

Doctoral Dissertation (Censored)

博士論文 (要約)

Systematics and molecular phylogeny of the Monostilifera

(phylum Nemertea)

(紐形動物門単針類の系統分類学的研究)

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DISCLAIMER

New taxon names and nomenclatural changes referred to in this dissertation are disclaimed and unavailable for nomenclatural purposes (ICZN Art. 8.3).

Part of this study has been published as the following papers:

- Hookabe, N., Kajihara, H. (2020). New record of the monostiliferous hoplonemertean *Gurjanovella littoralis* Uschakov, 1926 from Japan (Nemertea, Hoplonemertea, Monostilifera). *Check List* 16, 265–269.
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- Hookabe, N., Kohtsuka, H., Kajihara, H. (2021). A histology-free description of *Tetrastemma cupido* sp. nov. (Nemertea: Eumonostilifera) from Sagami Bay, Japan. *Marine Biology Research* 17, 467-474.
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- Hookabe, N., Kajihara, H., Chernyshev, A.V., Jimi, N., Hasegawa, N., Kohtsuka, H., Okanishi, M., Tani, K., Fujiwara, Y., Tsuchida, S., Ueshima, R. (2022b). Molecular phylogeny of the genus *Nipponnemertes* (Nemertea: Monostilifera: Cratenemertidae) and descriptions of 10 new species, with notes on small body size in a newly discovered clade. *Frontiers in Marine Science* 9, e906383.
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Abstract of Dissertation

Nemertea (ribbon worms) is one of the spiralian phyla. The body plan is characterized by unsegmented soft body with eversible proboscis housed in a fluid-filled chamber called the rhynchocoel. In the order Monostilifera, the proboscis is armed with a stylet apparatus composed of a single central stylet and accessory stylet pouches; the stylet is used for piercing the prey tissues to inject toxins and immobilize the prey animals. Although monostiliferans are known from almost all kinds of ecosystems—mostly inhabiting marine benthic habitats but also from brackish, freshwater, and terrestrial environments, the species diversity and phylogenetic relationships within the order are not well understood. In my dissertation, I aimed to elucidate phylogenetic relationships between monostiliferan families with molecular phylogenetic reconstruction using specimens freshly obtained during my extensive taxon sampling from deep-sea to terrestrial habitats. This dissertation is comprised of four chapters:

Chapter 1 is General Introduction for the monostiliferan classification, systematics, and phylogeny, dealing with general problems and current progress in the two major groups of Monostilifera, Cratenemertea and Eumonostilifera. Since the advent of molecular phylogenetic techniques and their application to nemertean systematics, it has been increasingly apparent that monostiliferans cannot be placed with certainty into any of existing families or genera solely based on morphological characters. Therefore, I examined external and internal morphology of approximately 60 specimens of Monostilifera obtained during my Ph. D course from Northwestern Pacific (Chapter 3) and confirmed the generic affiliation with molecular phylogenetic analyses (Chapter 2, 3).

Phylogenetic relationships within Monostilifera (especially Eumonostilifera) were not well resolved in the previous studies. In Chapter 2, I performed multi-locus molecular phylogenetic analyses using two mitochondrial (16S rRNA and cytochrome oxidase *c* subunit I) and three nuclear (18S rRNA, 28S rRNA, histone H3) genetic markers for 108 monostiliferan species. One notable finding of the phylogenetic study was the phylogenetic position of Malacobdellidae Blanchard, 1847 nested in Prosorhochmidae Bürger, 1895. To further evaluate the phylogenetic relationship between Malacobdellidae and Prosorhochmidae, I performed molecular phylogenetic analyses with the increased numbers of loci (13 protein coding regions, 16S rRNA, and 12S rRNA) obtained from the mitochondrial genome. The nested position of Malacobdellidae within Prosorhochmidae was recovered with both the analyses, based on

which I discuss familial/subfamilial assignment for the taxon name of Malacobdellidae, by comparing adult and larval morphology with Prosorhochmidae.

