

Caligus fajerae n. sp. (Copepoda: Caligidae) parasitic on the Pacific sierra *Scomberomurus sierra* Jordan & Starks (Actinopterygii: Scombridae) in the Pacific Ocean off Mexico

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Abstract A new species of parasitic copepod, *Caligus fajerae* n. sp. (Caligidae), is described from *Scomberomurus sierra* Jordan & Starks (Scombridae) caught off the northwestern coast of Mexico. The new species morphologically resembles *Caligus cybii* Bassett-Smith, 1898, *Caligus kanagurta* Pillai, 1961, *Caligus pelamydis* Krøyer, 1863 and *Caligus robustus* Bassett-Smith, 1898, all of which have been reported from scombrid hosts. *Caligus fajerae* n. sp. differs from these species by having spinules on the abdomen and caudal ramus, two processes on the proximal

antennular segment, fine striations on the claw of the antenna and maxilliped, a stouter and more recurved maxillulular dentiform process, shorter tines on the sternal furca, two additional patches of spinules on the distal endopodal segment of leg 2, a sclerotised lobe on the anteromedian surface of the leg 3 protopod and serrations on both margins of the first exopodal spine of leg 3. Analysis of the DNA sequences of the mitochondrial cytochrome *c* oxidase subunit 1 gene for *Caligus fajerae* n. sp. and 28 congeners, including *C. pelamydis* and *C. robustus*, showed that the new species grouped with *Caligus belones* Krøyer, 1863 (with 20% divergence), a species known to occur predominantly on needlefishes. *Caligus fajerae* n. sp. is the fifth species of *Caligus* reported from *S. sierra*. An updated host-parasite list for *Caligus* spp. on scombrids is provided.

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Introduction

Parasitic copepods of the family Caligidae Burmeister, 1835 are frequently found on marine and brackish water fishes. Caligids have been intensively studied given the negative impact that some species have on finfish aquaculture, mostly salmoniculture, in different parts of the world (Johnson et al., 2004; Costello, 2009). There are about 490 described species of caligids classified in 30 genera (Dojiri & Ho, 2013; Özak et al., 2017), of which *Caligus* Müller, 1785 is the most speciose genus with approximately 250 species. In their comprehensive studies of parasitic copepods on fishes of the family Scombridae (tunas and mackerels), an important group in commercial and sports fisheries (Collette 2001), Cressey & Cressey (1980) and Cressey et al. (1983) reported a total of 17 *Caligus* species from 45 host taxa (Table 1). Nine additional species have been recorded subsequently from various scombrids, bringing the total number of *Caligus* spp. from tunas and mackerels to 26 (Table 1). During a recent parasitological survey of the Pacific sierra *Scomberomorus sierra* Jordan & Starks (Scombridae) occurring off the north-western Pacific coast of Mexico, specimens of an undescribed species of *Caligus* were collected. This paper provides a detailed description of the new species based on adult females, as well as comparisons of the mitochondrial cytochrome *c* oxidase subunit 1 (*cox1*) gene sequence of the new species with that of selected congeners.

Materials and methods

A total of 109 Pacific sierra, caught and landed at Mazatlan Port on the north-western coast of Mexico, were purchased and then transferred in an icebox to the Aquatic Parasitology Laboratory at the Centro de Investigación en Alimentación y Desarrollo, Unidad Mazatlán (CIAD-Mazatlán), Sinaloa, Mexico, for parasitological examination. Copepod specimens were all collected from the hosts' body surface and were fixed and preserved in 96% ethanol. Selected specimens were later cleared in lactophenol for about 1 h before dissection of the appendages on a slide under an Olympus SZ61 dissection microscope. The body parts and appendages were mounted on slides in lactophenol and examined under a Leica DMLB

compound microscope with a series of magnifications up to 1000×. All drawings were made with the aid of a drawing tube attached to the compound microscope. Measurements were made using an ocular micrometer, and are given in millimeters as the range followed by the mean in parentheses. The type-material was deposited in the Colección de Parásitos de Peces del Noroeste del Pacífico (CPPNP) at CIAD- Mazatlán, Sinaloa, Mexico. Fish taxonomy and classification used herein follow FishBase (Froese & Pauly, 2017).

Two copepods were fixed in 96% ethanol and kept at 4°C until DNA extraction. Genomic DNA was extracted with the Animal and Fungi Preparation kit (Jena Bioscience, Jena, Germany). Primers used for the amplification reaction of the barcode region of the *cox1* gene through the Polymerase Chain Reaction (PCR) were LCO1490 (5'-GGT CAA CAA ATC ATA AAG ATA TTG G-3') and HCO2198 (5'-TAA ACT TCA GGG TGA CCA AAA AAT CA-3') (Folmer et al., 1994). Amplification reactions contained 0.12 µl of Biolase Taq Polymerase (Biolone, London, UK), 3 µl of 5× My Taq reaction buffer, 0.25 µl of each primer, 2 µl of template DNA and 9.38 µl of water to reach 15 µl. Reactions were accomplished with an Arktik™ Thermal Cycler (Thermo Fisher Scientific, Waltham, USA) using an amplification protocol of 94°C for 4 min, followed by 35 cycles of 94°C for 40 s, 48°C for 45 s, 72°C for 45 s and a final extension at 72°C for 7 min. Amplification reactions were conducted at the Laboratorio Nacional de Biodiversidad, Instituto de Biología, Universidad Nacional Autónoma de México. DNA sequences of complementary strands were edited and assembled using the software Geneious 5.1.7 (Biomatters Ltd. Auckland, New Zealand). Newly generated sequences were aligned together with *cox1* sequences for 28 species of the genus *Caligus* and *Lepeophtheirus salmonis* (Krøyer, 1837) (Caligidae) used as the outgroup (Table 2). Sequence alignment was performed in MUSCLE through the European Bioinformatics Institute webpage (<http://www.ebi.ac.uk/Tools/msa/muscle/>). The final data matrix included 36 terminals and 495 aligned nucleotides. A neighbor joining (NJ) analysis was performed in PAUP* 4.0 using the Kimura 2-parameter algorithm. Finally, *cox1* sequences were used to calculate genetic distances between the sequences with the Kimura 2-parameter algorithm in PAUP* 4.0. GenBank accessions of *cox1* sequences generated for the new species are given in Table 2.

