment with 7 elements; 2 spines (I, II) on medial margin, 3 spines (III–V) and 2 setiform elements (VI, VII), from medial to lateral on distal margin. Proportional lengths about 1:1:1:1:2:3:4 for 7 elements (I–VII). Five short spines (I–V) with serrated margin distally, and 2 long setiform elements also with minute serrations. Left antenna (Fig. 2d) with 7 setiform elements (4 stout, 3 slender); remnant of marginal serration of stout seta visible only by SEM (arrow 1, Fig. 6b). These elements distinctly longer than corresponding elements in right antenna.

Labrum (Fig. 2e) subtriangular; each side of rounded apical margin ornamented with row of several minute hairlike setules.

Mandible (Fig. 2f, g) consisting of coxa and elongated palp (basis, exopod, and endopod fused). Gnathobasic medial margin of coxa with double rows of minute spinules (anteriorly ending in 1 longer element) and single row of 2 conical spines and 2 sharp conical spines on common base (or 1 bifurcate spine), from anterior to posterior. Posterior 2 sharp conical spines widely separated from anterior 2 conical spines; anterior sharp conical spine with minute serration on anterior edge. Palp with 9 setae: exopodal region with 3 setae (1 long proximal; 2 much longer distal on common base); and endopodal region with 6 setae (2 medial [long, short], 2 long distolateral, and 2 long terminal).

Paragnath (Fig. 2e, PG) oblong and beanshaped, located medial to oral sclerite and close to lateral margin of labrum. Maxillule (Fig. 2h) consisting of precoxa and palp (coxa, basis and exopod fused; endopod 1-segmented). Precoxa with 6 plumose medial setae; 2 of them (indicated by black dots) rather stout. Epipodite of coxa represented by small lateral lobe with 1 plumose seta. Basis represented by proximal hairlike setules and 2 distal setae on medial margin. Exopodal region with 3 lateral setae (1 directed proximally, 2 directed distally) and 1 small distolateral projection. Endopod with 3 terminal setae.

Maxilla (Fig. 2i) with 7 major setae (I–VII) and 2 subordinate setae (on posterior surface of major seta III). Formula: I; II; III+2; IV; V; VI; VII. Five major setae (I–V) and 1 proximal subordinate seta plumose; remaining setae simple.

Maxilliped (Fig. 2j) consisting of coxa, basis, and endopod with elongated 2-segmented claw. Coxa with 1 short medial seta proximally, and basis with 2 short setae (anterior, posterior) midway on medial margin. Longer proximal portion of claw with 2 small setae on posterior side, and shorter distal portion notched proximally and slightly tridentate terminally. All setae simple.

In right and left legs 1–4 (Figs. 3a–d, 4a–d), protopod without articulation on body surface and partially sclerotized. Coxa without setal or spinal armature, and basis with 1 simple seta on lateral margin and row or patch of spinules (usually 5, Fig. 6e) on mediodistal margin.

Endopods 2-segmented, and slightly longer than wide; segmentation indistinct in legs 1 and 2 but distinct in legs 3 and 4. Endopods of right and left leg 1 (Fig. 3a, b) bearing setae. Endopods of right legs 2–4 (Figs. 3c, 4a, c) bearing setae plus 2 spiniform elements, each with serrated margin (short proximal and long distal, indicated by black dots); in leg 4, proximal spiniform element elongated. Endopods of left legs 2–4 (Figs. 3d, 4b, d) with only long setae; in leg 4, longest seta on second segment nearly 2.5 times as long as endopod. In legs 1–4, total number of armature elements on endopods identical on both sides.

Exopods of right legs 1–4 (Figs. 3a, c, 4a, c) 1-segmented, approximately trapezoidal; wider than long in legs 1–3 and slightly longer than wide in leg 4. These exopods about as long as endopods, bearing stout



Fig. 2. *Botryllophilus bamfieldensis*, new species, female. a, antennule, left, anterior; b, same specimen, fourth segment, posterior; c, antenna, right, anterior; d, antenna, left, anterior; e, labrum and paragnaths, anterior; f, mandible, left, anterior; g, gnathobasic margin of mandible, showing double rows of spinules, anteromedial; h, maxillule, left, posterior; i, maxilla, right, posterior.



Fig. 3. *Botryllophilus bamfieldensis*, new species, female. a, leg 1, right, anterior; b, leg 1, left, anterior; c, leg 2, right, anterior; d, leg 2, left, anterior.