Chapter 3 is partially excerpted from my publication in 2019–2022. This chapter is subdivided into 8 subchapters dealing with systematics of species/genera in 8 monostiliferan families: Cratenemertidae Friedrich, 1968 (Cratenemertea), Amphiporidae Örsted, Carcinonemertidae Sumner, Osburn & Cole, 1913, Oerstediiidae Chernyshev, 1993, Plectonemertidae Gibson, 1990a, Prosorhochmidae Bürger, 1895, Sacconemertidae Chernyshev, 2005, and Tetrastemmatidae Hubrecht, 1879 (Eumonostilifera). Before my studies, a total of 50 species of monostiliferans were recognized in Japan and has been increased to 70 by my studies, where I established 2 new genera (*Genrokunemertes* Hookabe et al. 2022d, *Rhombonemertes* Hookabe et al. 2022c) and 20 new species (*Genrokunemertes obesa* Hookabe et al. 2022d, *Nipponnemertes crypta* Hookabe et al. 2022b, *N. ganahai* Hookabe et al. 2022b, *N. jambio* Hookabe et al. 2022b, *N. kozaensis* Hookabe et al. 2022b, *N. lactea* Hookabe et al. 2022b, *N. neonilae* Hookabe et al. 2022b, *N. notoensis* Hookabe et al. 2022b, *N. ojimaorum* Hookabe et al. 2022b, *N. ornata* Hookabe et al. 2022b., *N. sugashimaensis* Hookabe et al. 2022b, *Proamphiporus kaimeiae* Hookabe et al. 2020, *Rhombonemertes rublinea* Hookabe et al. 2022c, *Tetrastemma album* sp. nov., *T. carneum* sp. nov., *T. cupido* Hookabe et al. 2021, *T. persona* sp. nov., *T. shohoense* sp. nov., and two unnamed forms) and proposed two new combinations (*Amphiporus tokmakovae* (Chernyshev, 1991) comb. nov. and *Proamphiporus rectangulus* (Strand, Herrera-Bachiller, Nygren & Kånneby, 2014)) and by species redescriptions. These studies include a new record of Plectonemertidae from Japan.

My systematic studies on Monostilifera in Northwestern Pacific revealed only a part of species diversity. Still, there are a number of undescribed/unreported monostiliferans awaiting formal description in NW Pacific, especially in deep sea or supratidal zones, where nemertean faunae have been scarcely understood. Previous molecular systematics on nemerteans are rather biased toward intertidal to shallow-water species. Therefore, morphological definition of each family and genus seems not necessary to cover the morphological characteristics of deep-sea or supratidal species. Further faunistic studies in various ecosystems without biased toward intertidal or shallow-water species are required not only to obtain a more accurate picture of nemertean diversity but also systematics.

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Chapter 1.

Introduction to Monostiliferans

1.1 The order Monostilifera

Ribbon worms (phylum Nemertea) are commonly known as predators or scavengers in a wide variety of marine, freshwater, and terrestrial ecosystems (McDermott and Roe 1985; Thiel and Kruse 2001). The phylum currently contains around 1300 species (Chernyshev 2021; Kajihara 2021a, b), of which about 594 species in 127 genera are members in the order Monostilifera (Kajihara 2021b). Nemerteans are characterized by possessing an eversible proboscis that is usually housed in a fluid-filled chamber called the rhynchocoel. The proboscis is mainly used for capturing prey such as polychaetes, molluscs, and small crustaceans (Coe 1943; McDermott and Roe 1985; Thiel and Kruse 2001). In monostiliferans, proboscis is generally armed with a stylet apparatus containing a single central stylet on the stylet basis (Gibson 1994). The stylet is used to stab and inflict wounds on prey, by which paralytic neurotoxins produced in the proboscis can be injected (Kem et al. 1971; Stricker 1981).

The Monostilifera can be divided into the suborders Cratenemertea and Eumonostilifera (Fig. 1) chiefly by a combination of three characters: the arrangement of the rhynchocoel musculature (a wickerwork of longitudinal and circular muscle fibres in Cratenemertea as well as in several eumonostiliferans *vs* separate inner longitudinal and outer circular muscle layers in Eumonostilifera), the vascular plug (single plug in Cratenemertea *vs* variously 0–2 plugs in Eumonostilifera), and the position of the cerebral organs (mostly extending behind brain in Cratenemertea *vs* without extending behind brain in Eumonostilifera) (Chernyshev 2021; Kajihara 2021b). The distinction between Cratenemertea and Eumonostilifera is supported by molecular phylogenetic analyses based on multiple loci of nuclear and mitochondrial genes (Andrade et al. 2012; Kvist et al. 2014, 2015; Chernyshev and Polyakova 2019).