Table 1 Species of *Caligus* reported from scombrid hosts

Species	Host	Reference
<i>Caligus amblygenitalis</i> Pillai, 1961	<i>Acanthocybium solandri</i> (Cuvier)	Prabha & Pillai (1986)
	<i>Euthynnus affinis</i> (Cantor)	Cressey & Cressey (1980)
	<i>Thunnus maccoyii</i> (Castelnau)	Hayward et al. (2009)
<i>Caligus asymmetricus</i> Kabata, 1965	<i>Auxis rochei</i> (Risso)	Ho & Lin (2004)
	<i>Auxis thazard</i> (Lacépède)	Venmathi Maran et al. (2016)
	<i>Cybiosarda elegans</i> (Whitley); <i>Euthynnus affinis</i> (Cantor); <i>Grammatorcynus bicarinatus</i> (Quoy & Gaimard); <i>Katsuwonus pelamis</i> (Linnaeus); <i>Sarda australis</i> (Macleay); <i>Sarda orientalis</i> (Temminck & Schlegel); <i>Scomberomorus commerson</i> (Lacépède); <i>Thunnus albacares</i> (Bonnaterre)	Cressey & Cressey (1980)
	<i>Grammatorcynus bilineatus</i> (Rüppell)	Pillai (1985)
<i>Caligus biseriodentatus</i> Shen, 1957	<i>Scomberomorus plurilineatus</i> Fourmanoir	Cressey et al. (1983)
	<i>Auxis thazard</i> (Lacépède); <i>Scomberomorus commerson</i> (Lacépède); <i>Scomberomorus guttatus</i> (Bloch & Schneider); <i>Scomberomorus lineolatus</i> (Cuvier); <i>Scomberomorus queenlandicus</i> Munro	Cressey & Cressey (1980)
	<i>Scomberomorus munroi</i> Collette & Russo; <i>Scomberomorus multiradiatus</i> Munro; <i>Scomberomorus semifasciatus</i> (Macleay)	Cressey et al. (1983)
<i>Caligus bonito</i> Wilson, 1905	<i>Allothunnus fallai</i> Serventy; <i>Cybiosarda elegans</i> (Whitley); <i>Grammatorcynus bicarinatus</i> (Quoy & Gaimard); <i>Katsuwonus pelamis</i> (Linnaeus); <i>Orcynopsis unicolor</i> (Geoffroy Saint-Hilaire)	Cressey et al. (1983)
	<i>Euthynnus affinis</i> (Cantor); <i>Euthynnus alleteratus</i> (Rafinesque); <i>Euthynnus lineatus</i> Kishinouye; <i>Gymnosarda unicolor</i> (Rüppell); <i>Sarda australis</i> (Macleay); <i>Sarda chiliensis</i> (Cuvier) (as <i>Sarda chiliensis chiliensis</i> and <i>S. chiliensis lineolatus</i>); <i>Sarda orientalis</i> (Temminck & Schlegel); <i>Sarda sarda</i> (Bloch); <i>Scomberomorus regalis</i> (Bloch); <i>Thunnus thynnus</i> (Linnaeus)	Cressey & Cressey (1980)
	<i>Scomberomorus maculatus</i> (Mitchill)	Pillai (1985)
<i>Caligus calotomi</i> Shiino, 1954	<i>Sarda orientalis</i> (Temminck & Schlegel)	Lin & Ho (2007)
<i>Caligus chistos</i> Lin & Ho, 2003	<i>Thunnus maccoyii</i> (Castelnau)	Hayward et al. (2008)
<i>Caligus coryphaenae</i> Steenstrup & Lütken, 1861	<i>Acanthocybium solandri</i> (Cuvier); <i>Auxis</i> sp.; <i>Euthynnus alleteratus</i> (Rafinesque); <i>Katsuwonus pelamis</i> (Linnaeus); <i>Thunnus alalunga</i> (Bonnaterre); <i>Thunnus albacares</i> (Bonnaterre); <i>Thunnus atlanticus</i> (Lesson); <i>Thunnus obesus</i> (Lowe); <i>Thunnus thynnus</i> (Linnaeus)	Cressey & Cressey (1980)
	<i>Auxis thazard</i> (Lacépède)	Venmathi Maran et al. (2016)
	<i>Euthynnus affinis</i> (Cantor); <i>Grammatorcynus bilineatus</i> (Rüppell)	Pillai (1985)
	<i>Sarda orientalis</i> (Temminck & Schlegel)	Cressey et al. (1983)

Table 1 continued

Species	Host	Reference
<i>Caligus cybii</i> Bassett-Smith, 1898	<i>Scomberomorus commerson</i> (Lacépède); <i>Scomberomorus koreanus</i> (Kishinouye); <i>Scomberomorus semifasciatus</i> (Macleay); <i>Scomberomorus sinensis</i> (Lacépède)	Cressey & Cressey (1980)
	<i>Scomberomorus guttatus</i> (Bloch & Schneider); <i>Scomberomorus lineolatus</i> (Cuvier)	Pillai (1985)
	<i>Scomberomorus plurilineatus</i> Fourmanoir	van der Elst & Collette (1984)
	<i>Scomberomorus queenslandicus</i> Munro	Cressey et al. (1983)
<i>Caligus diaphanus</i> von Nordmann, 1832	<i>Scomberomorus tritor</i> (Cuvier)	Cressey & Cressey (1980)
<i>Caligus elongatus</i> von Nordmann, 1832	<i>Scomber scombrus</i> Linnaeus	Williams & Williams (1996)
<i>Caligus epinepheli</i> Yamaguti, 1936	<i>Scomberoides commersonianus</i> Lacépède	Venmathi Maran et al. (2016)
<i>Caligus infestans</i> Heller, 1868	<i>Euthynnus affinis</i> (Cantor); <i>Scomberomorus maculatus</i> (Mitchill)	Pillai (1985)
	<i>Scomberomorus commerson</i> (Lacépède)	Cressey & Cressey (1980)
<i>Caligus kanagurta</i> Pillai, 1961	<i>Rastrelliger kanagurta</i> (Cuvier); <i>Sarda orientalis</i> (Temminck & Schlegel); <i>Thunnus tonggol</i> (Bleeker, 1851)	Cressey et al. (1983)
	<i>Scomber australasicus</i> Cuvier	Ho & Lin (2004)
	<i>Scomber japonicas</i> (Houttuyn)	Kim (1998) ^a
<i>Caligus macarovi</i> Gusev, 1951	<i>Auxis</i> sp.	Cressey & Cressey (1980)
	<i>Thunnus orientalis</i> (Temminck & Schlegel)	Nagasawa (2011)
<i>Caligus mutabilis</i> Wilson, 1905	<i>Katsuwonus pelamis</i> (Linnaeus); <i>Sarda sarda</i> (Bloch)	Williams & Williams (1996)
	<i>Scomber japonicas</i> (Houttuyn); <i>Scomberomorus brasiliensis</i> Collette, Russo & Zavala-Camin; <i>Scomberomorus cavalla</i> (Cuvier); <i>Scomberomorus maculatus</i> (Mitchill)	Cressey & Cressey (1980)
	<i>Scomberomorus sierra</i> Jordan & Starks	Causey (1960) ^b
<i>Caligus omissus</i> Cressey & Cressey, 1980	<i>Scomberomorus concolor</i> (Lockington); <i>Scomberomorus sierra</i> Jordan & Starks	Cressey & Cressey (1980)
<i>Caligus pelamydis</i> Krøyer, 1863	<i>Auxis</i> sp.; <i>Euthynnus affinis</i> (Cantor); <i>Euthynnus alleteratus</i> (Rafinesque); <i>Sarda australis</i> (Macleay); <i>Sarda chiliensis</i> (Cuvier) (as <i>Sarda chiliensis lineolatus</i>); <i>Sarda sarda</i> (Bloch); <i>Scomber japonicas</i> (Houttuyn); <i>Scomber scombrus</i> Linnaeus; <i>Scomberomorus nipponius</i> (Cuvier)	Cressey & Cressey (1980)
	<i>Grammatorcynus bicarinatus</i> (Quoy & Gaimard); <i>Scomberomorus sinensis</i> (Lacépède)	Cressey et al. (1983)
<i>Caligus phipsoni</i> Bassett-Smith, 1898	<i>Scomberomorus guttatus</i> (Bloch & Schneider)	Pillai (1985)

