

New Record of a Marine Fish Parasite *Nerocila trichiura* (Crustacea: Isopoda: Cymothoidae) from Japan, with its Confirmed Distribution in the Western North Pacific Ocean

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The cymothoid isopod *Nerocila trichiura* (Miers, 1877) is reported based on an ovigerous female from the ventral body surface of a flyingfish, *Cypselurus hiraii* Abe, 1953 (Belontiiformes: Exocoetidae), in the coastal Pacific waters of central Japan. This represents the first record of *N. trichiura* from Japan, and *C. hiraii* is a new host record for this isopod. *Nerocila trichiura* has been reported from the tropical and middle-latitude waters of the Indian and Atlantic oceans and was recorded in 1881 from the Philippines. This paper confirms that the species occurs in the western North Pacific Ocean.

Key Words: Parasitic crustacean, new country record, new host record, flyingfish, *Cypselurus hiraii*.

Introduction

The body surface-attaching cymothoid genus *Nerocila* Leach, 1818 currently comprise 43 valid species (Trilles 1975; Bruce 1987; Bokyo *et al.* 2019). Little information is available on the cymothoids of the genus infesting Japanese fishes, and only two nominal species have been reported from Japan to date (Yamauchi 2016): *Nerocila japonica* Schioedte and Meinert, 1881 from 17 species of wild fishes in 10 families (Cyprinidae, Mugillidae, Latidae, Lateolabridae, Sparidae, Embiotocidae, Terapontidae, Labridae, Gobiidae, Monacanthidae) from central and western Japan and from one species of farmed fish (Kyphosidae) from central Japan (Yamauchi and Nagasawa 2012; Nagasawa *et al.*, 2018; Nagasawa and Kawai, 2019); and *Nerocila phaiopleura* Bleeker, 1857 from eight species of wild fishes in six families (Clupeidae, Dussumieriidae, Engraulidae, Carangidae, Scombridae, Sphyrinaeidae) from central and western Japan and from one species of farmed fish (Scombridae) from central Japan (Mitani 1982; Bruce and Harrison-Nelson 1988; Nagasawa and Tensha 2016; Nagasawa and Shirakashi 2017; Nagasawa and Isozaki 2017; Nagasawa and Nakao 2017; Nagasawa and Kawai 2018).

Nerocila trichiura (Miers, 1877) is a skin parasite of flyingfishes (*e.g.*, Trille 1975, 1994; Bruce and Harrison-Nelson 1988). Recently, we collected a specimen of *N. trichiura* from a flyingfish, *Cypselurus hiraii*, in a small bay on the Pacific coast of central Japan. This represents the first and second records of *N. trichiura*, respectively, from Japan and the western North Pacific Ocean.

Materials and Methods

The infested host fish (*C. hiraii*) was caught on 16 July 2018 with a set net installed in Kowaura Bay off Maruwa (34°13'33"N, 136°29'00"E), Minami-Ise, Mie Prefecture, Japan. The isopod was removed from the fish and frozen with sea water. Later, it was thawed, fixed in 70% ethanol, and identified as *N. trichiura*. Since only one specimen was collected, it was examined without dissection using an Olympus SZX10 stereo microscope. It was also measured for body length (from the anterior extremity of the cephalon to the posterior end of the pleotelson) and various parts of the body. Drawings were made with the aid of a drawing tube fitted on the microscope. The specimen of *N. trichiura* has been deposited in the Crustacea (Cr) collection of the National Museum of Nature and Science, Tsukuba, Ibaraki Prefecture (NSMT-Cr). The scientific names of fishes mentioned in this paper follow Froese and Pauly (2018).

Nerocila trichiura (Miers, 1877)

[New Japanese name: Tobiuo-no-koban]

(Figs 1, 2)

Anilocra trichiura Miers, 1877: 677–678, pl. 69, fig. 6–6a; Clark and Presswell 2001, 155 [*Anilocra trichiura* reported by White (1847: 108) is a *nomen nudum*].