Fig. 4. *Botryllophilus bamfieldensis*, new species, female. a, leg 3, right, anterior; b, leg 3, left, anterior; c, leg 4, right, anterior; d, leg 4, left, anterior; e, leg 5, right, dorsolateral.

spines, each with marginal serration (Fig. 6c). Most spines (I–V, in case of leg 1) protruding from truncated distal margin (this corresponding to fused lateral and distal margins). Remaining single spine (VI, in case of leg 1) protruding from posterior surface (in legs 1–3) or medial margin (in leg 4); this spine not longer than distalmost spine in legs 1–4. Proportional lengths 1:0.64:0.64:0.7:1.4:1 for 6 spines (I–VI, in case of leg 1). Three short spines between proximalmost and distalmost spines on distal margin of leg 1 represented by 2 in legs 2 and 3 and by only 1 in leg 4.

Distal margin of ramus close to each spine (I–IV, in case of leg 1) somewhat protruded and with 3 or 4 conical spinules next to base of spine (Figs. 3a, 6c). Spinules on ramus close to distalmost spine (V, in case of leg 1) fused and protruded, forming mediodistal corner of ramus, this usually with slightly bifurcated apex. Spinules absent around spine protruded from posterior surface (in legs 1–3) or medial margin (in leg 4).

Exopods of left legs 1–4 (Figs. 3b, d, 4b, d) markedly elongated, sickle-shaped, about 3.5 times as long as wide and much longer than endopods. Exopods of legs 1 and 2 slightly longer than those of legs 3 and 4. Setiform elements arranged on distal fourfifths (legs 1, 2) or three-fifths (legs 3, 4) of lateral margin; each being slender and flexible (Fig. 6d). These elements (I–V, in case of left leg 1) corresponding to spines (Fig. 3a, I–V) on distal margin of right exopod. Single minute vestige visible on posterior surface near distalmost setiform element and corresponding to spine from posterior surface or medial margin of right exopods. Thus, 1 element lacking in exopods of left legs 1-4. Proportional lengths 1:1:1:2:3.5 for 5 setiform elements (I–V, in case of left leg 1). Distalmost element always longest in exopods of legs 1-4.

As in right exopods, lateral margin of ramus close to each of setiform elements (I-IV, in case of left leg 1) slightly protruded and with several conical spinules (Fig. 6d). Apex of ramus close to distalmost element (V, in case of left leg 1) conspicuously protruded, forming laterally curved, pointed apex (not bifurcated) of sickle-shaped ramus and corresponding to mediodistal corner (usually bifurcated) of right exopod. Formula for armature (legs 1-4):

(Right)	Coxa;	Basis (T)	Endopod (T)	Exopod (T)
P_1	0–0;	0–1(1)	0–1; 0–7(8)	VI-0(6)
P_2	0-0;	0-1(1)	0–1; II–4(7)	V-0(5)
P_3	0–0;	0–1(1)	0–1; II–2(5)	V-0(5)
P_4	0–0;	0–1(1)	0–1; II–2(5)	IV-0(4)
(Left)				
P.	0-0:	0–1(1)	0-1: 0-7(8)	0–5(5)
P_2	0-0;	0-1(1)	0-1; 0-6(7)	0-4(4)
P_{3}^{2}	0–0;	0–1(1)	0-1; 0-4(5)	0-4(4)
P	0-0:	0–1(1)	0-1: 0-4(5)	0–3(3)

0-0; 0-1....(1) 0-1; 0-4....(5) 0-3....(3)

Leg 5 (Fig. 4e) almost symmetrical, short. Length about 17% of that of urosome, 1.5 times as long as proximal width. Armature consisting of 3 short simple setae (1 at proximal fourth on dorsal margin, others almost terminal and terminal) and 1 long simple terminal seta; 2 short distal setae closely set, without conspicuous gap between them. Proportional lengths 1:1.3:1.3:4.5 from proximal to terminal; long terminal seta nearly as long as appendage.

Apparatus at gonopore (Fig. 5a) including 2 closely set small conical spines (larger proximal and smaller distal, both articulated) externally and 6 small sclerites internally on medial margin of cuticular flap covering gonopore. Dorsal surface between gonopores with 2 sensilla. Midventral transverse slit located near anterior margin of genital segment (Figs. 1c, 5b), representing opening of genital atrium with copulatory organs internally. Details of copulatory organs not studied.