Table 1 continued

Species	Host	Reference
<i>Caligus productus</i> Dana, 1852	<i>Acanthocybium solandri</i> (Cuvier); <i>Euthynnus affinis</i> (Cantor); <i>Euthynnus alleteratus</i> (Rafinesque); <i>Gymnosarda unicolor</i> (Rüppell); <i>Katsuwonus pelamis</i> (Linnaeus); <i>Sarda chiliensis</i> (Cuvier) (as <i>Sarda chiliensis lineolatus</i>); <i>Sarda orientalis</i> (Temminck & Schlegel); <i>Sarda sarda</i> (Bloch); <i>Scomberomorus cavalla</i> (Cuvier); <i>Scomberomorus regalis</i> (Bloch); <i>Scomberomorus tritor</i> (Cuvier); <i>Thunnus alalunga</i> (Bonnaterre); <i>Thunnus albacares</i> (Bonnaterre); <i>Thunnus atlanticus</i> (Lesson); <i>Thunnus thynnus</i> (Linnaeus)	Cressey & Cressey (1980)
	<i>Allothynnus fallai</i> Serventy; <i>Grammatocynus bicarinatus</i> (Quoy & Gaimard); <i>Scomberomorus munroi</i> Collette & Russo; <i>Thunnus maccoyii</i> (Castelnau)	Cressey et al. (1983)
	<i>Auxis thazard</i> (Lacépède); <i>Euthynnus lineatus</i> Kishinouye; <i>Scomberomorus maculatus</i> (Mitchill)	Pillai (1985)
	<i>Scomberomorus sierra</i> Jordan & Starks	Causey (1960) ^b
	<i>Thunnus obesus</i> (Lowe)	Ho & Lin (2004)
<i>Caligus pseudokalumai</i> Lewis, 1968	<i>Gymnosarda unicolor</i> (Rüppell)	Cressey & Cressey (1980)
<i>Caligus quadratus</i> Shiino, 1954	<i>Thunnus albacares</i> (Bonnaterre) [as <i>Germo albacora</i> (Lowe) and <i>Neothynnus macropterus</i> (Temminck & Schlegel)]	Pillai (1985)
<i>Caligus regalis</i> Leigh-Sharpe, 1930	<i>Euthynnus affinis</i> (Cantor)	Cressey & Cressey (1980)
	<i>Grammatocynus bicarinatus</i> (Quoy & Gaimard)	Cressey et al. (1983)
	<i>Katsuwonus pelamis</i> (Linnaeus)	Pillai (1985) ^c
<i>Caligus robustus</i> Bassett-Smith, 1898	<i>Thunnus albacares</i> (Bonnaterre) [as <i>Neothynnus macropterus</i> (Temminck & Schlegel)]	Pillai (1985)
<i>Caligus savala</i> Gnanamuthu, 1948	<i>Euthynnus affinis</i> (Cantor)	Cressey & Cressey (1980)
<i>Caligus serratus</i> Shiino, 1965	<i>Scomberomorus sierra</i> Jordan & Starks	Morales-Serna et al. (2013)
<i>Caligus tylosuri</i> (Rangnekar, 1956)	<i>Auxis rochei</i> (Risso); <i>Euthynnus affinis</i> (Cantor)	Lin & Ho (2007)

^aReported as *Caligus pelamydis*; ^b Roger Cressey planned to reexamine this material [see Cressey & Nutter (1987)], but did not manage to do this before his untimely death in 2001; ^c Reported as *Caligus euthynnus* Kurian, 1961

Order Siphonostomatoida Thorell, 1859

Family Caligidae Burmeister, 1835

Genus *Caligus* Müller, 1785

Caligus fajerae n. sp.

Type-host: *Scomberomorus sierra* Jordan & Starks (Scombridae), Pacific sierra.

Type-locality: off Mazatlan Port (23°12'N, 106°26'W), Mexican Pacific, in the State of Sinaloa, Mexico.

Type-material: Holotype female (CPPNP 1369) and 6 paratype females (CPPNP 1370–1372).

Site on host: Upper body surface.

Prevalence: 43%.

Representative DNA sequences: MF069191 and MF069192

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 *Caligus fajerae* n. sp. is urn:lsid:zoobank.org:act:C1D8F27D-9C11-43A5-9286-310F16515C21.

Etymology: The species is named in honor of Dr Emma Josefina Fajer Avila (CIAD, Mazatlán, Sinaloa, Mexico) for her work on fish parasitology.

Table 2 GenBank accession numbers for *cox1* sequences of caligid species used in the molecular analysis