Nerocila trichiura: Schioedte and Meinert 1881, 83–85, pl. VII (Cym. XIV), figs 1–2; Stebbing 1910, 84, 102; Barnard 1914, 372; Nierstrasz 1918, 111–113; Nierstrasz 1931, 125; Monod 1931, 6, fig. 1a–b; Barnard 1940, 491, 501; Barnard 1955, 5; Monod 1970, 10, 30, 65, pl. 3, fig. 2;

Trilles 1975, 315–316, pl. 2, fig. 10; Kensley 1978, 81, 83, fig. 33F–G; Kurochkin 1980, 286; Ellis 1981, 125; Bruce 1987, 406; Bruce and Harrison-Nelson 1988, 598–600, fig. 7; Trilles 1994, 101–102; Kensley 2001, 234; Trilles *et al.* 2011, 453, table 1; Trilles *et al.* 2013, 1275–1276, 1281, table 1, fig. 2k, fig. 7j; Rameshkumar *et al.* 2013, 89–91, tables 1–2, 4.

Nerocila sp. 1: Trilles 1979, 267, pl. 2, fig. 10 [see Bruce and Harrison-Nelson 1988 for synonymy].

Material examined. Oviparous female (NSMT-Cr 26315), 31.0 mm in body length, 12.7 mm in body width.

Description. Body 2.4 times as long as greatest width, widest at pereonite 6 and 7. Cephalon 0.5 times longer than wide, anterior margin slightly concave. Eyes indistinct. Coxae all with rounded point, not exceeding beyond posterior margin of pereonites; coxae 2–4 short; coxae 5–6 slightly longer than 2–4.

Pereonite 6 longest; pereonite 1 longer than 2–4; pereonites 5 and 7 slightly shorter than 6; pereonites 1–7 gradually increasing in width towards posterior. Pereonite 1 anterior margin concave. Posterior margins of pereonites 1–3 slightly concave; pereonites 4–6 almost straight; pereonite 7 weakly convex. Posterolateral angles of pereonites 1–6 bluntly rounded; pereonite 7 slightly pointed. Pleonite 1 widest; pleonites 2–4 decreasing in width; pleonite 5 slightly wider than 4. Ventrolateral margins of pleonites 1 and 2 both posteriorly directed and acute, extending to pleonites 3 and 4, respectively. Pleotelson 1.1 times as long as anterior width, lateral margins curving to medial point.

Pereopods 1–7 increasing in size towards posterior; pereopods 1–6 without robust setae; pereopod 7 with 3 robust setae on posterior margin of carpus, with 4 robust setae on posterior margin of propodus. Pereopod 7 basis 4.3 times as long as greatest width; ischium 0.3 times as long as basis; merus 0.9 times as long as ischium, 1.0 times as long as wide; carpus 0.6 times as long as ischium, 0.7 times as long as wide; propodus 1.2 times as long as ischium, 1.8 times as long as wide; dactylus slender, 2.1 times as long as propodus, 3.7 times as long as basal width.

Uropod 1.6 times as long as pleotelson; peduncle 0.4 times as long as rami, peduncle lateral margin without setae; rami extending beyond medial point of pleotelson, tapering posteriorly. Endopod apically rounded, lateral margin weakly arched, mesial margin slightly convex, 4.0 times long as greatest width. Exopod slender, characteristically long, 7.5 times long as greatest width, extending far beyond end of endopod, apically rounded, lateral margin almost straight, mesial margin straight.

Brood pouch formed from pair of large oostegites, enclosing embryos, arising from coxa 6 and anterior pairs of small oostegites.

Color. White when fresh/live; whitish yellow in ethanol preservative.

Host. *Cypselurus hiraii* [Japanese name: Hoso-tobiuo] (Beloniformes: Exocoetidae).

Site of infestation. Ventral body surface, below the base of the pectoral fins, of *C. hiraii* (Fig. 1A). A skin wound

(18.1 mm long, 12.8 mm wide) with exposed muscle was found at the attachment site (Fig. 1B).

Locality. Kowaura Bay, western North Pacific Ocean, off Maruwa, Minami-Ise, Mie Prefecture, Japan (locality 12 in Fig. 3).

Distribution. Recorded from the tropical and middle-latitude waters of the Indian, Atlantic, and western North Pacific oceans (Fig. 3, see Remarks for detailed information on the collection localities).

