



A new species of *Cantellius* and a redescription of *C. sextus* (Hiro, 1938) (Cirripedia, Balanomorpha Pyrgomatidae) from the elephant skin coral, *Pachyseris speciosa* (Dana, 1846) (Scleractinia, Agariciidae) from Taiwan

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Abstract

Two species of *Cantellius* from Taiwan were identified from the elephant skin coral, *Pachyseris speciosa* Dana, 1846. *Cantellius sextus* was found on the underside of the coral plate and a new species, *C. hoegi* sp. nov., on the upper surface. The microdistribution indicates the different requirements of the two species. In the present study, both arthropodal and opercular plate characters of these two species were investigated using SEM techniques. Morphologically, *C. hoegi* is close to *C. pallidus*. However, the tergal scutal margins of *C. pallidus* are curved, whilst those of *C. hoegi* are straight, and its spur is narrower than that of *C. pallidus*. The apex of the scutum and tergum of *C. hoegi* forms a honeycomb-like surface, most probably the 'imprint' of the chitinous primordial valves. The separation of *C. hoegi* from *C. pallidus* is also supported by molecular analyses of three mitochondrial gene sequences (COI, 16S, and 12S rRNA). *Cantellius hoegi* differs from *C. pallidus* and *C. sextus* by >5% in all three mitochondrial genes (COI, 16S, and 12S rRNA) sequenced. The values of sequence divergence in 12S are comparable to the inter-specific divergence reported for other *Cantellius* and *Savignium* species. Interestingly, *C. hoegi* is not the sister group of *C. pallidus*, a morphologically similar species, in the present study. Further studies should focus on molecular phylogenetic analysis of *Cantellius* species.

Key words: Coral barnacles, *Cantellius*, corals, *Pachyseris*, taxonomy

Introduction

The pyrgomatid (Crustacea, Cirripedia, Thoracica) genus *Cantellius* is the most speciose genus of its family, with 22 nominal species presently known (Ross & Newman 2000). *Cantellius* shows the most plesiomorphic characteristics within the coral-inhabiting barnacles, four shell wall plates and unmodified balanoid-type opercular valves. *Cantellius* occupies the largest number of host corals (Ogawa & Matsuzaki 1992). Some *Cantellius* species are widely distributed among different corals, while a few are restricted to a single host coral. Due to their modest appearance, commonly overgrown with sclerosept, and their small size, coral barnacles tend to be overlooked. Careful examination of the opercular plates and soft parts often reveals unrecognized species and genera (Achituv & Hoeksema 2003; Achituv & Newman 2002; Chan *et al.* 2007a, b; 2008a, b).

While examining corals collected in Taiwan, we encountered two species of *Cantellius* on the agariciide coral *Pachyseris speciosa* Dana, 1846, commonly known as elephant skin coral. The colonies of the coral are primarily laminar, unifacial and usually growing in horizontal directions (Veron 2000). One of the barnacle species was found mainly on the upper surface of the coral, facing the light; the second mainly on the side facing the substratum, which is less exposed to radiation. The barnacles found on the underside were

identified as *Cantellius sextus* (Hiro 1938) (= *Creusia spinulosa* var 6; subvar. 3, Darwin 1854), while those located on the upper surface of the coral were from an unidentified species.

Darwin (1854) based the description of his *Creusia spinulosa* var 6 on barnacles embedded in an unidentified coral from the Philippines. He noted that there are specimens with "shell absolutely identical" but with three different scuta. Darwin concluded that "it is impossible to separate the first and the second subvarieties, and it is almost equally certain that the third subvariety is only an exaggeration of the second". He, therefore, assigned all these subvarieties to var 6. Darwin depicted the scuta and terga of his second and third subvarieties (Darwin, 1854: Plate 14 Figs 6n-6o and 6p-6q, respectively). Hiro (1938) redescribed and named the second subvariety *Creusia spinulosa* forma *acuta*, and the third subvariety forma *sexta*. Hiro (1938) collected forma *acuta* on the coral *Acropora formosa*, while forma *sexta* was collected on two species of elephant skin coral, *Pachyseris rugosa* and *P. speciosa*. Our material, which resembles that of Darwin and Hiro, also came from the same coral genus (*Pachyseris*). Ross & Newman (1973) raised the status of forma *acuta* and forma *sexta* to the specific status.

Previous studies on the taxonomy of *Cantellius* were often based only on the morphology of the hard parts (parietes and opercular plates). In the present study, we used SEM to examine and describe the hard as well as arthropodal parts of the two species of *Cantellius* associated with *Pachyseris speciosa* (Dana, 1846), *C. sextus* and *C. hoegi* **sp. nov.** The taxonomic status of the new *Cantellius* species identified in the present study is further proved using molecular analysis in the sequence divergence of three mitochondrial genes (COI, 16S, and 12S rRNA).

Material and methods

A fragment of a colony of *Pachyseris speciosa* with its associated barnacles was collected by SCUBA at a 5–10 m depth, at Dong-Qing-Wan, Lanyu Island, Taiwan (22°01', 121°32'). Samples were preserved in 95% ethanol and stored at the Biodiversity Museum of the Academia Sinica, Taipei, Taiwan (ASIZCR). Barnacles were removed from the coral; hard parts were separated from soft parts and treated separately. The wall plates and opercular valves were immersed for approximately two hours in household bleach, rinsed in distilled water, and adhering chitin removed using needles and fine forceps. The hard parts were then air-dried. After gold coating, the wall plates and opercular plates were observed using a scanning electron microscope (SEM, Quanta 2000). The soft body of the barnacles was examined under the dissecting microscope. The procedure of preparation of soft parts for light microscopy investigation followed Newman *et al.* (1969). For SEM investigations, all cirri and the oral cone were dissected, critical pointed dried and gold coated prior to observation using SEM. Setal descriptions follow Chan *et al.* (2008 a).