Species	Host	Locality	GenBank ID	Reference
<i>Caligus aesopus</i> Wilson, 1921	<i>Caranx caballus</i> Günther	Pacific: off Mexico	KF483712	Morales-Serna et al. (2014)
<i>Caligus belones</i> Krøyer, 1863	Unspecified ^a	Atlantic: off Norway	AY861368	Øines & Heuch (2005)
<i>Caligus brevipedis</i> Bassett-Smith, 1896	na	na	KC345610	na
<i>Caligus callaoensis</i> Duran, 1980	<i>Cynoscion xanthulus</i> Jordan & Gilbert	Pacific: off Mexico	KF483717	Morales-Serna et al. (2014)
<i>Caligus centrodonti</i> Baird, 1850	Unspecified	Atlantic: off Norway	AY861370	Øines & Heuch (2005)
<i>Caligus chamelensis</i> Morales-Serna, Pinacho-Pinacho, Gómez & Pérez-Ponce de León, 2014	<i>Kyphosus elegans</i> (Peters)	Pacific: off Mexico	KF483680	Morales-Serna et al. (2014)
<i>Caligus cheilodactyli</i> Krøyer, 1863	<i>Sebastes oculatus</i> Valenciennes	Pacific: off Chile	KU317606	González et al. (2016)
<i>Caligus chorinemi</i> Krøyer, 1863	<i>Caranx caninus</i> Günther	Pacific: off Mexico	KF483710	Morales-Serna et al. (2014)
<i>Caligus clemensi</i> Parker & Margolis, 1964	<i>Oncorhynchus keta</i> (Walbaum)	Pacific: off Canada	AM235887	McBeath et al. (2006)
<i>Caligus confusus</i> Pillai, 1961	<i>Caranx caballus</i> Günther	Pacific: off Mexico	KF483699	Morales-Serna et al. (2014)
<i>Caligus curtus</i> Müller, 1785	Unspecified	Atlantic: off Norway	AY861366	Øines & Heuch (2005)
<i>Caligus diaphanus</i> von Nordmann, 1832	Unspecified	Atlantic: off Norway	EF065616	Øines & Schram (2008)
<i>Caligus elongatus</i> types 1 and 2	<i>Salmo salar</i> Linnaeus, <i>S. trutta</i> Linnaeus, <i>Pollachius pollachius</i> (Linnaeus), <i>P. virens</i> (Linnaeus), <i>Clupea harengus</i> Linnaeus	Atlantic: off Norway	AY386273, AY861365	Øines & Heuch (2005)
<i>Caligus fajerae</i> n. sp.	<i>Scomberomorus sierra</i> Jordan & Starks	Pacific: off Mexico	MF069191-MF069192	This study
<i>Caligus fugu</i> (Yamaguti, 1936)	<i>Takifugu rubripes</i> (Temminck & Schlegel)	Pacific: off Japan	KC569368	Freeman et al. (2013)
<i>Caligus gurnardi</i> Krøyer, 1863	Unspecified ^b	Atlantic: off Norway	AY861369	Øines & Heuch (2005)
<i>Caligus hoplognathi</i> Yamaguti & Yamasu, 1959	na	na	KR049058	na
	<i>Caranx caballus</i> Günther	Pacific: off Mexico	KF483707	Morales-Serna et al. (2014)
<i>Caligus longirostris</i> Heegaard, 1962	<i>Salmo salar</i> Linnaeus	Pacific: off Tasmania	HQ667343	Nowak et al. (2011)
<i>Caligus mutabilis</i> Wilson, 1905	<i>Epinephelus labriformis</i> (Jenyns)	Pacific: off Mexico	KF483686, KF483688, KF483689	Morales-Serna et al. (2014)
<i>Caligus nuenonae</i> Andrews, Bott, Battaglene & Nowak, 2009	<i>Latris lineata</i> (Forster)	Pacific: off Tasmania	EF452642	Andrews et al. (2009)
<i>Caligus pelamydis</i> Krøyer, 1863	Unspecified ^c	Atlantic: off Norway	AY861367	Øines & Heuch (2005)
<i>Caligus punctatus</i> Shiino, 1965	na	na	KR049057	na

Table 2 continued

Species	Host	Locality	GenBank ID	Reference
<i>Caligus quadratus</i> Shiino, 1954	Unspecified	Atlantic: off Norway	EF065619	Øines & Schram (2008)
	na	na	KR049059	na
<i>Caligus robustus</i> Bassett-Smith, 1898	<i>Caranx caballus</i> Günther	Pacific: off Mexico	KF483701	Morales-Serna et al. (2014)
<i>Caligus rogercresseyi</i> Boxshall & Bravo, 2000	<i>Salmo salar</i> Linnaeus	Pacific: off Chile	HQ667349	Nowak et al. (2011)
<i>Caligus sclerotinosus</i> Roubal, Armitage & Rohde, 1983	<i>Lutjanus colorado</i> Jordan & Gilbert	Pacific: off Mexico	KF483709	Morales-Serna et al. (2014)
<i>Caligus serratus</i> Shiino, 1965	<i>Calamus brachysomus</i> (Lockington)	Pacific: off Mexico	KF483694	Morales-Serna et al. (2014)
<i>Caligus tylosuri</i> (Rangnekar, 1956)	<i>Tylosurus pacificus</i> (Steindachner)	Pacific: off Mexico	KF483682	Morales-Serna et al. (2014)
<i>Caligus uniartus</i> (Ho, Kim, Cruz & Nagasawa, 2004)	<i>Siganus guttatus</i> (Bloch)	Indo-Pacific: off South Sulawesi	KC569367	Freeman et al. (2013)
<i>Lepeophtheirus salmonis</i> (Krøyer, 1837)	Unspecified	Atlantic: off Norway	AY861361	Øines & Heuch (2005)

Abbreviation: na, not available

^aAlthough the host was not specified by Øines & Heuch (2005), *C. belones* is known to occur predominantly on needlefishes (Belontiidae) (see Cressey & Collette, 1970); ^b Although the host was not specified by Øines & Heuch (2005), *C. gurnardi* is known to occur exclusively on searobins (Triglidae) (see Parker, 1965); ^c Although the host was not specified by Øines & Heuch (2005), *C. pelamydis* is known to occur predominantly on scombrids (see Cressey, 1991)

Description (Figs. 1–2)

Adult female. [Based on 10 specimens.] Body (Fig. 1A) 4.6–5.2 (4.9) long, excluding setae on caudal ramus. Cephalothoracic shield longer than wide, 1.9–2.4 × 1.5–1.6 (2.1 × 1.5), excluding marginal hyaline membranes. Fourth pediger 2.4 times wider than long. Genital complex bell-shaped, 1.25 times longer than wide. Abdomen composed of 2 indistinctly separated somites, 1–1.3 (1.2) long in total, and 0.6 times as long as cephalothorax; proximal somite nearly 2 times wider and 4 times longer than distal (anal) somite; latter with patch of tiny spinules near posteroventral margin (Fig. 1B). Caudal ramus (Fig. 1B) 1.5 times longer than wide, with patch of tiny spinules on proximolateral corner, setules on median edge, and 3 short and 3 long, plumose setae.

Antennule (Fig. 1C) 2-segmented; proximal segment larger than distal segment, with proximal conical process, bifid process on posterodistal corner, and 23 plumose and 4 naked setae; distal segment with 1 subterminal seta on posterior margin and 11 setae plus 2 aesthetascs on distal margin. Antenna (Fig. 1D) 3-segmented, situated on pedestal; first segment

(coxa) unarmed; second segment (basis) stout, subquadrate; terminal segment (endopod) a curved claw bearing 1 small, naked seta in proximal region and another one in middle region plus fine striations at tip. Postantennal process (Fig. 1D) small, subtriangular, with 2 papillae each bearing 3 sensilla; another similar sensilla-bearing papilla located posterior to tip of process. Mandible (Fig. 1E) composed of 4 sections, with 12 teeth on medial margin of distal blade and hyaline membrane on distal half of outer margin. Maxillule (Fig. 1F) comprising basal papilla with 3 unequal, naked setae and short, recurved dentiform process with tapered tip. Maxilla (Fig. 1G) 2-segmented, brachiform; proximal segment (lacertus) large, unarmed; distal segment (brachium) slender, carrying small subterminal hyaline membrane (flabellum) on inner edge, and short, finely serrated canna and long, finely serrated calamus distally. Maxilliped (Fig. 1H) subchelate, 3-segmented; proximal segment (corpus) robust, with tiny hyaline process on inner subdistal margin; middle segment (shaft) longer than distal segment (claw), with tiny hyaline process midway on posterior surface; claw with small, naked basal seta and fine striations at tip. Sternal furca

(Fig. 1I) with subquadrate box and short, pointed tines.

