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Morphological description and molecular characterisation of a new species of *Anilocra* Leach, 1818 (Crustacea: Isopoda: Cymothoidae) from India

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ABSTRACT

A fish parasitic isopod recently reported from India as *Anilocra leptosoma* Bleeker, 1857, was re-examined and morphologically compared to five closely related species: *A. capensis* Leach, 1818, *A. clupei* Williams and Bunkley-Williams, 1986, *A. leptosoma* Bleeker, 1857, *A. paulsikkeli* Welicky and Smit, 2019 and *A. pilchardi* Bariche and Trilles, 2006. This species was sequenced and compared to other known *Anilocra* species based on mitochondrial cytochrome *c* oxidase subunit I (COI) gene fragments. Both morphological and molecular data corroborate that the *Anilocra* species parasitising the clupeid fish *Tenualosa toli* (Valenciennes, 1847) from India should be recognised as a new species, and we describe *Anilocra grandmaae* n. sp. based on the holotype and paratype females. The key characters of *A. grandmaae* n. sp. include the body being less than 4.0 times as long as wide; antennula article 3 anterodistal margin expanded, 1.2–1.4 times as wide as long; pleonite 1 concealed by pereonite 7 and lateral margin posteriorly produced; pereopods 1–4 with three nodules on dactylus; endopod of pleopods 3–5 with a proximomedial lobe and folding; and pleotelson ovate, with lateral margins converging smoothly to a caudomedial point.

1. Introduction

Anilocra Leach, 1818, the most specious genus of body surface attaching cymothoids, currently includes 56 valid species. Leach (1818) established the genus with the description of three species (*Anilocra capensis* Leach, 1818; *Anilocra cuvieri* Leach, 1818; and *Anilocra mediterranea* Leach, 1818). Trilles (1975) synonymised *A. cuvieri* with *A. physodes* (Linnaeus, 1758) and, after 161 years, the type species for the genus (*A. cuvieri*) was designated by Kussakin (1979). Subsequently, *A. mediterranea* has also been synonymised with *A. physodes* (see Ellis 1981). A detailed generic diagnosis of *Anilocra* was provided by Bruce (1987).

Thus far, only two species of *Anilocra* have been reported from India, namely *Anilocra dimidiata* Bleeker, 1857 originally described from Malaysia and later found on the Indian oil sardine, *Sardinella longiceps* Valenciennnes, 1847 and *Anilocra leptosoma* Bleeker, 1857 originally from Java and subsequently collected in India from the Toli shad, *Tenualosa toli* (Valenciennes, 1847) (see Aneesh et al., 2019). Recently Welicky and Smit (2019), based on the drawings by Aneesh et al. (2019), suggested that the *A. leptosoma* from India may not be the original *A. leptosoma* as it differs from the *A. leptosoma* lectotype drawings by Bruce (1987) and that it shows some similarities with *A. capensis* parasitising the Hottentot seabream, *Pachymetopon blochii* (Valenciennes, 1830) from South Africa. Therefore, specimens of the aforementioned *Anilocra* sp. reported from India as *A. leptosoma* were re-examined. This study thus aimed to (1) use both morphological and molecular information based on fresh material collected from the Toli shad to determine the correct identity of the species identified by Aneesh et al. (2019) as *A. leptosoma* and (2) to provide a detailed morphological comparison of this Indian *Anilocra* sp. to some of the closely related species within the genus (such as *A. capensis*, *Anilocra clupei* Williams and Bunkley-Williams, 1986, *Anilocra paulsikkeli* Welicky and Smit, 2019 and *Anilocra pilchardi* Bariche and Trilles, 2006).

2. Materials and methods

2.1. Sampling sites

Fresh specimens of *Anilocra* were collected from the host fish *Tenualosa toli* from several fish landing centres in India (Fig. 1). These centres are from different localities along the Kerala coast of the Arabian Sea (Ayyikkara, 11°51′30″N, 75°22′27″E; Azhikkal, 11°56′36″N,

