

A new species of *Eisothistos* (Isopoda, Cymothoidea) and first molecular data on six species of Anthuroidea from the Peninsular Malaysia

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Abstract

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Eisothistos tiomanensis

Accalathura borradailei

Apanthura pariensis

Apanthura stocki

Expanathura corallis

Mesanthura quadrata

Pendanthura tinggiensis

Tinggianthura alba

COI

A new species of expanathurid (*Eisothistos tiomanensis* sp. n.) is described and illustrated. It was collected from Pulau Tioman, Malaysia and can be distinguished by the unique bipartite shape of its uropodal exopod. *Accalathura borradailei*, *Apanthura pariensis*, *A. stocki*, *Expanathura corallis* and *Mesanthura quadrata* in the Malaysian waters are recorded for the first time. Additionally, six sequences of cytochrome c oxidase subunit I (COI) genes are presented. These are the first molecular evidence of anthuroids from the waters of Malaysia.

Introduction

The Anthuroidea Leach, 1814 are a distinctive group of mainly marine isopods (Poore 2009). More than a decade ago, Poore (2001) and Brandt and Poore (2003) revised the taxonomic and evolutionary relationships of what had formerly been treated as a suborder, Anthuridea Monod, 1922. In essence, the former work was a cladistics analysis

of family and generic relationships which resulted in the recognition of six families while the latter work replaced Anthuridea with the superfamily Anthuroidea within the Cymothoidea Wägele, 1989.

DNA barcoding was first suggested just over two decades ago (Pecnikar and Busan 2014) and it has been utilized for swift reliable biological identifications. It is a taxonomic method that uses a short genetic marker in

an organism's DNA as a unique molecular identifier to a particular species (Hebert et al. 2003a). Hebert et al. (2003b) suggested that the integration of DNA barcoding into traditional taxonomic methods are more efficient in uncovering hidden biodiversity rather than relying on traditional methods alone. On the other hand, due to the increased attention on species barcoding, the issue with questionable and poor quality sequences are becoming more apparent which could possibly harm future systematics studies (Buhay 2009).

Anthuroids have been primarily studied using morphological methods, in particular, dissection of specimens and cladistic analysis. It is only in the recent years that molecular documentations have slowly emerged such as the work of Dreyer and Wägele (2001), Haye et al. (2004), Song and Min (2015a, 2015b) and Wetzer (2001, 2002). Although the Malaysian anthuroids have attracted appreciable taxonomic attention lately (Chew et al. 2014; Chew et al. 2016), no molecular data were provided in their studies. The present study is the first to report molecular properties of anthuroids from this region. In order to ensure that the sequences are reliable genes, molecular gene cloning method was implemented and the sequences were assigned to a Phred quality score of 30.

Methods

Source of specimens and DNA preservation

The specimens in this study were obtained from coral reef areas around Peninsular Malaysia. Coral rubble was collected into a 56 litres bucket with sea water via SCUBA diving and were moderately broken up. About 10 drops of 37 % formaldehyde were added and left to stand for about 30 minutes. Next, the samples were rinsed and washed with seawater passing through a 500 µm sieve. In the field, samples were fixed with about 10 % formalin in seawater. Molecular specimens were obtained in the same manner except that they were laced with about 1 litre of absolute ethanol and left to stand for about 15 minutes. They were placed directly into precooled absolute ethanol (0 °C) and kept in an icebox which was carried throughout the field sampling to maximize tissue preservation.

Identification and morphological study

At the laboratory, the specimens were sorted and conserved separately in 4 % formalin in water in vials. Specimens were then identified and new species was selected for dissection. Whole bodies and dissected appendages were mounted in glycerol and illustrated under a Leica DMLB light microscope equipped with a camera lucida. Materials are deposited in the Muzium Zoologi, Universiti Kebangsaan Malaysia, Malaysia. The following abbreviations are used: A, antenna; AM, appendix masculina; MD, mandible; MP, maxilliped; MX, maxilla; P, pereopod; PL, pleopod; PLT, pleotelson U, uropod; UN, uropod endopod; UX, uropod exopod; UKMMZ, Universiti Kebangsaan Malaysia Muzium Zoologi.

DNA extraction, PCR amplification, DNA fragment cloning and sequencing

Before extraction, residual ethanol was removed from the tissue by evaporation at room temperature. The samples were left opened covered only with a Kimwipes tissue until they were completely desiccated. DNA was extracted from whole organism using DNeasy Blood and Tissue Kit (Qiagen, USA).

The COI sequences were amplified using the Folmer et al. (1994) universal primers (LCO1490 and HCO2198). The PCR were performed based on the program: initial denaturation period at 95 °C for 2 minutes and 35 cycles of denaturation at 95 °C for 30 seconds, annealing at 48 °C for 45 seconds and extension at 72 °C for 90 seconds. A final extension was set at 72 °C for 5 minutes. PCR products were purified using the Qiaquick PCR purification Kit (Qiagen, USA).

Purified PCR products were cloned using *E. coli* Top 10 (Invitrogen) and Thermo Scientific CloneJET PCR Cloning Kit (Thermo Fisher Scientific, USA). Colonies containing the vector along with the cloned PCR fragments were picked with a sterile pipette tip and put in to 20 µl of water. This was mixed and 1 µl was taken as the DNA template for PCR colony screening. Successful clones were purified using the innuPREP Plasmid Rapid Kit (Analytik Jena AG, Germany) and sequences were generated on an automated DNA Sequencer ABI 3100 (Applied Biosystems Inc., USA) using the ABI PRISM Dye Terminator Cycle Sequencing Ready Reaction v3.0 kit (Applied Biosystem Inc., USA).

Bioinformatics screening

Raw COI sequences were assigned to Phred quality score of 30 in order to assess the quality (Ewing and Green 1998). Then, CrossMatch software is used to obtain clean sequences by eliminating the vector sequence (Gotoh 1982). All clones were assembled using the CAP contig assembly program in Bioedit software (Hall 1998).

Results and discussions

Family Anthuridae Leach, 1814

Genus *Apanthura* Stebbing, 1990

Apanthura forceps Negoescu & Brandt, 2001

Apanthura pariensis Negoescu, 1997

Figure 1

Apanthura pariensis Negoescu, 1997: 186-194, figs 6-10.