Molecular analysis. Total DNA was extracted from the adductor muscle of individual *Cantellius* species using QIAamp Tissue Kit (QIAGEN, Hilden, Germany). Partial sequences of mitochondrial genes, cytochrome *c* oxidase subunit I (COI), and 16S rRNA were amplified using the universal primers LCO1490 and HCO2198 (Folmer *et al.* 1994) for COI, and AR (Simon *et al.* 1994) and 1472 (Crandall & Fitzpatrick 1996) for 16S. For amplifying 12S rDNA, we used the primer set of Kocher *et al.* (1989) as modified by Mokady *et al.* (1994): 5'-GAAACCAGGATTAGATACCC-3' and 5'-TTTCCCGCGAGCGACGGGCG-3'. Amplifications were conducted in reaction with 40 ng of template DNA, 5 µl *Taq* DNA Polymerase Master Mix (1.5 mM MgCl₂; Ampliqon, Taipei, Taiwan), 2 µM of each primer, and ddH₂O to a total volume of 10 µl. Amplification was carried out by performing the PCR profile as follows: 2 min at 95°C for initial denaturing, then 35 cycles of 30 sec at 95°C, 1 min at 48°C, 1 min at 72°C, with a final extension of 5 min at 72°C. The PCR products were then purified using the DNA gel purification kit (Tri-I Biotech, Taipei, Taiwan). Nucleotide sequences were determined by direct sequencing of the purified PCR products on an ABI 3300 Genetic Analyzer with BigDye terminator cycle sequencing reagents (Applied Biosystems, Foster City, CA, USA). Sequences were aligned using CLUSTAL W (Thompson *et al.* 1994) with default gap weighting parameters, and manually minor adjustments. For a combined analysis, phylogenetic congruence among the

two datasets was tested under parsimony criterion using incongruent length difference (ILD) test (Farris *et al.* 1994) implemented in PAUP* v4.0 b10 as the partition-homogeneity test (Swofford 2000). Modeltest 3.7 (Posada & Crandall 1998) was used to determine the best-fit model of nucleotide substitution for the combined dataset. Three methods of phylogenetic inference were applied to the datasets: neighbor-joining (NJ), maximum parsimony and maximum likelihood (ML) using PAUP*, and Bayesian inference (BI) using MrBayes v.3.12 (Huelsenbeck & Ronquist 2001). Kimura 2-parameter distance was used with 1000 replicates used for bootstrapping in the NJ analysis. Maximum Parsimony (MP) analyses were performed using heuristic search and tree-bisection-reconnection (TBR) with 1000 random sequence addition replicates. Character states were unordered and equally weighted. Gaps were treated as missing data. Bootstrap support for the most parsimonious tree was evaluated using 1000 replicates with 100 random sequence addition replicates. For ML analysis, the heuristic search was performed with 100 random sequence addition replicates, and support for individual clades was obtained from 100 bootstrap (BP) replicates with 10 random sequence addition replicates. Bayesian analysis was run with four Markov Chain Monte Carlo (MCMC) chains for 1 000 000 generations starting from a random tree. The chain was sampled every 100 generations and the first 2000 trees were discarded as burn-in. A 50% majority-rule consensus tree was constructed from the remaining trees to estimate posterior probabilities (PP).

Results

Family Pyrgomatidae Gray 1825

Subfamily Pyrgomatinae Gray 1825

Genus *Cantellius* Ross & Newman 1973

Cantellius hoegi sp. nov.

(Figs 1–3)

Materials examined. Holotype, ASIZCR 000202, Dong-Qing-Wan, Lanyu Island, Taiwan (22°01', 121°32'), 5–10 m, coll. B.K.K. Chan; paratype, ASIZCR 000203-205, data same as holotype.

Diagnosis. Wall with approximately 20 internal ribs; lengths of scutal basal and tergal margins subequal; tergum with broad spur, external spur furrow shallow, spur length approximately 1/2 length of scutal margin.

Description. Parietes slightly projecting above coral surface, covered by coral skeleton overlying external ribs, number of ribs indistinct (Fig. 1A, B). Rostro-carinal diameter to 4 mm; wall of 4 parietes, approximately 20 internal radial ribs radiating from central orifice, some secondary ribs reaching 1/2 way between wall circumference and orifice (Fig. 1C); tubes between sheath and parietes largely filled. Orifice oval; 1/5 of rostro-carinal diameter. Sheath pigmented, orange, with concentric growth ridges extending more than 1/2 way down internal surface of parietes (Fig. 1C). Carina wider than rostrum. Basis white, solid, thin, shallow cup-shape.

Opercular valves thin, orange-red. Scutum triangular, basal margins slightly longer than tergal margins; externally growth ridges coarse, crossed by fine striae, alternate ridges forming teeth on occuludent margins (Fig 1D, F). Tergal portion of scutum inflected. Apical angle approximately 60°, 11–14 rows of round pits at apical angle forming honeycomb-like surface (Fig. 1E). Basal margins sinusoidal. Internally adductor ridge not extending beyond basal margin. Pit of lateral depressor muscle small; rostral tooth absent.

Tergum (Fig. 1G, I) with blunt spur, spur width approximately 1/2 tergal width, external furrow shallow, open from apex to spur base, angle between spur and basal margin obtuse. Pits similar to those on Scutum, forming honeycomb-like surface at apex (Fig. 1H). Sinusoidal, fine growth ridges on tergum surface. Tergal wing height approximately 1/2 tergal length (including spur). Internally small pits for tergal depressor muscle present (Fig. 1I).

Trophi: Mandibles with quadridentoid cutting edge (Fig. 2A), 2nd and 4th teeth bidentate, first 3 teeth occupying more than 3/4 length of cutting edge (Fig. 2C). Lower mandibular margin smooth, without fine setae or spines (Fig. 2A). Lateral side of mandible bearing serrulate setae with fine setules (Fig. 2B). Labrum with deep, V-shaped notch, 2 large teeth on each side of notch (Fig. 2D). Maxillule straight, row of 9 strong spines of differing sizes (Fig. 4E), serrulate setae at base of large spines (Fig. 2F). Mandibular palp and maxilla bearing serrulate setae with short and dense setules (Fig. 2G, H, I, J).