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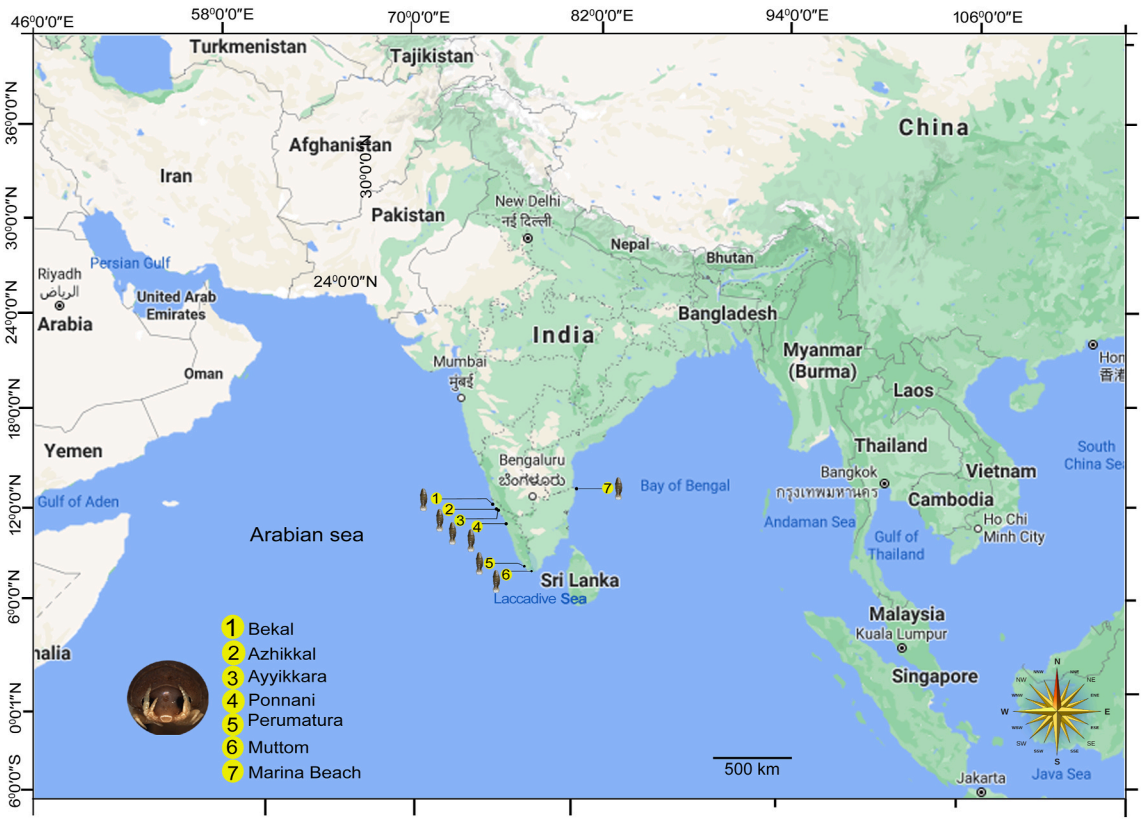

Fig. 1. Map of the distribution of *Anilocra grandmaae* n. sp., along the Kerala coast of the Arabian Sea (Ayyikkara, 11°51′30″N, 75°22′27″E; Azhikkal, 11°56′36″N, 75°18′36″E; Bekal, 12°24′05″N, 75°00′45″E; Perumatura, 8°37′40″N; 76°47′16″E; Ponnani, 10°46′57.9″N, 75°54′32″E) Muttom (south west coast, 8°07′48″N, 77°19′12″E) and the Chennai Coast in the Bay of Bengal (Marina Beach, 13°02′57″N, 80°16′58″E).

75°18′36″E; Bekal, 12°24′05″N, 75°00′45″E; Perumatura, 8°37′40″N; 76°47′16″E; Ponnani, 10°46′57.9″N, 75°54′32″E), the south west coast (Muttom, 8°07′48″N, 77°19′12″E) and the Chennai Coast in the Bay of Bengal (Marina Beach, 13°02′57″N, 80°16′58″E).

2.2. Parasite identification

The collected cymothoids were preserved in 95% ethanol for DNA studies and the remaining specimens were processed following the techniques described in Aneesh et al. (2019, 2020). One ovigerous female was designated as the holotype and one paratype was minimally dissected to conserve the specimens (the dissected appendages were kept in separate vials along with the said specimen). Methods for dissection, mounting, and drawings of appendages were according to the techniques described in Aneesh et al. (2019). The drawings of the observed mouthparts and appendages were performed using a Nikon SMZ1500 Stereo Zoom Microscope and a Nikon Eclipse80i Compound Microscope, both equipped with drawing tubes, following techniques from Hadfield and Smit (2020). Drawings were digital inked using Adobe Illustrator and a WACOM CTL-472/K0-c drawing pad. Descriptions of the species were made with the aid of the taxonomy software package DELTA (Descriptive Language for Taxonomy) (see Coleman et al., 2010), adapting a general Cymothoidae character data set originally developed by Hadfield et al. (2013). The specimens were microphotographed using the multi-focusing stereomicroscope Leica-M205A and image capturing software (Leica Application Suit).

