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New Species of *Metacaligus* (Caligidae, Copepoda) Parasitic on the Cutlassfish (*Trichiurus lepturus*) of Taiwan, with a Cladistic Analysis of the Family Caligidae

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ABSTRACT—*Metacaligus latus* n. sp. (Copepoda, Caligidae) is described from specimens found parasitic in the oral and gill cavities of the cutlassfish, *Trichiurus lepturus* Linnaeus, caught from the Strait of Taiwan and landed at Dong-Shih Fishing Port in Chiayi County, Taiwan. It is close to *M. uruguayensis* (Thomsen, 1949), but can be distinguished from the latter by the possession of shorter caudal rami in both sexes and wider cephalothorax and genital complex in the male. Cladistic (phylogenetic) analysis of the Family Caligidae, based on 23 selected morphological characters, revealed that *Metacaligus* Thomsen, 1949 is distantly related to *Caligus* Müller, 1758. It is a valid genus and occurs in sister-group relationship with *Apogonia* Cressey and Cressey, 1990.

Key words: parasitic Copepoda, *Metacaligus latus*, cutlassfish, caligid cladistics

INTRODUCTION

Two species of *Metacaligus* Thomsen, 1949 are known parasitic on the marine fishes of East Asia: *M. hilsae* (Shen, 1957) from China and *M. uruguayensis* Thomsen, 1949 from Taiwan (Lin and Ho, 2000). While the former is confined to China (Shen, 1957) the latter has been reported from Uruguay (Thomsen, 1949), India (Pillai, 1963), Sri Lanka (Kirtisinghe, 1964), Venezuela (Ho and Bashirullah, 1977) and Brazil (Boxshall and Montú, 1997). In this paper we report a new species of this genus recovered from the cutlassfish, *Trichiurus lepturus* Linnaeus, caught in the Strait of Taiwan.

In his work on the copepod parasites of marine fishes of Uruguay, Thomsen (1949) proposed the subgenus *Metacaligus* to accommodate a new species of *Caligus* found parasitic on the cutlassfish from Uruguay. It was called "*Caligus* (*Metacaligus*) *uruguayensis* n. sp." Furthermore, he included in this subgenus three other species of *Caligus*, namely, *C. afurcatus* Wilson, 1913; *C. enormis* Wilson, 1913; and *C. rufus* Wilson, 1908. Twenty-eight years later, on the occasion of the discovery of *C. (Metacaligus) uruguayensis* from the same species of host caught off Vene-

zuela, Ho and Bashirullah (1977) reevaluated Thomsen's (1949) concept of *Metacaligus* and elevated it to the generic rank. Moreover, after examining the type-material of the three species of *Caligus* included in the *Metacaligus* by Thomsen (1949), Ho and Bashirullah (1977) excluded *C. afurcatus* and *C. enormis*, and included *Caligus hilsae* Shen, 1957, in the genus *Metacaligus*.

The rationale for Ho and Bashirullah (1977) to elevate *Metacaligus* to the generic rank was the possession in common of a combination of three character states by *M. uruguayensis*, *M. rufus*, and *M. hilsae*. Those three common morphological properties are (1) absence of sternal furca, (2) absence of accessory processes on two middle terminal spines of second exopodal segment of leg 1, and (3) possession of only one outer spine (instead of two) on the terminal exopodal segment of leg 2. Since none of the three states of character are unique to *Metacaligus* and each of them is respectively found in some species of *Caligus*, Dojiri (1983) was skeptical about the recognition of *Metacaligus* as a genus distinct from *Caligus*. Therefore, in this report, in addition to providing the description of a new species of *Metacaligus*, a cladistic analysis based on the morphological characters of the Family Caligidae is attempted. It is believed this analysis shall provide means to explore phylogenetic relationships within the family and to examine if the establishment of *Metacaligus* as a genus separate from *Caligus* is justifiable.

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MATERIALS AND METHODS

The cutlassfish (*Trichiurus lepturus* Linnaeus) caught in the Strait of Taiwan and landed at Dong-Shih Fishing Port in Chiayi County was purchased and transferred in an icebox to National Chiayi University where the laboratory examination of the copepod parasites was carried out. The copepod parasites removed from the fish were preserved in 70% ethanol. In studying the parasites, the specimens were cleared in 85% lactic acid for 1 to 2 hours before making dissection in a drop of lactic acid. The hanging-drop method devised by Humes and Gooding (1964) was employed to examine the dissected parts and appendages under the compound microscope. All drawings were made with the aid of a camera lucida. Measurements of the body and body parts were taken from 10 randomly selected specimens. The measurements given in the description are the average with its range shown in the parentheses that follow.

In this paper only qualitative characters are used in the cladistic analysis. Also, autapomorphic characters are excluded because they do not aid in a search for the wanted phylogenetic relationships, except for help in boosting consistency index of the generated cladogram(s). The type-species of each valid caligid genus is selected to extract the wanted homologous characters for the OTU. Based on Benz's (1993) cladogram for the siphonostomatoids parasitic on the vertebrates, *Euryphorus* Milne Edwards, 1840 and *Trebisius* Krøyer, 1838 were utilized as the outgroup to polarize the transformation of the identified homologous character states of the Caligidae. Moreover, in coding multistate characters, when a transformation series containing a single basal bifurcation (dichotomous transformation) was encountered, we followed the method of "internal rooting" proposed by O'Grady and Deets (1987). In this case, the state of character that received a code of "0" is not plesiomorphic.

The computer program HENNIG86 Version 1.5 (Farris, 1988) was used to analyze the phylogenetic relationships between the genera of the Caligidae. The commands "mhennig*" and "bb*" were employed to produce multiple, shortest trees through performance of extended branch swapping.

DESCRIPTION

Metacaligus latus sp. nov.
(Figs. 1–4)

Material examined: 261 , 105 , and 20 chalimus larvae attached to adults of both sexes were recovered from oral and gill cavities of cutlassfish, *Trichiurus lepturus* Linnaeus, caught in Strait of Taiwan and landed at Dong-Shih Fishing Port on following dates in 2001: 1 and 1 from 1 fish on 31 March; 59 , 19 and 3 larvae from 27 fishes on 27 April; and 201 , 85 , and 17 larvae from 62 fishes on 22 June. One holotype female, 1 allotype male, and 60 paratypes (30 and 30) were selected from the last collection and deposited in the National Museum of Natural History, Smithsonian Institution in Washington, D.C.

Female: Body (Fig. 1A) 3.29 (2.42–3.72) mm long, excluding setae on caudal rami. Cephalothoracic shield subcircular, 1.13 (0.82–1.30) mm long and 1.01 (0.78–1.32) mm wide, excluding marginal hyaline membranes. Fourth pediger 2.21 times wider than long. Genital complex globular, 1.13 (0.86–1.40) mm long and 1.11 (0.82–1.42) mm wide.

