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***Botryllophilus conicus* n. sp. (Copepoda: Cyclopoida: Ascidicolidae) associated with a compound ascidian from the Strait of Gibraltar**

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Abstract

Within morphotype B of the genus *Botryllophilus* Hesse, 1864, there are four recognisable species. In this paper, *B. conicus* n. sp. is described and illustrated from material associated with *Aplidium conicum* (Olivi, 1792). The new species is compared with congeneric species of this morphotype.

Introduction

Illg & Dudley (1980) listed 25 species for *Botryllophilus* Hesse, 1864, but only ten of them are characterised in detail. Some of these species, such as *B. ruber* Hesse, 1864 and *B. banyulensis* Brément, 1909 are still inadequately described and need detailed redescription. The 15 remaining species are indeterminable. In the past, the systematics of this genus was largely based on the morphology and structure of the legs, which are rather variable. The establishment of a new species of *Botryllophilus* must be based on the structure of the antennules, mouth-parts (chiefly mandible and maxillule) and fifth legs, which are more stable within species (Stock, 1970).

Ooishi & Illg (1988) tried to solve these taxonomic problems. They divided the ten recognisable species into two morphotypes (A and B). These have been defined by the characters of the urosome, including the anal segment and caudal rami, the rostrum, almost all the appendages, and the apparatus at the oviducal aperture. A new species of *Botryllophilus*, *B. abbotti* was described

by Ooishi & Illg (1989) and placed into the morphotype A, increasing the number of recognisable species of this genus to 11.

The majority of species included in morphotype B, with the exception of *B. randalli* Stock, 1970, are found in European waters.

In this work a new species of *Botryllophilus*, *B. conicus*, is described and compared with the species of morphotype B, where this new species belongs.

Materials and methods

Compound ascidians, *Aplidium conicum* (Olivi, 1792), were collected on stones from the infralittoral zone (6 m depth). These specimens were maintained in separate glass bottles. The copepods were taken and preserved in 4% formalin in seawater. The specimens were stained with cotton blue and dissected under a stereomicroscope. Semi-permanent mounts were made using lactophenol. Five specimens were dissected and mounted for light-microscopy. Nine specimens

were prepared for scanning electron microscopy. All figures have been drawn with the aid of a camera lucida. The letter following each figure legend refers to the scale at which it was drawn.

Family Ascidicolidae Thorell, 1859

Subfamily Botryllophilinae Sars, 1921

Genus *Botryllophilus* Hesse, 1864

***Botryllophilus conicus* n. sp. (Figs 1–4)**

Type-material: 68♀♀ from *Aplidium conicum* (Olivi, 1792) at Isla Verde, Algeciras Bay, Cádiz, Spain, 17 July 1991. Holotype and 3 paratypes deposited in the Museo Nacional de Ciencias Naturales de Madrid (Spain) (MNCNM 2004/325) (MNCNM 2004/326). Remaining paratypes in the authors' collection.

Description

Female. Body (Fig. 1a) divided into cephalosome, metasome and urosome, with their proportional lengths measured along body axis c. 1:2.4:2. Cephalosome and all pedigerous segments of metasome with indistinct articulations. Pedigerous segments wider than long. Fifth legs transformed into typical postero-dorsally directed horns (Fig. 1b). Ovisacs (Fig. 1c) $850 \times 475 \mu\text{m}$, longer than wide. Urosome apparently 8-segmented due to telescopic folding. Genital segment wider than long, 2 dorso-lateral oviducal apertures, each with cuticular flap. Outer edge of cuticular flap provided with 3 tooth-like processes (Fig. 1e). Anal segment (Fig. 1f) wider than long with some patches of small spinules on ventral surface. Caudal ramus $150 \times 100 \mu\text{m}$.

Prominent rostrum (Fig. 1a) with sclerotised margins on both sides, each with several teeth.

Antennule (Fig. 2a) 5-segmented gradually tapering and ornamented with naked setae. First segment largest, occupying nearly half of total length. Second segment wider than long; 3rd, 4th and 5th segments as long as wide. First segment with 7 large and 2 smaller setae on anterior mar-

gin. Second segment somewhat eccentrically implanted, with 3 large and one smaller smooth setae; 3rd and 4th segments with 3 setae. Last segment with 2 short subterminal and 5 terminal setae. Most of larger setae implanted on basal protuberances.