Fish taxonomy and host nomenclature follow FishBase (Froese and Pauly, 2021) and Fricke et al. (2021). The lectotype and several fresh specimens of *A. capensis*, holotype and paratype of *A. paulsikkeli* were also examined. The non-type females of *Anilocra* sp. were re-examined from the Indian Museum. The holotype and paratypes are deposited in Western Ghat Field Research Centre of Zoological Survey of India, Kozhikode, India (ZSI/WGRC).

2.3. Molecular analysis

Genomic DNA was extracted from a pereopod and pleopod of two specimens following the protocol for animal tissue extraction of the NucleoSpin® Tissue Genomic DNA Tissue Kit (Macherey-Nagel, Düren, Germany). A targeted part of the mitochondrial cytochrome *c* oxidase subunit I (COI) gene (approximately 680 bp) of these specimens was subjected to PCR amplification with the aid of a ProFlex™ PCR thermal cycler (Applied Biosystems by Life Technologies). and universal invertebrate primers LCO1490 (5′-GGTCAACAAATCATAAAGATATTGG-3′) and HC02198 (5′ TAAACTTCAGGGTGACCAAAAAATCA-3′) (Folmer et al., 1994). PCR reactions were performed with volumes of 25 µl, using 12.5 µl Thermo Scientific DreamTaq PCR master mix, 1.25 µl of each primer, 7 µl of PCR-grade nuclease-free water and 3 µl of DNA. Conditions for the PCR were as follows: initial denaturation at 94 °C for 5 min; followed by 35 cycles of a 94 °C denaturation for 30 s, annealing at 47 °C for 50 s with an end extension at 72 °C for 2 min; and ending with a final extension of 72 °C for 10 min. PCR products were sequenced in both directions by a commercial sequencing company, Inqaba Biotechnical Industries (Pty) Ltd, Pretoria, South Africa. The bioinformatics software platform, Geneious R7.1.3 (Biomatters, Auckland, New Zealand; Kearse et al., 2012), was used to assemble the sequences.

Comparative sequences of *Anilocra* species from GenBank were downloaded and aligned to one sequence from the current study. These sequences included: LC159542 (*Anilocra* sp. 1, from Japan); KY562744 (*Anilocra brillae*); MK450445 (*Anilocra capensis*); KY562743 (*Anilocra chromis*); LC159540 (*Anilocra clupei*); KY562753 (*Anilocra haemuli*); MK652476 (*Anilocra physodes*); LC159541 (*Anilocra prionuri*). Nucleotide genetic divergence in percentage (p-distance) and base-pair differences among the different species were determined using MEGA7

Fig. 2. (A) *Anilocra capensis* Leach, 1818Leach, 1818 lectotype (43.0 mm; BMNH 1979.329:207b). (B) *Anilocra paulsikkeli* Welicky and Smit, 2019, paratype (32.0 mm; SAMC-A091295). (C) *Anilocra grandmaae* n. sp. paratype (34.0 mm; ZSI/WGRC/IR. INV./12329).

(Kumar et al., 2016).

3. Results

3.1. Taxonomy

Suborder: Cymothoida Wagele, 1989.
Superfamily: Cymothooidea Leach, 1814.
Family: Cymothoidae Leach, 1814.
Genus: *Anilocra* Leach, 1818.

Anilocra Leach, 1818: 348, 350; Desmarest (1825): 306; Milne-Edwards (1840): 255; Dana (1853): 747; Schioedte and Meinert, (1881): 100; Gerstaecker (1882): 231; Richardson (1905): 25; Hale (1926): 210; Schultz (1969): 153; Kensley (1978): 78; Kussakin (1979): 281; Brusca (1981): 140; Brusca and Iverson (1985): 45. Bruce, (1987): 89; Trilles (1975): 303; Trilles (1994): 55; Thatcher and Blumenfeldt, (2001): 270; Welicky et al. (2017): 24; Aneesh et al., (2019): 444.

Canolira Leach, 1818: 350.

Epichthyes Herklots, 1870: 122.

Type species: *Anilocra physodes* (Linnaeus, 1758), and the type specimen is held at the British Museum of Natural History (see Trilles, 1975; Ellis, 1981; Bruce, 1987).

Diagnostic characters: The identification of *Anilocra* is still based on Bruce (1987). A detailed generic diagnosis was recently provided by Welicky and Smit (2019).

3.1.1. Remarks

Anilocra Leach, 1818 can be separated from other similar external-attaching cymothoid genera by: cephalon posterior margin straight or smoothly curved, rostrum folded back and lying between antennula bases; antennula shorter than antenna, antennula articles 4–8 short; mandible palp article 3 shorter than article 2; maxilla medial lobe partially fused to lateral lobe; coxae 5–7 shorter than respective pereonite and posteriorly round; pleonites 1 and 2 with ventrolateral margins not produced; dorsal body surface strongly vaulted and posterior of pleon about 0.7 width of pereon (see Bruce, 1987).