Materials examined. 43 females, 2 males, UKMMZ-1585, Pangkor Laut, Pulau Pangkor, Perak, Malaysia, 4°11'22.32"N; 100°32'50.22"E (DMS), C. Melvin, 15 April 2014, coral rubble, ~3 m; 9 females, UKMMZ-1586, Pantai Kok, Pulau Langkawi, Kedah, Malaysia, 6°21'56.05"N; 99°40'31.13"E (DMS), C. Melvin, 4 November 2013, intertidal; 8 females, 2 males, UKMMZ-1587, Pantai Kok, Pulau Langkawi, Kedah, Malaysia, 6°21'56.05"N; 99°40'31.13"E (DMS), C. Melvin, 8 March 2015, intertidal.

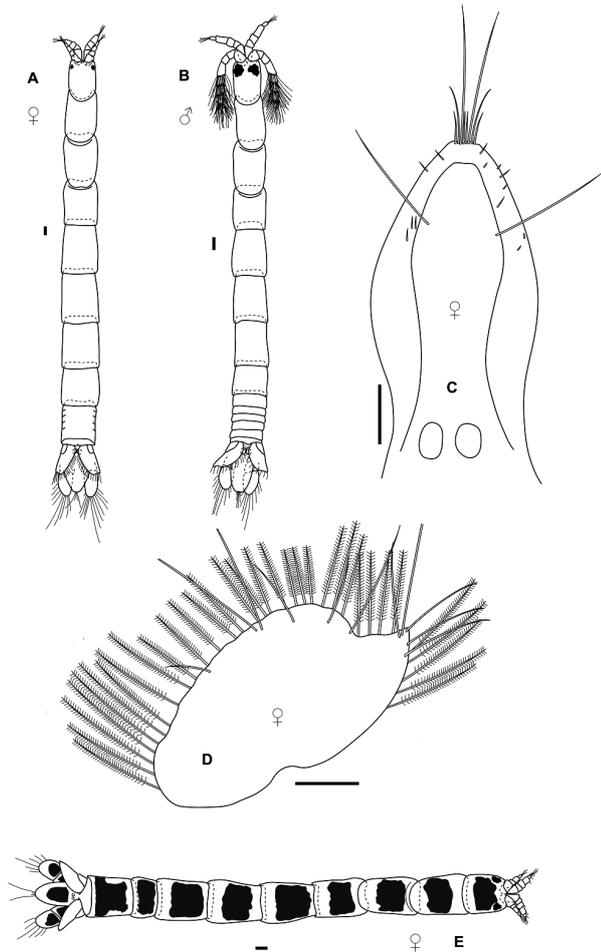


Figure 1. A–D. *Apanthura pariensis*. A. female. B. male. C. pleotelson. D. uropodal exopod. E. *Mesanthura quadrata*. All scales represent 0.1 mm.

Distribution. Pari Island, Java Sea, Indonesia (type locality); Pulau Pangkor, Malaysia; Pulau Langkawi, Malaysia.

Molecular data. A 726 base pairs of COI sequence (GenBank: MF680510) was acquired from one individual of *A. pariensis*. No insertion or deletion in the sequence alignment.

Remarks. Negoescu (1997) described in detail the female and manca materials. Fortunately, the male specimens were collected for the first time in the present study. The materials were found particularly from the west coast of Peninsula Malaysia. This is the first record of such species from this country.

***Apanthura stocki* (Müller, 1991)**

Figure 2

Amakusanthura stocki Müller, 1991: 595–600, figs. 30–56.
Apanthura stocki Müller, 1992a: 166.

Materials examined. 1 female, 1 male, UKMMZ-1576, Mentinggi, Pulau Tinggi, Johor, Malaysia, 2°16'21.67"N;

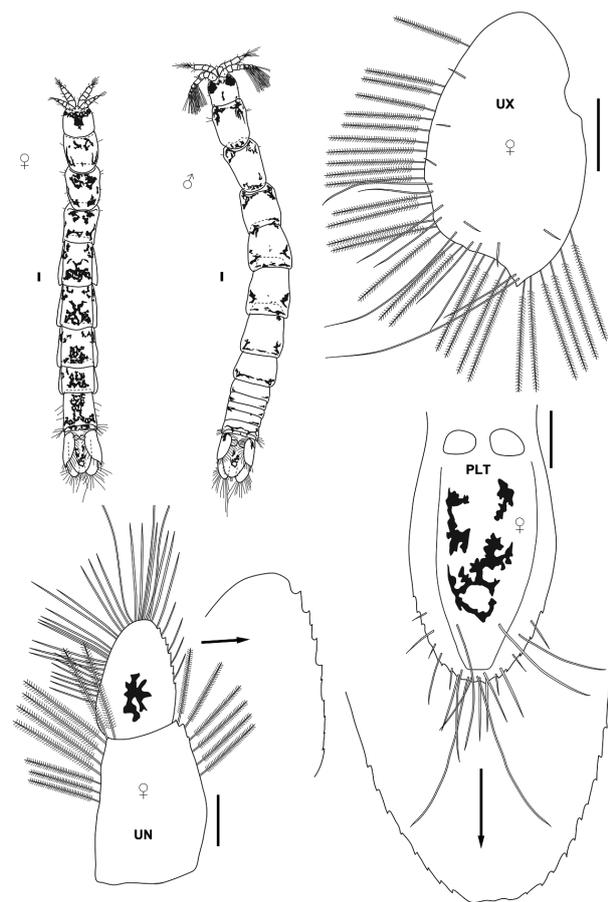


Figure 2. *Apanthura stocki*. All scales represent 0.1 mm.

104°7'18.61"E (DMS), C. Melvin, 19 April 2013, coral rubble, ~3 m; 3 females, 1 male, UKMMZ-1577, Kampung Pasir Panjang, Pulau Tinggi, Johor, Malaysia, 2°17'37.96"N; 104°6'1.97"E (DMS) E, C. Melvin, 18 December 2012, coral rubble, ~3 m; 14 females, UKMMZ-1578, Sebirah Kechil, Pulau Tinggi, Johor, Malaysia, 2°18.622"N; 104°05.616'E (DDM), C. Melvin, 18 April 2013, coral rubble, ~3 m; 13 females, 1 male, UKMMZ-1579, Pulau Seri Buat, Pahang, Malaysia, 2°41'13.59"N; 103°55'25.99"E (DMS), C. Melvin, 19 April 2014, coral rubble, ~7 m; 4 females, UKMMZ-1580, Kampung Pasir Panjang, Pulau Tinggi, Johor, Malaysia, 2°17'37.96"N; 104°6'1.97"E (DMS), C. Melvin, 15 June 2015, coral rubble, ~3 m; 27 females, UKMMZ-1581, Batu Bonchek, Pulau Dayang, Johor, Malaysia, 2°28'40.90"N; 104°30'19.12"E (DMS), C. Melvin, 26 July 2016, coral rubble, ~3 m; 6 females, UKMMZ-1582, Teluk Rha, Pulau Aur, Johor, Malaysia, 2°28'17.24"D; 104°30'53.14"E (DMS), C. Melvin, 27 July 2016, coral rubble, ~3 m.