Abdomen stocky and 1-segmented, 0.56 (0.40–0.82) mm long and 0.23 (0.18–0.38) mm wide. Caudal ramus (Fig. 3C) 1.63 (0.13×0.08 mm) times longer than wide; with 3 short, subterminal setae and 3 long, terminal setae in addition to a setule-bearing papilla on dorsal surface. Egg sac longer than body, 3.9 mm and containing 35 eggs.

Antennule (Fig. 1B) 2-segmented; proximal segment with 27 setae on anterodistal surface, distal segment with 1 subterminal seta on posterior margin and 11 setae plus 2 aesthetascs on distal margin. Antenna (Fig. 1C) 3-segmented; proximal segment smallest and unarmed; second segment rectangular and also unarmed; distal segment stocky claw bearing 1 seta in proximal region and another one close to middle region. Postantennal process (Fig. 1C) small and bluntly pointed, carrying 2 basal papillae with each bearing 4 setules; another similar papilla located nearby on sternum. Mandible (Fig. 1D) 4-segmented; with 12 teeth on medial margin of distal blade. Maxillule (Fig. 1C) comprising short, stout, dentiform process and prominent basal papilla with 3 naked setae. Maxilla (Fig. 1E) 2-segmented; proximal segment (lacertus) large and unarmed; slender, distal segment (brachium) carrying a large, subterminal, hyaline membrane on outer edge; both subterminal canna and terminal calamus fringed with fine spinules. Maxilliped (Fig. 2A) 3-segmented; proximal segment (corpus) largest but unarmed; middle (shaft) and distal (claw) segments fused to form subchelum, with small, blunt knob in basal region. Sternal furca absent.

Armature on rami of legs 1–4 as follows (Roman numeral indicating spines and Arabic numeral, setae):

	Exopod	Endopod
Leg 1	1-0; III,1,3	(vestigial)
Leg 2	I-1; I-1; I,1,5	0-1; 0-2; 6
Leg 3	I-0; I-1; III,4	0-1; 6
Leg 4	I-0; IV	(absent)

Leg 1 (Fig. 2B) protopod with long, plumose outer seta and another small, plumose inner seta, in addition to a papilla bearing 2 long setules near outer margin of coxa; vestigial endopod small conical protrusion; first segment of exopod with row of spinules on posterior edge and short spiniform seta at outer distal corner; middle 2 of 4 terminal elements on last segment of exopod without accessory process but pinnate bilaterally; large pecten at base of each distal 3 spines; 3 plumose setae on posterior margin about half length of terminal element 4. Leg 2 (Fig. 2C) coxa small, with large, plumose, inner seta on posterior edge and small papilla tipped with a short seta on ventral surface; basis with short, naked outer seta; posterior margin of basis with long, narrow membrane and long marginal setule near midway of this membrane; outer spines on exopod (Fig. 2D) differentially constructed, with largest one on proximal segment and smallest one on middle segment. Leg 3 (Fig. 3A) protopod (apron) with small, plumose, outer seta and long, plumose inner seta in addition to an outer and a posterior marginal membrane; one short and one long setule with each at tip

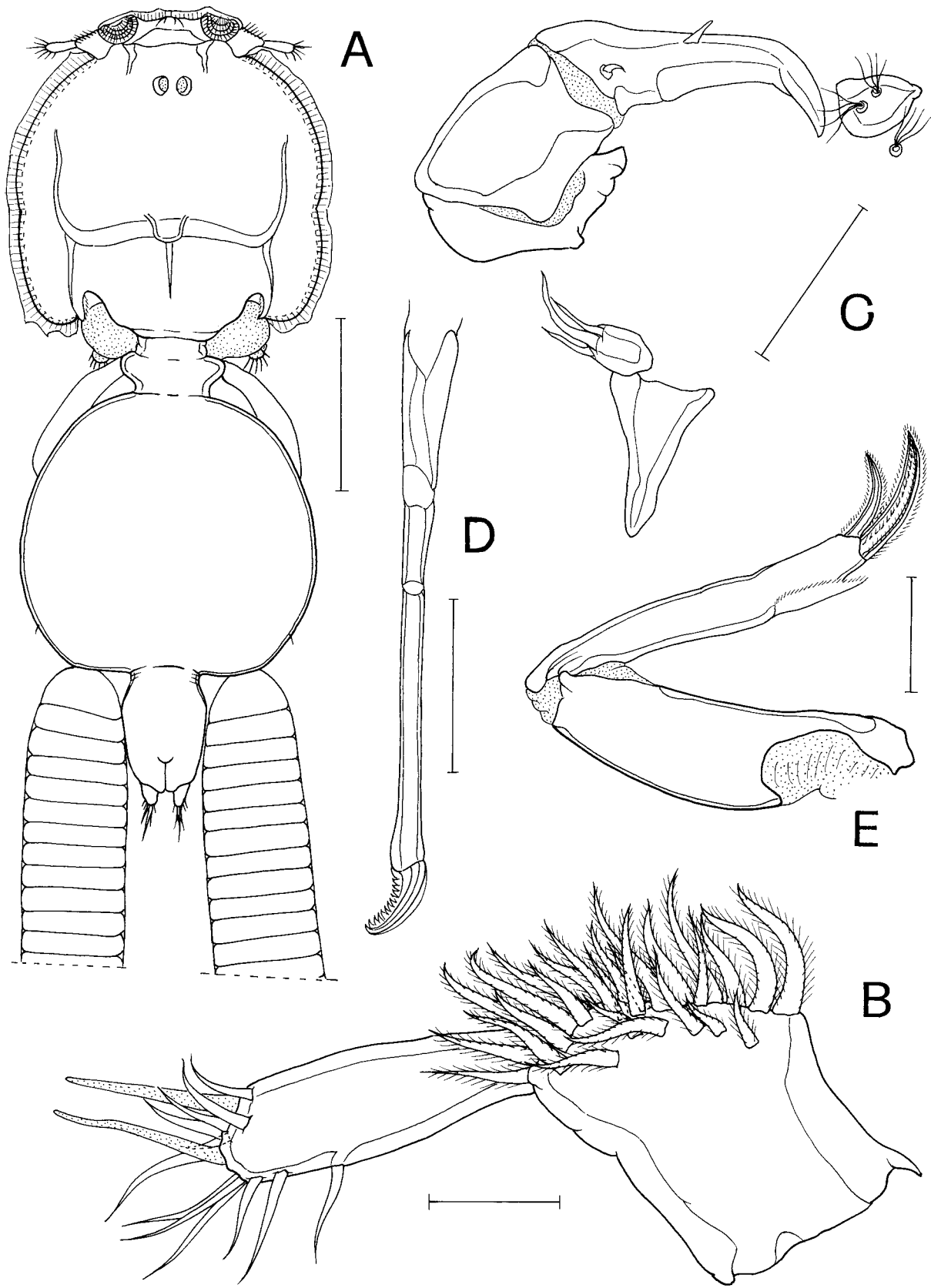


Fig. 1. *Metacaligus latus* sp. nov., female. A, habitus, dorsal. B, antennule, dorsal. C, antenna, postantennary process and maxillule, ventral. D, mandible. E, maxilla. Scale bars: 0.8 mm in A; 0.06 mm in B; 0.1 mm in C, E; 0.98 mm in D.