Species of *Anilocra* can be well separated from the recently described external-attaching genus *Bambalocra* (Bruce et al., 2019) by: coxae ventral in position (vs. coxae lateral in position in *Anilocra*), posterolateral margins of pereonites 6 and 7 posteriorly produced (vs posterolateral margins of pereonites 5–7 not produced in *Anilocra*), pleopods 3–5 endopods with weak lobes, pleopods hardly visible (vs pleopod 5 with prominent folded fleshy lobes; pleopods clearly visible in dorsal view in *Anilocra*) (see Bruce et al., 2019).

Anilocra grandmaae n. sp.

Anilocra leptosoma.—Aneesh et al., (2019): 443–450, Figs. 1–4.

All material examined: 28 ♀♀, all from the body surface of *Tenualosa toli* collected from various fish landing centres such as Azhikkal (3 ♀♀), Ayyikkara (2 ♀♀), Bekal (5 ♀♀), Perumatura (5 ♀♀) and Ponnani (8 ♀♀) (Kerala coast); Muttom (2 ♀♀) (south west coast); and Marina Beach (3 ♀♀) (Bay of Bengal), India.

Holotype: 1 ♀ (31.0 mm TL; 10.0 mm W), on the base of the dorsal fin of *Tenualosa toli*from Ponnani, Kerala coast, India, August 2018, coll. PT Aneesh (ZSI/WGRC/IR. INV./12328).

Paratypes: 1 ♀ partially dissected (34.0 mm TL; 10.2 mm W), ZSI/WGRC/IR. INV./12329; 1♀ (30.0 mm TL; 10.0 mm W), ZSI/WGRC/IR. INV./12330; 1♀ (37.0 mm TL; 11.0 mm W), ZSI/WGRC/IR. INV./14611, all with same information as holotype. 1♀ (35.0 mm TL; 11.0 mm W) from Perumatura, Kerala April 2019, coll. PT Aneesh (ZSI/WGRC/IR. INV./14612); 1♀ (35.0 mm TL; 11.0 mm W) from Perumatura, November 2019, coll. PT Aneesh (ZSI/WGRC/IR. INV./14613); 1♀ (31.0 mm TL; 10.0 mm W) from Ayyikkara, November 2019, coll. PT Aneesh (ZSI/WGRC/IR. INV./14614); 1♀ (28.0 mm TL; 8.5 mm W) from Muttom, Tamil Nadu January 2020, coll. PT Aneesh (ZSI/WGRC/IR. INV./14615).

Additional material: All from the body surface of *Tenualosa toli*, Ayyikkara, Malabar coast of the Arabian Sea, India, March 2017, coll. PT Aneesh and AK Helna; 2 ♀♀ [1 ovig. (24.0 mm; 7.0 mm W) and 1 non-ovig. (24.0 mm; 7.2 mm W)]; 1 ♀ ovig. (33.0 mm; 10.0 mm W).

Representative DNA sequences: Two newly generated mitochondrial cytochrome *c* oxidase subunit I (COI) partial sequences of *A. grandmaae* have been submitted to NCBI GenBank database (http://www.ncbi.nlm.nih.gov/genbank/) with the following accession numbers: MW803168 (Isolate 1: 681 bp long); and MW803171 (Isolate 2: 682bp long).

ZooBank registration: The Life Science Identifier (LSID) pf the article is urn:lsid:zoobank.org:pub:AF44C1CB-2518-495A-B5C3-E78CE1FD415C. The LSID for the new name *Anilocra grandmaae* n. sp. is urn:lsid:zoobank.org:act:205E3E1D-A859-48B6-A2A7-02B2829F3369.

Etymology: The species name is derived from 'Grandma' in honour of the recently deceased grandmother of PTA, as a tribute to her memory. She always supported him in pursuing his education as well as his research career. This species is dedicated to all grandmothers.