Right antenna (Fig. 2c) 3-segmented. First segment elongate, slightly curved and unarmed. Second segment small, triangular and likewise unarmed. Third segment elongate, straight, as long as first segment. Proportional lengths 3.7:1:2.8. Last segment with 2 spiniform elements on proximal half of inner margin, one short spine subterminally on inner margin and 4 stiff setae of gradually increasing length on distal margin; latter with proportional lengths from medial to lateral of c. 1:1.1:1.25:1.75. Row of small spinules extend from subterminal spine to longest distal seta.

Left antenna (Fig. 2b) similar to right in segmentation, but proportional lengths of 4 elements of 3rd segment are 1:1:1.3:1.36. All 7 elements of armature somewhat more setiform than those of right antenna and subterminal spine stronger. Subterminal element with serrate margins.

Labrum (Fig. 2g) subtriangular, with smooth margin.

Coxa of mandible (Fig. 2d) with clearly demarcated masticatory portion, dorsal to ventral with small seta, 2 short triangular teeth and one long sharp tooth. Basis of palp unarmed. Exopod indistinctly articulated, with one subterminal plumose seta and 2 terminal plumose setae. Endopod 2-segmented; 1st segment more or less rectangular and unarmed; 2nd segment with wide proximal part and narrower distal part, separated by distinct constriction. Distal part of 2nd segment with one slender seta, one long plumose seta subterminally and 4 long plumose setae terminally.

Maxillule (Fig. 2e) consisting of precoxa with setiferous gnathobase and palp apparently comprising coxa, basis and fused rami. Precoxal gnathobase bearing 5 strong setae. Epipodite represented by small rounded lateral lobe with single small, naked seta at tip. Basis with 2 long medial setae directed distally and hairs on proximal margin. Endopodite with 3 plumose setae.