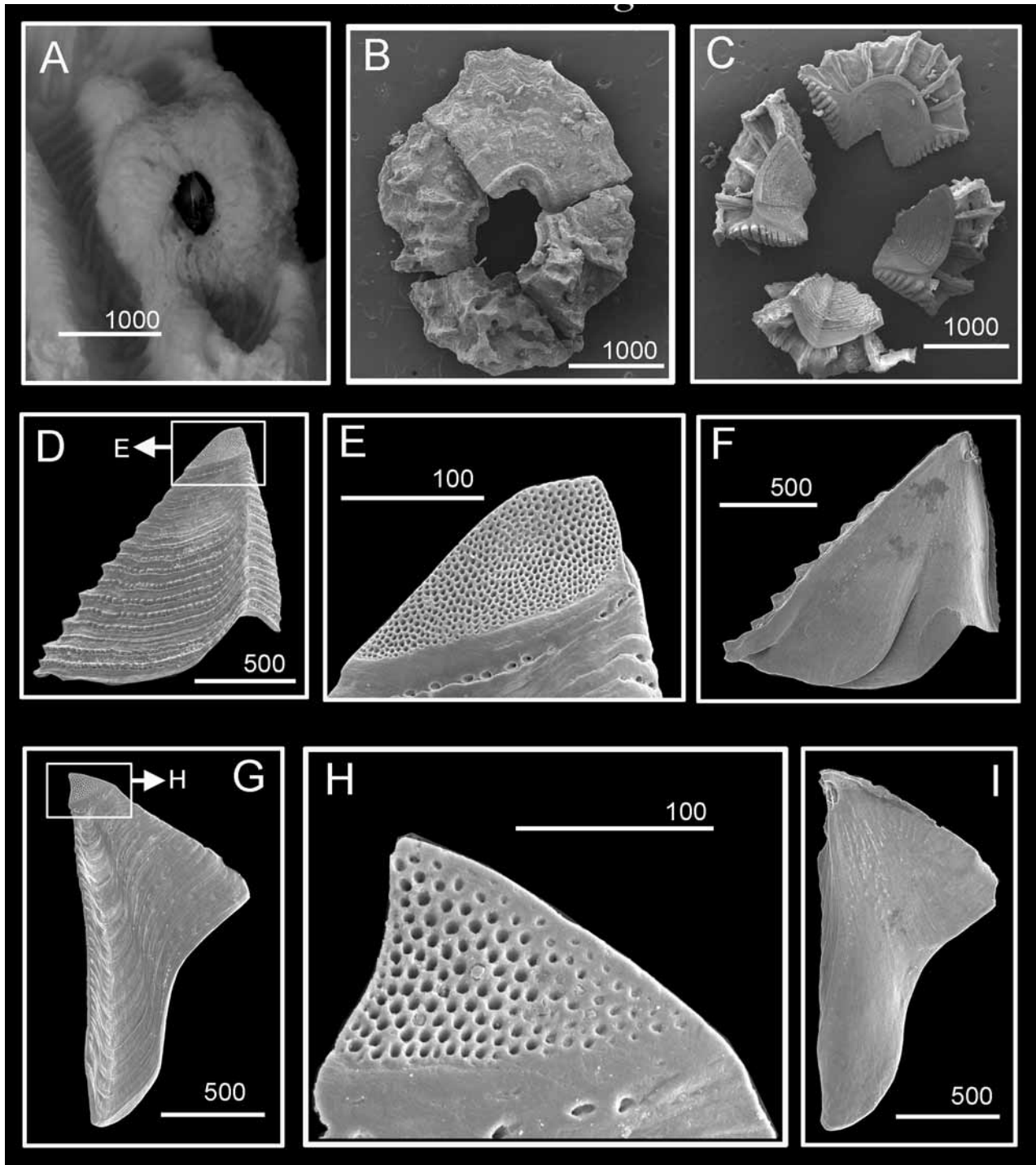


FIGURE 1. *Cantellius hoegi* sp. nov. Hard parts. A) Stereomicrograph of barnacle specimen (fixed in 95% ethanol) covered by coral sclerosept; B) outer view of barnacle with coral deposition removed; C) inner view of parietes; D) scutum, outer view; E) apical end of scutum showing honeycomb-like structure; F) scutum, inner view; G) tergum, outer view; H) apical end of scutum showing honeycomb-like structure; and I) tergum, inner view.