Distribution. Sri Lanka (type locality); Pulau Besar, Malaysia; Pulau Tinggi, Malaysia; Pulau Seri Buat, Malaysia.

Molecular data. A 712 base pairs of COI sequence (GenBank: MF680509) was acquired from one indi-

vidual of *A. stocki*. No insertion or deletion in the sequence alignment.

Remarks. The Malaysian *A. stocki* differs slightly from the original material by its weakly serrated inner margin of uropodal endopod and acute distal margin of uropodal exopod in female.

Genus *Mesanthura* Barnard, 1914

Mesanthura quadrata Kensley & Schotte, 2000

Mesanthura quadrata Kensley & Schotte, 2000: 2080-2083, figs. 17–18.

Materials examined. 3 immature females, UKMMZ-1595, Pulau Seri Buat, Pahang, Malaysia, 2°41'13.59"N; 103°55'25.99"E (DMS), C. Melvin, 19 April 2014, coral rubble, ~7 m; 1 immature female, UKMMZ-1596, Batu Malang, Pulau Tioman, Pahang, Malaysia, 2°54'15.44"N; 104°6'1.08"E (DMS), C. Melvin, 18 April 2014, coral rubble, ~7 m; 1 female, UKMMZ-1597, Kampung Pasir Panjang, Pulau Tinggi, Johor, Malaysia, 2°17'37.96"N; 104°6'1.97"E (DMS), C. Melvin, 15 June 2015, coral rubble, ~3 m; 1 female, UKMMZ-1598, Sebirah Kechil, Pulau Tinggi, Johor, Malaysia, 2°18.622'N; 104°05.616'E (DDM), C. Melvin, 15 June 2015, coral rubble, ~3 m; 29 juveniles, UKMMZ-1599, Kampung Pasir Panjang, Pulau Tinggi, Johor, Malaysia, 2°17'37.96"N; 104°6'1.97"E (DMS), C. Melvin, 13 October 2012, artificial substrate unit, ~3 m; 1 female, UKMMZ-1560, Sebirah Kechil, Pulau Tinggi, Johor, Malaysia, 2°18.622'N; 104°05.616'E (DDM), C. Melvin, 18 April 2013, coral rubble, ~3 m.

Distribution. Mahé Island, Seychelles (type locality); Pulau Seri Buat, Malaysia; Pulau Tioman, Malaysia; Pulau Tinggi, Malaysia.

Molecular data. n/a.

Remarks. *Mesanthura quadrata* is recorded for the first time from the Southeast Asia region.

Genus *Pendantura* Menzies & Glynn, 1968

Pendantura tinggiensis Chew, Rahim & Mohd Yusof, 2016

Pendantura tinggiensis Chew et al., 2016: 232-238, figs 2–8.

Distribution. Pulau Tinggi, Malaysia.

Molecular data. A 709 base pairs of COI sequence (GenBank: MF680512) was acquired from one individual of *P. tinggiensis*. No insertion or deletion in the sequence alignment.

Remarks. The material used in this study is taken from the study of Chew et al. (2016).

Genus *Tinggianthura* Chew, Abdul Rahim & Haji Ross, 2014

Tinggianthura alba Chew, Abdul Rahim & Haji Ross, 2014

Tinggianthura alba Chew et al., 2014: 1-11, figs 3–9.

Distribution. Pulau Tinggi, Malaysia.

Molecular data. A 658 base pairs of COI sequence (GenBank: MF680513) was acquired from one individual of *T. alba*. No insertion or deletion in the sequence alignment.

Remarks. The material used in this study is taken from the study of Chew et al. (2014).

Family Expanathuridae Poore, 2001

Genus *Eisothistos* Haswell, 1884

Eisothistos tiomanensis sp. n.

<http://zoobank.org/2C36E902-F854-409D-9FEA-98C41EB299F8>

Figure 3–5

Holotype. Adult male, UKMMZ-1559, Labas, Pulau Tioman, Pahang, Malaysia, 2°53'13.71"N; 104°3'54.65"E (DMS), C. Melvin, 18 April 2014, coral rubble, ~15 m.

Description. Holotype, adult male.

Total body length 2.2 mm (tip of rostrum to base of telson), approximately 9.9 times as long as greatest width. Cephalon with minute rostrum; eyes grossly enlarged, containing many ommatidia laterally. Pereonites 1–7 progressively shorter, pereonites 2–3 longest, each 1.3 times as long as pereonite 1, pereonites 4–5 each the same length as pereonite 1, pereonite 6 about 0.7 times length of pereonite 5, pereonite 7 half as long as pereonite 5, pereonites 4–6 anteriorly narrow, subdistally widest, pereonite 7 anteriorly narrow, widest posteriorly. Pleonites 1–5 progressively shorter with pleonites 1–2 of similar length, pleonite 3 about 0.8 times as long as pleonite 2, pleonites 4 about 0.7 times as long as pleonite 3, pleonite 5 shortest half as long as pleonite 4, posterior margin of pleonite 6 with a medial cleft.

Antenna 1 peduncle article 1 subquadrate, articles 2–3 progressively shorter; flagellum of 9 articles with short proximal article bearing numerous aesthetascs, articles 2–9 progressively narrower, articles 2–6 each bearing 2 aesthetascs and 1 seta, articles 7–8 each bearing 1 aesthetasc distally, terminal article with 3 apical setae.

Antenna 2 peduncle articles equal, articles 1–2 each bearing 2 setae, article 3 with 1 seta, article 4 elongate, 2.4 times as long as wide, article 5 longest, twice as long as article 4; flagellum of 6 articles, progressively smaller, articles 1–4 each with 1 distal aesthetasc, article 5 with 2 distal setae, terminal article with 4 apical setae.

Mouthparts reduced.