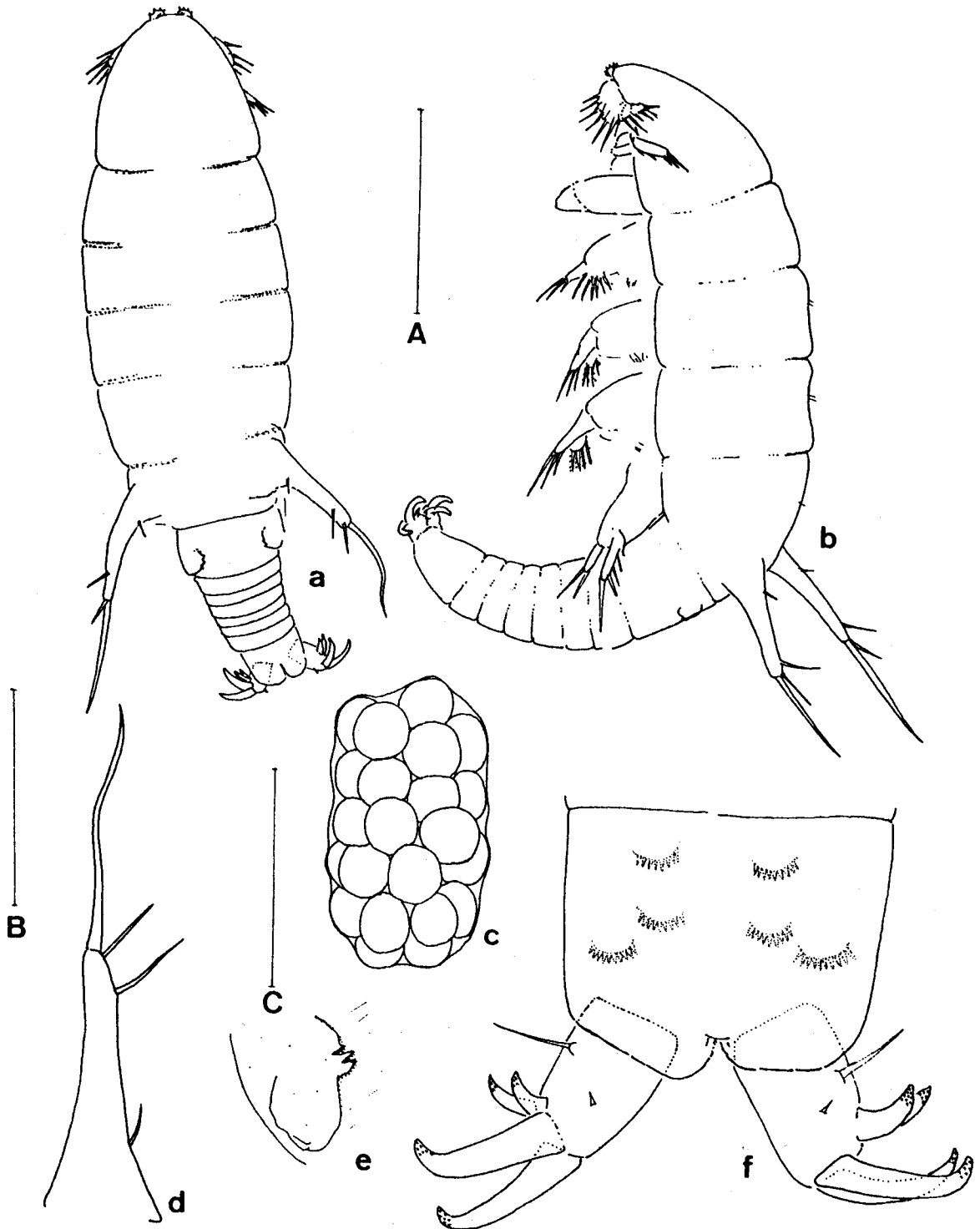


Fig. 1. *Botryllophilus conicus* n. sp., female: a, habitus, dorsal (A); b, habitus, lateral (A); c, egg sac (A); d, Leg 5 (B); e, genital aperture (C); f, Anal segment and caudal ramus, dorsal view (B). Scale-bars: A, 600 μm ; B, 200 μm ; C, 100 μm .