Legs 1 to 3 (Fig. 2A–C) biramous; leg 4 (Fig. 2D) uniramous. Armature on rami of legs 1–4 as follows (Roman and Arabic numerals indicating spines and setae, respectively):

	Exopod	Endopod
Leg 1	I-0; III+1, 3	vestigial
Leg 2	I-1; I-1; II, I, 5	0-1; 0-2; 6
Leg 3	I-0; I-1; II, I, 4	0-0; 0-1; 6
Leg 4	I-0; I-0; III	absent

Leg 1 (Fig. 2A) intercoxal sclerite naked and elongate. Protopod with 1 long, outer plumose seta, 1 small, inner plumose seta and 1 outer papilla bearing 2 setules. Exopod 2-segmented; first segment elongate, with inner row of setules and small, outer distal spine furnished with tiny membrane at its base; second segment carrying 3 unequal apical spines each with inner row of teeth and pectinate membrane at base, 1 naked apical seta (as long as third spine) and 3 large, inner plumose setae. Endopod represented by small lobe tipped with 2 tiny processes.

Leg 2 (Fig. 2B) intercoxal sclerite subquadrate, with large hyaline membrane along distal margin. Coxa with large, inner plumose seta and 1 long setule on anterior surface. Basis with small, outer naked seta, 1 inner setule and hyaline membrane on outer posterior surface and along inner edge. Exopod 3-segmented; first segment with 1 large, outer serrated spine, 1 inner plumose seta, inner row of setules, pectinate membrane at base of outer spine and hyaline membrane on outer posterior surface; second segment with 1 large, serrated outer spine, 1 pore on anterior surface, inner row of setules and 1 inner plumose seta; third segment with 2 small outer spines (proximal spine with row of tiny teeth; distal spine with row of tiny teeth and flange along both margins), 1 long apical spine furnished with membrane along outer margin and setules along inner margin and 5 inner plumose setae. Endopod 3-segmented; first segment with long inner plumose seta; second segment with numerous fine spinules on outer surface, inner row of setules and 2 long, inner plumose setae; third segment with outer patch of fine spinules, 2 patches of short denticles on

anterior surface, short inner row of setules and 6 plumose setae.

Leg 3 (Fig. 2C) protopod (apron) with 1 small, outer plumose seta, 1 long, inner plumose seta, 1 anteromedian sclerotised lobe, corrugated surface along outer proximal margin, 1 longitudinal and 1 patch of spinules near outer margin, 2 posteromedian setules and hyaline membranes on outer and posterior margins. Exopod 3-segmented; first segment with several sensilla and pectinate membrane at base of long (almost as long as second segment) serrated spine; second segment with outer row of setules, 1 short outer spine and 1 long, plumose inner seta; terminal segment with outer row of setules, 3 short outer spines and 4 plumose setae. Endopod 3-segmented; first segment forming broad, well developed velum fringed with setules along posterior margin; second segment with long, inner plumose seta; third segment with outer row of setules and 6 plumose setae.

Leg 4 (Fig. 2D) protopod large, with small plumose seta at outer distal corner. Exopod 3-segmented; first segment with sensillum on mid-lateral margin and conspicuous pectinate membrane at base of long, serrated outer spine; second segment ornamented with conspicuous pectinate membrane along posterior margin and armed with long, serrated outer spine; distal segment with 3 serrated spines (innermost spine longer than both middle and outermost spines) and pectinate membrane at base of each spine.

Leg 5 (Fig. 2E) represented by unisetose papilla and trisetose lobe on posterolateral margin of genital complex.

Molecular results

The *cox1* sequences successfully obtained from the two specimens of *Caligus fajerae* n. sp. were identical and grouped with *Caligus belones* Krøyer, 1863 based on the NJ analysis (Fig. 3). Nonetheless, the genetic divergence between the new species and *C. belones* is 20.69%. *Caligus robustus* and *C. pelamydis* are morphologically similar to *C. fajerae* n. sp. (see below), but they did not group with the new taxon and genetically differ from it in 19.43 and 21.19%, respectively. In general, the genetic divergence between *Caligus fajerae* n. sp. and the 26 congeners included in the alignment ranged between 22–24%.