Anal segment with only hairlike sensilla on dorsal and ventral surfaces, without spinules or pubescence (Fig. 5c-e). Short caudal ramus sclerotized, longer than wide, with 2 simple setae (lateral, dorsal) and 4 curved stout spines (Figs. 5c-e, 6f, g). Medial (MS), dorsal (DS), and ventral (VS) spines sharply pointed, but lateral spine (LS) rounded at apex. Spines with spinular serration.

Male.—Body (Fig. 7a, b) 1 mm long (excluding terminal caudal setae), representing cyclopoid swimming form. Ratio of length to greatest width of prosome 2.2:1. Ratio of length of prosome to that of urosome 1.18:1. In preserved specimens, inner and outer terminal caudal setae directed laterally and medially, respectively, thus crossing distally as seen in males of other species (preserved) of this genus.

Cephalosome (Fig. 7a) subtriangular, bearing symmetrical appendages through maxillipeds (Fig. 8a, b). Triangular rostrum with pointed apex directed ventrally. Anterior sur-



Fig. 5. *Botryllophilus bamfieldensis*, new species, female. a, genital segment, showing apparatus at gonopore on each side, dorsal; b, opening of genital atrium with copulatory organs internally, ventral; c, anal segment and left caudal ramus, dorsal; d, anal segment with left caudal ramus, left, lateral; e, anal segment (posterior portion) with left caudal ramus, ventral.

face (Fig. 8c) ornamented with many minute sensilla. Among these, 3 pairs distinctive and widely separated on both sides.

Metasome (Fig. 7a, b) 4-segmented, bearing legs 1–4 of swimming type, but endopod of leg 1 modified. Legs symmetrical, unlike those of female. Greatest width 0.20 mm in first segment and narrowest (0.13 mm) in fourth segment.

Urosome 6-segmented (Fig. 7c): short segment bearing fifth legs; enlarged genital segment enclosing pair of ovoid spermatophores and bearing sixth legs; 3 abdominal segments; and anal segment bearing caudal rami. Ventral surface of each abdominal segment with 3 or 4 transverse rows of minute denticles.

In life, body opaque and mostly white, but slightly pinkish; large red eye (depicted by dotted line, Fig. 7a) internally near anterior distal margin of cephalosome; gut slightly yellowish.

Antennule (Fig. 8a, b, d) 4-segmented. Ventral margin of first segment hemispherically expanded and second to fourth segments narrower and shorter. First segment with 9 setae on distal margin and at least 87 aesthetes on ventral surface; second segment with 5 setae and 1 aesthete; third segment with 2 setae and 1 aesthete; fourth segment with 10 setae and 2 aesthetes. Total number (26) of setae resembling that (28) in female.

Antenna (Fig. 8e) 4-segmented. Second segment proportionally more elongated than in female. Fourth segment with 7 armature elements: 2 spines (minutely serrated on distal half) on medial margin and 5 setiform elements (with minute serrations) on apical margin.

Mandible (Fig. 9a, MD) consisting of re-



Fig. 6. *Botryllophilus bamfieldensis*, new species, female, scanning electron micrographs. a, cephalosome, ventral, scale bar = $86 \mu m$; b, antennule and antenna (arrow 1 indicating stout seta with remnant of serration), left, anterior, scale bar = $27 \mu m$; c, stout spine and spinules on exopod of right leg 1, anterior, scale bar = $8.8 \mu m$; d, short setiform element and spinules on exopod of left leg 1, anterior, scale bar = $8.6 \mu m$; e, spinules on mediodistal margin of basis of left leg 1, anterior, scale bar = $12.0 \mu m$; f, left caudal ramus with 4 spines, ventral, scale bar = $30 \mu m$; g, spines of left caudal ramus, showing serrated margin of medial caudal spine (MS), ventral, scale bar = $17.3 \mu m$; h, hairlike sensillum on body, surface view, scale bar = $7.5 \mu m$.