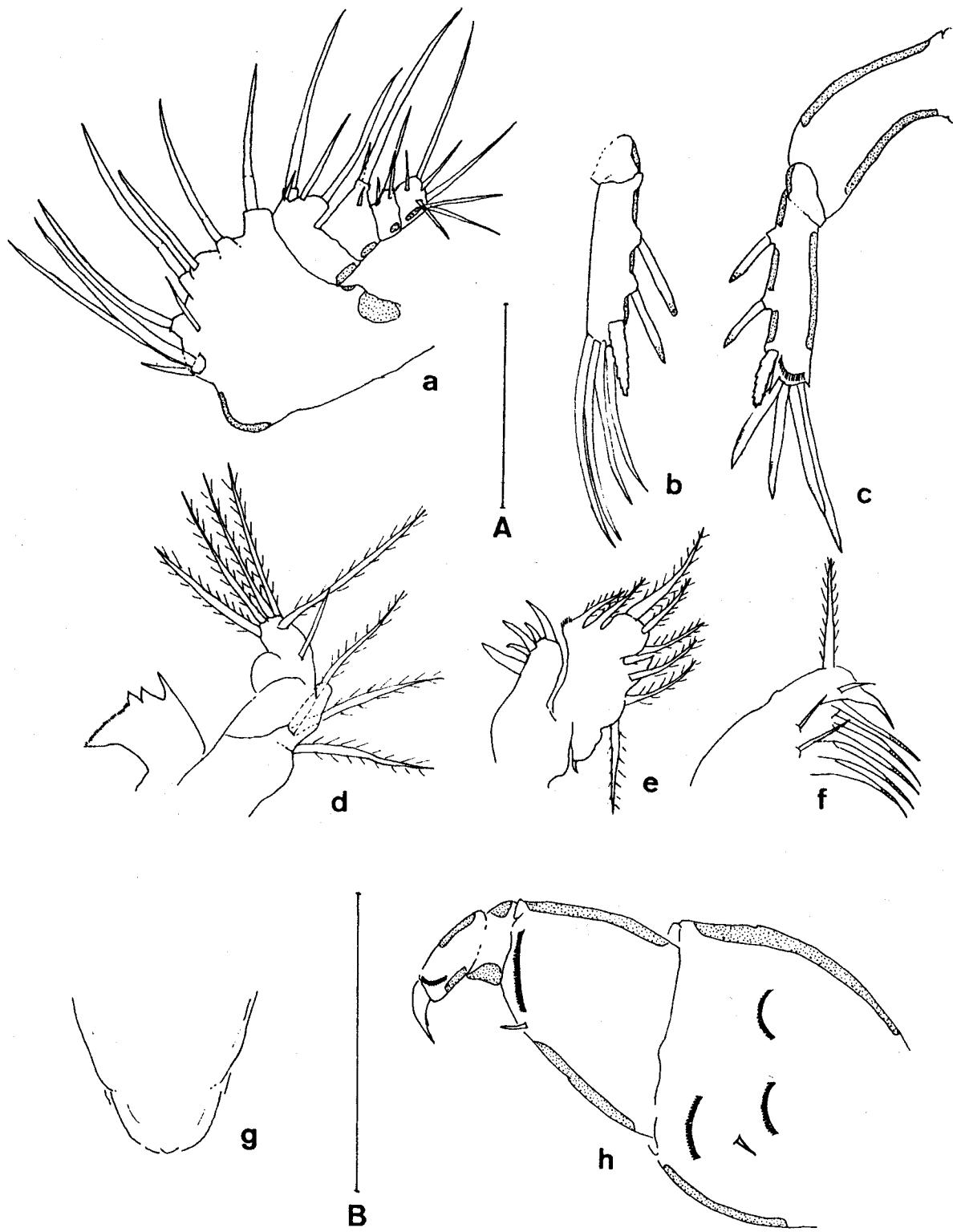


Fig. 2. *Botryllophilus conicus* n. sp.: a, antennule (A); b, antenna, left, last segment (A); c, antenna, right (A); d, mandible (A); e, maxillule (A); f, maxilla (A); g, labrum (A); h, maxilliped (B). Scale-bars: A, 100 μ m; B, 100 μ m.

Exopodite comprising small process protruding from lateral margin and 2 long plumose setae laterally and one long plumose seta proximally.

Maxilla (Fig. 2f) indistinctly segmented, with suggestion of smooth basal segment and distal portion armed with 5 long inner setae, 2 shorter setae slightly set apart near bases of 2nd and 4th inner setae, one short inner seta with one small additional seta at base, and one long plumose seta on lateral margin near apex.

Maxilliped (Fig. 2h) 3-segmented. First segment wider than long with 3 rows of spinules and one short naked seta. Second segment about as long as wide with one short naked seta and row of spinules at distal margin. Third segment with constriction suggesting 2 portions, one trapezoidal basal segment and one subrectangular segment with curved claw.

Asymmetry between right and left legs pronounced. Protopods of legs weakly bimerous. Coxa unarmed. Basis ornamented with slender seta on lateral margin. Row or patch of conical spinules on medio-distal corner of basis. First and 2nd endopodites weakly bimerous. Third and 4th endopodites bimerous. First and 2nd exopodites unimerous. Third and 4th exopodites bimerous.

Right first endopodite (Fig. 3a; Table I) approximately as long as wide, with one long plumose seta on 1st segment and 7 on 2nd. Right first exopodite wider than long with 7 strong spines; one spine on external lateral margin with patch of minute spinules at base; 3 slightly serrated spines on medial margin, each with small spinule at base; 2 curved spines on internal margin with spinule at base; and one more spine on posterior surface. Shape of left 1st endopodite (Fig. 3b) very similar to right with one long plumose seta on 1st segment and 6 on 2nd. Left first exopodite longer than wide, with 6 spiniform setae, longer than those of right endopodite, except for 3rd setae on outer margin. Patch of minute spinules at base of 1st and 2nd setae, remaining ones with one or 2 spinules.