1.2 Problems in systematics of Cratenemertea

Molecular systematics within Cratenemertea is still in progress. Although 28 species in three families have been recognized in this taxon, phylogenetic relationships within Cratenemertea

are not well resolved due to a lack of molecular data. So far, species in two genera, *Nipponnemertes* Gibson & Crandall, 1989 and *Uniporus* Brinkmann, 1914–1915, and unidentified specimens collected from the Far Eastern seas of Russia, have been included in molecular phylogeny (e.g., Kvist et al. 2015; Chernyshev and Polyakova 2018a, b, 2019). Recent molecular phylogenetic analyses suggest that ‘Cratenemertea sp. 25DS’ (Chernyshev and Polyakova 2019) and ‘Cratenemertidae sp. IZ-45644’ (Kvist et al., 2015) are the sister taxon to *Uniporus* (Chernyshev and Polyakova 2019). ‘Cratenemertea sp. 25DS’ is likely to be a planktonic juvenile of ‘Cratenemertidae sp. IZ-45644’ (Chernyshev and Polyakova 2019). Although ‘Cratenemertea sp. 25DS’ is morphologically similar to *Achoronemertes scorebyi* (Wheeler, 1934) and *Korotkevitschia pelagica* (Korotkevitsch, 1961) (Chernyshev and Polyakova 2019), a comparison of both morphological and molecular data is required for accurate species identification.

Another issue that needs to be investigated with the molecular approach is the generic distinction between *Cratenemertes* Friedrich, 1955 and *Nipponnemertes*. Friedrich (1955) established *Cratenemertes* for a species originally described by Bürger (1890), *Amphiporus amboinensis* Bürger, 1890; in Friedrich (1968), the generic name was attributed to species with a small cerebral organ posteriorly fused with dorsal cerebral ganglia. Whether smaller cerebral organs in *Cratenemertes*—the only difference between the two genera—are a phylogenetic signal or not need to be verified with molecular data.

1.3 Problems in Systematics of Eumonostilifera

Since the advent of molecular phylogenetic techniques and their application to nemertean systematics, it has been increasingly apparent that eumonostiliferans cannot be placed with certainty into any of existing genera solely based on morphological characters except for those with an apparent synapomorphy, such as Takakura’s duct [a specialized vas deferens, possibly an evolutionary co-option from the mid-dorsal blood vessel] in *Carcinonemertes* Coe, 1902 and *Ovicides* Shields, 2001, posterior sucker in *Malacobdella* Blainville, 1827, statocysts in *Ototyphlonemertes* Diesing, 1863, and post-cerebral eyes in *Pheroneonemertes* Gibson, 1990a and *Zygonemertes* Montgomery, 1897. While anatomical/histological characters were employed in eumonostiliferan systematics in the pre-molecular era (e.g., Friedrich 1955), many of genera and families were circumscribed by morphological characters. Many of these taxa, however, have turned out to be non-monophyletic in a number of molecular analyses during the

past two decades (Thollessen and Norenburg 2003; Strand and Sundberg 2005a, b; Sundberg et al. 2007; Kajihara et al. 2011; Andrade et al. 2012; Kajihara and Kuris 2013; Taboada et al. 2013, 2018; Kvist et al. 2014, 2015; Strand et al. 2014; Krämer and Döhren 2015; Chernyshev and Polyakova 2019; Chernyshev et al. 2021).

There are about 600 species in 127 genera classified in the order Monostilifera (Kajihara 2021b). Most of the monostiliferan genera are represented by molecular sequences from specimens reliably identified as the type species or likely congener(s), especially for *Amphiporus* Ehrenberg, 1831 and *Tetrastemma* Ehrenberg, 1828, the oldest established, species-rich, and taxonomically problematic genera within Eumonostilifera. Sequences derived from a specimen identified as *Amphiporus lactiflorens* (Johnston, 1828)—the type species of *Amphiporus*; see Gibson and Crandall (1991) and International Commission on Zoological Nomenclature (1992)—collected in Wales, and thus can be practically regarded as a topotype, were determined by Andrade et al. (2012); this topotype is likely conspecific with another specimen earlier collected in the White Sea and identified as *A. lactiflorens* (Thollessen and Norenburg 2003). The taxonomic identity of the type species of *Tetrastemma*, *T. flavidum* Ehrenberg, 1828, is unclear due to the inconsistencies in morphology with other species in Amphiporina (Kajihara 2021). Therefore, the Swedish specimen labelled ‘isolate T3’ in Strand and Sundberg (2005a, b) has been proposed to be temporarily regarded as *T. flavidum* (Kajihara 2021b).