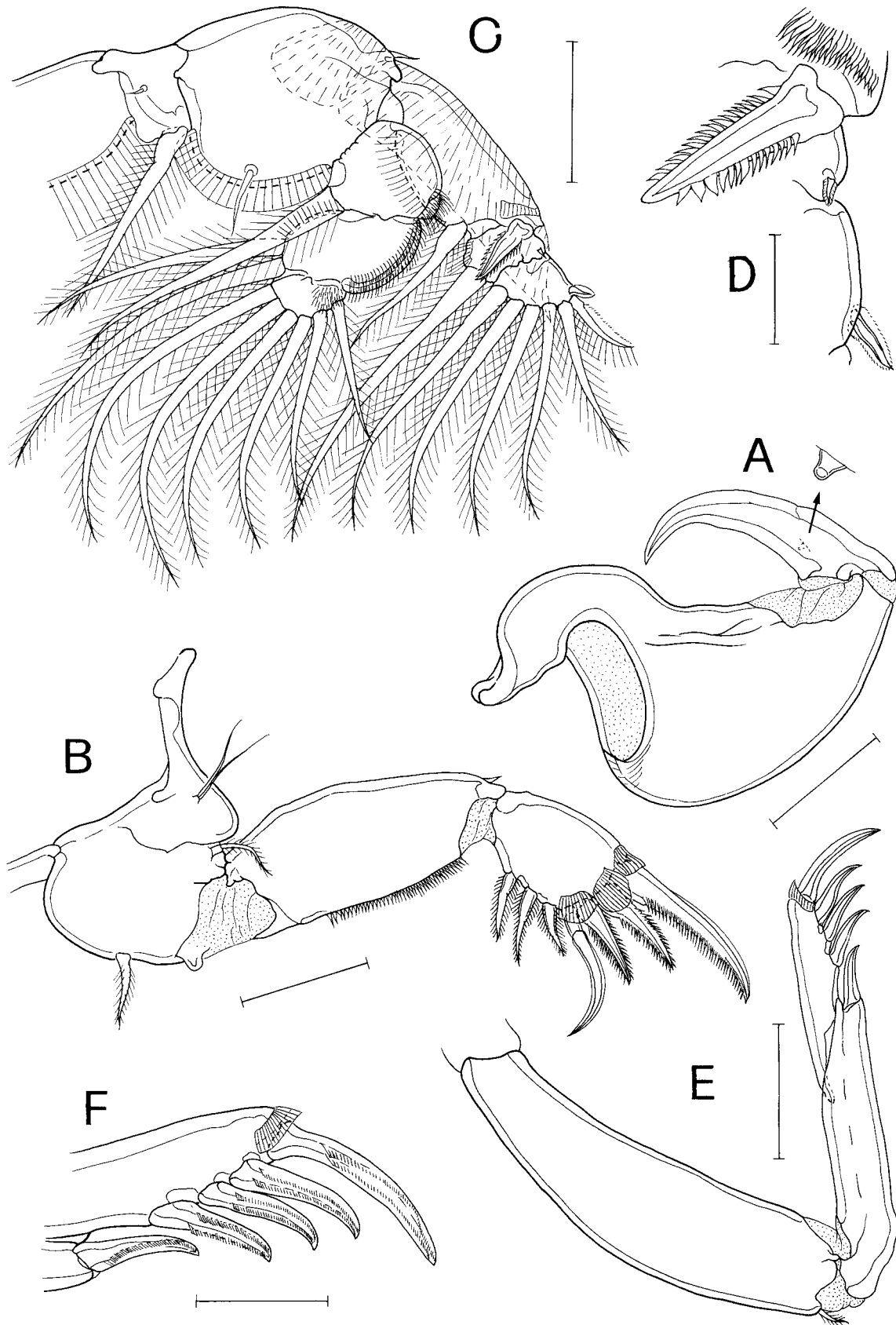


Fig. 2. *Metacaligus latus* sp. nov., female. A, maxilliped. B, leg 1. C, leg 2. D, outer spines of leg 2 exopod. E, leg 4. F, terminal part of leg 4 exopod. Scale bars: 0.2 mm in A, E; 0.1 mm in B, F; 0.15 mm in C; 0.04 mm in D.