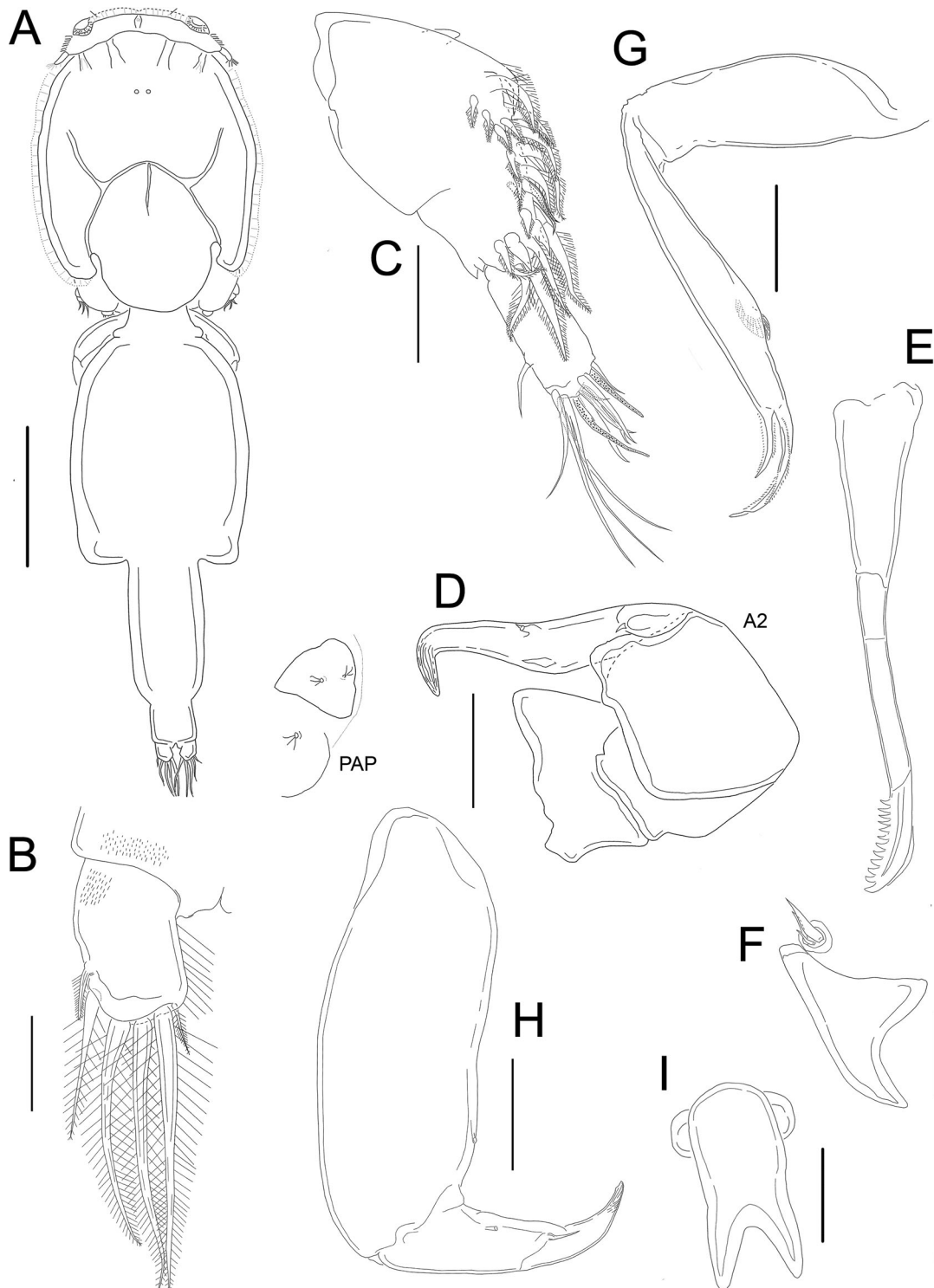


Fig. 1 *Caligus fajerae* n. sp., adult female. A, Habitus, dorsal view; B, Right caudal ramus, ventral view; C, Left antennule, ventral view; D, Right antenna (A2) and postantennal process (PAP), ventral view; E, Mandible; F, Left maxillule, ventral view; G, Right maxilla, anterior view; H, Left maxilliped, posterior view; I, Sternal furca, ventral view. Scale-bars: A, 1 mm; B–I, 0.1 mm

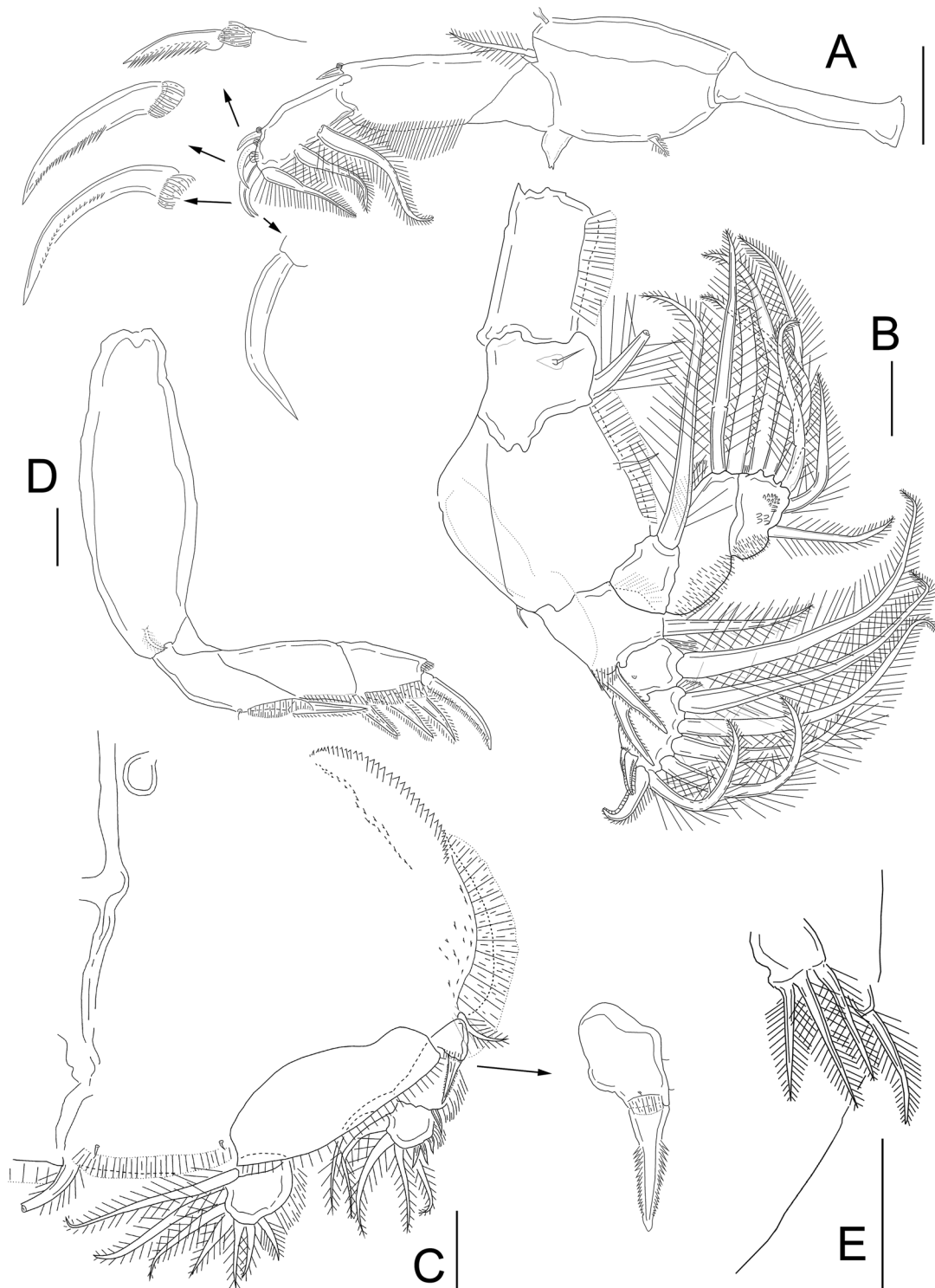


Fig. 2 *Caligus fajerae* n. sp., adult female. A, Right leg 1 with detail of apical elements on second exopodal segment, anterior view; B, Right leg 2, anterior view; C, Left leg 3 with detail of first exopodal segment, ventral view; D, Right leg 4, ventral view; E, Left leg 5, ventral view. Scale-bars: A–D, 0.1 mm; E, 0.05 mm

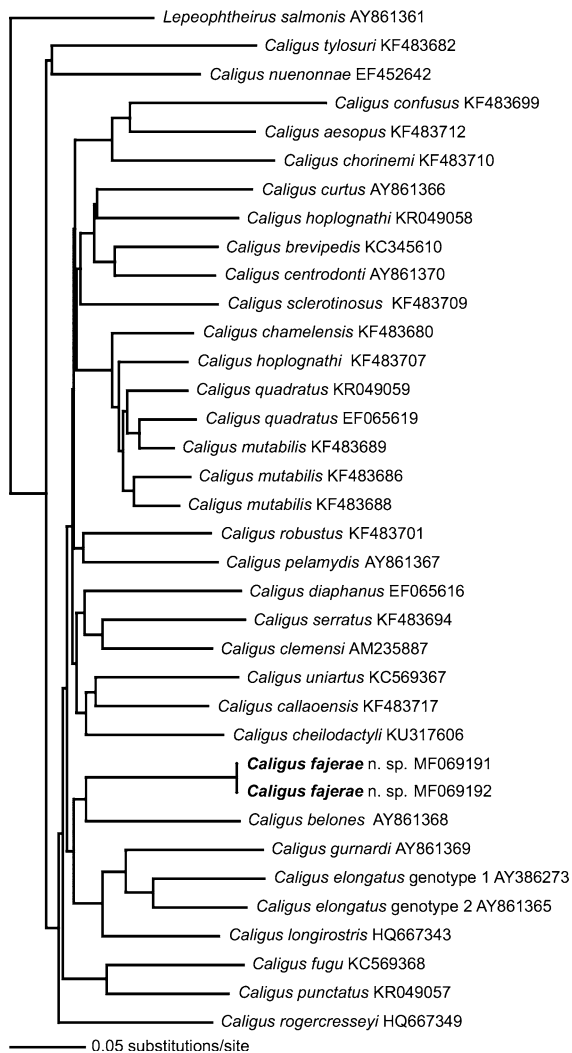


Fig. 3 Neighbor-joining phylogram showing relatedness among *cox1* gene sequences for species of *Caligus* and *Lepeophtheirus* (see Table 2 for details)