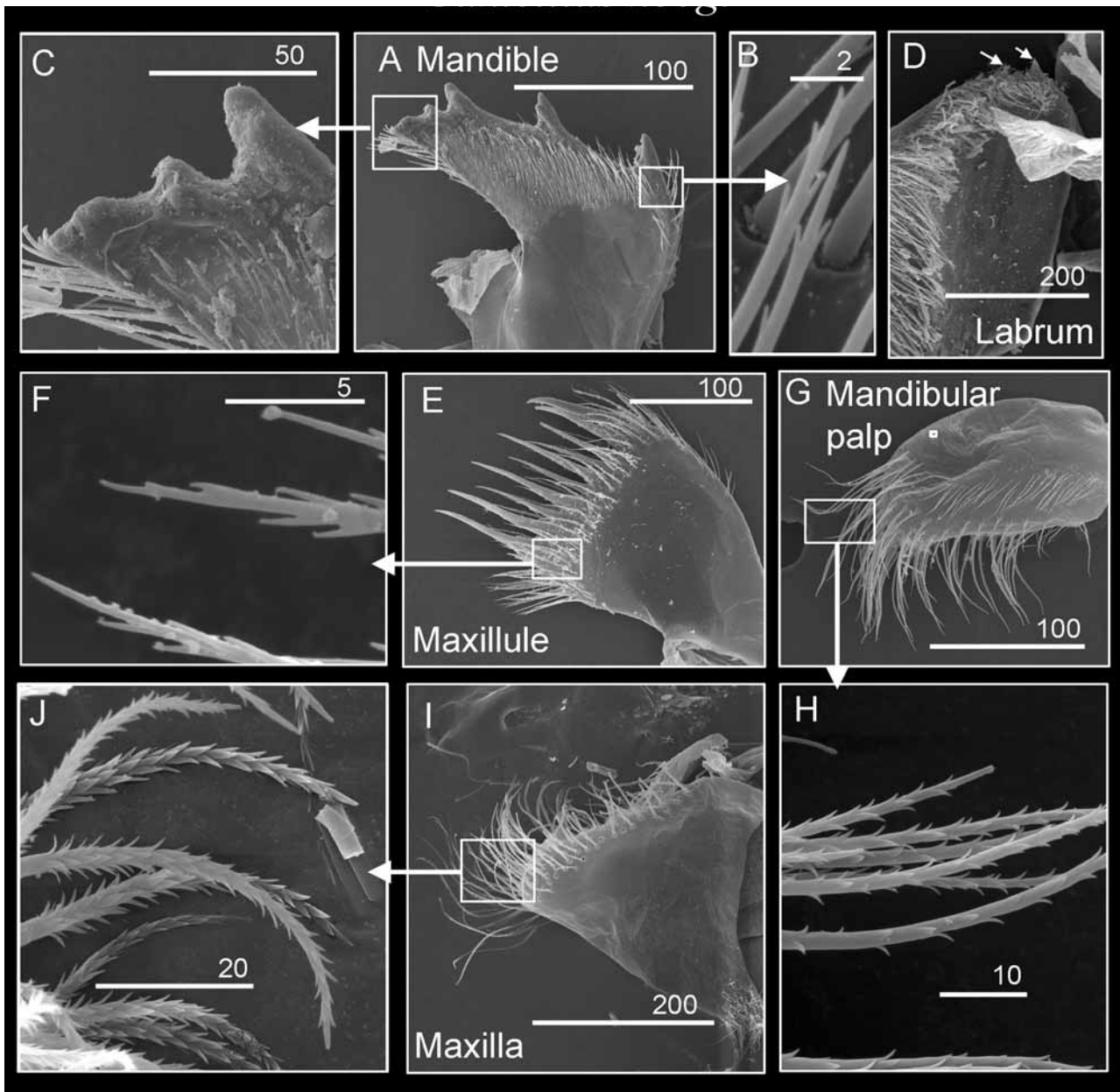


FIGURE 2. *Cantellius hoegi* sp. nov. Trophi. A) Mandible, general view; B) serrulate-type setae on mandible; C) lower margin of mandible; D) left side of labrum; arrows indicate marginal teeth; E) maxillule, general view; F) serrulate-type setae on maxillule; G) mandibulatory palp, general view; H) serrulate setae on mandibulatory palp; I) maxilla, general view; and J) serrulate-type setae on maxilla. Scale bar in μm .

Cirri: Cirrus I with rami unequal, exopodite of 13 articles, endopodite of 5 (Fig. 3A). Setae on rami serrulate (Fig. 3B) with dense setules. Protopodite bearing serrulate setae with sparsely distributed setules (Fig. 3C). Cirrus II with exopodite and endopodite similar length, 6–7 articles (Fig. 3D), rami bearing serrate setae with 3 rows of setules (Fig. 3E, F), setules distally curved, sharply pointed (Fig. 3F). Cirrus III with exopodite and endopodite thin (Fig. 3G), rami bearing serrate setae with 3 rows of setules (Fig. 3H). Protopod bearing plumose setae with long setules (Fig. 3C, I). Cirri IV–VI similar (Fig. 3J), articles of rami with 2–3 pairs of thin, serrulate setae, distal pair longest (Fig. 3K).

Etymology. The species is named in honor of Jens T. Høeg's (University of Copenhagen, Denmark) in recognition of his contribution to the study of cirripede phylogeny.

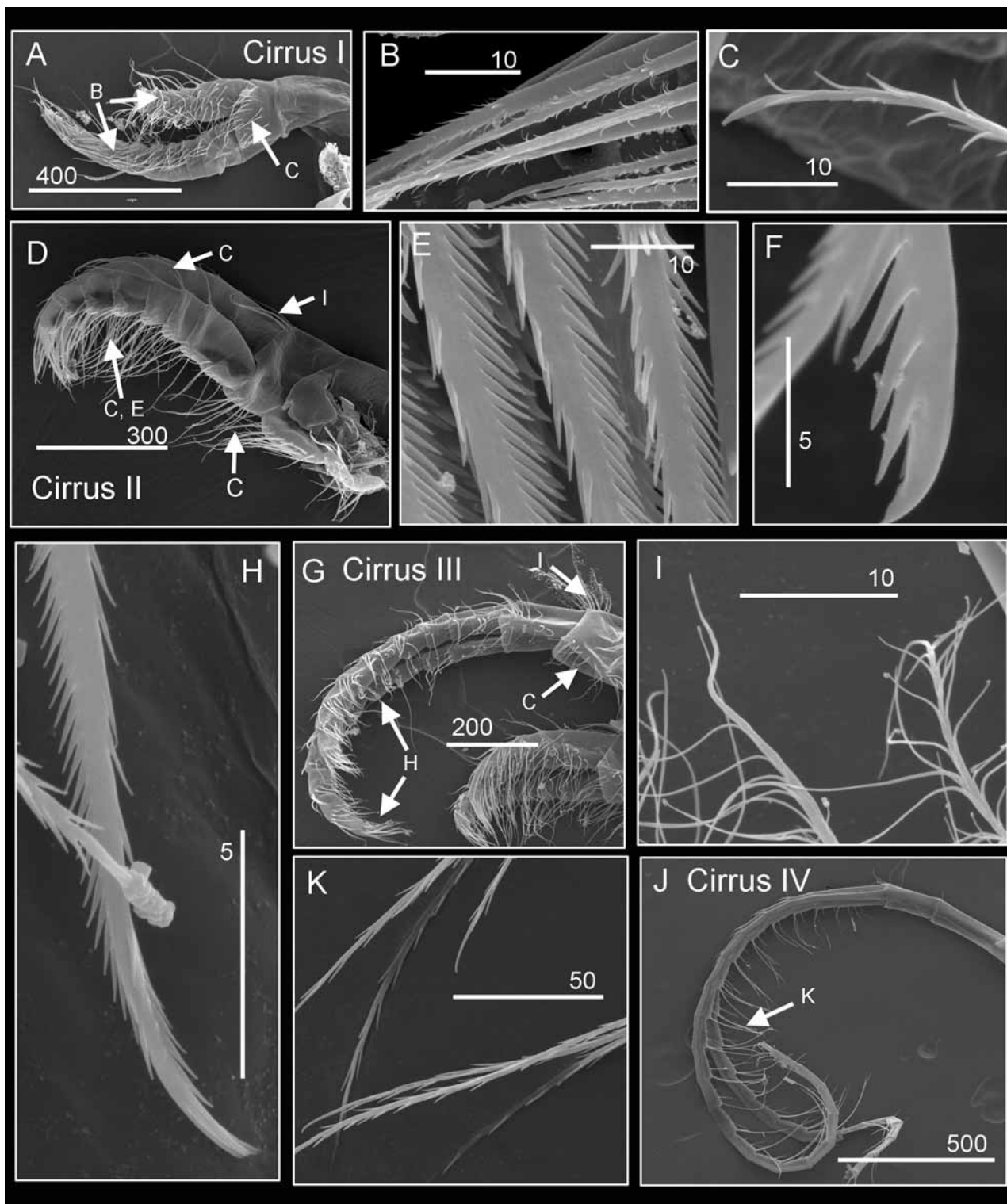


FIGURE 3. *Cantellius hoegi* sp. nov. Cirri. A) Cirrus I, general view; B) serrulate setae on rami of cirrus I; C) serrulate setae on protopod of cirri I and III and rami of cirrus II, D) cirrus II, general view, E) serrate setae with three rows of setules, located on rami of cirrus II; F) tip of serrate setae in E; G) cirrus III, general view; H) serrate setae on rami of cirrus III; I) plumose setae with long setules on protopod of cirrus III; J) cirrus IV, general view; and K) serrulate setae on rami of cirrus IV. Cirri V and VI not shown as they are similar to cirrus IV. Scale bar in μm .