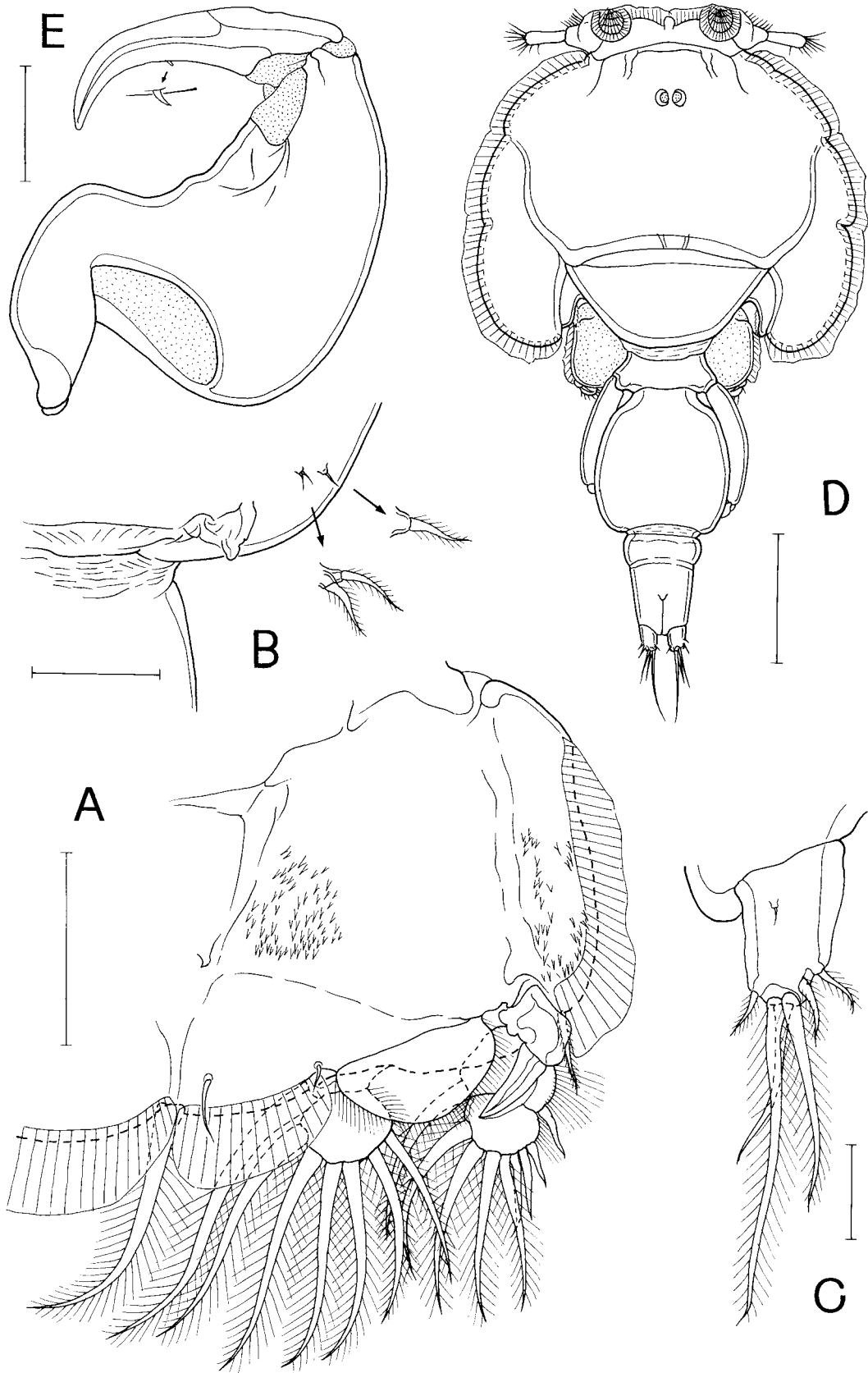


Fig. 3. *Metacaligus latus* sp. nov. Female: A, leg 3. B, posterolateral corner of genital complex, ventral. C, caudal maul, dorsal. Male: D, habitus, dorsal. E, maxilliped. Scale bars: 0.15 mm in A, E; 0.25 mm in B; 0.05 mm in C; 0.5 mm in D.

of a papilla just inner to baseline of posterior marginal membrane; 2 patches of denticles on ventral surface; velum well developed and fringed with marginal setules. Leg 4 (Fig. 2E) protopod with small, pinnate seta at outerdistal corner; pecten at base of innermost outer spine on exopod (Fig. 2F). Leg 5 (Fig. 3B) represented by 2 papillae on postero-

lateral margin of genital complex, one tipped with a pinnate seta and another one, 2 pinnate setae.

Male: Body (Fig. 3D) short and broad, 2.31 (1.72–2.70) mm long, excluding setae on caudal rami. Cehalothoracic shield subcircular, 1.27 (0.96–1.48) mm long and 1.22 (1.00–1.56) mm wide, excluding hyaline marginal membranes. Fourth

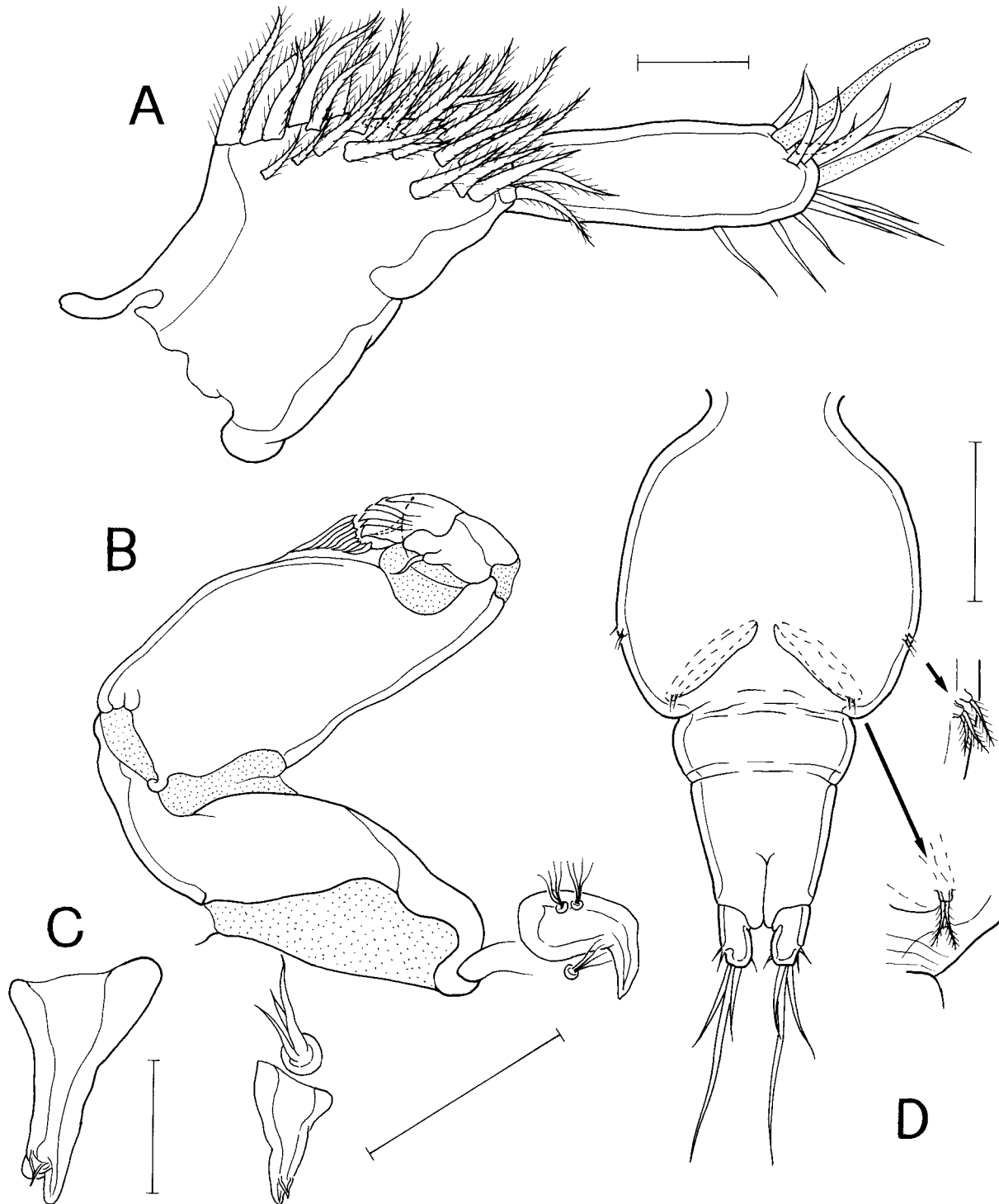


Fig. 4. *Metacaligus latus* sp. nov., male. A, antennule, dorsal. B, antenna, postantennary process and maxillule, ventral. C, dentiform process of maxillule, ventral. D, genital complex and abdomen, ventral. Scale bars: 0.05 mm in A, C; 0.15 mm in B; 0.3 mm in D.

pediger 4.44 times wider than long. Genital complex shaped as a jar, 0.51 (0.36–0.58) mm long and 0.55 (0.44–0.60) mm wide. Abdomen 2-segmented; proximal segment short, 0.09 (0.06–0.12) mm long and 0.30 (0.22–0.38) mm wide and anal segment about as long as wide, 0.26 (0.20–0.34) × 0.24 (0.18–0.28) mm. Caudal ramus 1.29 (0.09×0.07 mm) times longer than wide and armed as in female. Antennule (Fig. 4A) generally as in female, but proximal segment armed with 29 setae. Antenna (Fig. 4B) with corrugated, adhesion pad on mediolateral surface of middle segment; terminal segment with lamellate cuticular tip and 2 basal setae with one on each side. Postantennary process (Fig. 4B) stronger than in female, with prominent terminal hook. Dentiform process of maxillule (Fig. 4C) bearing subterminal tine. Maxilliped (Fig. 3E) with small medial seta in middle region of subchelum. Leg 5 as in female but leg 6 represented by a papilla bearing 2 setae located in distal region of ventral ridge on genital complex (Fig. 4D).