Remarks. *Nerocila trichiura* was first listed, without any description, as “*Anilocra trichiura*, n. sp.”, one of the crustaceans in the collections of the British Museum, London (White 1847, see Clark and Presswell 2001 and the above synonym list). The specimen reported was a female from Mauritius (Indian Ocean) and, using this specimen, Miers (1877) formally described *A. trichiura*. Later, the species was transferred to *Nerocila* and redescribed by Schioedte and Meinert (1881). The male from the Atlantic Ocean was also described by Nierstrasz (1918). The dorsal and lateral views of an oviparous female from Congo were shown by Monod (1931). A photograph of the dorsal view of an oviparous female from an unknown locality was given by Trilles (1975: pl. 2, fig. 2). Bruce and Harrison-Nelson (1988) illustrated the holotype (fig. 7A–E) from Mauritius and the females (fig. 7F–I) from Senegal and the Indian Ocean south of India. Recently, Trilles *et al.* (2013) also provided a photograph (fig. 2k) and an illustration (fig. 7j) showing the dorsal view of a female from the Tamil Nadu coast of India. The female specimen collected in the present study corresponds well to the descriptions, figures, and photographs of *N. trichiura* given by Miers (1877), Schioedte and Meinert (1881), Monod (1931), Trilles (1975), Bruce and Harrison-Nelson (1988), and Trilles *et al.* (2013). *Nerocila trichiura* is characterized by having short coxae with rounded point and the posterolateral angles of pereonites 1–6 bluntly rounded. The color of a fresh specimen of *N. trichiura* from India was white without any band (Trilles *et al.* 2013: fig. 2k), which is also confirmed in this study (Fig. 1C).

There is considerable variation in the ratio of the uropod to the pleotelson between specimens examined in the previous and present studies: 1.9 in the specimen from Mauritius (holotype) (Bruce and Harrison-Nelson 1988: fig. 7A); 2.4 in the specimen from an unspecified locality (Schioedte and Meinert 1881: pl. 7, fig. 6); 2.6 in the specimen from Congo (Monod 1931: fig. 1a); 1.7 in the specimen from South Africa (Kensley 1978: fig. 33G); 2.4 in the specimen from Senegal; 1.9 in the specimen from off south of India (Bruce and Harrison-Nelson 1988: fig. 7E, I); 2.3 in the specimen from India (Trilles *et al.* 2013: fig. 7j); and 1.6 in the specimen from Japan (Fig. 2D). The number of robust setae on the posterior margin of propodus and carpus of pereopod 7 also varies: 6 and 2 in the specimen from Mauritius; 3 and 2 in the specimen from Senegal (Bruce and Harrison-Nelson 1988: fig. 7E, H); and 4 and 3 in the specimen from Japan (Fig. 2E).

Nerocila trichiura is similar to *N. exocoeti* Pillai, 1954, which parasitizes beloniform fishes including exocoetids (flyingfishes) in the Indian and western Pacific oceans