With the progress of the taxonomic revisions accompanied with phylogenetic approaches, new-species description for *Amphiporus* and *Tetrastemma* can be accomplished even without histology. For this, the most recent example is a description of *Tetrastemma freyae* Chernyshev, Polyakova, Vignesh, Jain, Sanjeevi, Norenburg & Rajesh, 2020. It was differentiated from the congeners based on the body coloration pattern as well as the shape of stylet basis. The morphological diagnosis free from histology was supported by the COI genetic distances between the congeners comparable with intraspecific divergences among hoplonemertean. The approaches would help to give taxonomic identities to species that are almost impossible to recollect due to the hard-to-reach habitat such as deep-sea bottom and promise to accelerate elucidation of uncovered nemertean species diversity.

1.4 Aims of the Present Study

Monostiliferan classification solely based on morphological characteristics (especially internal morphology) has turned out to be not necessarily stable. Instead, a DNA-based approach is now in commonplace to untie tangled knots of monostiliferan systematics. In my dissertation, I aimed to reveal monostiliferan interfamilial phylogenetic relationships based on extensive taxon sampling from terrestrial, semiterrestrial, inter- or subtidal, and deep-sea environments in Northwestern Pacific (Chapter 2). With the specimens newly obtained, species identification and taxonomic revisions were performed for 8 families in Monostilifera based on morphological observation and molecular phylogeny (Chapter 3).

Chapter 2.

Molecular Phylogeny of the Monostilifera

第2章については、5年以内に雑誌等で刊行予定のため、非公開とする。

Chapter 3.

Systematics of Selected Monostiliferan Families and Genera

This chapter is taxonomic parts for specimens used in the monostiliferan molecular phylogeny (Chapter 2), including species descriptions and taxonomic revisions.

3.1. Cratenemertidae Friedrich, 1968 (CRATENEMERTEA)

This subchapter deals with molecular systematics of the genus *Nipponnemertes* Friedrich, 1968 published in Hookabe et al. (2022b).

3.1.1. Introduction

Species in the genus *Nipponnemertes*—18 are currently recognized as valid—represent the majority of the suborder Cratenemertea (Gonzalez-Cueto et al. 2017; Kajihara 2021b). They are marine benthic, usually feeding on small crustaceans such as amphipods (Brunberg 1964; Berg 1972a; McDermott 1984; McDermott 1993; McDermott 1997) and caridean shrimps (Kajihara et al. 2015) by sucking out body fluid and internal tissue of those prey organisms. *Nipponnemertes* was established by Friedrich (1968) for the seven nominal species *Amphiporus bergendali* Gering, 1912, *Amphiporus drepanophoroides* Griffin, 1898, *Amphiporus pacificus* Coe, 1905, *Amphiporus occidentalis* Coe, 1905, *Amphiporus punctatulus* Coe, 1905, *Cratenemertes danae* Friedrich, 1957, and *Cratenemertes madagascarensis* Kirsteuer, 1965 without type designation, and thus was unavailable under Article 13.3 of the International Code of Zoological Nomenclature (International Commission on Zoological Nomenclature 1999). To make *Nipponnemertes* nomenclaturally available, Gibson and Crandall (1989) designated *A. drepanophoroides* as the type species and provided a morphological diagnosis for the genus; the authorship of *Nipponnemertes* is thus ascribed to Gibson and Crandall (1989). Phylogenetic relationships among some members of *Nipponnemertes* were inferred by Kajihara et al. (2015), where four described—*Nipponnemertes bimaculata* (Coe, 1901), *Nipponnemertes ogumai* (Yamaoka, 1947), *Nipponnemertes pulchra* (Johnston, 1837), and *Nipponnemertes punctatula* (Coe, 1905)—and two undescribed congeners—*Nipponnemertes* sp. 1 (MCZ DNA105622) and *Nipponnemertes* sp. 2 (MCZ DNA105589) (Andrade et al. 2012)—were analyzed. The resulting tree indicated that the six species were divided into two groups, which seemed to be

characterized by the presence and absence of a cephalic patch (Kajihara et al. 2015); however, it has never been confirmed based on a more comprehensive dataset.

From the Northwest Pacific in eastern Asia, including the Sea of Okhotsk, the Sea of Japan, and the East and South China Seas, four species of *Nipponnemertes* have been recorded: *Nipponnemertes arenaria* (Uschakov, 1927), *N. bimaculata*, *N. ogumai*, and *N. punctatula* (Kajihara 2007a; Kajihara 2017; Chernyshev 2020). Besides, several authors reported unidentified forms of *Nipponnemertes* from Japanese (Yamaoka 2005; Kajihara 2017), Vietnamese (Chernyshev 2016), and Far East Russian waters (Chernyshev 2020), suggesting there is hidden nemertean diversity.