Etymology: The species name *latus* is a Latin for “broad” or “wide”. It alludes to the possession of broad cephalothoracic shield and genital complex in both sexes of this species.

Remarks: Three species of *Metacaligus* are currently known, they are: *M. rufus* (Wilson, 1908), *M. uruguayensis* (Thomsen, 1949) and *M. hilsae* (Shen, 1957). The last species differs from all other species of *Metacaligus*, including the present new species, in the possession of a long, 4-segmented abdomen, which is longer than its genital complex (Shen, 1957). The first species differs from the present species in having the middle segment of the exopod of leg 2 equipped with a large outer spine, which is as large as the outer spine on the basal segment of the same ramus, and the outermost spine at the tip of leg 1 exopod longer than the segment carrying it (Wilson, 1908; Ho and Bashirullah, 1977). Thus, *M. uruguayensis* bears the closest resemblance to the present new species. Nevertheless, *M. latus* is distinguishable from *M. uruguayensis* in the possession of

the following four characters: (1) broader cephalothoracic shield and genital complex in both sexes, (2) shorter caudal ramus in both sexes, (3) papillae on the postantennary process tipped with 4 setules in both sexes, and (4) the anal somite in male distinctly longer than the proximal abdominal somite.

Both *M. uruguayensis* and *M. latus* are found in the gill cavities of the cutlassfish taken from the Strait of Taiwan. However, in our past two years of examination of the cutlassfishes landed at Dong-Shih Fishing Market, these two congeners have never been found co-existing on a fish host. Another intriguing point to mention is about the occurrence of the chalimus larvae of *M. latus*. On 27 April and 22 June 2001, chalimus larvae in various stages of development (from chalimus II to chalimus IV) were recovered attached randomly to the cephalothorax, genital complex or abdomen of the adult of both sexes. This is unusual in that the chalimus larval stages are always found attaching directly to the fish host. Of the 20 chalimus larvae, 8 were found attached to the female and 12, to the male.

VALID CALIGID GENERA

In their work on the parasitic copepods of Brazilian fishes, Boxshall & Montú (1997) provided a key to 29 genera of the family Caligidae. It is the latest published work showing the recognized genera of the Caligidae. However, it is worthy to point out that while Dojiri (1983) considered *Caligopsis* and *Diphyllogaster* insufficiently known and relegated *Heniochophilus* to a synonym of *Anuretes*, these three genera were recognized valid by Boxshall & Montú (1997). Further difference between those two works is the treatment of *Midias* Wilson, 1911. This genus was not found in the key given by Boxshall & Muntú (1997) but considered valid by Dojiri (1983).

According to Dojiri's (1983) redescription of *Midias lobodes*, based on Wilson's (1911) type material kept at the

Table 1. The valid caligid genera.

<i>Abasia</i> Wilson, 1908	<i>Mappates</i> Rangnekar, 1958
<i>Anchicaligus</i> Stebbing, 1900	<i>Metacaligus</i> Thomsen, 1949
<i>Anuretes</i> Heller, 1865	<i>Midias</i> Wilson, 1911
<i>Apogonia</i> Cressey & Cressey, 1990	<i>Paralebion</i> Wilson, 1911
<i>Arrama</i> Dojiri & Cressey, 1991	Parapetalus Steenstrup & Lütken, 1861
<i>Belizia</i> Cressey, 1990	<i>Parechetus</i> Pillai, 1961
<i>Caligodes</i> Heller, 1865	<i>Pseudanuretes</i> Yamaguti, 1936
<i>Caligus</i> Müller, 1759	<i>Pseudechetus</i> Prabha & Pillai, 1979
<i>Caritus</i> Cressey, 1967	<i>Pseudocaligus</i> Scott, 1901
<i>Dartevellia</i> Brian, 1939	<i>Pseudolepeophtheirus</i> Markewitsch, 1940
<i>Echetus</i> Krøyer, 1863	<i>Pupulina</i> van Beneden, 1892
<i>Hermilius</i> Heller, 1868	<i>Sciaenophilus</i> van Beneden, 1852
<i>Kabataella</i> Prabha & Pillai, 1988	<i>Sinocaligus</i> Shen, 1957
<i>Lepeophtheirus</i> Nordmann, 1832	<i>Synestius</i> Steenstrup & Lütken, 1861

Smithsonian Institution, this parasite of the great barracuda, *Sphyræna barracuda* (Walbaum), is not attributable to *Caligus* as considered by Kabata (1979) and followed by Boxshall and Montú (1997). Thus, in following Dojiri's (1983) opinion, *Midias* is treated in this paper as a valid genus of the Caligidae.

In their preliminary review of the 19 species of *Anuretes* Ho and Lin (2000) concurred with Dojiri's (1983) treatment to relegate *Heniochophilus* to a junior synonym of *Anuretes*. Our examination of the original works of *Caligopsis* by Markewitsch (1940) and *Diphyllogaster* by Brian (1899) showed that both genera had not been fully characterized for making an evaluation of their taxonomic status. Thus, both *Caligopsis* and *Diphyllogaster* are considered *incertae sedis* and, therefore, 28 genera of caligid copepods are recognized in this paper. They are listed in Table 1.

CLADISTIC ANALYSIS

Inasmuch as all caligids are known of their females and the male is unknown in seven genera, the homologous characters utilized in the following analysis are confined to the adult females. Furthermore, in search for homologous characters in a genus comprising several species with different states of character, the ones possessed by the type-species are selected disregarding their status in the order of the character transformation. For instance, in the genus *Caligus* where there are more than 200 species with many of them bearing 3-segmented exopod on leg 4, the state of 2-segmented ramus is selected for this character even though 3-segmented state is more primitive. This is simply because the type-species of this genus, *C. curtus* Müller, 1785, has a 2-segmented exopod on its leg 4.

The characters selected for performing the analysis of phylogenetic relationships between the 28 caligid genera (Table 1) are listed in Appendix A with the code for each state indicated in the parentheses that follow. All together 36 apomorphic states were recognized and the coding of those 36 apomorphic states are given in Appendix B. It is from this list an input file was created to conduct the cladistic analysis.

Ten of the 23 selected characters exhibit multiple states of character transformation. The series of transformation

were coded in an *ordered* fashion that reflects a predetermined direction of character evolution (see Appendix A). Inasmuch as an analysis with *ordered* character coding has been criticized as running the risk of predetermining the topology of the resultant cladogram (O'Grady and Deets, 1987; Dojiri and Deets, 1988; Ho, 1991; Ho, 1994), both *ordered* and *unordered* analysis were performed to search for the best resolved cladogram(s) of the Caligidae.

