

Two new species of *Sagum* Wilson, 1913 (Copepoda: Siphonostomatoida: Lernanthropidae) parasitic on reef fishes off the Ryukyu Islands

Daisuke Uyeno D · Tohru Naruse

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Abstract Two new species of parasitic copepods, *Sagum gurukun* n. sp. and *S. bitaro* n. sp. (Siphonostomatoida: Lernanthropidae), are described based on specimens collected from the gill filaments of the double-lined fusilier *Pterocaesio digramma* (Bleeker) (Actinopterygii: Perciformes: Caesionidae) and the five-lined snapper *Lutjanus quinquelineatus* (Bloch) (Actinopterygii: Perciformes: Lutjanidae), respectively, caught off Iriomote-jima Island, the Ryukyu Islands, East China Sea, Japan. The findings bring the number of species of *Sagum* to 14. The two new species are similar in the morphology of the head and the legs 3 and 4 to the three congeners, *S. folium* Ho,

Liu & Liu, 2011, *S. paracaesionis* Izawa, 2014 and *S. vietnamiensis* Kazachenko, Kovaleva, Nguyen & Ngo, 2017. *Sagum gurukun* n. sp. is differentiated from the three congeners by the proportions of the caudal rami, the absence of leg 5, and the shape of the legs 1 and 2. *Sagum bitaro* n. sp. differs from the three congeners and *S. gurukun* n. sp. by the presence of the conical leg 5 armed with an apical seta. A key to the species of the genus *Sagum*, based on the available information on female morphology in previous publications and the present study, is provided. To date, a total of 19 species of lernanthropid copepods considered valid are known in Japanese waters.

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D. Uyeno (⊠)

Graduate School of Science and Engineering, Kagoshima University, 1-21-35 Korimoto, Kagoshima 890-0065, Japan

e-mail: duyeno@sci.kagoshima-u.ac.jp

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T. Naruse

Tropical Biosphere Research Center, Iriomote Station, University of the Ryukyus, 870 Uehara, Taketomi, Okinawa 907-1541, Japan Introduction

Lernanthropidae Kabata, 1979 is one of the large groups, comprising over 150 species, of gill parasites from marine actinopterygian fishes (Ho et al., 2011). Most species have been found from temperate to tropical waters, and its diversity decreases in higher latitudes (Boxshall & Halsey, 2004). The family was established by Kabata (1979b) based on *Lernanthropus* de Blainville, 1822 with other four genera, *Aethon* Krøyer, 1837, *Lernanthropodes* Bere, 1936, *Norion* von Nordmann, 1864 and *Sagum* Wilson, 1913, which were members of Anthosomatidae Yamaguti, 1963 (junior synonym of the Dichelesthiidae Milne Edwards, 1840). Subsequently, *Lernanthropinus* Ho



& Do, 1985 and Lernanthropsis Ho & Do, 1985 have been transferred to the family, and Mitrapus Song & Chen, 1976 omitted by Kabata (1979b) was retained again by Ho & Do (1985). Recently, Chauvanium Kazachenko, Kovaleva, Nguyen & Ngo, 2017 was established (Kazachenko et al., 2017). Hence, nine genera are recognised as valid to date. In Japanese waters, 17 valid and two unidentified species of five genera are known (Nagasawa & Uyeno, 2011; Izawa, 2014). In this study, two new species of Sagum are described based on specimens collected from the double-lined fusilier Pterocaesio digramma (Bleeker) (Actinopterygii: Perciformes: Caesionidae) and the five-lined snapper Lutjanus quinquelineatus (Bloch) (Actinopterygii: Perciformes: Lutjanidae). Both are consumed as food fish, and P. digramma is one of the major fishery resources off the Ryukyu Islands, East China Sea, Japan. A key to the species based on female morphology is provided.