Pereopod 1 basis 3.3 times as long as greatest width, anterior margin with 1 medial seta, posterior margin with 1 subdistal seta and fine setae; ischium 3.2 times as long

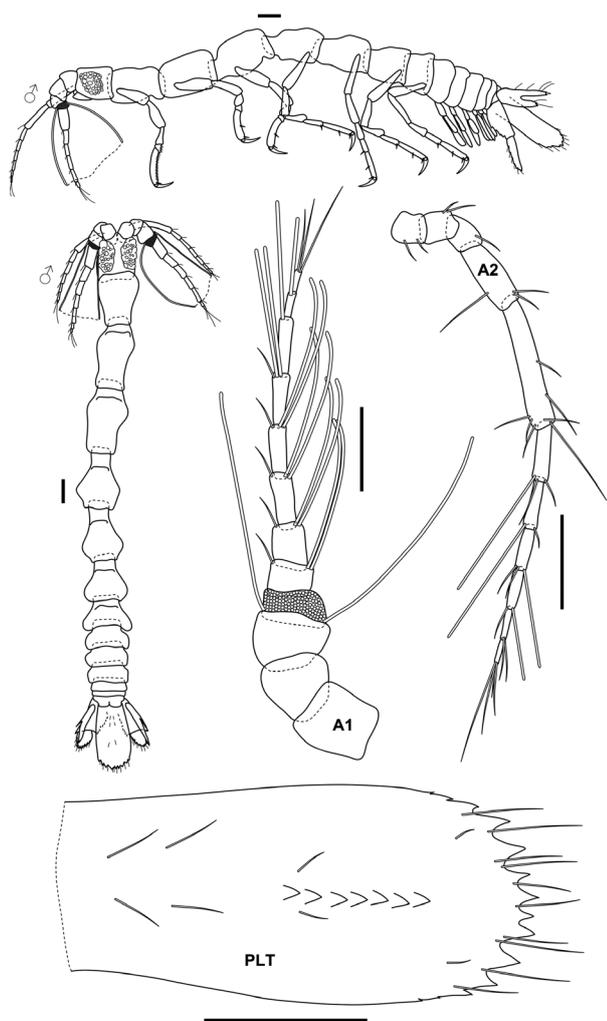


Figure 3. *Eisothisistos tiomanensis* sp. n. All scales represent 0.1 mm.

as greatest width, posterior margin subdistally with 1 seta and fine setae; merus anterior margin convex distally, bearing 2 setae, posterior margin with fine setae; carpus anterior margin half the length of posterior margin, posterior margin with 1 seta, with 4 submarginal spinules; propodus 4.4 times as long as greatest width, palm weakly concave with 21 submarginal spinules, anterior margin with 1 distal seta, palmar margin with 1 posterodistal robust seta; unguis half as long as propodus.

Pereopod 2 basis 3.7 times as long as greatest width, posterior margin with 1 subdistal seta and fine setae; ischium 3.1 times as long as greatest width, anterior margin with 2 setae, posterior margin with 2 setae and fine setae; merus anterior margin convex, bearing 1 seta, posterior margin with fine setae; carpus anterior margin half the length of posterior margin, posterior margin with 1 seta and fine setae; propodus 3.9 times as long as greatest width, anterior margin with 2 setae, palmar margin with 1 medial robust seta and 1 posterodistal robust seta, fine setae and subdistal palmar comb; unguis half the length of propodus.

Pereopod 3 basis 3.3 times as long as greatest width, anterior margin with 2 setae and fine setae, posterior mar-

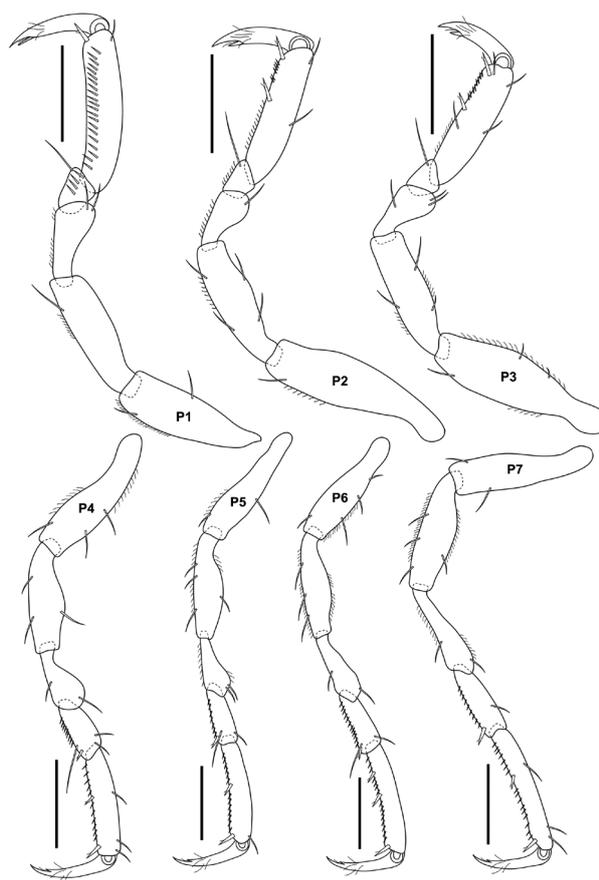


Figure 4. *Eisothisistos tiomanensis* sp. n. All scales represent 0.1 mm.

gin with 1 seta and fine setae; ischium 3.2 times as long as greatest width, anterior margin with 2 setae and fine setae, posterior margin with 1 seta and fine setae; merus anterior margin convex bearing 2 setae, posterior margin with 1 seta, carpus anterior margin one third length of posterior margin, posterior margin with 1 seta; propodus 3.6 times as long as greatest width, anterior margin with 2 setae, palmar margin with fine setae, 2 setae, subdistal palmar comb, 1 medial robust seta and 1 posterodistal robust seta; unguis half as long as propodus.

Pereopod 4 basis 3.9 times as long as greatest width, anterior margin with 2 setae and fine setae, posterior margin with 1 seta and fine setae; ischium 3 times as long as greatest width, anterior margin with 1 seta, posterior margin with 2 setae; merus anterior margin convex, bearing 1 seta, posterior margin with 1 seta, carpus linear twice as long as wide anterior margin with 2 setae, posterior margin with setae comb, 1 seta and 1 robust seta; propodus 4.5 times as long as greatest width, anterior margin with 3 setae, palmar margin bordered with setae comb, 1 medial robust seta and 1 posterodistal robust seta; unguis 0.8 times as long as propodus.