Cantellius sextus (Hiro, 1938)

Figs 4–6

Pyrgoma (*Creusia*) *spinolosa* var. 6 subvar. 3. -Darwin, 1854: 379, figs 6n-6q

Creusia spinolosa forma *sexta* n. Nom. - Hiro, 1938: 398 Fig.7

Cantellius sextus. - Ross & Newman, 1973

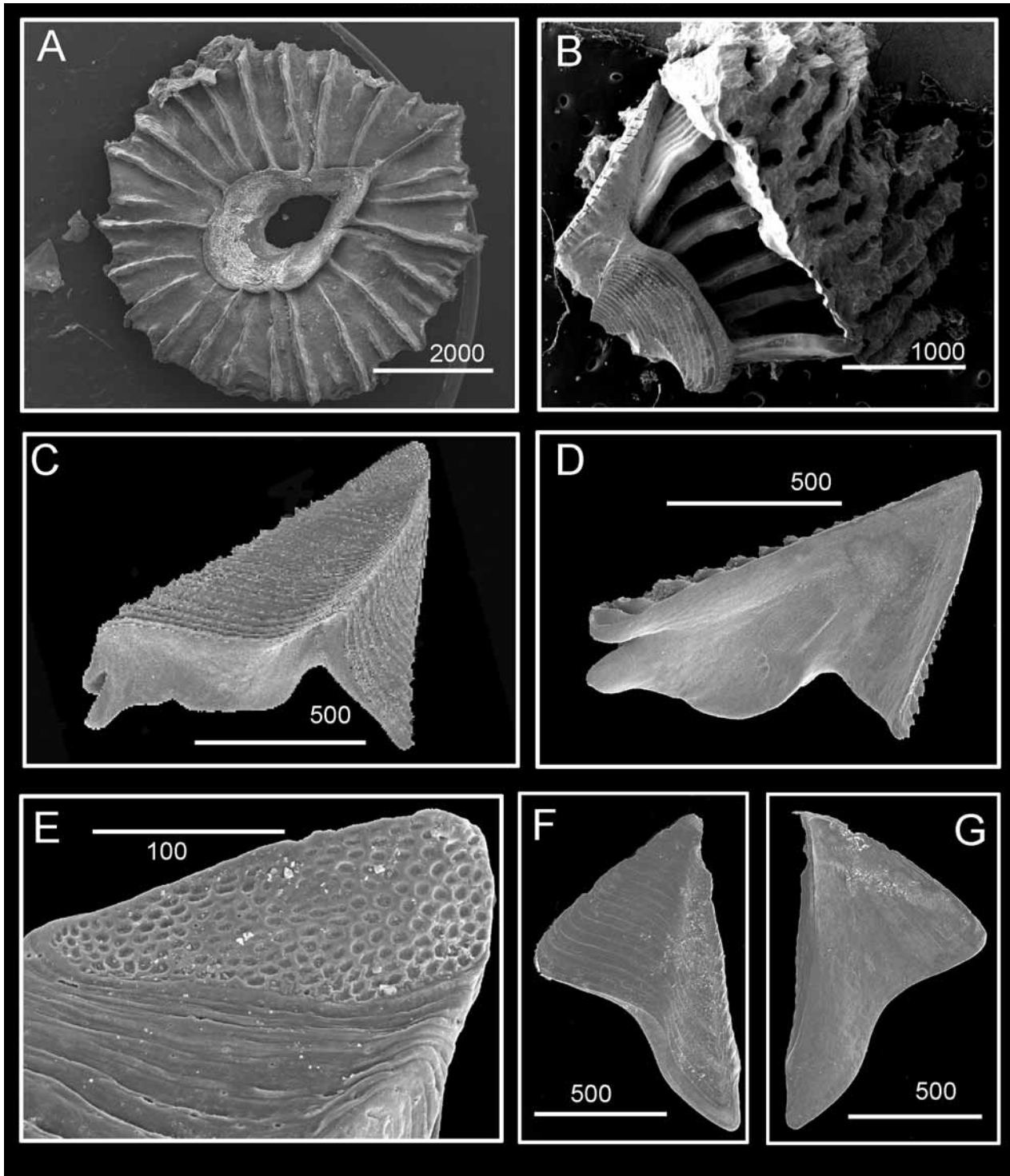


FIGURE 4. *Cantellius sextus*. Hard parts. A) Inner view of parietes under SEM; B) inner view of one of the paries; C) scutum, outer view; D) scutum, inner view; E) apical end of scutum showing honeycomb-like structure; F) tergum, outer view; and G) tergum, inner view.