Analysis with ordered coding

In employing the computer program HENNIG86 to perform the cladistic analysis, the algorithm "mhennig*" uses the available data, such as the matrix in Appendix B, to construct initial cladogram(s) and saves all shortest trees (cladograms) after running a limited branch-swapping. Five such trees were produced in this analysis. Then, with the application of "bb*" command the computer uses those five initial trees to perform extended branch-swapping to produce multiple shortest trees. As a result of this combined algorithms, a total of 668 equally parsimonious trees were generated, with a length of 96 steps, a consistency index (CI) of 40 and a retention index (RI) of 55. Close examination of these 668 trees revealed that there are seven groups of cladograms distinguished by the number of nodes that they carry.

As shown in Table 2, the number of nodes in those seven groups of trees runs from 20 to 26, with 1/3 (226/668 trees, 33.83 %) of the obtained cladograms showing 23 nodes on the tree. The highest number of nodes on a tree is 26 and only three of the 668 trees show the best resolution like this. It is called the best resolved because only one place (node) on these trees shows a polytomy where there are three branches (clades). There are ten least resolved trees with 20 nodes on the cladogram, which displays five places (nodes) with 17 clades (branches) occurring in polytomy, three nodes with three clades and two nodes with four clades (see Table 2).

In the output file of the analysis performed by employing the algorithms "mhennig*" and "bb*" with the *ordered* coding, the three best-resolved trees were designated as Trees 27, 50 and 280. They are reproduced in Fig. 5, with Tree 50 shown in its entirety. As to Trees 27 and 280 only the parts different from Tree 50 are shown. The difference between

Table 2. Number of trees in each of the seven groups of cladograms (identified with the number of nodes that they carry) obtained in the analysis with *ordered* coding.

Number of tree	Number of node	Number of branches on polytomous nodes
10 trees	20 nodes	17 branches (3 nodes each with 3 branches, 2 nodes each with 4 branches)
72 trees	21 nodes	14 branches (2 nodes each with 3 branches, 2 nodes each with 4 branches)
191 trees	22 nodes	11 branches (1 node with 3 branches, 2 nodes each with 4 branches)
226 trees	23 nodes	10 branches (2 node each with 3 branches, 1 node with 4 branches)
127 trees	24 nodes	7 branches (1 node with 3 branches, 1 node with 4 branches)
39 trees	25 nodes	4 branches (1 node with 4 branches)
3 trees	26 nodes	3 branches (1 node with 3 branches)

TREE 50

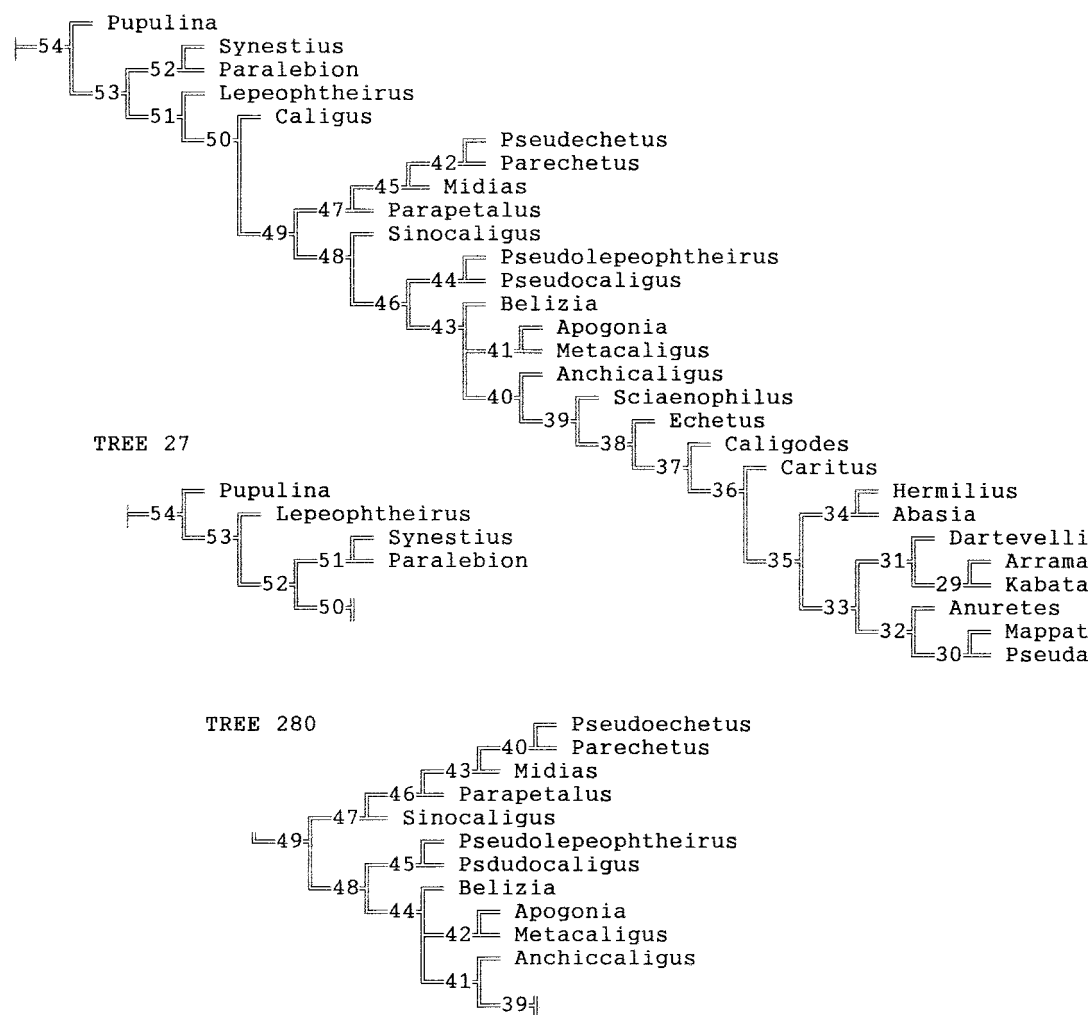


Fig. 5. Caligid phylogeny produced through analysis of *ordered* coding. Tree 50 shows entirety but for Trees 27 and 280 only the parts different from Tree 50 are shown. Note: the names of *Dartevelia* and *Kabataella* on Node 31 and *Mappates* and *Pseudanuretes* on Node 32 are truncated on Tree 50. Transformations of character states at each node on Tree 50 are: 7(0) 7(1), 12(0) 12(1) at Node 54; 3(0) 3(1), 21(0) 21(1), 22(0) 22(1) at Node 53; 9(1) 9(0) at Node 52; 7(1) 7(0) at Node 51; 1(0) 1(1) at Node 50; 10(0) 10(1) at Node 49; 23(0) 23(1) at Node 48; 10(1) 10(2) at Node 47; 10(1) 10(0) at Node 46; 12(1) 12(0) at Node 45; 23(1) 23(3) at Node 44; 13(0) 13(1) at Node 43; 6(0) 6(1), 16(0) 16(1) at Node 42; 16(0) 16(1) at Node 41; 14(0) 14(1) at Node 40; 9(1) 9(0) at Node 39; 11(0) 11(1) at Node 38; 14(1) 14(0) at Node 37; 9(0) 9(1) at Node 36; 1(1) 1(0) at Node 35; 2(0) 2(1) at Node 34; 20(0) 20(1), 23(1) 23(2) at Node 33; 5(0) 5(1), 15(0) 15(1), 21(1) 21(2) at Node 32; 22(1) 22(2) at Node 31; 4(0) 4(1), 9(1) 9(2) at Node 30; and 4(0) 4(1), 16(0) 16(1), 17(0) 17(1), 19(0) 19(1) at Node 29.