Pereopod 5 basis 4.1 times as long as greatest width, anterior margin with 1 seta, posterior margin with 1 seta and fine setae; ischium 4.1 times as long as greatest width, anterior margin with 1 seta, posterior margin with

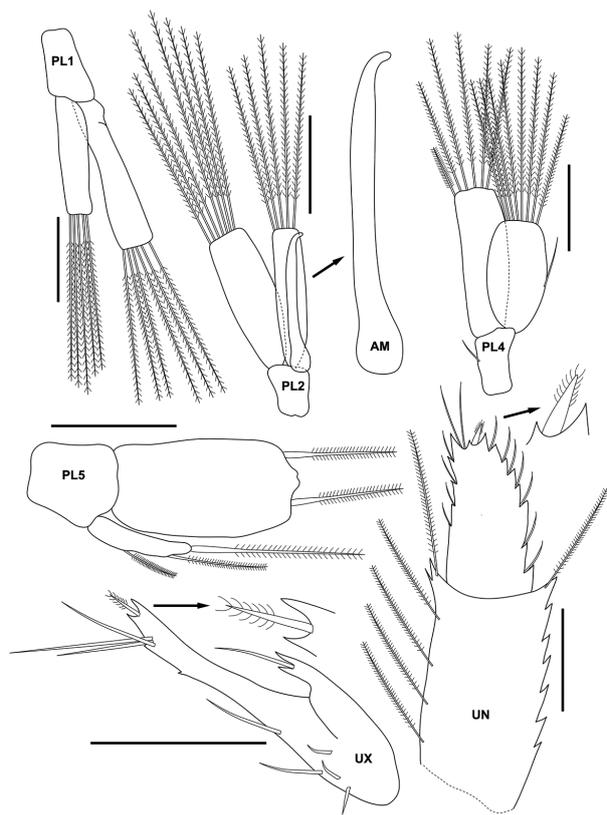


Figure 5. *Eisothistos tiomanensis* sp. n. All scales represent 0.1 mm.

3 setae and fine setae; merus anterior margin subdistally convex, bearing 2 setae, posterior margin with fine setae; carpus linear 2.4 times as long as greatest width, anterior margin with 1 seta, posterior margin with setae comb, 1 distal seta and 1 distal robust seta; propodus 4.6 times as long as greatest width, anterior margin with 1 distal seta, palmar margin bordered with setae comb, 1 medial robust seta and 1 posterodistal robust seta; unguis 0.8 times the length of propodus.

Pereopod 6 basis 4 times as long as greatest width, anterior margin with 3 setae and fine setae, posterior margin with 1 subdistal seta; ischium 3.8 times as long as greatest width, anterior margin with fine setae, posterior margin with 3 setae and fine setae; merus anterior margin convex, bearing 1 seta, posterior margin with 1 seta; carpus linear 2.6 times as long as greatest width, anterior margin with 1 seta, posterior margin bordered with setae comb, distally with 1 seta and 1 robust seta; propodus 5.3 times as long as greatest width, palmar margin bordered with setae comb, proximally with 1 robust seta, medially with 1 robust seta and posterodistally with 1 robust seta; unguis 0.7 times the length of propodus.

Pereopod 7 basis 4.1 times as long as greatest width, anterior margin with 1 seta, posterior margin with 1 seta; ischium 3.2 times as long as greatest width, anterior margin with 1 seta and fine setae, posterior margin with 3 setae and fine setae; merus slender 3.6 times as long as greatest width with anterior margin slightly convex, bear-

ing 1 seta, posterior margin with 1 seta; carpus linear 3.2 times as long as greatest width, anterior margin with 1 seta, posterior margin with setae comb and 1 robust seta; propodus 7.5 times as long as greatest width, anterior margin with 3 setae, palmar margin bordered with setae comb, 1 medial robust seta and 1 posterodistal robust seta; unguis half as long as propodus.

Pleopod 1 with elongated rami, sympod rectangular; exopod slender 4.3 times as long as greatest width, apex truncated bearing 6 plumose setae; endopod shorter than exopod 0.75 times the length of exopod, apex truncated bearing 5 plumose setae.

Pleopod 2 sympod subrectangular; exopod 3 times as long as wide, apex truncated bearing 7 plumose setae; endopod slender 4.8 times as long as greatest width, apex truncated bearing 5 plumose setae; appendix masculina almost as long as endopod, ending in a somewhat hook-like with a rounded end.

Pleopod 3 not available.

Pleopod 4 sympod subrectangular, inner margin with 1 seta; exopod broadened 1.8 times as long as greatest width, apex truncated bearing 8 plumose setae, medial lateral margin with 1 seta; endopod as long as exopod 1.3 times as long as exopod with apical margin truncated bearing 6 plumose setae.

Pleopod 5 shorter than others with sympod subquadrate; exopod much smaller than endopod half as long as endopod bearing 3 plumose setae; endopod broadened 1.8 times as long as greatest width bearing 2 plumose setae, apical margin medially with two small protuberances.

Uropod sympod with serrated inner margin, inner distal margin raised with an acute apex, outer distal margin raised with 2 acute serrations apically; endopod elongate with serrated margin bearing 13 setae, apically with 1 short robust plumose sensory spine; exopod bipartite with 5 setae proximally, medially with a central elongated spike having 3 subdistal setae and apical margin concave bearing 1 short plumose sensory spine, outer margin with a short slender spike bearing 1 seta apically, inner margin obsolete.

Pleotelson 2.2 times as long as greatest width, narrowest anteriorly, widest near posterior end, dorsal surface with 4 pairs of setae and a middorsal row of 7 obscure denticles, subdistal margin with 2 obscure teeth on each side, posterior margin strongly dentate with 10 teeth and 5 pairs of setae.

Etymology. This species is named after the type locality, Pulau Tioman, Malaysia.

Molecular data. n/a.

Remarks. Only one male specimen is available. The male *Eisothistos* in general, differs from the female by its grossly enlarged eyes, antenna 1 with short basal article bearing numerous aesthetascs, reduced mouthparts, pleonites 1–3 elongate and pleopods 1–3 with elongated peduncles and rami (Knight-Jones and Knight-Jones 2002;

Poore 2001; Poore and Lew Ton 2002; Wägele 1979). Though they appear distinctive, their tail-fan shape are more conserved within a species. Hence, it is possible to determine the identity of *Eisothistos* species based on a male specimen. Moreover in this study, the good condition of preserved material and the unique characteristic of its tail-fan, justify a description.