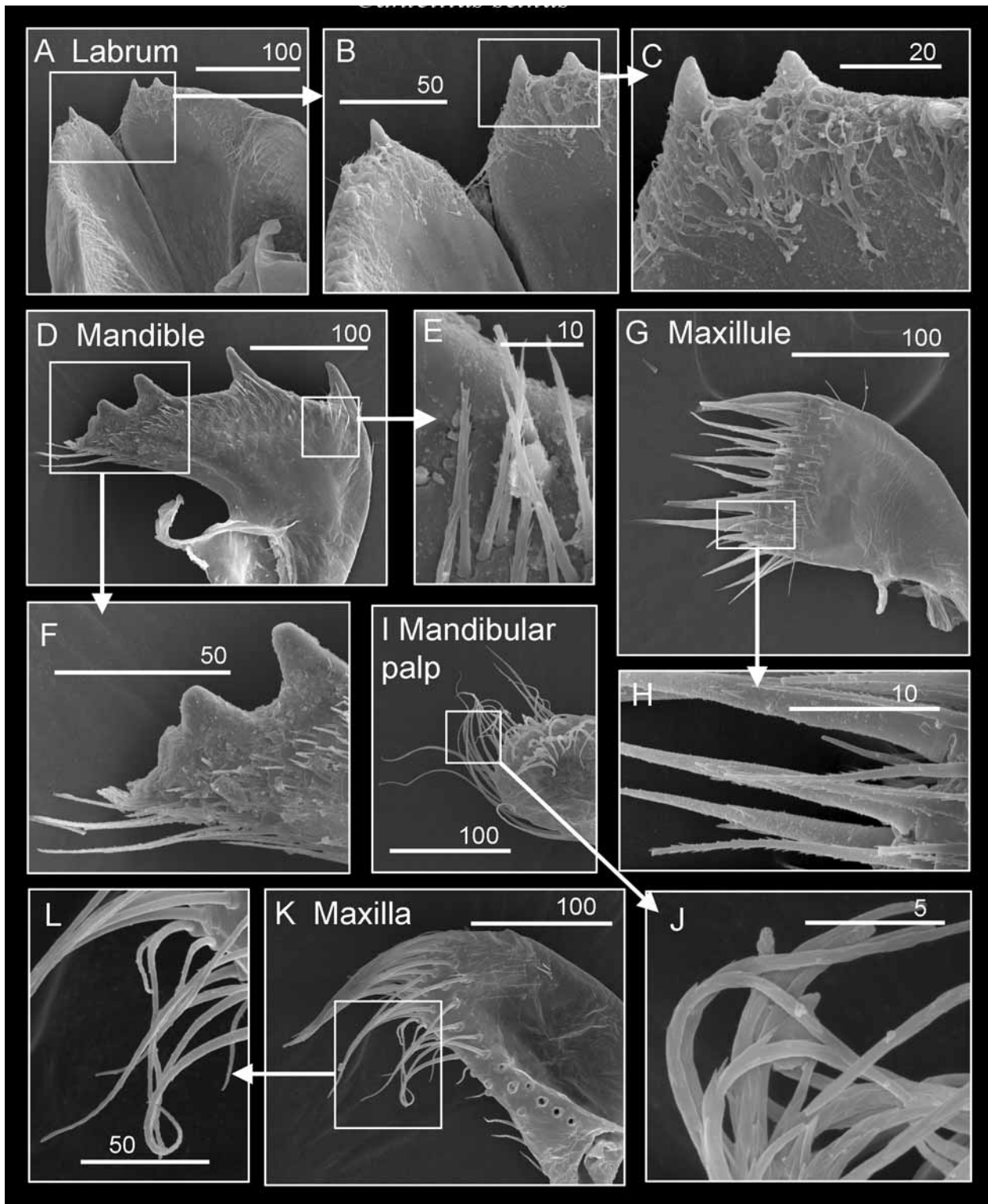


FIGURE 5. *Cantellius sextus*. Trophi. A) Labrum, general view; B) labrum, showing sharp, deep notch; C) large teeth on the edge of labrum; D) mandible, general view; E) serrulate setae on the side of mandible; F) lower margin of mandible; G) maxillule, general view; H) serrulate setae on maxillule; I) mandibulatory palp; J) serrulate setae on mandibulatory palp; K) maxilla; and L) serrulate setae on maxilla. Scale bar in μm .

Materials examined. ASIZCR 2006-2008, Dong-Qing-Wan, Lanyu Island, Taiwan ($22^{\circ}01'$, $121^{\circ}32'$), 5–10 m, coll. B.K.K. Chan.

Diagnosis. Wall with approximately 23 internal ribs (Fig. 4A, B); basal margin of scutum longer than tergal margin (Fig. 4C), adductor plate extending beyond basal margin (Fig. 4D). Tergum with broad spur, external spur furrow shallow; spur length approximately 1/2 length of scutal margin (Fig. 4F, G).

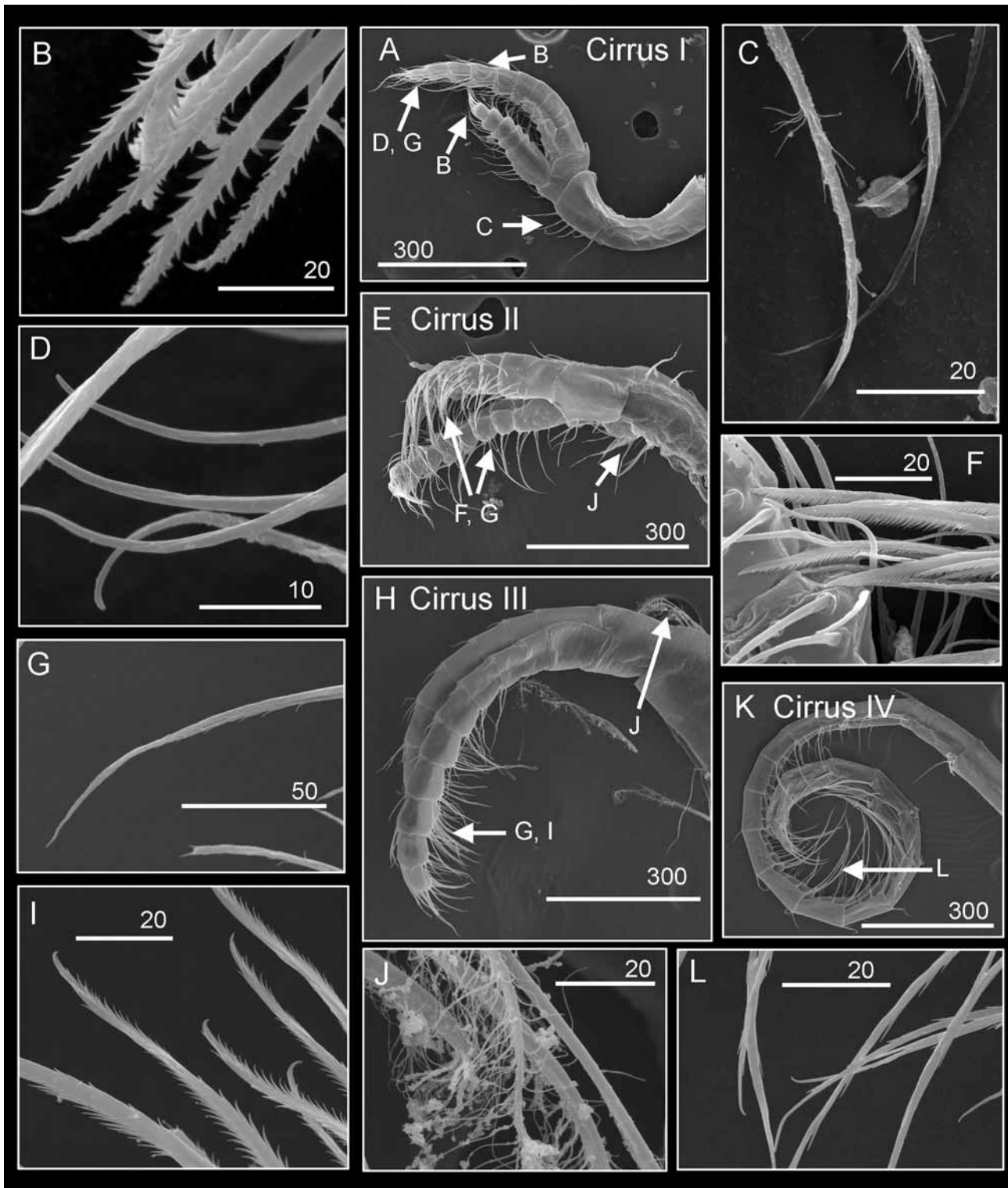


FIGURE 6. *Cantellius sextus*. Cirri. A) Cirrus I, general view; B) serrulate setae on endopod of cirrus I; C) serrulate setae on exopod of cirrus I; D) plumose setae on rami of cirrus I; E) cirrus II, overview; F) bidentate serrate setae on rami of cirrus II; G) serrulate setae on rami of cirrus III; H) cirrus III, general view; I) serrate setae on rami of cirrus III; J) plumose setae with long, loose setae on protopod of cirrus III; K) cirrus IV, general view; L) serrulate setae on rami of cirrus IV. Cirrus V and VI not shown as they are similar to cirrus IV. Scale bar in μm .