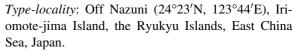
Materials and methods

Host fishes were collected by rod and line fishing off Iriomote-jima Island, the Ryukyu Islands, Japan. Copepods were carefully removed from gill filaments using forceps and dissection scissors and preserved in 80% ethanol. After soaking in lactophenol for 24 h, appendages of the copepods were then dissected using sharpened tungsten needles and observed under the compound microscope based on the modified method of Humes & Gooding (1964). The drawings were made with the aid of a drawing tube. Morphological terminology follows by Huys & Boxshall (1991). Measurements in micrometres are shown as the range followed by the mean in parentheses. Type-material is deposited in the crustacean collection of the National Museum of Nature and Science, Tsukuba (NSMT) and the University of the Ryukyus Museum, Fujukan (RUMF), Okinawa, Japan.

Family Lernanthropidae Kabata, 1979 Genus Sagum Wilson, 1913

Sagum gurukun n. sp.

Type-host: Pterocaesio digramma (Bleeker) (Actinopterygii: Perciformes: Caesionidae).



Other localities: off Nupan-zaki Cape (24°18′N, 123°39′E), Iriomote-jima Island, the Ryukyu Islands, East China Sea, Japan, 22 m depth, 10.ix.2017.

Type-material: Holotype (NSMT-Cr 25845): adult female, ex gill filaments of the type-host caught at a depth of 13 m depth on 15.v.2016. Allotype: adult male (NSMT-Cr 25846), collection data as for the holotype. Paratypes: 2 adult females and 4 adult males (NSMT-Cr 25847), collection data as for the holotype; 1 adult female and 2 adult males (RUMF-ZC-4741), collection data as for the holotype.

Voucher material examined: 3 adult females and 3 adult males (NSMT-Cr 25848), ex gill filaments of the type host.

Site in host: Both sexes attach on gill filaments (Fig. 1).

ZooBank registration: To comply with the regulations set out in article 8.5 of the amended 2012 version of the *International Code of Zoological Nomenclature* (ICZN, 2012), details of the new species have been submitted to ZooBank. The Life Science Identifier (LSID) for *Sagum gurukun* n. sp. is urn:lsid:zoobank. org:act:42699408-78A9-4C43-A45A-C1B5940EF067. *Etymology*: The specific name of the new species "gurukun" refers to the general local name for fusiliers including the type-host, *Pterocaesio digramma*, in Okinawa, Japan.



Fig. 1 Sagum gurukun n. sp., adult female attached on a gill filament of *Pterocaesio digramma* (Bleeker). Scale-bar: 1,000 µm



Japanese name for species: Gurukun-hitogatamushi.

Description (Figs. 2–5)

Adult female [Based on the holotype.] Body (Fig. 2A, B) 2,867 long, composed of cephalosome (head) and trunk composed of first pedigerous somite (short neck), fused middle part (second to fourth pedigerous somites), and small urosome. Head longer than wide, $1,110 \times 763$, wider posteriorly, with posterolateral corners ventrally bent. Trunk bearing pair of conical, protruded shoulders and dorsal plate completely covering urosome and caudal rami. Urosome (Fig. 2C) fused into a single unit. Genital complex (Fig. 2C) wider than long, 198×291 . Abdomen (Fig. 2C) wider than long, 205×223 . Caudal rami (Fig. 2C, D) bulge-like, 1.39 times longer than wide, 214×154 , bearing 5 short setae.

Antennule (Fig. 2E) fused into one segment bearing 7 setae on anterior margin and 10 + 2 aesthetascs on distal part. Parabasal flagellum (Fig. 2F) composed of rod-like basal part with seta-like distal part. Antenna (Fig. 2H) 2-segmented, subchelate; corpus (coxa and basis) robust, bearing tiny element; subchela (endopod) incurved bearing small element. Postantennal process (Fig. 2G) represented by 2 protrusions. Mandible with 8 teeth on distal tip (Fig. 2I). Maxillule (Fig. 3A) bilobate; longer outer lobe bearing 3 elements; smaller wrinkled inner lobe bearing apical element. Maxilla (Fig. 3B) 2-segmented; lecertus (syncoxa) unarmed; brachium bearing tiny element and terminal blade with serrated margin. Maxilliped (Fig. 3C) 2-segmented, subchelate; corpus bearing tiny basal element; subchela bearing tiny median seta, conical subterminal process, and terminal claw.