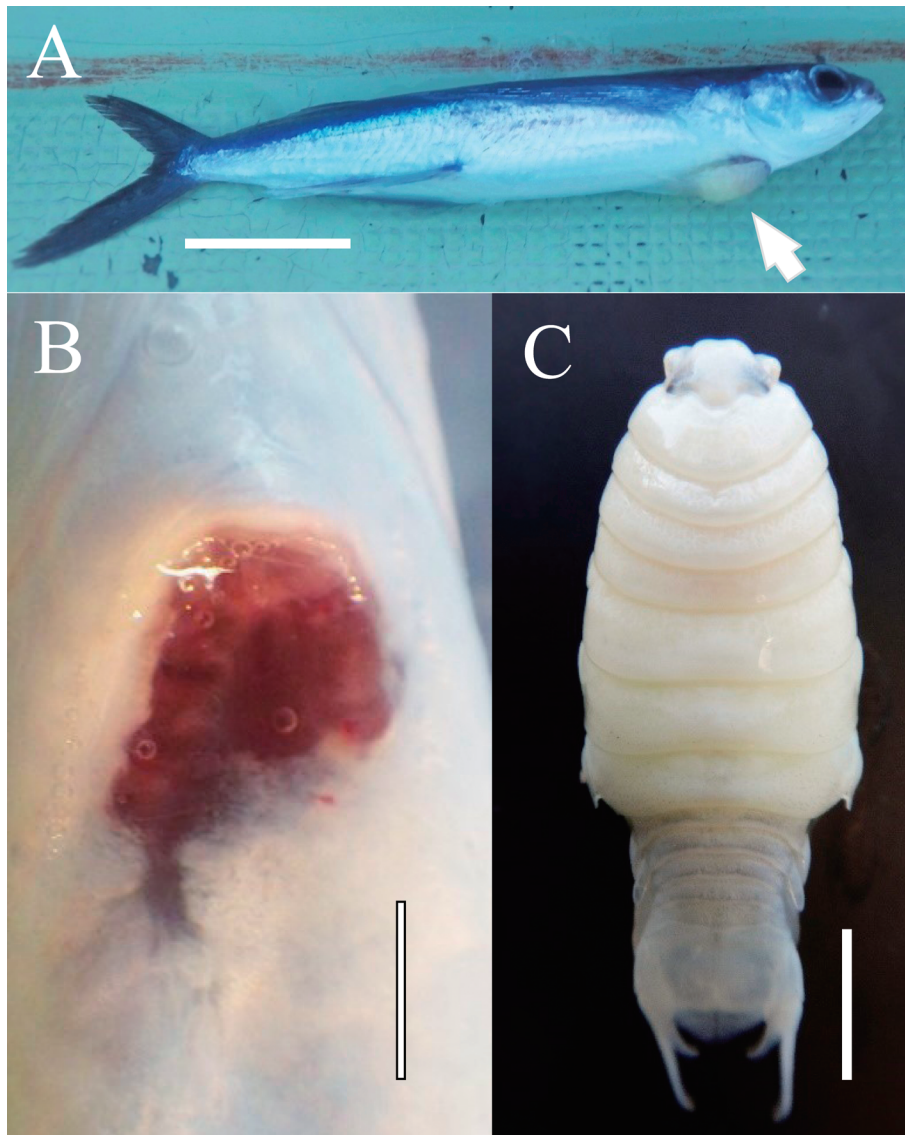


Fig. 1. *Nerocila trichiura*, ovigerous female, NSMT-Cr 26315. A, *Cypselurus hiraii* (252 mm in total length) infested with *N. trichiura* (arrow); B, skin wound at attachment site; C, *N. trichiura*, dorsal view, fresh specimen. Scale bars: A, 50 mm; B, C, 10 mm.

(Bruce and Harrison-Nelson 1988; Trilles *et al.* 2013; Aneesh *et al.* 2017). In the redescription of *N. exocoeti*, Aneesh *et al.* (2017) stated that this species can be distinguished from *N. trichiura* by having the posterior margin of coxae 5–7 acute and the body color (steel blue) when fresh. The difference in body color between two species (white in *N. trichiura* vs. steel blue in *N. exocoeti*) is closely related to the difference in their infestation site: *N. trichiura* attaches to the white-colored ventral body surface of the host (Fig. 1A), while *N. exocoeti* to the dark-colored dorsal body surface of the host (see Sivasubramanian *et al.* 2011: figs 1, 2; Aneesh *et al.* 2017: fig. 6G, H).

Nerocila trichiura can be also differentiated from the two congeneric species, *N. japonica* and *N. phaiopleura*, occurring in Japanese waters. *Nerocila japonica* has a wider body with two submedian pale longitudinal bands than *N. trichiura* (Yamauchi and Nagasawa 2012). The body shape of *N. phaiopleura* is similar to that of *N. trichiura*, but the former species has the large eyes and dark brown or black stripes on

the uropod exopod and lateral sides of the posterior pereonites and the pleon (Nagasawa and Tensha 2016). The known hosts of *N. japonica* and *N. phaiopleura* are coastal marine fishes but do not contain exocoetids (see the Introduction for references).

The collection of *N. trichiura* in this study represents its first and second records in Japan and the western North Pacific Ocean, respectively. The species has so far been reported from the Indian Ocean [Mauritius-type locality (Miers 1877; Bruce and Harrison-Nelson 1988), Great Chagos (Stebbing 1910), Durban, South Africa (Barnard 1955; Kensley 1978), 10°20'S, 70°00'E (Bruce and Harrison-Nelson 1988), Comoro Islands (Kensley 2001), Tamil Nadu coast, India (Trilles *et al.* 2013; Rameshkumar *et al.* 2013)]; the Atlantic Ocean [31°N, 76°W (Schioedte and Meinert 1881), Banana and an unknown locality, Congo (Nierstrasz 1918; Monod 1931), the West Indies (Trilles 1979, reported as *Nerocila* sp. 1, which was regarded as *N. trichiura* by Bruce and Harrison-Nelson 1988), Dakar Harbor, Senegal