Fig. 7. *Botryllophilus bamfieldensis*, new species, male. a, body form, dorsal; b, same specimen, left, lateral; c, urosome (abdominal segments with many transverse rows of denticles), ventral.

duced small coxa and elongated palp (basis, exopod, and endopod fused). Medial margin of coxa with at least 3 short simple setae (numbers 1–3, Fig. 9a). Palp 6 times as long as wide, narrowed at distal one-seventh. Single long plumose seta midway on lateral margin, exceeding distal margin of palp and representing exopodal region. Narrowed distal portion with 5 simple elements (1 short medial seta, 1 small mediodistal seta, 2 long terminal setae, and 1 distolateral setule) and representing endopodal region.

Maxillule (Fig. 9a, MX1) reduced to small unsegmented structure with integumental wrinkles; distal margin bilobed. Inner lobe with 4 simple setae along apical margin; outer lobe without armature. Maxilla (Fig. 9b) reduced to subtriangular, saclike structure. Sclerotized distal portion slightly protruded medially, with 5 simple setae (numbers 1–5). Stout posterior seta (number 1) on prominence at margin of appendage. Small posterior seta (number 2) protruding from anterior side of prominence.

Maxilliped (Fig. 9c) comparable to that of female in structure and armature, but proportionally elongated. Two elongated medial setae with minute serrations (longer anterior, shorter posterior) on basis, differing from shorter simple setae of female.

Legs 1–4 (Figs. 9d, g, 10a, b) consisting of coxa, basis, and rami; intercoxal sclerite present. Endopods 2-segmented in legs 1 and



Fig. 8. *Botryllophilus bamfieldensis*, new species, male. a, cephalosome, ventral; b, cephalosome, left, lateral; c, rostrum, anterior; d, third and fourth segments of antennule, right, anterior; e, antenna, right, anterior.

4, and 3-segmented in all others. Coxa with 1 short plumose seta on mediodistal corner in legs 3 and 4. Basis with 1 short simple seta on lateral margin in legs 1–4 and with several setules on mediodistal margin in legs 2–4. Except for modified endopod of leg 1, armature

of other legs consisting of plumose setae and spines, each spine with serrated hyaline margin.

Endopod of leg 1 (Fig. 9d) 0.4 times as long as exopod. Proximal segment massive, as long as wide, and inserted on posterior side of basis. Distal segment longer than wide,



Fig. 9. *Botryllophilus bamfieldensis*, new species, male. a, mandible (numbers 1–3 showing setae on coxa) and maxillule, right, anterior; b, maxilla, numbers 1–5 showing setae, right, anterior; c, maxilliped, right, posterior; d, leg 1, right, anterior; e, endopod of right leg 1, arrow pointing to embedded portion of proximal rodlike spine, anterior; f, endopod of right leg 1, posterior; g, leg 2, right, anterior.

much narrower than proximal segment, directed laterally. Armature (Fig. 9e, f) including setules, setae, and variously modified spines. Basal segment with 1 plumose seta on medial margin and setules on lateral margin. Distal segment with 3 rodlike spines (2 small, 1 long) and 2 stout bifurcate spines (larger medial, smaller lateral), from proximal to dis-



Fig. 10. *Botryllophilus bamfieldensis*, new species, male. a, leg 3, right, anterior; b, leg 4, right, anterior; c, leg 5 on first urosomal segment, right, anterior (for leg); d, leg 6 on cuticular flap covering gonopore, right, anterior (for leg); e, anal segment and right caudal ramus with armature, dorsal; f, right caudal ramus, showing ornamentation (rows of denticles, 2 minute tubercles near distal margin, and 1 small spinule on distal margin), ventral.

tal on anterior surface, and 4 subequal plumose setae, obliquely arranged on distal two-thirds of posterior surface. Base of proximalmost rodlike spine (arrow) apparently embedded in segment (Fig. 9d, e). Formula for armature (legs 1–4):

	Coxa;	Basis (T)	Endopod (T)	Exopod (T)
P_1	0–0;	0–1(1)	0-1; V-4(10)	I-0; I-1; IV-4(11)
P_2	0–0;	0–1(1)	0-1; 0-2; III-3(9)	I-0; I-1; III-5(11)
P_3	0-1;	0–1(2)	0-1; 0-2; II-2(7)	I-0; I-1; III-5(11)
P ₄	0-1;	0-1(2)	0–1; II–3(6)	I-0; I-1; III-5(11)

Leg 5 (Fig. 10c) consisting of proximal portion (protopod) with 1 simple lateral seta and small free segment (exopod) with 2 simple apical setae (shorter proximal, longer distal).