Leg 1 (Fig. 3D) composed of intercoxal sclerite, coxa, basis and both rami; intercoxal sclerite rod-like, unarmed; coxa covered with numerous spinules; basis covered with numerous spinules, bearing naked outer seta and inner pinnate seta; exopod unsegmented, bearing 5 blunt spines on distal margin; endopod represented by process with round distal margin. Leg 2 (Fig. 3E) lacking intercoxal sclerite; protopod fused to pedigerous somite, bearing simple outer seta; exopod represented by rod-like process bearing 5 blunt, conical spines; endopod represented by hemispherical protrusion. Leg 3 (Fig. 2B) foliate, represented by modified rami; exopod not reaching middle of dorsal plate. Leg 4 (Fig. 2B) bilobate bearing simple outer

seta near base (Fig. 3F); protopod and cylindrical both rami fused. Leg 5 absent.

Adult male [Based on the allotype.] Sexual dimorphism distinctly present in body shape and swimming legs. Body (Fig. 4A, B) 1,116 long, composed of cephalosome (head) and trunk composed of first pedigerous somite (short neck) and remaining fused part (second to anal somites). Head 593×635 , bearing pair of protrusions on posterolateral corners. Trunk barrel-shaped, 485×488 , covered with numerous small spinules on posterior half of lateral surface (Fig. 4C), bearing pair of hemispherical protrusions on posterolateral corners. Caudal rami (Fig. 4D) longer than wide, 49×26 , bearing 5 short setae.

Antennule (Fig. 4E) elongate, fused into one segment bearing 5 setae on anterior margin and 10 + 2 aesthetascs on distal part. Parabasal flagellum, antenna, mandible, maxillule, maxilla and maxilliped as in female. Postantennal process (Fig. 4F) represented by 2 adjacent protrusions.

Leg 1 as in female. Leg 2 (Fig. 4G) composed of intercoxal sclerite, protopod and both rami; intercoxal sclerite split into 2 parts, unarmed; protopod bearing naked outer seta; exopod indistinctly 2-segmented, bearing bilobed distal tip with 5 spines and patches of spinules and short hairs; endopod represented by conical process bearing patch of short hairs on distal part. Legs 3 and 4 each represented by pair of bifid cylindrical processes with simple seta near bases (Fig. 4H, I). Leg 5 absent.

Variability

The morphology of the female paratypes and other material is as in the holotype. The measurement of the body parts is as follows: body length 2,374-3,846 (2,859) (n=6); head length 915-1172 (1,017) (n=6); head width 562-995 (698) (n=6); genital complex length 397 (n=1); genital complex width 432 (n=1); abdomen length 135 (n=1); abdomen width 254 (n=1); caudal ramus length 135 (n=1); caudal ramus width 123 (n=1). Caudal ramus 1.10 times longer than wide (n=1).

The morphology of the male paratypes and other material is as in the allotype. The measurement of the body parts (n = 9) is as follows: body length 962-1,245 (1,154); head length 546-709 (642); head width 414-662 (539); genital segment length 434-517