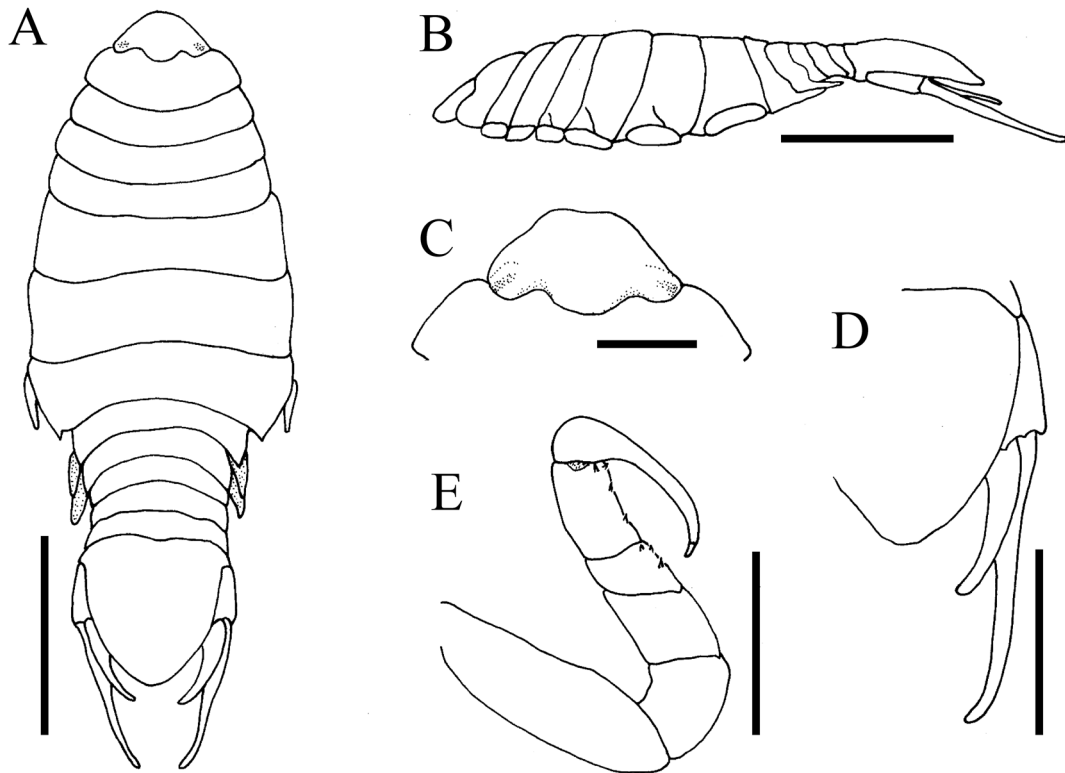


Fig. 2. *Nerocila trichiura*, ovigerous female, NSMT-Cr 26315. A, dorsal view; B, lateral view; C, cephalon and pereonite 1, dorsal view; D, pleotelson and right uropod, dorsal view; E, pereopod 7. Scale bars: A, B, 10 mm; C, E, 2 mm; D, 5 mm.