Leg 6 (Fig. 10d) represented by 2 subequal simple setae near distal margin of cuticular flap covering gonopore; transverse row of denticles present posterior to each seta.

Caudal ramus (Fig. 10e, f) nearly 4 times as long as wide, 1.3 times as long as anal segment; ventral surface with several rows of minute denticles, 2 small tubercles near distal margin, and 1 small spinule on distal margin. Armature: 2 setae (lateral, dorsolateral) near middle of ramus, 1 simple stout spine at distolateral corner, and 2 long setae (longer inner, shorter outer) terminally.

Etymology.—The species is named for the Bamfield area where the new *Botryllophilus* species, the first to be described from the west coast of Canada, was found.

DISCUSSION

In the female morphotype A of the genus *Botryllophilus*, legs 1–4 have been given the following diagnosis: right and left exopods 1-segmented; endopods of right legs 2–4 including 2 or 3 spinelike elements besides setae; exopod of right leg 4 bearing 4 spines (Ooishi and IIIg, 1988). The diagnosis was based on characters for legs 1–4 of six unnamed species from the North Pacific Ocean. Characters of legs of known species from European seas were not used, because the right and left legs 1–4 of these copepods had not been adequately described in the original papers. Characters of legs of *B. bamfieldensis* fit the diagnostic features mentioned above.

Botryllophilus norvegicus is the only European species of type A in which legs 1–4 have recently been redescribed (Ooishi,

1996). The formula for leg armature of this species distinctly differs from that for *B. bam-fieldensis*. Thus, the characters for legs of *B. norvegicus* deviate from the diagnosis.

These facts mean that there are two patterns for leg armature in species of morphotype A: pattern I is represented by *B. bamfieldensis* and pattern II is represented by *B. norvegicus*. Although details for legs 1–4 are still lacking for the remaining five European species, it is possible to compare the armature of some of the legs in different species. As a result of a comparative study, it is clear that their legs also have the same armature patterns.

These two patterns suggest that all members of type A (1 new, 8 already described) can be divided into two subgroups: 1 and 2. In the comparative study of the armature of legs in these subgroups, only the total number (T) of setae (se) and spines (sp) on the endopods (endo) and exopods (exo) is used; setiform elements and spiniform elements are included in counts of setae and spines, respectively.

Subgroup 1 of Type A Species.—The formula for leg armature (pattern I) represented by *B*. *bamfieldensis* for subgroup 1 is as follows:

	(Rig	(Right)		(Left)	
	Endo	Exo(sp)	Endo(se)	Exo(se)	
P ₁	8 (se)	6	8	5	
P,	7 (se & sp)	5	7	4	
P ₃	5 (se & sp)	5	5	4	
P_4	5 (se & sp)	4	5	3	

The formulas for legs 1–4 of *B. abbotti* (based on Ooishi and Illg, 1989) and *B. koreensis* (based on Seo and Lee, 1995) correspond to this pattern. In *B. abbotti*, the endopod of left leg 4 was shown to have an additional seta (thus the formula was T = 6, instead of T = 5). The number of elements on endopods is usually the same in each leg; thus, the additional element on only the left side is considered to be simply a variation in the number of elements.

It should be noted that the formula (endo T = 5; exo T = 4) for right leg 4 is identical in the three Pacific species.

Among six known European species, three have the same formula for right leg 4 (with a similar shape of the ramus): *B. bergensis* (Schellenberg, 1921: 11, fig. 9b), *B. brevipes* Sars (Sars, 1921, pl. XXXII, p4), and *B. inaequipes* (Hansen, 1923: 25, pl. III, 2e). Besides the formula for right leg 4, these three species have the same formulas for certain other legs as mentioned below.

In *B. bergensis* (Schellenberg, 1921: 11, fig. 9a), the exopod of right leg 1 was shown to have only five spines (T = 5). It is thought, however, that one spine was inadvertently omitted from Schellenberg's figure. The missing spine apparently corresponds to the spine inserted on the posterior side of the ramus in *B. bamfieldensis* (Fig. 3a, VI); thus, the exopod of right leg 1 probably has six spines (T = 6) as in pattern I. The formula (T = 5, 4, 4) for the exopods to that in pattern I.