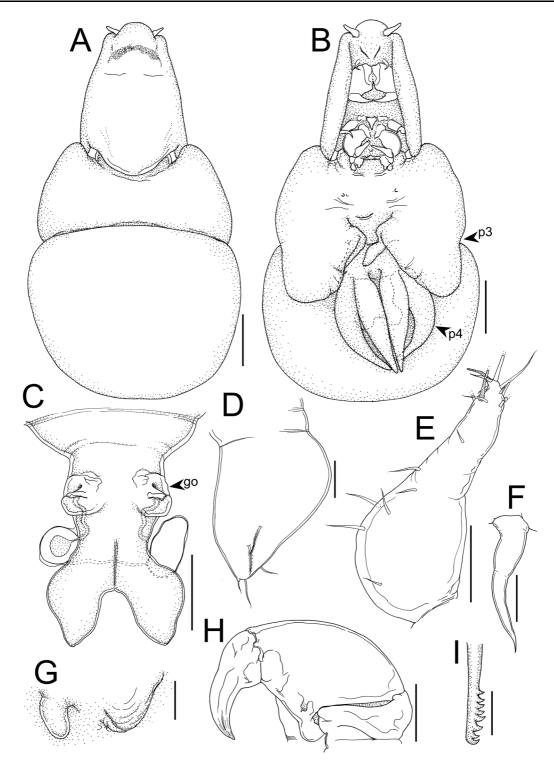


Fig. 2 *Sagum gurukun* n. sp., adult female, holotype (NSMT-Cr 25845). A, Habitus dorsal view; B, Habitus, ventral view; C, Urosome, dorsal view; D, Right caudal ramus, dorsal view; E, Right antennule, anterior part; F, Left parabasal flagellum, anterior part; G, Left postantennal process; H, Right antenna, anterior part; I, Distal part of right mandible. *Abbreviations*: go, genital opening; p3, leg 3; p4, leg 4. *Scale-bars*: A, B, 400 μm; C, 200 μm; D, 50 μm; E, F, 40 μm; G, 30 μm; H, 100 μm; I, 10 μm



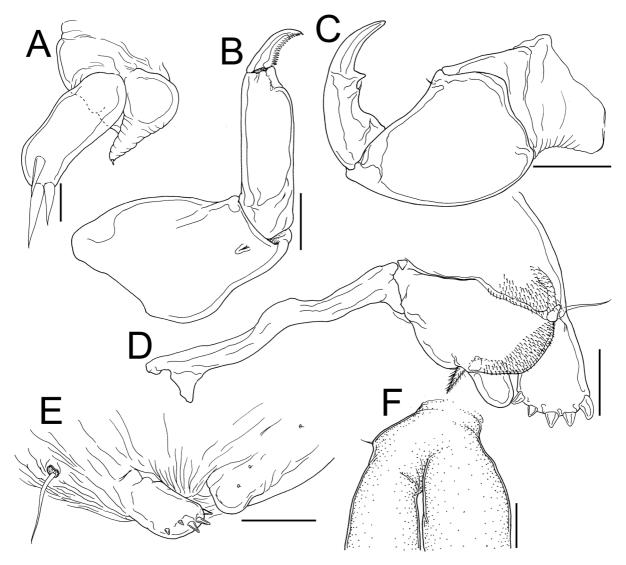


Fig. 3 *Sagum gurukun* n. sp., adult female, holotype (NSMT-Cr 25845). A, Right maxillule, anterior part; B, Right maxilla, anterior part; C, Left maxilliped, anterior part; D, Left leg 1, anterior part; E, Right leg 2, anterior part; F, Basal part of right leg 4. *Scale-bars*: A, B, 20 μm; C, 100 μm; D, 40 μm; E, 30 μm; F, 100 μm

(491); genital segment width 447–522 (495); caudal ramus length 34–46 (43); caudal ramus width 20–30 (26).

Remarks

Sagum gurukun n. sp. differs from all 12 congeners based on female characters. Seven congeners [S. angulatum (Krøyer, 1863); S. enneacentri Pillai, 1985; S. epinepheli (Yamaguti & Yamasu, 1960); S. flagellatum Wilson, 1913; S. posteli Delamare Deboutteville & Nunes-Ruivo, 1954; S. texanum Pearse,

1952; and *S. vespertilio* Kabata, 1979] are distinguished from the new species by having a pair of lateral processes or protrusions on the head (*vs* head without lateral processes or protrusions) (e.g. Krøyer, 1863; Wilson, 1913; Pearse, 1952; Delamare Deboutteville & Nunes-Ruivo, 1954; Yamaguti & Yamasu, 1960; Kabata, 1979a; Pillai, 1985). *Sagum foliaceum* (Goggio, 1905) differs from the new species in having the flat and foliaceous leg 4 (*vs* subcylindrical) (Hewitt, 1968). *Sagum petersi* (Beneden, 1857) is separated from the new species by having the endopod of leg 3 bifid with a tapering process (*vs* not bifid)