Discussion

Among the more than 250 species of *Caligus* considered valid, *Caligus fajerae* n. sp. resembles *Caligus cybii* Bassett-Smith, 1898, *Caligus kanagurta* Pillai, 1961, *Caligus pelamydis* Krøyer, 1863 and *Caligus robustus* Bassett-Smith, 1898 by having: (i) a long, indistinctly 2-segmented abdomen; (ii) no accessory process on the proximal segment of the antenna; (iii) a postantennal process composed of a broad base and short tip; (iv) no accessory process on the three apical spines of leg 1; (v) spinules on the last two endopodal segments of leg 2 and on the ventral surface of the leg

3 protopod; and (vi) a 3-segmented leg 4 exopod with an armature of I-0; I-0; III and furnished with a conspicuous pectinate membrane at the base of each exopodal spine. With regard to the aforementioned fourth character, we note here that Ho & Lin (2007) described two of the three apical spines of leg 1 as being bifid in their single adult female specimen of *C. robustus* collected from the bigeye trevally *Caranx sexfasciatus* Quoy & Gaimard captured off Taiwan. Whether this feature represents intra- or interspecific variation remains to be determined. *Caligus fajerae* n. sp. can be readily distinguished from *C. cybii*, *C. kanagurta*, *C. pelamydis* and *C. robustus* by the presence of a cluster of spinules on the posteroventral surface of the abdomen and on the anteroventral surface of the caudal ramus, a proximal conical process and posterodistal bifid process on the proximal antennular segment, fine striations on the tip of the antennal and maxillipedal claw, a stouter and more recurved maxillary dentiform process, shorter tines on the sternal furca, two additional patches of spinules on the distal endopodal segment of leg 2, a sclerotised lobe on the anteromedian surface of the leg 3 protopod and serrations on both margins of the first exopodal spine of leg 3.

Our molecular analysis revealed that *C. fajerae* n. sp. grouped with *C. belones*, and then with a group formed by *C. gurnardi* Krøyer, 1863, *C. elongatus* von Nordmann, 1832 and *C. longirostris* Heegaard, 1962. This grouping was unexpected because the latter four species were collected from non-scombrid hosts from either off Norway or Tasmania (Table 2), and possess a relatively short unsegmented abdomen and a 2-segmented exopod on leg 4 unlike *C. fajerae* n. sp. In contrast, *C. pelamydis* and *C. robustus*, two species morphologically similar to *C. fajerae* n. sp. and reported from scombrid hosts as noted above, grouped together separately from *C. fajerae* n. sp. based on the NJ analysis. Unfortunately, no *cox1* sequences of *C. cybii* and *C. kanagurta* are yet available in public repositories, making the evaluation of their shared morphological characters with the new species (e.g. small postantennal process, indistinctly 2-segmented abdomen and 3-segmented exopod on leg 4) not possible at this time. As *cox1* sequences have been obtained for a small fraction of *Caligus* spp., increased taxon sampling is needed to improve our understanding of the phylogenetic relationships within *Caligus*.

Four species of *Caligus* have been reported previously from *Scomberomorus sierra*: *C. omissus* Cressey & Cressey, 1980 from off Mexico, Panama, Colombia, Ecuador and Peru (Cressey & Cressey 1980; Morales-Serna et al., 2012, 2015) and *C. mutabilis* Wilson, 1905, *C. productus* Dana, 1852 and *C. serratus* Shiino, 1965 from off Mexico (Causey 1960; Morales-Serna et al., 2013). Pillai (1985) reported *C. productus* from *S. sierra* from off India, but the host identity is most likely erroneous as *S. sierra* is distributed from southern California to Chile (Froese & Pauly, 2017). In this study, *C. omissus* and *C. fajerae* n. sp. were found frequently on *S. sierra*, the former on the gills and the latter on the skin.

Thirty-two species of *Caligus* have been reported hitherto from Mexico, of which 23 are from the Mexican Pacific and three from the Mexican Caribbean (Morales-Serna et al., 2012, 2014; Suárez-Morales & Gasca, 2016). Therefore, the discovery of *C. fajerae* n. sp. represents the 33rd species of *Caligus* recorded from Mexico and the 24th species of *Caligus* for the Pacific Ocean off Mexico.

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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.

References

- Andrews, M., Bott, N., Battaglene, S., & Nowak, B. (2009). A new species of copepod (Siphonostomatoida: Caligidae) parasitic on the striped trumpeter, *Latris lineata* (Forster), from Tasmania. *Zootaxa*, 1971, 59–68.
- Causey, D. (1960). Parasitic Copepoda from Mexican coastal fishes. *Bulletin of Marine Science*, 10, 323–337.
- Collette, B. B. (2001). Scombridae. Tunas (also, albacore, bonitos, mackerels, seerfishes, and wahoo). In: Carpenter, K. E. & Niem, V. H. (Eds), *FAO species identification guide for fishery purposes. The living marine resources of the Western Central Pacific, vol. 6. Bony fishes, part 4 (Labridae to Latimeriidae)*. Rome: FAO, pp. 3721–3756.
- Costello, M. J. (2009). The global economic cost of sea lice to the salmonid farming industry. *Journal of Fish Diseases*, 32, 115–118.
- Cressey, R. (1991). Parasitic copepods from the Gulf of Mexico and Caribbean Sea, III: *Caligus*. *Smithsonian Contributions to Zoology*, 497, 1–53.
- Cressey, R. F., & Collette, B. B. (1970). Copepods and needlefishes: a study in host-parasite relationships. *Fishery Bulletin*, 68, 347–432.
- Cressey, R., & Cressey, H. B. (1980). Parasitic copepods of mackerel- and tuna-like fishes (Scombridae) of the world. *Smithsonian Contributions to Zoology*, 311, 1–186.
- Cressey, R. F., Collette, B. B., & Russo, J. L. (1983). Copepods and scombrid fishes: A study in host-parasite relationships. *Fishery Bulletin*, 81, 227–265.
- Cressey, R. F., & Nutter, P. (1987). Reidentification of David Causey's *Caligus* collections (Crustacea: Copepoda). *Proceedings of the Biological Society of Washington*, 100, 600–602.
- Dojiri, M., & Ho, J.-S. (2013). *Systematics of the Caligidae, Copepods Parasitic on Marine Fishes*. Crustaceana Monographs, 18. Leiden: Brill.
- Folmer, O., Black, M., Hoeh, W., Lutz, R., & Vrijenhoek, R. (1994). DNA primers for amplification of mitochondrial cytochrome *c* oxidase subunit I from diverse metazoan invertebrates. *Molecular Marine Biology and Biotechnology*, 3, 294–299.
- Freeman, M. A., Anshary, H., & Ogawa, K. (2013). Multiple gene analyses of caligid copepods indicate that the reduction of thoracic appendage in *Pseudocaligus* represents convergent evolution. *Parasites & Vectors*, 6, 336.
- Froese, R., & Pauly, D. (Eds) (2017). FishBase. World Wide Web electronic publication. Retrieved April 8, 2017, from <http://www.fishbase.org>, version (02/2017).
- González, M. T., Castro, R., Muñoz, G., & López, Z. (2016). Sea lice (Siphonostomatoida: Caligidae) diversity on littoral fishes from the south-eastern Pacific coast determined from morphology and molecular analysis, with description of a new species (*Lepeophtheirus confusum*). *Parasitology International*, 65, 685–695.
- Hayward, C. J., Aiken, H. M., & Nowak, B. F. (2008). An epizootic of *Caligus chistos* on farmed southern bluefin tuna *Thunnus maccoyii* off South Australia. *Diseases of Aquatic Organisms*, 79, 57–63.
- Hayward, C. J., Bott, N. J., & Nowak, B. F. (2009). Seasonal epizootics of sea lice, *Caligus* spp., on southern bluefin tuna, *Thunnus maccoyii* (Castelnau), in a long-term farming trial. *Journal of Fish Diseases*, 32, 101–106.
- Ho, J.-S., & Lin, C.-L. (2004). *Sea lice of Taiwan (Copepoda: Siphonostomatoida: Caligidae)*. Keelung: Sueichan Press.
- Ho, J.-S., & Lin, C.-L. (2007). Three species of *Caligus* Müller, 1785 (Copepoda: Caligidae) parasitic on *Caranx* spp. (Teleostei: Carangidae) off Taiwan. *Systematic Parasitology*, 68, 33–43.
- ICZN (2012). International Commission on Zoological Nomenclature: Amendment of articles 8, 9, 10, 21 and 78 of the International Code of Zoological Nomenclature to expand and refine methods of publication. *Bulletin of Zoological Nomenclature*, 69, 161–169.
- Johnson, S. C., Treasurer, J. W., Bravo, S., Nagasawa, K., & Kabata, Z. (2004). A review of the impact of parasitic copepods on marine aquaculture. *Zoological Studies*, 43, 229–243.