The same formula in pattern I is seen in two other species. In *B. brevipes* Sars (Sars, 1921, pl. XXXII, p^1 , p^2), the formula for left leg 1 is T = 8 (endopod) and T = 5 (exopod), and for left leg 2 it is T = 7 (endopod) and T = 4 (exopod); in *B. inaequipes* (Hansen, 1923: 25, pl. III, 2d, 2f), the formula for left leg 1 is T = 5 (exopod), and for left leg 4 it is T = 5 (endopod) and T = 3 (exopod).

In *B. bergensis* (Schellenberg, 1921: 11), however, the formulas given for the endopods of legs 1 and 2 (T = 9 in both) and for the exopod of left leg 4 (T = 4) differ from those in pattern I. In *B. inaequipes* (Hansen, 1923: 25), the endopod of leg 1 was also shown to have nine setae. It is difficult to say, at present, that these differences are characteristic of these two species.

The terminology for elements of armature on the exopods of left legs differs; they have been called setae, setiform elements, spines, and spiniform projections. In *B. bamfieldensis*, the short proximalmost element shown by SEM is setiform (Fig. 6d), not spiniform, and is identical in morphology to the remaining short or long elements on the same margin of the ramus. I think that other species also have similar elements on the left exopods.

For these reasons, it is considered that the six species (3 European, 3 Pacific) can be assembled to form a subgroup of species of type A. Redescriptions of the three European species are left for future studies, however.

Dudley and Illg (1991) recorded, as *B*. sp., an unnamed *Botryllophilus* species from the Woods Hole region. This unnamed species is included in this subgroup, because the right legs 1 (fig. 26) and 4 (fig. 39) have the same formula for armature as in pattern I. Kim (1998) recently reported a Korean species as *B. abbotti*. The Korean specimen, however, differs from *B. abbotti*. The specimen from Korea (fig. 390E) does not have the folded gnathobasic margin of the mandible that is species-specific for *B. abbotti* from the west coast of U.S.A. (Ooishi and Illg, 1989, fig. 2i–k). In any case, the Korean specimen is also included in this subgroup, because it has the same armature formula for legs 1-4 (except for the endopod of left leg 3).

In similar species of this subgroup that have the same formula for leg armature and the same arrangement (or composition) of the elements, further detailed morphological studies are needed for the rami (exopods, endopods) and/or for armature or ornamentation (setae, spines, spinules, etc.). *Botryllophilus bamfieldensis* will then be distinguished from other species by the morphology of its legs.

Subgroup 2 of Type A Species.—The formula for leg armature (pattern II) represented by *B. norvegicus* (according to Ooishi, 1996) for subgroup 2 is as follows:

	(Right)		(Left)	
	endo(se)	exo(sp)	endo(se)	exo(se)
P_1	8	7* (6+1)	8	7* (6+1)
P_2	8	6* (5+1)	8	6* (5+1)
P ₃	6	6* (5+1)	6	6* (5+1)
P_4	6	6* (5+1)	6	6* (5+1)

In this formula, 6* means that one considerably smaller element is included in it. Schellenberg (1921) used 5+1, instead of 6*, for legs 3 and 4 that have a small element on the exopod of each leg. In my reexamination, such a small element has been confirmed even for legs 1 and 2. Therefore, the formulas (6 and 5, respectively) that he presented for legs 1 and 2 are here interpreted to be 6+1 and 5+1.

When compared with the formula of pattern I, the number (8) of setae of endopods of leg 1 is the same as that of pattern I, but all others have 1–3 additional elements. For instance, the armature formula for right leg 4 (endopod with 6 setae; exopod with 6* spines) shows that these rami, respectively, have 1 and 2 additional elements.

The remaining two European species, *B. brevipes* Brément (= *B. brev.*) and *B. banyulensis* (= *B. bany.*), also have an additional element, at least on certain legs. The following formulas for both species are based on Brément (1909: 71–76, figs. VII–XI):

	(Rig	ht)	(Left)	
	endo(se)	exo(sp)	endo(se)	exo(se)
B. brev. P_1	8	7	8	6
P_4	6	5	6	4
B. bany. P_1	8	6	8	5
P ₃	6	6		
P_4	6	5	6	4

In both species, the total number of elements (T = 8) on the endopods of leg 1 on both sides is the same as that of *B. norvegicus* as well as that of pattern I. In addition, in *B. banyulensis*, the formula for the exopods of right leg 1 (T = 6) and left leg 1 (T = 5) is comparable to that in pattern I. However, all other rami or legs of these two species have an additional element.