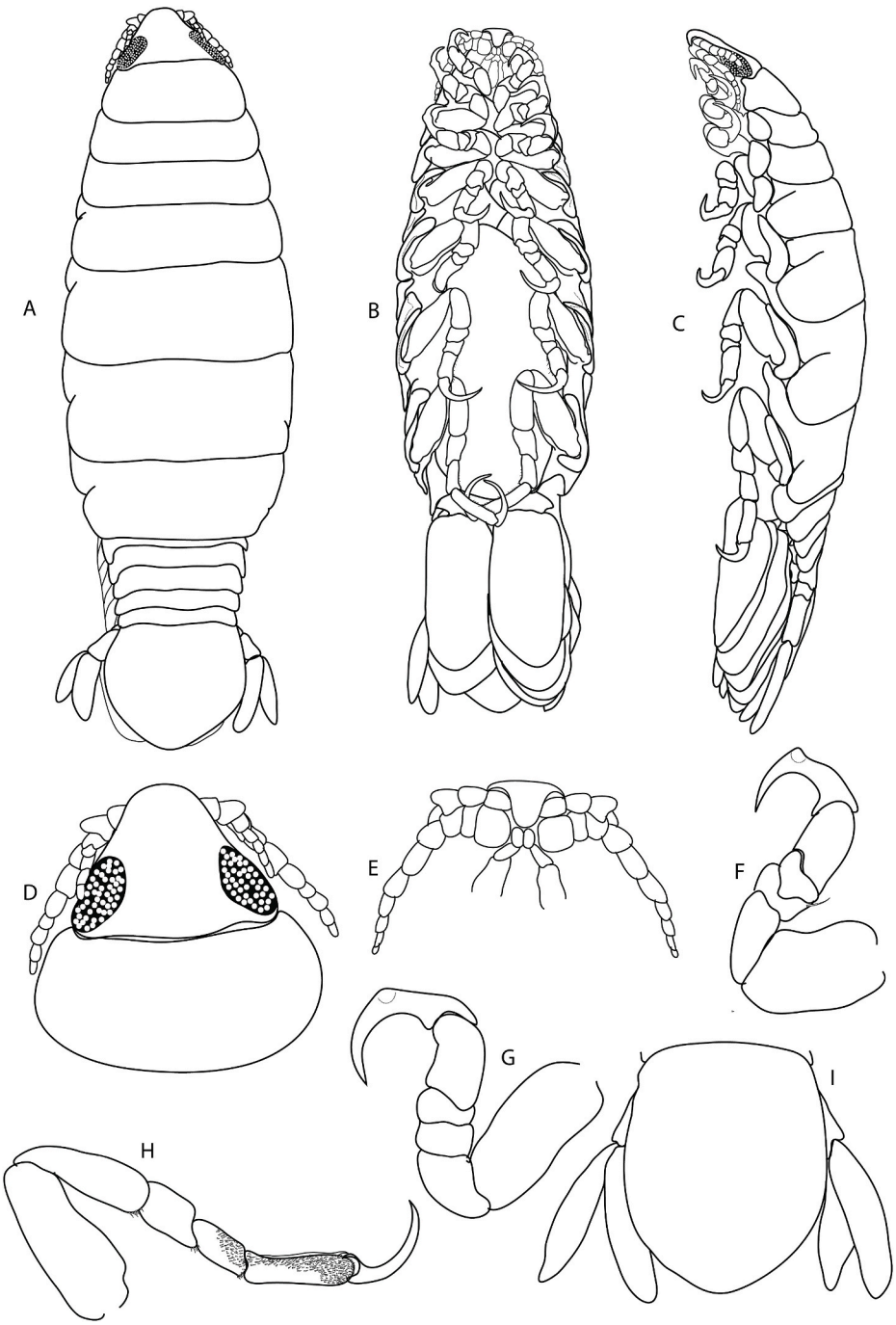

Fig. 3. *Anilocra grandmaae* n. sp. ovigerous female, holotype (34.0 mm; ZSI/WGRC/IR. INV./12328) female. (A) Dorsal view. (B) Ventral view. (C) Lateral view. (D) Cephalon dorsal. (E). Cephalon ventral. (F) Pereopod 1., (G) Pereopod 4. (H) Pereopod 7, (I). Pleotelson and uropods.

3.1.2. Description

Female (Figs. 2–6): *Body* elongate, dorsal surfaces smooth, strongly arched longitudinally or with medial longitudinal ridge present, 3.1–3.4 times as long as greatest width, widest at pereonite 6, most narrow at pereonite 1, body lateral margins convex. *Cephalon* 1.4–1.9 times wider than long, visible from dorsal view, sub triangular; frontal margin gradually tapering anterior to eyes. Rostrum narrowly rounded, domed and broad; folded back and lying between antennula bases ventrally. *Eyes* oval with distinct margins or relatively well-developed, one eye 0.2 times width of cephalon. 0.5 times as the length of the cephalon. Each eye made up of ~ 5–6 transverse rows of ommatidia, each row with ~18–22 ommatidia. *Coxae* not visible in dorsal view. Coxae of pereonite 2 more or less equal length to respective pereonite. Coxa 3 equal or slightly shorter than pereonite 3. Coxae 4–6 half the length of corresponding pereonite. Coxa 7 more than half the length of corresponding pereonite. *Pereonite* 1 smooth, anterior border straight, anterolateral angle narrowly rounded, anterolateral margins extend to base of eyes. Posterior margins of pereonites smooth and straight. Pereonite 6 longest and widest, 5 shorter than 6, 7 slightly shorter than 5 and longer than pereonite 4, pereonite1 and 4 subequal in length. Posterolateral margin of pereonites 4–7 obtusely rounded. Pereonite 7 partially overlapping pleonite 1. Pleonites posterior margin smooth, medially produced; pleonite 1 largely concealed by pereonite 7, slightly visible in dorsal view. Pleonite 2 not overlapped by pereonite 7; posterolateral angles rounded. Pleonites 1–5 progressively getting smaller in width; pleonite 5 narrowest, posterior margin produced medially. *Pleotelson* 1.1–1.3 times