Right and left endopodites of 2nd legs (Fig. 3c,d; Table I) with one long plumose seta on 1st segment and 7 on 2nd. Right exopodite (Fig. 3c) longer than wide with 6 spines, 5 terminal and

one on posterior surface. Second and 3rd terminal spines (on lateral margin) small; 4th wider and serrated than others. Left exopodite (Fig. 3d) of 2nd leg longer than wide, with 5 spiniform setae; 2nd setae on lateral margin much smaller than others. All setae with patch of spinules at base.

Right and left endopodites of 3rd legs (Fig. 4a,b; Table I) nearly identical, with one long plumose seta on first segment and 6 on second. Right exopodite (Fig. 4a) bimerous. First segment with spine on lateral margin. Second segment with 2 terminal short spines, 2 long curved terminal spines and one curved spine on posterior surface. All elements with patch of spinules at base. Left exopodite (Fig. 4b) bimerous, with one seta-like element on 1st segment and 4 increasing in length in 2nd (2 terminal seta-like longer than others). All elements with patch of spinules at base.

Right and left endopodites of 4th legs (Fig. 4c,d; Table I) with one plumose seta on segment and 5 on 2nd. Terminal setae thickest and longest. Right exopodite of 4th leg with one spine on 1st segment and 4 gradually increasing in length on 2nd; 2 smaller spines serrate; 3rd spine on lateral margin strong, curved and serrate on its lateral margin; and one more spine on posterior surface of segment. Left exopodite of 4th leg (Fig. 4d) similar to left exopodite of 3rd leg.

Fifth legs (Fig. 1d) elongate, symmetrical in shape. Armature consisting of one proximal small seta, one subterminal seta and 2 terminal setae, one long and other short. Long terminal seta as long as appendage.

Oviduct apertures (Fig. 1e) with 3 spines.

Caudal rami (Fig. 1f) symmetrical, with 4 terminal spines curving ventrally. Each spine slightly denticulate at tip. Lateral margin with long smooth seta and one more small dorsal seta.

Male. Unknown.

Etymology: The specific name, *conicus*, refers to the specific name of the host.