Concerning right leg 4 in these two species, the formula for the endopod (T = 6) and exopod (T = 5) resembles that for *B. norvegicus* (endo T = 6, exo T = 6* or 5+1), which has additional elements in both rami. As seen in the formula for *B. brevipes*, the exopods of right and left leg 1 and both rami of left leg 4 have an additional element. In the formula for *B. banyulensis*, there is an additional element in both rami of legs 3 (right) and 4 (left). Therefore, I think that the formulas for leg armature of these two species conform, in general, to pattern II, and that all three species form subgroup 2 of type A.

In subgroup 2, the three species are distinguished from each other with respect to their exopods. The number of armature elements on the exopods of right and left legs 1-4 of *B. norvegicus* is the same, whereas it is not the same in the other two species. The number of armature elements of the exopods of right and left leg 1 of *B. brevipes* Brément is slightly different from that of *B. banyulensis*, as discussed above.

In species of morphotype B, right leg 4 was characterized by an endopod with six setae and an exopod with six or five spines (Ooishi and Illg, 1988). This formula resembles that for the right leg 4 (endo T = 6; exo T = 5) of *B. brevipes* Brément and *B. banyulensis*.

In fact, *B. brevipes* Brément (in spite of having a 5-segmented urosome) had been incorrectly assigned to type B by Ooishi and Illg (1988) because of the similar formula for the right leg 4. My correction of this was given as a personal communication in the paper by Conradi *et al.* (1994) and later emended by me (Ooishi, 1996) when I placed this species in type A. It was also noted (Ooishi and Illg, 1988) that characters for some structures of certain species belong to or deviate from those of both types A and B, and such aberrant characters may possibly be diagnostic features for the species or for other taxa; *B. banyulensis* was treated as such a species.

An erroneous or confused treatment for *B. brevipes* Brément and *B. banyulensis* has been corrected, for the present, by confining them, together with *B. norvegicus*, to subgroup 2. Redescriptions of the former two species and descriptions of new species of this subgroup are needed in order to explain diagnostic features of subgroup 2 with respect not only to legs but to all other appendages.

Characteristics of the Female.—Botryllophilus bamfieldensis is easily distinguished from its five congeners of subgroup 1 by the elongated exopods (more than 3 times as long as wide and much longer than endopods) of left legs 1–4. In the five other species, these left exopods are not remarkably elongated. For example, the ratio of length to width of the exopod of left leg 1 of *B. abbotti* (Ooishi and Illg, 1989: fig. p) is 2.7:1.

The following features are also distinctive: the cephalosome has a sharply pointed apex with characteristically arranged sensilla (it is not sharply pointed in the five other species); the metasome is weakly five-segmented (it is distinctly segmented in *B. inaequipes*, and there is no indication of segmentation in the four other species).

The arrangement of setae on the first (with 8 setae) and second (with 5 setae) segments of the antennule appears to be characteristic, differing from that of B. abbotti (first segment with 9 setae; second segment with 4 setae). The setal number on the third segment (with 3 setae) and fourth segment (with 12 setae) is the same in both species; thus, the total number (28) of setae on the antennule is also the same. It is apparent that an additional long independent seta (Figs. 2a [indicated by black dot], 6b) on the second segment of B. bam*fieldensis* is homologous to the similarly long seta protruding from the distal corner of the first segment (close to the second segment) of B. abbotti (Ooishi and Illg, 1989: fig. 2e). This seta is short and placed in the same position in some other species (manuscript in

preparation). It seems that the position or length of this seta varies according to species.

Characteristics of the Male.—The male retains characters of the female of type A in having four postgenital segments (3 abdominal, 1 anal) and an antenna with seven armature elements. However, the antennae of the male are symmetrical and different from the asymmetrical right and left antennae of the female with respect to the arrangement of setae and spines. The antenna of the male is comparable to that of the male of *B. abbotti* in appearance and arrangement of elements (2 spines, 5 setae).

The endopod of leg 1 of *B. bamfieldensis* (with 5 plumose setae, 5 spines [3 rodlike, 2 forked]) is characteristic, being different from that of *B. abbotti* (4 setae [1 plumose, 3 simple], 4 spines [2 simple, 2 forked]). Two forked spines are commonly present in these two species of type A; in the male of *B. ruber* (type B), the endopod of leg 1 bears only stout curved spines (Ooishi, 1999).