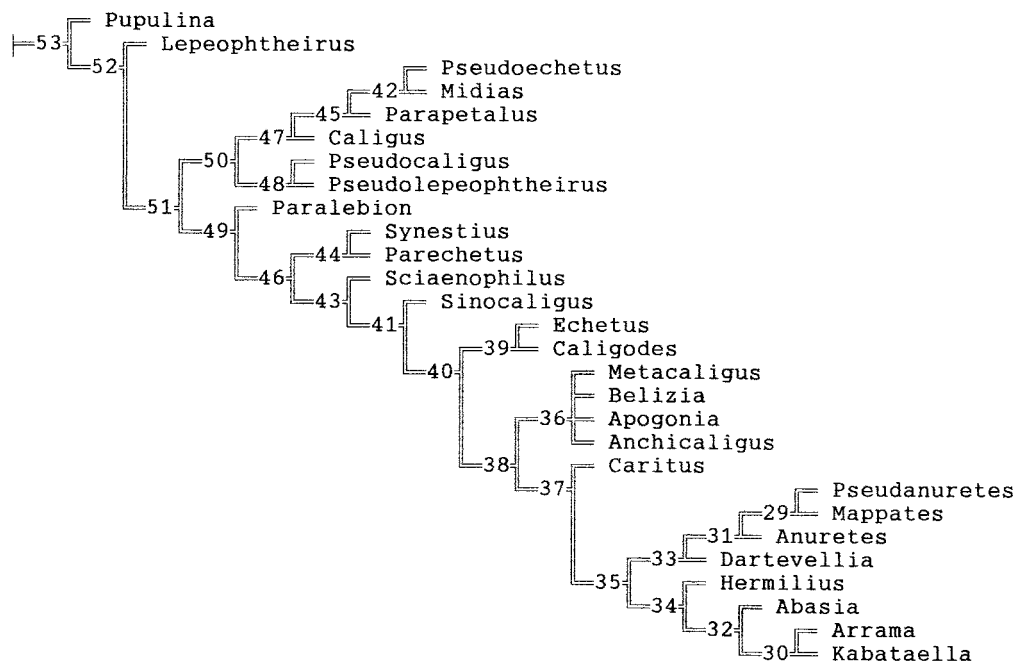
Tree 50 and Tree 27 is the displacement of *Lepeophtheirus*, moving one step (node) closer to the stem, and between Tree 50 and Tree 280, the displacement of *Sinocaligus*, jumping from Node 48 to Node 47. It should be emphasized that the topology of the 18 genera (*Pseudolepeophtheirus*, *Pseudocaligus*, *Belizia*, *Apogonia*, *Metacaligus*, *Anchicaligus*, *Sciaenophilus*, *Ecuetus*, *Caligodes*, *Caritus*, *Hermilius*, *Abasia*, *Dartevelia*, *Arrama*, *Kabataella*, *Anuretes*, *Mappates*, and *Pseudanuretes*) on Clade (Node) 46 of Tree 50 is exactly the same between these three best-resolved trees.

Analysis with *unordered* coding

Unlike the previous analysis, application of the algorithm “mhennig*” produced four initial, shortest trees (cladograms), and, with the execution of “bb*” it generated 3,165 equally parsimonious trees. The length of these trees is 87, with a consistency index (CI) of 44 and a retention index (RI) of 55. Although the tree length and CI are improved over the previous analysis with *ordered* coding, the value of RI remained unchanged and the overall resolution of the obtained trees is not as high.

Nine groups of cladograms are recognized with the number of nodes on a giving tree varying from 16 to 25. In

TREE 496



TREE 3061

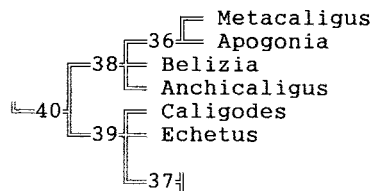


Fig. 6. Caligid phylogeny produced through analysis of *unordered* coding. Tree 496 shows entirety but for Tree 3061 only the parts different from Tree 496 are shown.

other words, the best resolved tree (with 25 nodes) is not as good as those produced in the previous analysis and the least resolved tree (with 16 nodes) is worse than those found in the previous analysis. While there are 11 best resolved trees (Trees 439, 454, 493, 496, 498, 3044, 3046, 3054, 3056, 3059 and 3061) there is but one least-resolved tree (Tree 1180). Those 11 best-resolved trees can be divided into two groups with one carrying a polytomy comprising four branches (Trees 493, 496, 3056 and 3059) and the other, two polytomies with each carrying 3 branches (Trees 439, 454, 498, 3044, 3046, 3054 and 3061). One tree from each of these two groups was reproduced in Fig. 6. Tree 496 is shown with its entirety and Tree 3061, only the parts that are different. The least resolved tree, Tree 1180, has five polytomies, with two nodes containing three branches and another three nodes, five branches.

Caligid phylogeny

It is apparent from the above two sections that analysis of caligid phylogeny with *ordered* coding generated better-resolved trees, producing three cladograms with each carrying 26 nodes. Having only one node with three branches,

any of these three trees are next best to the completely resolved tree where every node has dichotomy. Since Tree 50 occupies the central role with only one difference from each of the two other 26-node trees, it is adopted as a working hypothesis for the phylogeny of the Caligidae.

Examination of the transformation of each character on Tree 50 (see Appendix A) shows that Character 10 (outgrowth on the abdomen) changed at Node 49 from the plesiomorphy ("0", smooth and cylindrical) to an apomorphy ("1", bearing lateral aliform structures), and at Node 46 it reverted to the original plesiomorphy. It is this character reversal (homoplasy) of the abdominal outgrowth that serves as a synapomorphy to unite those 18 genera on Clade 46 to a monophyletic clade.

The absence of sternal furca (apomorphy of Character 13) is a synapomorphy at Node 43 (Table 3) and that places the afore-mentioned 18 genera in a monophyly excluding *Pseudocaligus* and *Pseudolepeophtheirus*. It is interesting to point out that the sternal furca is present in the species of both *Anuretes* and *Hermilius*, but these two genera are shown on the three trees in Fig. 5 as the descendents of an ancestor lacking the sternal furca.