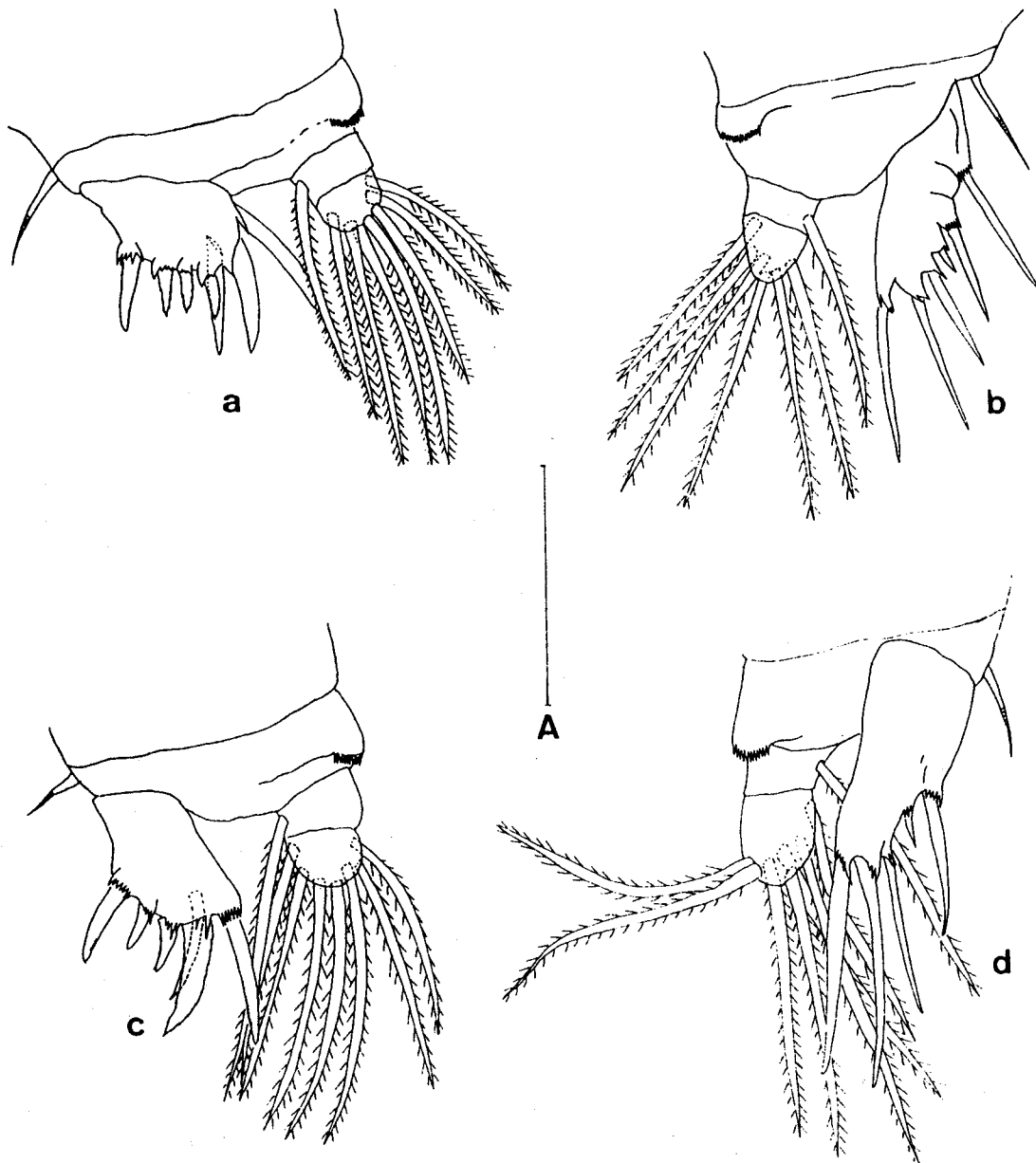


Fig. 3. *Botryllophilus conicus* n. sp.: a, leg 1, right (A); b, leg 1, left (A); c, leg 2, right (A); d, leg 2, left (A). Scale-bar: A, 100 μ m.

Discussion

Lang (1948) pointed out that the degree of asymmetry of the legs is not an adequate character for establishing a new species because of the great diversity in the structure of legs in the genus *Botryllophilus*. However, in accord with Stock

(1970) and Illg & Dudley (1980), we have never observed such a striking variation within one species. Nevertheless, we will not use these differences for distinguishing species, since the limits of intraspecific variability have not been established.

The new species is included in a group designated by Ooishi & Illg (1988) as type B for fe-