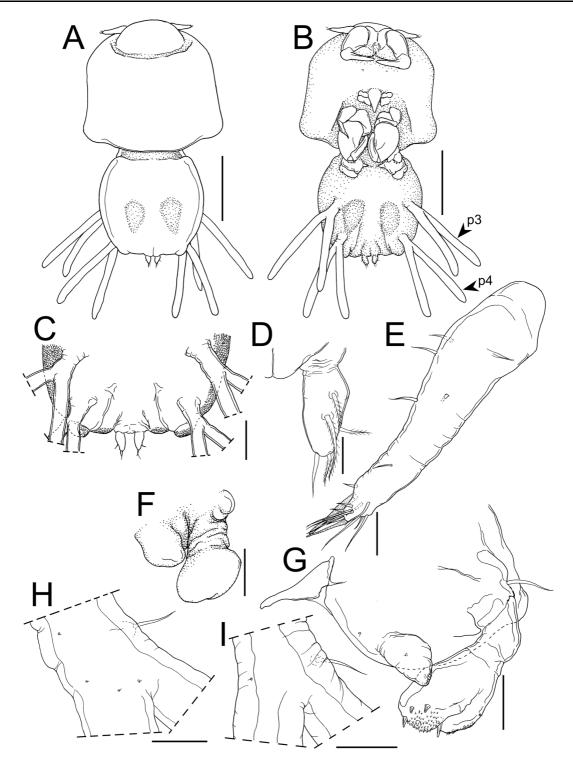


Fig. 4 *Sagum gurukun* n. sp., adult male, allotype (NSMT-Cr 25846). A, Habitus, dorsal view; B, Habitus, ventral view, C, posterior part of trunk, ventral. D, right caudal ramus, dorsal. E, left antennule, anterior. F, left postantennal process. G, left leg 2, anterior. H, basal part of left leg 3, anterior. I, basal part of left leg 4, anterior. *Abbreviations*: p3, leg 3; p4, leg 4. *Scale-bars*: A, B, 300 μm; C, 100 μm; D, E, 20 μm; F–I, 30 μm



(Kabata, 1979a). Sagum folium Ho, Liu & Lin, 2011 differs from S. gurukun n. sp. in having leg 5 (vs leg 5 absent) (Ho et al., 2011). Sagum vietnamiensis Kazachenko, Kovaleva, Nguyen & Ngo, 2017 is differentiated from the new species by having slender caudal rami five times longer than wide (vs less than 1.5 times) (Kazachenko et al., 2017). Although S. paracaesionis Izawa, 2014 shares many characters with the new species, the former is differentiated by having the following characters: caudal rami with two setae (vs five setae); leg 2 composed of a distinct intercoxal sclerite, basis and coxa (vs without distinct intercoxal sclerite, coxa and basis); and the endopods of legs 1 and 2 bearing an apical seta and a rugose distal expansion, respectively (vs without armature) (Izawa, 2014).

Sagum bitaro n. sp.

Type-host: Lutjanus quinquelineatus (Bloch) (Actinopterygii: Perciformes: Lutjanidae).

Type-locality: Off Nupan-zaki Cape (24°18′N, 123°39′E), Iriomote-jima Island, the Ryukyu Islands, East China Sea, Japan.

Type-material: Holotype (NSMT-Cr 25849): adult female, ex gill filaments of the type-host caught at a depth of 22 m on 10.ix.2017. Paratypes: 3 adult females (NSMT-Cr 25850), collection data as for the holotype.

Site in host: Female attaches on gill filaments.

ZooBank registration: To comply with the regulations set out in article 8.5 of the amended 2012 version of the *International Code of Zoological Nomenclature* (ICZN, 2012), details of the new species have been submitted to ZooBank. The Life Science Identifier (LSID) for *Sagum bitaro* n. sp. is urn:lsid: zoobank.org:act:779564F2-2842-422F-BB2E-BDD6 5B43F6EB.

Etymology: The specific name of the new species "bitaro" refers to one of the local names generally used for medium-sized snappers including the typehost, L. quinquelineatus, in Okinawa, Japan.

Japanese name for species: Bitaro-hitogatamushi.