In the study of the male of *B. ruber*, it was pointed out that except for the endopod of leg 1, all other legs are exactly the same, with respect to the armature formulas and arrangement (spines and setae), as those of the male of *B. abbotti*. The same features are found in the male of *B. bamfieldensis*.

ACKNOWLEDGEMENTS

I am greatly indebted to John McInerney (1991) and Andrew Spencer (1993) for providing facilities and equipment at Bamfield Marine Station. Louis Druehl helped in arranging my visit to the Station. Dawn Renfrew and Dave Hutchinson helped in collecting specimens of the ascidian host by diving. Fu-Shiang Chia (University of Alberta) made it possible for me to work at the Station. Gretchen Lambert (California State University, Fullerton) identified the ascidian host. I acknowledge Vivian Gotto (Queen's University of Belfast), Patricia L. Dudley (Barnard College, Columbia University), and Geoff Boxshall (Natural History Museum, London) for their critical comments on the manuscript; Norimitsu Watabe (University of South Carolina, Columbia) for help in use of SEM; and Il-Hoi Kim (Kangreung National University) for Botryllophilus specimens from Korea and also information concerning these. I thank Dennis Willows

(Friday Harbor Laboratories) for use of facilities and equipment. Eugene N. Kozloff and Craig Staude gave much help in preparation of the manuscript.

LITERATURE CITED

- Brément, E. 1909. Contribution à l'étude des copépodes ascidicoles du Golfe du Lion.—Archives de Zoologie Expérimentale et Générale 1 (Notes et Revue 5): 61–89.
- Conradi, M., P. J. López-González, and J. C. García-Gómez. 1994. *Botryllophilus conicus* n. sp. (Copepoda: Cyclopoida: Ascidicolidae) associated with a compound ascidian from the Strait of Gibraltar.—Systematic Parasitology 29: 97–104.
- Dudley, P. L., and P. L. Illg. 1991. Marine flora and fauna of the eastern United States. Copepoda, Cyclopoida: Archinotodelphyidae, Notodelphyidae, and Ascidicolidae.—NOAA Technical Report NMFS 96: 1–39.
- Hansen, H. J. 1923. Crustacea Copepoda. II. Copepoda parasita and hemiparasita.—Danish Ingolf-Expedition 3(7): i–ii, 1–92.
- Illg, P. L., and P. L. Dudley. 1980. The family Ascidicolidae and its subfamilies (Copepoda, Cyclopoida), with descriptions of new species.—Mémoires du Muséum National d'Histoire Naturelle 117: 1–192.
- Kim, I.-H. 1998. Cirripedia, symbiotic Copepoda, Pycnogonida.—Illustrated Encyclopedia of Fauna & Flora of Korea 38: 1–1038. [In Korean.]
- Ooishi, S. 1996. Two ascidicolid copepods, *Botryllophilus macropus* Canu and *B. norvegicus* Schellenberg, from British waters.—Journal of Crustacean Biology 16: 169–191.
- ———. 1999. Female and male *Botryllophilus ruber* (Copepoda: Cyclopoida) associated with the compound ascidian *Botryllus schlosseri*.—Journal of Crustacean Biology 19: 556–577.
- —, and P. L. Illg. 1988. Two morpho-types of *Botryllophilus* (Cyclopoida, Ascidicolidae).—Hydrobiologia 167/168: 561–566.
- —, and —, 1989. Botryllophilus abbotti new species (Copepoda: Cyclopoida: Ascidicolidae) associated with a compound ascidian from the Monterey Peninsula.—Bulletin of Marine Science 45: 447–466.
- Sars, G. O. 1921. An account of the Crustacea of Norway with short descriptions and figures of all the species. Vol. 8. Copepoda, Monstrilloida & Notodelphyoida. Pp. 1–91. Bergen Museum, Bergen, Norway.
- Schellenberg, A. 1921. Neue Norwegische Notodelphyiden.—Kongelige Norske Videnskabers Selskabs Skrifter 1921, 3: 1–11.
- Seo, I.-S., and K.-S. Lee. 1995. Copepod associated with ascidians from Korea. II. A new species of *Botryllophilus* (Copepoda, Cyclopoida, Ascidicolidae) from simple ascidians at Chindo Island in the South Sea, Korea.—Korean Journal of Systematic Zoology 2: 79–86.

RECEIVED: 22 October 1999. ACCEPTED: 2 March 2000.