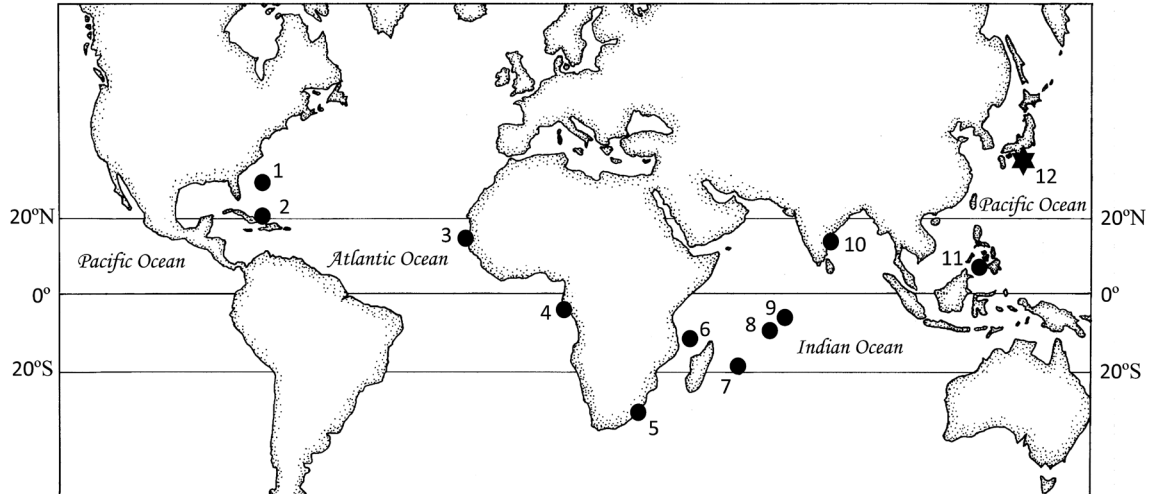


Fig. 3. Map showing the localities where *Nerocila trichiura* was collected in the previous (circles) and present (star) studies. 1, 31°N, 76°W (Schioedte and Meinert 1881); 2, the West Indies (Trilles 1979); 3, Dakar Harbor, Senegal (Bruce and Harrison-Nelson 1988); 4, Banana and an unknown locality, Congo (Nierstrasz 1918; Monod 1931); 5, Durban, South Africa (Barnard 1955; Kensley 1978); 6, Comoro Islands (Kensley 2001); 7, Mauritius (type locality, Mier 1877; Bruce and Harrison-Nelson 1988); 8, 10°20'S, 70°00'E (Bruce and Harrison-Nelson 1988); 9, Great Chagos (Stebbing 1910); 10, Tamil Nadu coast, India (Trilles *et al.* 2013; Rameshkumar *et al.* 2013); 11, Zamboanga, Philippines (Schioedte and Meinert 1881); and 12, Kowaura Bay, Japan (this paper).

(Bruce and Harrison-Nelson 1988)]; and the Pacific Ocean [Zamboanga (reported incorrectly as Zamboango), Philippines (Schioedte and Meinert 1881)] (Fig. 3). Barnard (1914) collected the species from an unknown locality in South Africa, and Trilles (1975) also recorded it from an unknown locality. These authors questioned Schioedte and Meinert's record of *N. trichiura* from the Philippines, but the

present study has confirmed that the species actually occurs in the western North Pacific Ocean.

The fact that we rediscovered *N. trichiura* in the western North Pacific Ocean 137 years after Schioedte and Meinert (1881) reported it from the Philippines clearly shows that the past faunal study of cymothoid isopods of this region was insufficient. There is no record of *N. trichiura* in the

literature on those isopods from China (Yu and Li 2003), Hong Kong (Bruce 1982, 1990), and Japan (Yamauchi 2016). We need more study to clarify the fauna of cymothoid isopods in the western North Pacific Ocean.

As shown in Fig. 3, *N. trichiura* occurs in the tropical waters of the Indian, Atlantic, and western North Pacific oceans, excluding three localities in the middle latitudes: the western North Atlantic off the southeast U.S.A. (locality 1 in Fig. 3), Durban, South Africa (locality 5), and Kowaura Bay, Japan (locality 12), which are affected by the Gulf Stream, the Agulhas Current, and the Kuroshio, respectively. This indicates that *N. trichiura* is a tropical species in the three oceans, where the species also occurs in the middle-latitude waters affected by the warm currents.

The known hosts of *N. trichiura* are the two-wing flyingfish, *Exocoetus volitans* Linnaeus, 1758 (Beloniformes: Exocoetidae) (Stebbing 1910, reported as *Exocoetus evolans* Linnaeus, 1766; Nierstrasz 1918; Monod 1970; Bruce and Harrison-Nelson 1988; Trilles *et al.* 2013; Rameshkumar *et al.* 2013) and unidentified species of *Exocoetus* (Barnard 1914, 1955; Monod 1931). Although “*Exocoeti evolantis*” was used by Schioedte and Meinert (1881) for the host of the isopod, this scientific name is not valid (*cf.* Froese and Pauly 2018), and the host reported is probably *E. volitans*. In a review of the parasites of flyingfishes from the world oceans, Kurochkin (1980) listed *E. volitans*, *E. evolans*, *E. sp.*, and the backsail flyingfish, *Cheilopogon nigricans* (Bennet, 1840), as the hosts of *N. trichiura*. In the present study, *Cypselurus hiraii* was found to harbor *N. trichiura* and is a new host record for the isopod. Based on the past and present records, flyingfishes (Exocoetidae) are considered to be preferred hosts for *N. trichiura*.