Description (Figs. 5–6)

Adult female [Based on the holotype.] Body (Fig. 5A, B) 2,260 long, composed of cephalosome (head) and trunk composed of first pedigerous somite (short

neck), fused middle part (second to fourth pedigerous somites) and small urosome. Head wider than long, 751×869 , with posterolateral corners. Trunk bearing pair of conical, protruded shoulders and dorsal plate completely covering urosome and caudal rami. Urosome (Fig. 5C) fused into a single unit. Genital complex (Fig. 5C) wider than long, 179×265 . Abdomen (Fig. 5C) wider than long, 152×189 . Caudal rami (Fig. 5C) 2.12 times longer than wide, 200×94 , bearing 5 short setae.

Antennule (Fig. 5D) indistinctly 3-segmented, bearing 6 anterior and single posterior setae and 10 + 2 aesthetascs on distal part. Parabasal flagellum (Fig. 5E) composed of rod-like basal part and seta-like distal part. Antenna (Fig. 5F) 2-segmented, subchelate; corpus (coxa and basis) robust, bearing tiny element near base; subchela (endopod) incurved bearing small element. Postantennal process (Fig. 5G) knob-shaped with additional protrusion. Mandible (Fig. 5H) slender, rod-like with 8 teeth on distal tip. Maxillule (Fig. 5I) bilobate; larger outer lobe rodlike, bearing three apical elements; smaller inner lobe bearing single apical element. Maxilla (Fig. 5J) 2-segmented; lecertus (syncoxa) robust, unarmed; brachium bearing tiny element and terminal blade with serrated margin. Maxilliped (Fig. 5K) 2-segmented, subchelate; corpus bearing tiny basal element; subchela bearing tiny median seta, subterminal conical process and terminal claw with row of spinules.

Leg 1 (Fig. 6A) composed of intercoxal sclerite, coxa, basis and both rami; intercoxal sclerite rod-like, unornamented; coxa covered with numerous spinules; basis covered with numerous spinules, bearing naked outer seta and inner pinnate seta; exopod unsegmented, covered with numerous spinules on outer margin, bearing 5 blunt spines and row of spinules on distal margin; endopod represented by process with rounded distal margin. Leg 2 (Fig. 6B) lacking intercoxal sclerite; protopod fused to pedigerous somite, bearing simple outer seta on small protrusion; exopod represented by wrinkled process bearing 5 blunt, conical spines; endopod represented by wrinkled protrusion with blunt distal margin. Leg 3 (Fig. 5B) foliate, represented by both rami modified; exopod not reaching middle of dorsal plate. Leg 4 (Fig. 6C) bilobate, bearing hemispherical protrusion with simple outer seta on outer margin near base (Fig. 6D); protopod and subcylindrical rami fused.