Description. Parietes (shell): Slightly projecting above the coral surface covered by coral skeleton overlying external ribs, number of ribs indistinct. Rostro-carinal diameter to 4 mm; wall of 4 parietes, approximately 23 internal radial ribs radiating from central orifice, some secondary ribs reaching 1/2 way between wall circumference and orifice (Fig. 4A, B). Tubes between sheath and parietes largely filled (Fig. 4B). Orifice oval, 1/5 of rostro-carinal diameter (Fig. 4A). Sheath pigmented, orange, with concentric growth ridges extending less than 1/2 way down internal surface of parietes. Carina wider than rostrum.

Opercular valves white, thin. Scutum (Fig. 4 C, D) triangular, externally growth ridges coarse, crossed by fine striae, alternate ridges forming teeth on occludent margin (and on tergal margins). Length of tergal margins approximately 2/3 occludent margin. Tergal portion of scutum strongly inflected. Apical angle approximately 60°, approximately 13–16 rows of pits at apical angle forming honeycomb-like surface (Fig 4E). Basal margins sinusoidal. Internally adductor plate extending beyond basal margin forming conspicuous basal ledge (Fig. 4C). Basi-occludent angle with distinct rostral tooth, pit of lateral depressor muscle small (Fig 4D).

Tergum (Fig. 4F, G) with blunt spur, spur width approximately 1/2 tergal width, external furrow shallow, open from apex to spur base, angle between spur and basal margin obtuse. Pits similar to those on scutum, at apex, forming honeycomb-like surface. Sinusoidal fine growth ridges on tergum surface. Tergal wing height approximately 1/2 tergal length (including spur). Internally small pits for tergal depressor muscle (Fig. 4F).

Trophi: Labrum with deep V-shaped notch (Fig 5A, B), 2 large teeth on each side of notch (Fig. 5d, F). Mandible quadridentoid, teeth simple; lower margin straight, without setae or spines (Fig. 5D, F).

Setae on surface of mandible serrulate with short setules (Fig. 5 E). Maxillule without obvious notch, approximately 9 strong spines (Fig 5G), serrulate setae with short setules at base of spines (Fig 5 H). Mandibular palp and maxilla bearing serrulate setae with short, sparse setules (Fig. 5 I–L).

Cirri: Cirrus I with rami unequal, exopodite longer than endopod, 9 and 5 articles, respectively (Fig. 6A); exopodite bearing thin, serrulate setae with sparse setules (Fig. 6B, D, G), endopodite bearing thick, serrulate setae with dense, short setules (Fig. 6B). Protopodite bearing plumose setae with sparse setules (Fig. 6C). Cirrus II with exopodite and endopodite similar lengths, 7–8 articles (Fig. 6E). Rami bearing serrate, bidentate setae (Fig. 6F). Protopod bearing serrulate setae with fine setules (Fig. 6J). Cirrus III with exopodite and endopodite similar lengths, approximately 10 articles (Fig. 6H), rami bearing serrate setae with 3 rows of setules (Fig. 6E). Protopod bearing plumose setae with long, loose setules (Fig. 6G, I). Cirrus IV–VI similar, rami subequal, articles bearing 3–4 pairs of serrulate setae with short setules (Fig. 6K, L).

Molecular analysis

Three individuals from both *Cantellius sextus* and the tentative new species were sequenced for three genes. 16S and 12S sequences for *Cantellius pallidus* and two other coral barnacles used as the outgroup, *Wanella milliporae* and *Nobia grandis*, were obtained from GenBank (Accession Numbers, 16S: AM497880, AM497907 and AM497898; 12S AM497881, AM497908 and AM497899, for the three species respectively), while we sequenced the COI for the three species. All sequences were deposited in GenBank (Accession Numbers, FJ379299–FJ379318; Table 1). COI is the most variable marker among the three genes analyzed. The 639 bp aligned sequence contains 171 variable sites, of which 107 are parsimony informative. The aligned 16S fragments consist of 476 bp, of which 43 of the 72 variable sites are parsimony informative. The aligned 12S rRNA sequences include 325 bp, of which 32 of the 67 polymorphic sites are parsimony informative. There is no significant incongruence between data from the three genes as revealed by ILD test ($P = 1$). Therefore, we concatenated the sequences of the three genes, resulting in a dataset with 1440 characters for analysis. The best-fit model selected for the combined dataset using Modeltest was GTR+I+G. The overall topology recovered from the other four phylogenetic inference methods result in trees that are congruent. Therefore, results from all three analyses are shown together based on the ML tree, with bootstrap values ≥ 50 and PP value ≥ 0.5 shown on the corresponding branches (Fig. 7).

TABLE 1. Sampling location, voucher ID (only applicable for *Cantellius hoegi* **sp. nov.** and *C. sextus*) and Gene bank accession numbers for the barnacle specimens used in the present study. NIL – no data available.

Barnacle species	Sampling location	Voucher ID	GenBank accession nos. COI	16S	12S
<i>Cantellius hoegi</i> sp nov	Kenting, Taiwan	ASIZCR000203-205	FJ379314 - FJ379316	FJ379308 - FJ379310	FJ379302 - FJ379304
<i>Cantellius sextus</i>	Kenting, Taiwan	ASIZCR000206-208	FJ379311 - FJ379313	FJ379305 - FJ379307	FJ379299 - FJ379301
<i>Cantellius pallidus</i>	Kenting, Taiwan	NIL	FJ379317	AM497880	AM497881
<i>Nobia grandis</i>	Kenting, Taiwan	NIL	FJ379318	AM497898	AM497899
<i>Wanella milliporae</i>	Red Sea	NIL	EU854770	AM497907	AM497908

In all analyses, individuals of *Cantellius hoegi* were consistently clustered into distinct sister clades to the other two *Cantellius* species with high nodal support (>90% for all analyses). Individuals of *Cantellius hoegi* were differentiated by ~10–11% (uncorrected p-distance) in COI and 5–7% in the 16S and 12S from the other two *Cantellius* species. These values were much greater than sequence divergences within each species ($\leq 1.7\%$). The three *Cantellius* species analysed in the present study formed a monophyletic group, with *C. pallidus* and *C. sextus* being sister taxa.