Metacaligus and *Sinocaligus* are the only two genera of caligids known to have two outer spines on the terminal segment of the exopod of leg 2 (Character 18 coded with "0"), but they are not in a sister group relationship through the sharing of this unique apomorphy. In other words, according to the adopted caligid phylogeny, the evolution of the loss of one outer spine on the terminal segment of leg 2 exopod had occurred independently in two lineages, one lead to the development of *Metacaligus* and another one, the genesis of *Sinocaligus*. The same phenomenon is found on the character state of 16(2) (absence of 3 medial plumose setae on terminal segment of leg 1 exopod). This derived character is present in only three of the 28 recognized caligid genera: *Apogonia*, *Kabataella* and *Parechetus*. But, the result of analysis did not recognize it as a synapomorphy of the three genera.

CONCLUSION

From Tree 50 in Fig. 5 it is apparent that *Metacaligus* and *Apogonia* are sister groups on Clade 41, which is in turn on a monophyletic clade comprising the above-mentioned 18 genera on Clade 46. Although these 18 genera are united by a synapomorphy of bearing a smooth, cylindrical abdomen (Character 10 coded with "0"), which is also the state of character found in *Caligus*, cladistic analysis based on the principle of parsimony did not identify the latter as a member of this 18-genera monophyletic clade. In other words, *Metacaligus* is only remotely affiliated with *Caligus* on the caligid phylogeny. Accordingly, treatment of *Metacaligus* as a genus of Caligidae separate from *Caligus* is justified.

The sister-taxa relationship between *Apogonia* and *Metacaligus* is not only found in the results of analysis with *ordered* coding (see Fig. 5) but also in the analysis with *unordered* coding (see Fig. 6). So far as we are aware, only one species of *Apogonia*, *A. stocki* Cressey and Cressey, is known. It is a parasite of the dwarf cardinalfish (*Apogon mosavi* Dale) occurring in the Caribbean Sea (Cressey and Cressey, 1990). It is intriguing to point out that a species of *Metacaligus*, *M. uruguayensis* (Thomsen), is also known from the Caribbean Sea (Ho and Basirullah, 1977).

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Appendix A. Characters and character states used in the cladistic analysis of Caligidae. Numbers in parentheses denote the numerical coding of the character states. Coding in Characters 2, 8, 9, and 18 employed “internal rooting” proposed by O’Grady and Deets (1987). *Euryphorus* and *Trebius* were utilized as the outgroup in polarization of the character states transformation.

01	Lunules absent (0) or present (1).
02	Cephalothoracic shield a flat plate (1), with lateral margins folded ventrad (2), or developed into aliform expansion (0).
03	Ventral apron of leg 3 absent (0) or present (1).
04	Posterior sinuses on cephalothoracic shield present (0) or absent (1).
05	Fourth pediger exposed (0) or covered by posterior extension of cephalothoracic shield (1).
06	Fourth pediger short and distinctly wider than long (0) or elongated into a neck-like structure (1).
07	Posterolateral corners of genital complex smoothly rounded (0), protruded into a digitiform process (1), or protruded into two digitiform process (2).
08	Genital complex with smooth margin (1), with lateral expansion (0), or with posterolateral expansion (2).
09	Abdomen distinctly shorter (1) or longer (0) than cephalothoracic shield, or greatly reduced (2).
10	Abdomen smooth and cylindrical (0), bearing lateral aliform expansion (1), with posterolateral digitiform process (2), or carrying posterolateral aliform expansion (3).
11	Postantennal process present (0) or absent (1).
12	Dentiform process of maxillule with accessory tine (0), a simple process without accessory tooth (1), or absent entirely (2).
13	Sternal furca present (0) or absent (1).
14	Outermost element on terminal segment of leg 1 situated at outer corner (0) or transposed to a suterminal location (1).
15	Innermost element on terminal segment of leg 1 situated at inner corner (0) or translocated between bases of middle two elements (2).
16	Setae on posterior margin of leg 1 exopod large and plumose (0) or reduced (1).
17	Leg 2 exopod 3-segmented (0) or 2-segmented (1).
18	Armature on terminal segment of leg 2 exopod II, I, 5(1); I, I, 5(0); II, 6(2); or I, 6(3).
19	Leg 2 endopod 3-segmented (0) or 2-segmented (1).
20	Leg 3 exopod 3-segmented (0) or 2-segmented (1).
21	Basal spine of exopod of leg 3 straight and pointing outward (0), inflated and bent like a claw (1), or fused to protopod (2).
22	Leg 3 endopod 3-segmented (0), 2-segmented (1), 1-segmented (2), or absent (3).
23	Leg 4 exopod 3-segmented (2), 2-segmented (1), 1-segmented (1), or absent (3).

Appendix B. Data matrix of 23 characters and their states in 28 genera of Caligidae as used in the cladistic (phylogenetic) analysis. The question mark “?” indicates an unknown state. Due to the application of “internal rooting” (O’Grady and Deets, 1987) those characters coded with “1” in the outgroup is treated as plesiomorphic and “0” in the ingroup, apomorphic.

Genus (OTU)	Characters				
	5	10	15	20	23
outgroup	01000	00110	00000	00100	000
<i>Abasia</i>	02100	00110	11110	10100	111
<i>Anchicaligus</i>	11100	00110	02110	00100	111
<i>Anuretes</i>	01101	00110	01001	00101	211
<i>Apogonia</i>	10100	01110	01100	20100	111
<i>Arrama</i>	02010	00110	11100	11?1?	??3
<i>Belizia</i>	11100	00110	01100	00100	111
<i>Caligodes</i>	11000	01100	11100	00101	111
<i>Caligus</i>	11100	00110	01000	00100	110
<i>Caritus</i>	11100	00110	11100	00100	120
<i>Dartevellia</i>	01100	00113	10100	00101	032
<i>Echetus</i>	11100	10100	11110	00100	101
<i>Hermilius</i>	02100	00110	11000	00100	111
<i>Kabataella</i>	00210	00110	11111	21211	120
<i>Lepeophtheirus</i>	01100	00110	01000	00100	110
<i>Mappates</i>	01111	00120	11111	00101	212
<i>Metacaligus</i>	11100	00110	01100	10000	111
<i>Midias</i>	11100	00112	00000	00100	110
<i>Paralebion</i>	01100	01100	01000	00100	110
<i>Parapetalus</i>	11100	00012	01000	00100	110
<i>Parechetus</i>	11100	10203	01000	20100	110
<i>Pseudanuretes</i>	01111	00120	12101	10101	112
<i>Pseudechetus</i>	11100	12112	00000	10100	110
<i>Pseudocaligus</i>	11100	00110	01000	00100	113
<i>Pseudolepeophtheirus</i>	01100	00110	01000	00100	113
<i>Pupulina</i>	01000	01110	01100	00300	000
<i>Sciaenophilus</i>	11100	00100	01110	00100	110
<i>Sinocaligus</i>	11100	00001	01000	00000	111
<i>Synestius</i>	11100	02100	01000	10100	110