Fig. 4. *Anilocra grandmaae* n. sp. ovigerous female paratypes. (A–B) Dorsal and ventral view of ovigerous female (35.0 mm; ZSI/WGRC/IR. INV./14612). (C–D) Dorsal and ventral view of non-ovigerous female (28.0 mm; ZSI/WGRC/IR. INV./14615).

Fig. 5. *Anilocra grandmaae* n. sp. ovigerous female (34.0 mm; ZSI/WGRC/IR. INV./12329). (A). Cephalon frontal view. (B). Pleotelson and uropods.

as long as anterior width, dorsal surface with two sub-medial depressions, lateral margins convex, not folded. Posterior margin converging to caudomedial point. Anterodorsal depression and medial longitudinal ridge are weakly developed.

Antennula approximately the same size as antenna, length shorter than antenna, extending slightly beyond midpoint of eye, bases moderately separated. Antennula consisting of 8 articles; article 3 anterodistal angle strongly produced; 1.2–1.3 times as wide as long; with no setae. *Antenna* with 10 articles; extending to middle or posterior of pereonite 2; article terminating without setae. *Mandibular* molar process present, ending in an acute and slightly curved incisor processes. Mandible palp article 3 shorter than article 2; article 3 with 11–14 serrate setae, terminal setae are longer. *Maxillula* simple with four terminal robust setae; two setae are slightly larger than others. *Maxilla* medial lobe partly fused to lateral lobe; median lobe with three recurved robust setae, lateral lobe with one recurved robust seta. *Maxilliped* without oostegial lobe in non-ovigerous female; article 3 with three terminal recurved spines. In ovigerous female, maxilliped article 1 modified into large, tri-lobed, oostegial lobes.

Pereopods 1–4 short with three nodules on dactylus (prominent in pereopods 2–4), anterior nodule large; distal side gently inclined, proximal side steep. Pereopods 1–3 distal margin of merus with one small spine; 6–7 distolateral margin of carpus and lateral surface of propodus with many minute spines. *Pereopods* generally gradually increasing in size, pereopod 7 longer than other pereopods.

Pleopods visible in dorsal view, exopod larger than endopod. *Proximomedial lobe* present in endopod of pleopods 3–5. *Pleopods* 1–2 endopod without proximomedial lobe, pleopod 3 proximomedial lobe moderately coiled, pleopod 4–5 proximomedial lobe well coiled. Pleopods 1–2 endopod without folds; pleopod 3–5 endopods with folds. Pleopod 3 endopod with three folds, pleopod 4 with three to four small folds, pleopod 5 multiple folded. Peduncle lobes absent. *Uropod* as long as pleotelson, peduncle 0.5 times longer than rami, peduncle lateral margin with a single seta; rami reaches up to the pleotelson apex, marginal setae absent, apices rounded. *Endopod* apically rounded, partially hidden behind the pleotelson, lateral margin weakly convex, medial margin weakly convex. *Exopod* slightly shorter than endopod, apically rounded, lateral margin weakly convex, medial margin straight,

Fig. 6. *Anilocra grandmaae* n. sp., ovigerous female paratype (34.0 mm; ZSI/WGRC/IR. INV./12329). (A) Antennula. (B) Antenna. (C) Mandible. (D) Maxilla. (E) Maxillula. (F) Maxilliped. (G) Distal segment of maxilliped palp. (H) Pereopod 2. (I) Pereopod 3. (J) Pereopod 5. (K) Pereopod 6. (L) Brood pouch. (M–Q), Pleopod 1–4. (R) Uropod.

Table 1
Nucleotide genetic divergence among COI sequences of *Anilocra grandmaae* n. sp. and *Anilocra* spp. available in GenBank. Values below the diagonal are expressed in percentage (p-distance) while values above the diagonal represent number of differences in nucleotides.