- Kim, I.-H. (1998). *Illustrated encyclopedia of fauna & flora of Korea. Vol. 38. Cirripedia, symbiotic Copepoda, Pycnogonida*. Seoul: Ministry of Education.
- Lin, C.-L., & Ho, J.-S. (2007). Six species of sea lice (Copepoda, Caligidae) new to Taiwan. *Journal of the Fisheries Society of Taiwan*, 34, 41–67.
- McBeath, A. J. A., Penston, M. J., Snow, M., Cook, P. F., Bricknell, I. R., & Cunningham, C. O. (2006). Development and application of real-time PCR for specific detection of *Lepeophtheirus salmonis* and *Caligus elongatus* larvae in Scottish plankton samples. *Diseases of Aquatic Organisms*, 73, 141–150.
- Morales-Serna, F. N., Caña-Bozada, V., Mera-Loor, G., Loor-Andrade, P., Fajer-Ávila, E. J., & Ho, J.-S. (2015). New records of sea lice (Copepoda: Caligidae) from marine fishes in Jaramijó, an area with potential for sea-cage aquaculture in Ecuador. *Zootaxa*, 3920, 366–380.
- Morales-Serna, F. N., Gómez, S., & Pérez-Ponce de León, G. (2012). Parasitic copepods reported from Mexico. *Zootaxa*, 3234, 43–68.
- Morales-Serna, F. N., Hernández-Inda, Z. L., Gómez, S., & Pérez-Ponce de León, G. (2013). Redescription of *Caligus serratus* Shiino, 1965 (Copepoda: Caligidae) parasitic on eleven fish species from Chamela Bay in the Mexican Pacific. *Acta Parasitologica*, 58, 367–375.
- Morales-Serna, F. N., Pinacho-Pinacho, C. D., Gómez, S., & Pérez-Ponce de León, G. (2014). Diversity of sea lice (Copepoda: Caligidae) parasitic on marine fishes with commercial and aquaculture importance in Chamela Bay, Pacific coast of Mexico by using morphology and DNA barcoding, with description of a new species of *Caligus*. *Parasitology International*, 63, 69–79.
- Nagasawa, K. (2011). *Caligus macarovi* (Copepoda, Caligidae) from Pacific bluefin tuna, *Thunnus orientalis*, cultured in Japan. *Crustaceana*, 84, 1145–1147.
- Nowak, B. F., Hayward, C. J., González, L., Bott, N. J., & Lester, R. J. G. (2011). Sea lice infections of salmonids farmed in Australia. *Aquaculture*, 320, 171–177.
- Øines, Ø., & Heuch, P. A. (2005). Identification of sea louse species of the genus *Caligus* using mtDNA. *Journal of the Marine Biological Association of the UK*, 85, 73–79.
- Øines, Ø., & Schram, T. (2008). Intra- or inter-specific differences in genotypes of *Caligus elongatus* Nordmann 1832. *Acta Parasitologica*, 53, 93–105.
- Özak, A. A., Yanar, A., & Boxshall, G. A. (2017). The discovery of *Caligus macrurus* Heller, 1865 (Copepoda: Caligidae) in the Mediterranean Sea, and the recognition of *Sciænoophilus* van Beneden, 1852 as a junior synonym of *Caligus* Müller, 1785. *Systematic Parasitology*, 94, 97–109.
- Parker, R. R. (1965). A review and redescription of *Caligus gurnardi* Krøyer, 1863 (Copepoda, Caligidae). *Crustaceana*, 9, 93–103.
- Pillai, N. K. (1985). *Fauna of India: Parasitic copepods of marine fishes*. Calcutta: Technical and General Press.
- Prabha, C., & Pillai, N. K. (1986). Additions to the copepods parasitic on the marine fishes of India. 4. On twenty six species of caligids. *Records of the Zoological Survey of India Occasional Paper*, 79, 1–139.
- Suárez-Morales, E., & Gasca, R. (2016). A new species of *Caligus* Müller, 1785 (Copepoda: Siphonostomatoida: Caligidae) from coral reef plankton in the Mexican Caribbean. *Zootaxa*, 4174, 424–436.
- van der Elst, R. P., & Collette, B. B. (1984). Game fishes of the east coast of southern Africa. 2. Biology and systematics of the queen mackerel *Scomberomorus plurilineatus*. *Investigational Report. Oceanographic Research Institute, Durban*, 55, 1–12.
- Venmathi Maran, B. A., Cruz-Lacierda, E. R., Ohtsuka, S., & Nagasawa, K. (2016). New records of Caligidae (Copepoda, Siphonostomatoida) from the Philippines. *Zootaxa*, 4174, 237–248.
- Williams, E. H., & Bunkley-Williams, L. (1996). *Parasites of offshore big game fishes of Puerto Rico and the western Atlantic*. Mayaguez: Antillean College Press.