The uropodal exopod form in the *Eisothistos* can be divided into two types. While most possess the tripartite uropodal exopod shape, several others are described with the uncommon bipartite uropodal exopod shape. *Eisothistos tiomanensis* sp. n. belongs to the latter group with *Eisothistos anomala*, *E. corinellae*, *E. macquariensis* and *E. minutus* (Kensley 1980; Poore and Lew Ton 2002; Sivertsen and Holthuis 1980). It differs in having a unique two slender spine-like structure in the uropodal exopod rather than one central slender spike-like structure along with a proximal lobe as in *E. anomala*, *E. corinellae* and *E. macquariensis*. *Eisothistos minutus* on the other hand, possesses a form similar to the present species. Sivertsen and Holthuis (1980) described it as “a narrow elongate process on the protopod, it ends in two teeth; the protopod seems to have a second similar process laterally, the nature of this process is not clear”. Nevertheless, there is no strong robust seta on the apex of the central spike and a seta on the lateral spike. Additionally, there are no teeth present on the dorsal surface of the pleotelson in *E. minutus* while there are 7 obscure denticles present in the new Malaysian species.

Eisothistos sp. n. is readily distinguishable from the other Malaysian species, *E. besar* Müller, 1992 by its bipartite uropodal exopod (tripartite in *E. besar*) and the surface of pleotelson with 7 obscure middorsal teeth (4 distinct middorsal teeth in *E. besar*).

Genus *Expanathura* Wägele, 1981

***Expanathura collaris* (Kensley, 1979)**

Panathura collaris Kensley, 1979a: 823-827, figs 7–9; Kensley and Poore 1982: 635; *Expanathura collaris* Wägele, 1981d: 89, 121–122; Negoescu 1999: 214–220, figs 9–11; Negoescu and Wägele 1984: 118; Negoescu and Brandt 2001: 121–129, figs 14–18; Poore and Lew Ton 2002: 26-32, figs 16–19, 20a.

Materials examined. 54 females, 3 males, UKM-MZ-1567, Sebirah Kechil, Pulau Tinggi, Johor, Malaysia, 2°18.622'N, 104°5.616'E (DDM), C. Melvin, 16 May 2013, coral rubble, ~3 m; 12 females, UKMMZ-1568, Mentinggi, Pulau Tinggi, Johor, Malaysia, 2°16'21.67"N; 104°7'18.61"E (DMS), C. Melvin, 19 April 2013, coral rubble, ~3 m; 56 females, 9 males, UKMMZ-1569, Batu Malang, Pulau Tioman, Pahang, Malaysia, 2°54'15.44"N; 104°6'1.08"E (DMS), C. Melvin, 18 April 2014, coral rubble, ~7 m; 35 females, 2 males, UKMMZ-1570, Labas, Pulau Tioman, Pahang, Malaysia, 2°53'13.71"N; 104°3'54.65"E (DMS), C. Melvin, 18 April 2014, coral rubble, ~15 m; 14 females, 4 males, PLAy2-iso4d, Pantai Kok, Pulau Langkawi, Kedah, Malaysia, 6°21'56.05"N;

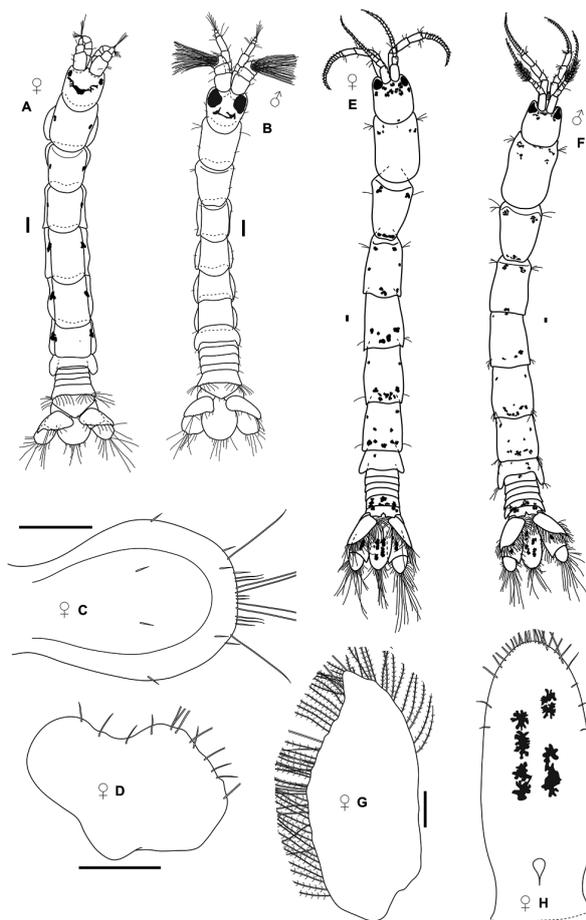


Figure 6. A–D. *Expanathura collaris*. A. female. B. male. C. pleotelson. D. uropodal exopod. E–H. *Accalathura borra-dailei*. E. female. F. male. G. uropodal exopod. H. pleotelson. All scales represent 0.1 mm.

99°40'31.13"E (DMS), C. Melvin, 8 March 2015, coral rubble, intertidal; 7 females, 1 male, PDYx1-iso4e, Batu Bonchek, Pulau Dayang, Johor, Malaysia, 2°28'40.90"N; 104°30'19.12"E (DMS), C. Melvin, 26 July 2016, coral rubble, ~3m.

Distribution. Fiji (type locality); Cook Island; Chesterfield and Melish Reefs, Moorea, Coral Sea; Lord Howe I, Tasman Sea; Papua New Guinea; Northern Territory, Queensland, Australia; Pulau Dayang, Pulau Tinggi, Malaysia; Pulau Tioman, Malaysia; Pulau Langkawi, Malaysia.

Molecular data. A 631 base pairs of COI sequence (GenBank: MF680512) was acquired from one individual of *E. collaris*. No insertion or deletion in the sequence alignment.

Remarks. *Expanathura collaris* is a widespread species dwelling in coral reef rubble especially within the South-eastern Pacific and Australian region (Kensley 1979a; Negoescu 1999; Negoescu and Brandt 2001; Poore and Lew Ton 2002). This is the first record of *E. collaris* from the Southeast Asia region.

Family Leptanthuridae Poore, 2001

Genus *Accalathura* Barnard, 1925

Accalathura borradailei (Stebbing, 1904)

Calathura borradailei Stebbing, 1904: 700, pl. 49A; Chilton 1924: 881
Accalathura borradailei Barnard, 1925: 149; Pillai 1966: 157-158, fig 3.