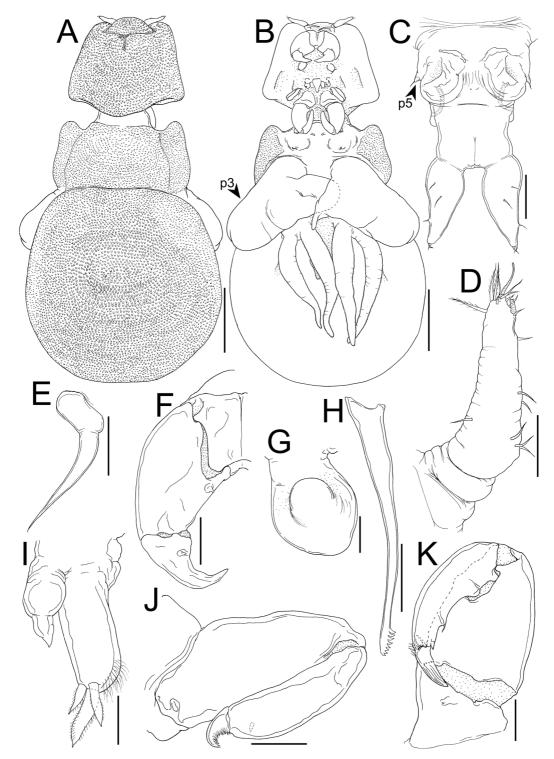


Fig. 5 Sagum bitaro n. sp., adult female, holotype (NSMT-Cr 25849). A, Habitus dorsal view; B, Habitus, ventral view; C, Urosome, dorsal view; D, Left antennule, anterior part; E, Left parabasal flagellum, anterior part; F, Right antenna, anterior part; G, Right postantennal process, anterior part; H, Right mandible, anterior part; I, Left maxillule, anterior part; J, Left maxilla, anterior part; K, Left maxilliped, posterior part. Abbreviations: p3, leg 3; p5, leg 5. Scale-bars: A, B, 400 μm; C, 100 μm; D, J, 40 μm; E, K, 50 μm; F, 70 μm; G, H, 20 μm; I, 30 μm



Leg 5 (Figs. 5C, 6E) represented by wrinkled conical process with apical seta.

bent. The measurement of the body parts (n = 3) is as follows: body length 2,492–2,715 (2,610); head length 692–823 (747); head width 560–569 (564).

Variability

The morphology of the female paratypes is as in the holotype, except with posterolateral corners ventrally

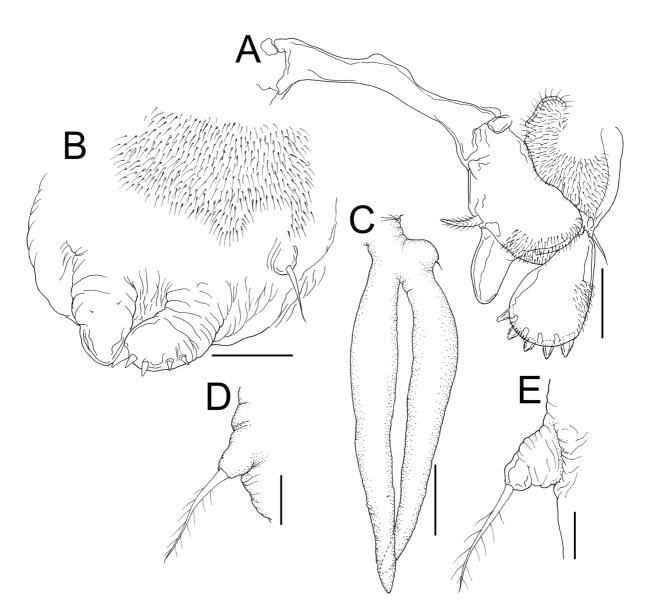


Fig. 6 *Sagum bitaro* n. sp., adult female, holotype (NSMT-Cr 25849). A, Left leg 1, anterior part; B, Left leg 2, anterior part; C, Left leg 4, anterior part; D, Seta near basal part of left leg 4, posterior part; E, Right leg 5, ventral view. *Scale-bars*: A, 30 μm; B, 40 μm; C, 200 μm; D, E, 10 μm



Remarks

Sagum bitaro n. sp. shares the following characters for the females with *S. folium*, *S. paracaesionis*, *S. vietnamiensis* and *S. gurukun* n. sp.: head without lateral processes or protrusions; endopod of leg 3 not bifid; leg 4 not foliaceous (see Ho et al., 2011; Izawa, 2014; Kazachenko et al., 2017; present study). Among these five species, only *S. bitaro* n. sp. and *S. folium* possess leg 5. That of the new species is represented by a conical process with an apical seta (*vs* an elongate rod with a median element in *S. folium*) (Ho et al., 2011).