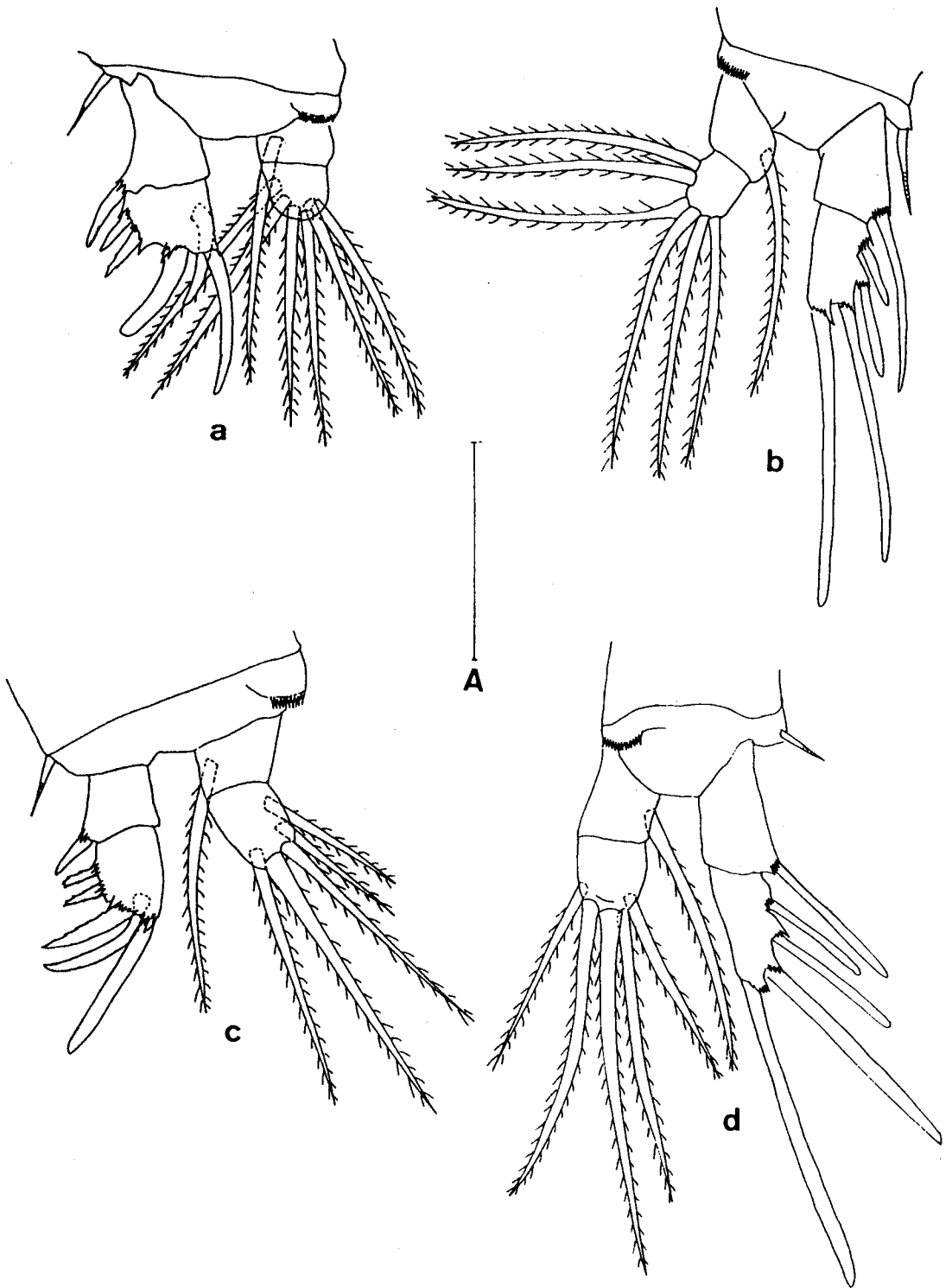


Fig. 4. *Botryllophilus conicus* n. sp.: a, leg 3, right (A); b, leg 3, left (A); c, leg 4, right (A); d, leg 4, left (A). Scale-bar: A, 100 μ m.

Table 1. *Botryllophilus conicus* n. sp. Armature of legs. (Roman numerals, spines; Arabic numerals, setae).

	Right		Left					
	Exopod		Endopod		Exopod		Endopod	
	1	2	1	2	1	2	1	2
Leg 1	VII	–	1-0	7	VI	–	1-0	6
Leg 2	VI	–	1-0	7	V	–	1-0	7
Leg 3	I-0	V	1-0	6	I-0	IV	1-0	6
Leg 4	I-0	V	1-0	5	I-0	IV	1-0	5

males of unrecorded species of the genus *Botryllophilus*. Type B includes *B. ruber* Hesse, 1864, *B. macropus* Canu, 1891, *B. brevipes* Brément, 1909, *B. aspinosus* Schellenberg, 1922 and *B. randalli* Stock, 1970 (*B. brevipes* may be included in morphotype A according to S. Ooishi, pers. comm.). *B. conicus* n. sp. is distinguishable from these species, among other features, by its long fifth legs and the shape and armature of the maxillules. Stock (1970) point out that there are only three species, *B. africanus* Schellenberg, 1922, *B. ruber* and *B. indicus* Schellenberg, 1922, with long legs like *B. randalli* and *Botryllophilus* sp. of Stock (1970). Among these species only *B. ruber* and *B. randalli* have been adequately characterised (a redescription of *B. ruber* is in preparation by Ooishi); the remaining species are indeterminate. The antennae of *B. ruber* and *B. macropus* (according to Stock, 1970) have three spines on the medial margin of the third segment (Ooishi & Illg, 1988), while *B. conicus* n. sp. has only two. *B. aspinosus* differs from the new species in the hooked and curved leg 5 and *B. randalli* differs in the shape and armature of the maxillules among others characters.

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