		1	2	3	4	5	6	7	8	9
1	**MW803168 *Anilocra grandmaae* n. sp.**		46	71	75	72	78	61	147	152
2	LC159540 *Anilocra clupei*	7		66	75	70	76	55	142	156
3	LC159541 *Anilocra prionuri*	11	10		58	51	51	54	150	161
4	KY562743 *Anilocra chromis*	12	12	9		47	48	62	151	158
5	KY562744 *Anilocra brillae*	12	11	8	8		21	53	155	156
6	KY562753 *Anilocra haemuli*	12	12	8	8	3		57	152	156
7	LC159542 *Anilocra* sp. 1	9	9	8	10	8	9		153	157
8	MK652476 *Anilocra physodes*	23	22	23	24	25	24	24		93
9	MK450445 *Anilocra capensis*	24	24	25	25	25	25	24	14	

Table 2
Inter-specific morphological character comparison between *Anilocra grandmaae* n. sp. and closely related species collated from original descriptions and, where applicable, redescriptions (see Leach, 1818; Bleeker, 1857; Williams and Bunkley-Williams, 1986; Bariche and Trilles, 2006; Welicky and Smit, 2019).

Characters	*A. grandmaae* n. sp.	*A. capensis*	*A. clupei*	*A. leptosoma*	*A. paulsikkeli*	*A. pilchardi*
Antennula	8 articles	8 articles	8 articles	8 articles	9 articles	8 articles
Antenna	10 articles	8 articles	10 articles	9 articles	9 articles	10 articles
Spines on pereopods	Pereopods 6–7 with many (more than 30) tiny spines	Pereopods 3, 7 with few (7–16) larger spines	Pereopods 7 with few (9–12) larger spines	Absent	Pereopods 6–7 with few (7–10) spines	Few (6–10) larger spines on pereopods 5–7
Pleopod folds	Pleopods 3–5	Only pleopod 5	Pleopods 4–5	Pleopods 3–5	Pleopods 4 and 5	Pleopods 4–5
Proximo-medial lobe of pleopod endopod	Present in endopod of pleopod 3–5.	–		Present in endopod of pleopod 3–5.	–	Present in endopod of pleopod 4–5.
Pleotelson	Widest at median point, narrow anteriorly and posteriorly.	Narrow	Narrow	Concave to median point	Posteriorly wider	Lateral margins nearly straight, strongly turned up; posterior margin scarcely bisinuate with broad caudomedial lobe
Uropod length	As long as pleotelson	1.4 times longer than pleotelson	1.3 times longer than pleotelson	As long as pleotelson	1.1 times longer than pleotelson	Extending beyond posterior of pleotelson
Uropod rami	Subequal	Exopod 1.3 times longer than endopod	Subequal	Subequal	Subequal	Subequal
Frons	Truncate	Truncate/rounded	Truncate	Pointed	Pointed	Sub-truncate
Antennula article 3	Expanded	Not expanded	Not expanded	Not expanded	Very sharp	Anterodistal angle strongly produced
Pleonite 1	Small	Normal	Normal	Small	Small	Normal
Posterior margin of pleonites	Straight	Rounded	Straight	Straight	Straight	Posterolateral margins smoothly rounded
Nodules on pereopod dactylus	1–4	Absent	1–4	2, 4		1–4

distally convex, terminating without setae.

Brood pouch with five pairs of overlapping oostegites, attached to the base of pereopods 1–5. Oostegites of pereonite 5 are larger than that of pereopod 1–4. Posterior sterna pocket present. Number of eggs or larvae per brood pouch ranges from 160 to 320, according to the size of the female.

3.2. Molecular analysis

Two 100% similar COI sequences (Isolate 1: 681 bp long, no stop codons on frame 3, Invertebrate mitochondrial code; Isolate 2: 682 bp long, no stop codons on frame 2, Invertebrate mitochondrial code) for *Anilocra grandmaae* n. sp. were newly generated. One sequence was compared to other known *Anilocra* spp. sequences available on GenBank (Table 1). The alignment was 646 bp, no stop codons, translation on frame 2, using invertebrate mitochondrial code. Nucleotide genetic divergence (p-distance) among *Anilocra grandmaae* n. sp. and other available *Anilocra* spp. ranged from 7% (*A. clupei* with 46 base pair differences) to 24% (*A. capensis* with 152 base pair differences).

4. Discussion

Anilocra grandmaae n. sp. is characterised by: body less than 4.0 times as long as wide; antennula article 3 anterodistal margin expanded, 1.2–1.4 times as wide as long; pleonite 1 visible but largely concealed by pereonite 7, lateral margin posteriorly produced; pereopods 1–4 with three prominent nodules on dactylus; endopod of pleopods 3–5 with proximomedial lobe and endopod of pleopods 3–5 with multiple folds; pleotelson ovate, lateral margins converging smoothly to a caudomedial point.