Cypselurus hiraii is distributed in the western North Pacific Ocean, including the southern Sea of Japan and the East China Sea (Aizawa and Doiuchi 2013). The species migrates for spawning to the southwestern Sea of Japan off western Japan from May to July (Kawano *et al.* 1995; Kawano 2004) but no information is available on its migration to the coastal Pacific waters of Japan. In Kowaura Bay, where we collected *N. trichiura* in this study, *C. hiraii* is commercially caught during summer months with coastal set nets but its catch is low (S. Isozaki, unpublished). *Nerocila trichiura* is not a common fish parasite in the bay.

In this study, the specimen of *N. trichiura* was found to be attached to the ventral body surface below the base of the pectoral fins of the host fish (Fig. 1A). A similar attachment site was previously reported for the species (Stebbing 1910; Monod 1970). A skin wound was found at the attachment site and the host's muscle was exposed (Fig. 1B). The wound was located under the anterior part of the body of *N. trichiura*. The observed disease conditions were most probably induced by the skin feeding and deep insertion of the pereopod's dactyli of the species. Similar skin wounds are also found on marine fishes infested by *N. phaiopleura* in Japanese waters (Nagasawa and Tensha 2016; Nagasawa and Shirakashi 2017; Nagasawa and Isozaki 2017; Nagasawa and Kawai 2018).

Nerocila madrasensis described by Ramakrishna and

Ramaniah (1978) from *Hemiramphus* sp. (Beloniformes: Hemiramphidae) in India was previously suggested to be a junior synonym of *N. trichiura* (Trilles 1994; Trilles *et al.* 2011), but the former taxon has currently been relegated to synonymy of *N. exocoeti* (Aneesh *et al.* 2017). *Nerocila* sp. reported by Daniel and Rama Rao (1967) from the African sailfin flyingfish, *Parexocoetus mento* (Valenciennes, 1847) (Beloniformes: Exocoetidae), in India was also suggested to be a junior synonym of *N. trichiura* (Trilles 1994; Trilles *et al.* 2011). *Nerocila* sp. reported by Monod (1976) was suggested by Kurochkin (1980) to be identical as *N. trichiura*. However, as indicated by Monod (1976: figs 14, 15) and Trilles (1994), the isopod is probably *N. phaiopleura*. According to Bruce and Harrison-Nelson (1988), *Nerocila excisa* (Richardson, 1914) may be conspecific with *N. trichiura*, the holotype of *N. excisa* was so damaged that it was impossible to clarify the synonymy of both species.

To date, three species of cymothoid isopods have been reported from flyingfishes in Japan (Yamauchi 2016): *Mothocya melanosticta* (Schioedte and Meinert, 1884) from the sailfin flyingfish, *Parexocoetus brachypterus* (Richardson, 1846) (Bruce 1986; Nunomura 1992, 2005, 2011); *Ceratothoa guttata* (Richardson, 1910) (reported as *Mothocya katoi* Nunomura 1992; Nunomura 1992, 2005, 2011, see Hadfield *et al.* 2015 for synonymy) from *P. brachypterus* (reported as *P. brachypterus brachypterus*, Nunomura 1992, 2005); and *Glossobius auritus* Bovallius, 1885 (reported as *Glossobius ogasawarensis* Nunomura 1992; Nunomura 1992, 2011, see Nunomura 2005 and Martin *et al.* 2015 for synonymy) from the Japanese flyingfish, *Cheilopogon agoo* (Temminck and Schlegel, 1846) (reported as *Cypselurus agoo*; Bruce and Bowman 1989) and the narrowhead flyingfish, *Cypselurus angusticeps* Nichols and Breder, 1935 (reported incorrectly as *C. angusticeps*; Nunomura 1992, 2005). *Nerocila trichiura* is the fourth species of cymothoid isopod reported from flyingfishes in Japanese waters.

In this study, the individual of *Cypselurus hiraii* infested with *N. trichiura* was collected in Kowaura Bay, central Japan, from which two other species of cymothoid isopods have been reported: *Ceratothoa verrucosa* (Schioedte and Meinert, 1883) from the crimson seabream, *Evynnis tumifrons* (Temminck and Schlegel, 1843) (Nagasawa and Isozaki 2016), and *Nerocila phaiopleura* from the Japanese barracuda, *Sphyraena japonica* Bloch and Schneider, 1801, the Japanese jack mackerel, *Trachurus japonicus* (Temminck and Schlegel, 1844), and the round herring, *Etrumeus micropus* (Temminck and Schlegel, 1846) (Nagasawa and Isozaki 2017).

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