Discussion

Species of Sagum have been found from gills of species in six families of perciform fishes: Caesionidae, Gempylidae, Labridae, Lethrinidae, Lutjanidae and Serranidae. Of 14 congeners considered valid, six species (S. angulatum, S. enneacentri, S. epinepheli, S. flagellatum, S. petersi and S. posteli) have been recorded from fishes in the Serranidae. Hence, the family is the most major host group (e.g. Wilson, 1913; Delamare Deboutteville & Nunes-Ruivo, 1954; Kabata, 1979b; Pillai, 1985; Ho et al., 2011). Sagum gurukun n. sp. found on the caesionid fish Pterocaesio digramma resembles in appearance S. folium, S. paracaesionis and S. bitaro n. sp., all found on lutjanid fishes [Paracaesio caerulea (Katayama), P. xanthura (Bleeker) and Lutjanus quinquelineatus (Bloch), respectively] (i.e. sharing following female characters: head without lateral processes or protrusions; leg 4 bearing subcylindrical rami). Further, these characters are shared with S. vietnamiensis found on an unidentified host (Kazachenko et al., 2017). Several species of the Caesionidae are often difficult to be distinguished from the Lutjanidae because of the morphological resemblance (e.g. Carpenter, 2001). Further, results of recently conducted molecular-based analysis strongly supported the fact that the Caesionidae is a sister group of the Lutjanidae (e.g. Tavera et al., 2012). Several results indicate that Caesionidae is a junior synonym of Lutjanidae because the clade of caesionids is nested within *Lutjanus* spp. (e.g. Miller & Cribb, 2007; Guo et al., 2016). Hence, sharing morphological characters in S. bitaro n. sp., S. folium, S. gurukun n. sp. and S. paracaesionis might reflect the fact that their hosts are closely related.

To date, four species of *Sagum* including two previously recorded species, *S. epinepheli* and *S. paracaesionis*, and a total of 19 species of Lernanthropidae considered valid are known in Japanese waters (Nagasawa & Uyeno, 2011; Izawa, 2014; present study). Below we provide a key to the species of the genus based on the available information from previous publications and the present study. As pointed out by Ho et al. (2011), morphological information on many species of *Sagum* are insufficient. Thus, further study to determine the validity of each species is needed as a priority.

Key to the species of *Sagum* based on female morphology

Head with pair of lateral processes or lobes; rami of leg 4 constricted with lamelliform or bulbous basal part and filiform distal part Head without distinct such structures; rami of leg 4 subcylindrical and without distinct constrictions 8 Head bearing pair of distinctly pointed lateral 2b Head bearing pair of lateral wing-like lobes 6 3a Lateral processes of head small and conical; leg 4 well developed, distinctly extending beyond posterior margin of dorsal plate S. enneacentri 3b Lateral processes of head well developed; leg 4 not reaching posterior margin of dorsal plate4 Lateral processes of head large, triangular, and basal width almost same as head length; short neck (first pedigerous somite) with pair of small lateral processes S. vespertilio Lateral processes of head beak-like, curved backwards; short neck without lateral processes 5 5a Leg 1 with large outer process on protopod S. angulatum 5b Leg 1 without large outer process on protopod S. epinepheli



6a	Pair of broad and long, wing-like lateral
	protrusions on trunk almost reached posterior
C 1.	tip of dorsal plate
6b	Pair of lateral protrusions on trunk almost
_	reaching middle of dorsal plate
7a	Head with pair of small anterior protrusions
	S. posteli
7b	Head without anterior protrusions
8a	Distal tip of leg 4 clearly extending beyond
	posterior margin of dorsal plate
8b	Distal tip of leg 4 not or rarely extending beyond
	posterior margin of dorsal plate
9a	Endopod of leg 3 bilobate, bearing elongate
	process-like inner lobe
9b	Endopod of leg 3 without such structures
	S. foliaceum
10a	Leg 5 present
10b	Leg 5 absent
11a	Leg 5 represented by bent blunt process with
	setulate papilla on median surface
	S. folium
11b	Leg 5 represented by wrinkled conical process
	with apical seta
12a	Caudal rami slender, five times longer than wide
12b	Caudal rami about two times or less than two
120	times longer than wide
13a	Leg 2 with distinct intercoxal sclerite, basis, and
134	coxa; endopods of legs 1 and 2 bearing apical
	seta and expansion, respectively
13b	Leg 2 without distinct intercoxal sclerite, basis,
130	
	and coxa; endopods of legs 1 and 2 unarmed
	S. gurukun n. sp.

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Compliance with ethical standards

Conflict of interest The authors declare that they have no conflict of interest.

Ethical approval All applicable institutional, national and international guidelines for the care and use of animals were followed.

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