This cymothoid is the first *Anilocra* species described from India and parasitises the clupeid fish *Tenualosa toli*. This species was originally identified as *A. leptosoma* by Aneesh et al. (2019), however, as previously mentioned, Welicky and Smit (2019) noted that the species illustrated by Aneesh et al. (2019) did not conform to the redescription of the lectotype of *A. leptosoma* by Bruce (1987) and shared characteristics with *A. capensis*. Following morphological comparisons (see Table 2), this species is here confirmed to be a new species, *Anilocra grandmaae* n. sp. and the redescription of *A. leptosoma* of Aneesh et al. (2019) should be disregarded. When comparing *A. grandmaae* to *A. capensis,* the following suit of characters can be noted: only pleopod 5 has folds in *A. capensis* (vs pleopods 3–5 with folds in *A. grandmaae* n. sp.); uropods 1.35 times longer than pleotelson (vs uropods as long as pleotelson in *A. grandmaae* n. sp.); uropod rami exopod 1.3 times longer than endopod (vs sub equal in *A. grandmaae* n. sp.); pereopod dactylus without nodules (vs dactylus of pereopods 1–4 with prominent nodules). Furthermore, the molecular analysis showed 152 base pair differences and only 76% similarity (Table 1).

Even though the new species is closely related to the other slender-bodied species from the "*alloceraea* group" such as, *A. clupei*, *A. leptosoma*, *A. paulsikkeli* and *A. pilchardi*, it can be readily separated by the combinations of characters. *Anilocra grandmaae* n. sp. can be differentiated from *A. pilchardi* by: pereopods 6 and 7 with more than 30 tiny spines in *A. grandmaae* n. sp. (vs few (6–10) bigger spines on pereopods 5–7 in *A. pilchardi*); pleopod 3–5 with folds and proximomedial lobe (vs folds and proximomedial lobe in pleopod 4–5 in *A. pilchardi*); pleotelson widest at median point, narrow anteriorly and posteriorly in *A. grandmaae* n. sp. (vs pleotelson lateral margins nearly straight, strongly turned up; posterior margin scarcely bisinuate with broad caudomedial lobe in *A. pilchardi*); uropod as long as pleotelson in *A. grandmaae* n. sp. (vs uropod extending beyond posterior of pleotelson in *A. pilchardi*).

Similarly, the new species can also be well separated from *A. paulsikkeli* and *A. leptosoma* by pereopods 6 and 7 with more than 30 tiny spines in *A. grandmaae* n. sp. (vs few large spines in *A. paulsikkeli* and spines absent in *A. leptosoma*); rostrum truncate in *A. grandmaae* n. sp. (vs pointed in both *A. paulsikkeli* and *A. leptosoma*); antennula article 3 expanded in *A. grandmaae* n. sp. (vs very sharp in *A. paulsikkeli* and not expanded in *A. leptosoma*); pleotelson widest at median point, narrow anteriorly and posteriorly *A. grandmaae* n. sp. (vs posteriorly wider in *A. paulsikkeli* and pleotelson concave to median point in *A. leptosoma*).

Even though *A. grandmaae* n. sp. is phylogenetically closer to *A. clupei* in the molecular analysis, both species can be well separated by: pleopod 3–5 with folds and proximomedial lobe in *A. grandmaae* n. sp.

(vs folds and proximomedial lobe present in pleopod 4–5 in *A. clupei*); pereopods 6 and 7 with more than 30 tiny spines in *A. grandmaae* n. sp. (vs few (9–12) bigger spines on pereopods 7 in *A. clupei*); pleotelson widest at median point, narrow anteriorly and posteriorly in *A. grandmaae* n. sp. (vs pleotelson narrow 1.8 times as long as wide and lateral margins nearly straight in *A. clupei*); uropod as long as pleotelson in *A. grandmaae* n. sp. (vs uropod extending beyond posterior of pleotelson in *A. clupei*).

The comparison of genetic distances among *A. grandmaae* n. sp. and the available *Anilocra* species on GenBank showed values higher than what is normally found within a genus. Nonetheless, this is still consistent with the findings of Welicky and Smit (2019). Additional sequences (COI and additional genes) of *Anilocra* spp. would provide more information on genetic distances among species that could be linked with their morphological aspects. The use of integrative taxonomy for *Anilocra* can aid a better circumscription of species, as in the case of the present study, enabling to draw more conclusions on the actual diversity and distribution of the genus.

Declaration of competing interest

I can confirm that all the authors have read the manuscript and accept responsibility for its contents and agree to conform to the 'Ethics in Publication' documents. All authors also do not have competing interest to declare.

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