Materials examined. 35 females, 3 males, UKM-MZ-1612, Kampung Pasir Panjang, Pulau Tinggi, Johor, Malaysia, 2°17'37.96"N; 104°6'1.97"E (DMS), C. Melvin, 28 February 2013, coral rubble, intertidal; 34 females, UKMMZ-1613, Kampung Pasir Panjang, Pulau Tinggi, Johor, Malaysia, 2°17'37.96"N; 104°6'1.97"E (DMS), C. Melvin, 18 December 2012, coral rubble, intertidal; 1 female, UKMMZ-1614, Kampung Pasir Panjang, Pulau Tinggi, Johor, Malaysia, 2°17'37.96"N; 104°6'1.97"E (DMS), C. Melvin, 15 June 2015, coral rubble, intertidal; 20 females, UKMMZ-1615, Sebirah Kechil, Pulau Tinggi, Johor, Malaysia, 2°18.622'N; 104°05.616'E (DDM), C. Melvin, 15 June 2015, coral rubble, intertidal; 14 females, 1 male, UKMMZ-1616, Batu Bonchek, Pulau Dayang, Johor, Malaysia, 2°28'40.90"N; 104°30'19.12"E (DMS), C. Melvin, 26 July 2016, coral rubble, ~3 m.

Distribution. Maldives, Fadifolu (type locality); Thailand; Chilka Lake, India; Quilon, India; Pulau Tinggi, Malaysia.

Molecular data. A 684 base pairs of COI sequence (GenBank: MF680508) was acquired from one individual of *E. collaris*. No insertion or deletion in the sequence alignment.

Remarks. This is the first record of *Accalathura borradailei* from the waters of Malaysia.

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References

- Barnard KH (1914) Contributions to the crustacean fauna of South Africa. 3. Additions to the marine Isopoda, with notes on some previously incompletely known species. *Annals of the South African Museum* 10(11): 325a–358a, 359–440. <http://biodiversitylibrary.org/page/1554824>
- Barnard KH (1925) A Revision of the Family Anthuridae (Crustacea Isopoda), with Remarks on certain Morphological Peculiarities. *Journal of Linnean Society London (Zoology)* 36: 109–160. <https://doi.org/10.1111/j.1096-3642.1925.tb01849.x>
- Brandt A, Poore GCB (2003) Higher classification of the flabelliferan and related Isopoda based on a reappraisal of relationships. *Invertebrate Systematics* 17: 893–923. <http://dx.doi.org/10.1071/IS02032>
- Buhay JE (2009) “COI-like” sequences are becoming problematic in molecular systematic and DNA barcoding studies. *Journal of Crustacean Biology* 29(1): 96–110. <https://doi.org/10.1651/08-3020.1>
- Chew M, Abdul Rahim A, Haji Ross OB (2014) *Tinggianthura alba*: a new genus and species of Anthuridae (Isopoda, Cymothoidea, Anthuroidea) from Pulau Tinggi, Johor, Malaysia with an updated key to the genera of Anthuridae. *PLoS ONE* 9(6): e99072. <https://doi.org/10.1371/journal.pone.0099072>
- Chew M, Rahim ABA, Mohd Yusof NYB (2016) Two new species of *Pendanthura* (Isopoda, Cymothoidea, Anthuroidea) from the east coast of Peninsular Malaysia with an identification key to the species of *Pendanthura*. *Bulletin Marine Science* 92(2): 229–242. <https://doi.org/10.5343/bms.2015.1056>
- Chilton C (1924) Fauna of the Chilka Lake. *Tanaidacea and Isopoda. Memoirs of the Indian Museum* 5: 875–895.
- Dreyer H, Wägele JW (2001) Parasites of crustaceans (Isopoda: Bopyridae) evolved from fish parasites: Molecular and morphological evidence. *Zoology* 103: 157–158.
- Ewing B, Green P (1998) Base-calling of automated sequencer traces using Phred II: Error probabilities. *Genome Research* 8: 186–194. <https://doi.org/10.1101/gr.8.3.186>
- Folmer OM, Black M, Hoeh R, Lutz R, Vrijenhoek R (1994) DNA primers for amplification of mitochondrial cytochrome c oxidase subunit I from diverse metazoan invertebrates. *Molecular Marine Biology Biotechnology* 3: 294–299.
- Gotoh O (1982) An improved algorithm for matching biological sequences. *Journal of Molecular Biology* 162(3): 705–708. [https://doi.org/10.1016/0022-2836\(82\)90398-9](https://doi.org/10.1016/0022-2836(82)90398-9)
- Hall TA (1998) BioEdit: A user-friendly biological sequence alignment editor and analysis program for Windows 95/98/NT. *Nucleic Acid Symposium Series* 41: 95–98.
- Haswell WA (1884) On a new crustacean found inhabiting the tubes of *Vermilia* (Serpulidae). *Proceedings of the Linnean Society of New South Wales* 9(3): 676–680. <http://biodiversitylibrary.org/page/6602850>
- Haye PA, Kornfield I, Watling L (2004) Molecular insights into Cumacean family relationships (Crustacea, Cumacea). *Molecular Phylogenetics Evolution* 30(3): 798–809. <https://doi.org/10.1016/j.ympev.2003.08.003>
- Hebert PDN, Cywinska A, Ball SL, deWaard JR (2003a) Biological identifications through DNA barcodes. *Proceedings of the Royal Society B* 270: 313–321. <https://doi.org/10.1098/rspb.2002.2218>
- Hebert PDN, Ratnasingham S, deWaard JR (2003b) Barcoding animal life: cytochrome c oxidase subunit I divergences among closely related species. *Proceedings of Biology Society* 270: S96–S99. <https://doi.org/10.1098/rsbl.2003.0025>
- Kensley B (1979) New species of anthurideans from the Cook and Fiji Islands (Crustacea: Isopoda: Anthuridae). *Proceedings of the Biological Society Washington* 92: 814–836. <http://biodiversitylibrary.org/page/35514535>
- Kensley B (1980) Records of anthurids from Florida, Central America, and South America (Crustacea: Isopoda: Anthuridae). *Proceedings of the Biological Society of Washington* 93: 725–742. <http://biodiversitylibrary.org/page/34599608>
- Kensley B, Poore GCB (1982) Anthurids from the Houtman Abrolhos Islands, Western Australia (Crustacea: Isopoda: Anthuridae). *Proceedings of the Biological Society of Washington* 95: 625–636. <http://biodiversitylibrary.org/page/34964939>