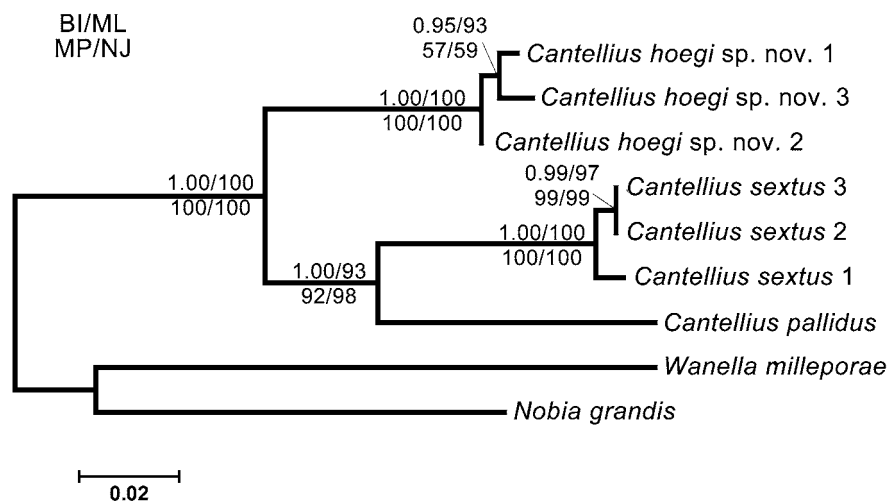


FIGURE 7. Maximum likelihood tree of combined COI, 16S and 12S dataset. The percentages of bootstrap replicates for NJ, MP, and ML and posterior probability of BI are shown above the corresponding branch for the node with values >50%. The numbers represent different individuals of the same species analyzed.

Discussion

The colony of the hermatypic coral *Pachyseris speciosus* from Taiwan, which was examined in the present study, contains two species of *Cantellius*. Comparison with other species of *Cantellius* reveals that the barnacles attached to the upper side of the *Pachyseris* coral is a new barnacle species. *Cantellius hoegi* is morphologically close to *C. pallidus* from *Pocillopora damicornis* (Broch 1931; Ross & Newman 1973). *Cantellius hoegi* can be distinguished from *C. pallidus* by characters of the scutal margins of the tergum, curved in *C. pallidus* and straight in *C. hoegi* are, and the tergal spur is narrower in *C. hoegi* than in *C. pallidus*. The species status of *C. hoegi* is supported by the molecular analyses of three mitochondrial

markers. *Cantellius hoegi* differs from the other two *Cantellius* species by being >10% in COI and >5% in 16S and 12S rRNA gene sequences. These values are comparable to the inter-specific sequence divergence in 12S reported for other *Cantellius* and *Savignium* species (Mokady *et al.* 1999; Mokady & Brickner 2001). Interestingly, *C. hoegi* is not the sister group of *C. pallidus*, a morphologically similar species.

Our findings emphasize the importance of using high-resolution devices for the study of barnacle morphology. Examination of the opercular valves under a scanning electron microscope (SEM) revealed morphological features not previously seen in opercular valves. The honeycomb structures found in the two species from *Pachyseris* were not yet reported to be observed in other species of *Cantellius*, even those studied by SEM technique (Achituv 2001; Achituv & Newman 2002; Achituv & Hoeksema 2003; Achituv 2004; Achituv & Langsam 2004). The value of the honeycomb structure as a diagnostic feature of *Cantellius* is not clear. It is possible that it can also be found in other barnacles but it is being eroded in mature animals. Evaluating the importance of this structure in taxonomy requires extensive work on other species of *Cantellius*, but finding uneroded opercular valves is rare. Darwin (1851: 22; 1854: 129) described primordial valves in lepadomorphs occurring on the scuta, terga and carina. These valves are chitinous and are lined by hexagonal prisms. When calcification commences, the first layer of shell is deposited under and around the primordial valves. Therefore, it seems that the honeycomb structure is the 'imprint' of primordial valves in calcareous matter, while in mature specimens, this structure is lost due to erosion. Similar structures were described by Newman (1987) in the early stages of *Chionelasmus darwini*.

The importance of using high-resolution techniques as a diagnostic tool has been noted by Achituv (2001), who described rimmed cylinders used for the attachment of depressor muscle on the inner side of the terga of *C. alphonsei*. In *C. septimus*, whose morphology is very similar to *C. alphonsei*, pits without rims for the attachment of this muscle are present. Chan *et al.* (2007a, b; 2008b) used the SEM technique to study the morphological variation of *Tetraclita* and *Hexechamaesipho pilsbryi* along their geographical distribution range. The colony of *Pachyseris*, the host coral of *C. hoegi* and *C. sextus*, is composed of horizontal and upright, irregularly folded plates. The host provides different niches with different microenvironments, for its inhabiting barnacles. The two species of *Cantellius* reported in this study use the different microenvironments, being found on the same coral colony but on different surfaces. *Cantellius hoegi* was found on the upper surface of the plate, which is more exposed to radiation and water energy, while *C. sextus* occurred on the underside. This microdistribution indicates the different requirements of the two species.

Soong and Chang (1983) reported the presence of two species of *Cantellius* from *Pachyseris*, *C. sextus* on *P. rugosa* and *C. pallidus* from *P. speciosa*, both from the south of Taiwan. It is most likely that the specimens they reported belong to *C. hoegi* but only the use of high-resolution devices, which were not used by these authors, could disclose the differences between *C. pallidus* and *C. hoegi*.

The available data on the distribution of *C. sextus* indicate that it is limited to one host coral, *Pachyseris*. The species has been reported in *Pachyseris* from the Malay Archipelago (Hiro 1935), Palao (Hiro 1938), and Taiwan (Soong & Chang 1983). In the present study, *Pachyseris* from Taiwan also revealed *C. sextus* on the underside of the coral. Therefore, we infer that *C. sextus* exhibits a high degree of host specificity to *Pachyseris* and suggest that its distribution may be related to the Kuroshio Current that passes through northern Philippine waters and continues into the western Pacific.

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