- Kensley B, Schotte M (2000) New species and records of anthuridean crustaceans from the Indian Ocean. *Journal of Natural History* 34: 2057–2121. <https://doi.org/10.1080/002229300750022358>
- Knight-Jones EW, Knight-Jones P (2002) Four new species of *Eisothistos* (Anthuridae: Isopoda) from tubes of *Spiribidae* (Serpuloidea: Polychaeta). *Journal of Natural History* 36: 1397–1419. <https://doi.org/10.1080/00222930110052454>
- Leach WE (1814) *Crustaceology*. Edinburgh Encyclopaedia; conducted by David Brewster L.L.D. 7: 383–437, pl. 221. <http://biodiversitylibrary.org/page/37187640>
- Menzies RJ, Glynn PW (1968) The common marine isopod Crustacea of Puerto Rico: A handbook for marine biologists. *Studies on the Fauna of Curacao and other Caribbean Islands* 27(104): 1–133. <http://decapoda.nhm.org/pdfs/4429/4429.pdf>
- Monod T (1922) Sur un essai de classification rationnelle des isopodes. *Bulletin de la Société Zoologique de France* 47: 134–140. <https://doi.org/10.5962/bhl.part.3467>
- Müller HG (1991) New species and records of *Amakusanthura*, *Cyathura* and *Haliophasma* from Sri Lanka (Crustacea: Isopoda: Anthuridae). *Revue Suisse de Zoologies* 98(3): 589–612. <https://doi.org/10.5962/bhl.part.79806>
- Müller HG (1992a) Anthuridae of the genera *Apanthura* and *Cyathura* from Malaysia coral reefs, with description of two new species (Crustacea: Isopoda: Anthuridae). *Zoologischer Anzeiger* 228: 156–166.
- Müller HG (1992b) *Eisothistos besar* n. sp. from a coral reef in the Tioman Archipelago, first member of the genus from southeast Asia (Crustacea: Isopoda). *Revue Suisse de Zoologies* 99(2): 369–376. <https://doi.org/10.5962/bhl.part.79835>
- Negoescu I (1997) Isopoda Anthuridea results of the Zoological Expedition organized by “Grigore Antipa” Museum in the Indonesia Archipelago (1991). I. Pericarida (Crustacea). *Travaux du Museum National d’Histoire Naturelle Grigore Antipa* 38: 177–251. <https://www.travaux.ro/web/pdf/638-38-177-251%20Negoescu%20%20I.pdf>
- Negoescu I (1999) Isopoda Anthuridae (Crustacea) from Fiji Islands. Three new species. First record of primary and secondary males in Paranthuridae family. *Travaux du Museum d’Histoire Naturelle “Grigore Antipa”* 41: 199–280. <https://www.travaux.ro/web/pdf/393-41-199-280%20Negoescu%20I.pdf>
- Negoescu I, Wägele JW (1984) World list of the anthuridean isopods (Crustacea, Isopoda, Anthuridea). *Travaux du Museum d’Histoire Naturelle “Grigore Antipa”* 25: 99–146. <https://www.travaux.ro/web/pdf/25-TMNHNGA-99-146.pdf>
- Pecnikar ZF, Busan EV (2014) 20 years since the introduction of DNA barcoding: from theory to application. *Journal of Applied Genetics* 55: 4–52.
- Pillai NK (1966) Littoral and parasitic isopods from Kerala: Family Anthuridae.-1. *Journal of the Bombay Natural History Society* 63(1): 152–161. <http://biodiversitylibrary.org/page/47950451>
- Poore GCB (2001) Families and genera of Isopoda Anthuridea. In: Kensley B, Brusca RC (Eds) *Isopod systematics and evolution*. Balkema, Rotterdam. *Crustacean Issues* 13: 63–173.
- Poore GCB (2009) *Leipanthura casuarina*, new genus and species of anthurid isopod from Australian coral reefs without a “five-petalled” tail (Isopoda, Cymothoidea, Anthuroidea). *Zookeys* 18: 171–180. <http://dx.doi.org/10.3897/zookeys.18.198>
- Poore GCB, Lew Ton HM (2002) Expananthuridae (Crustacea: Isopoda) from the Australian region. *Zootaxa* 82: 1–60. <https://doi.org/10.11646/zootaxa.82.1.1>
- Sivertsen E, Holthuis LB (1980) The Marine Isopod Crustacea of the Tristan da Cunha archipelago. *Gunneria* 35: 1–128. https://www.ntnu.no/c/document_library/get_file?uuid=a84d7dcc-ce88-47f7-9d44-9c2fc3b-7181c&groupId=10476
- Song JH, Min GS (2015a) New record of three isopod species (Crustacea: Malacostraca: Isopoda) from South Korea. *Korean Journal of Environmental Biology* 33(2): 182–188. <https://doi.org/10.11626/KJEB.2015.33.2.182>
- Song JH, Min GS (2015b) Two new species, *Caenanthura koreana* sp. nov. and *Apanthura koreaensis* sp. nov. (Crustacea: Isopoda: Anthuridae) from South Korea. *Zootaxa* 3937(2): 362–376. <https://doi.org/10.11646/zootaxa.3937.2.7>
- Stebbing TRR (1900) On Crustacea brought by Dr. Willey from the South Seas. In: Willey A (Ed.) *Zoological Results based on material from New Britain, New Guinea, Loyalty Islands and Elsewhere collected during the years 1895, 1896 and 1897 Part V*. University Press, Cambridge, 605–690. <http://biodiversitylibrary.org/page/21340469>
- Stebbing TRR (1904) Marine crustaceans. XII. Isopoda, with description of a new genus. In Gardiner JS (Ed.) *The Fauna and Geography of the Maldive and Laccadive Archipelagoes Being the Account of the Work Carried on and of the Collections made by an Expedition during the years 1899 and 1900*. University Press, Cambridge, 600–721. <http://biodiversitylibrary.org/page/10778816>
- Wägele JW (1979) Morphologische Studien an *Eisothistos* mit Beschreibung von drei neuen Arten (Crustacea, Isopoda, Anthuridea). *Mitteilungen aus dem Zoologischen Museum der Universität Kiel* 1(2): 1–19.
- Wägele JW (1981) Zur Phylogenie der Anthuridea (Crustacea, Isopoda). Mit Beiträgen zur Lebensweise, Morphologie, Anatomie und Taxonomie. *Zoologica* 132: 1–127.
- Wägele JW (1989) Evolution und phylogenetisches System der Isopoda. Stand der Forschung und neue Erkenntnisse. *Zoologica (Stuttgart)* 140: 1–262.
- Wetzer R (2001) Hierarchical analysis of mtDNA variation and the use of mtDNA for isopod (Crustacea: Peracarida: Isopoda) systematics. *Contributions to Zoology* 70: 23–39.
- Wetzer R (2002) Mitochondrial genes and isopod phylogeny (Peracarida: Isopoda). *Journal of Crustacean Biology* 22: 1–14. <https://doi.org/10.1163/20021975-99990204>