With the advent of molecular tools—DNA barcoding and molecular phylogeny—in monostiliferan systematics (e.g., Andrade et al. 2012; Kvist et al. 2014; Kvist et al. 2015; Leasi et al. 2016; Hookabe et al. 2020; Hookabe et al. 2021b), we are now in a position to obtain a more accurate picture of the species diversity. Some of the diagnostic characters traditionally used in monostiliferan taxonomy are often ambiguous and thus need to be revisited with molecular phylogeny. In the present study, we describe 10 new species of the genus from Japanese and Far East Russian waters. Since previous studies on molecular phylogenetic study in *Nipponnemertes* were biased toward shallow-water species (e.g., Kajihara et al. 2015) despite the bathymetrically wide distribution of this taxon, we extensively sampled specimens from intertidal to bathyal waters by various methods: by hand for intertidal species, SCUBA, dredging, sledging, and a remotely operated vehicle (ROV) equipped with a slurp gun. In the description part, histological observation (including cerebral organ, which is an important character for distinguishing *Nipponnemertes* from *Cratenemertes*) was performed to characterize the internal morphology of each taxon. Using molecular data obtained from the newly discovered species and described species from Japanese and Far East Russian waters, phylogenetic analyses based on partial sequences of nuclear and mitochondrial genes are performed to assess phylogenetic relationships as well as species boundaries within the genus.

3.1.2 Materials and Methods

Taxon sampling and morphological examination

In the present study, we collected cratenemertids from Japanese and Far East Russian waters in 2014–2021 by multiple methods (Fig. 6 and Table 2). Specimens were photographed with digital still cameras NIKON D5600 (NIKON, Japan) or OM-D E-M1 Mark II (Olympus, Japan)

while they were alive. Worms were anaesthetized in 7.5% MgCl₂ and gently placed between a slide glass and cover slip with a drop of anaesthetic solution so that ocelli and stylet apparatus could be observed under a light microscope. Afterwards, worms were picked up from the slides and the posterior tips were preserved in 99% ethanol for DNA extraction while the rest of the body was pre-fixed in 10% formalin in seawater, then post-fixed in Bouin's fluid for 24 hours, and transferred to 70% ethanol. For histological preparation, samples preserved in 70% ethanol were dehydrated in a series of ethanol solutions (70%, 80%, 90%, 95%, and 99% ethanol), cleared in xylene, and embedded in Surgipath Paraplast paraffin (Leica, Germany). Paraffin blocks were serially sectioned at 7 µm thickness and stained with Mallory's trichrome method (Gibson 1994). Samples were collected by SCUBA in Shizuoka, Kagoshima, and Okinawa, Japan, immediately transported to the laboratory. The living worms were photographed with a digital camera. To examine the morphology of proboscis, worms are physically stimulated, with the middle part of the body pressed by a glass cell spreader or otherwise dissected under a dissecting microscope. After anaesthetized by dropwise addition of MgCl₂ solution isotonic to seawater, the posterior tip of body was dissected and preserved in 99% ethanol for DNA extraction while the rest of the body was fixed with 10% formalin in seawater and then preserved in 70% ethanol. Voucher specimens have been deposited in Invertebrate Collection of the Hokkaido University Museum (ICHUM), Sapporo, Japan (Table 2).

DNA extraction, PCR amplification, and sequencing

Total DNA was extracted using the DNeasy Blood & Tissue Kit (Qiagen, Germany) according to the manufacturer's protocol. Each partial sequence was amplified with primer pairs summarized in Table 3 according to the following PCR protocol: preheating at 94°C for 2 min; 35 cycles of 94°C for 40 s, 52°C for 60–75 s, and 72°C for 60 s; then a final extension at 72°C for 7 min. PCR products were purified using Exo-SAP-IT (Thermo Fisher Scientific, USA). Sequencing reaction was performed with BigDye Terminator ver. 3.1 Cycle Sequencing Kit (Thermo Fisher Scientific, USA); for partial sequences of 18S rRNA (18S) and 28S rRNA (28S), internal primers listed in Table 3 were additionally used. The resulting dye-labelled products were purified with the EDTA/ethanol precipitation method and sent to GENEWIZ (Tokyo, Japan) for nucleotide sequencing. Sequences newly obtained in this study have been deposited in DNA Data Bank of Japan (DDBJ) under accession numbers listed in Table 2. Type and voucher specimens have been deposited in Invertebrate Collection of the Hokkaido

University Museum (ICHUM), Sapporo, Japan and Zoological Museum of Far East Federal University, Vladivostok, Russia.

Sequence alignment and phylogenetic analyses

Sequencing chromatographs were checked and edited with GeneStudio Pro ver. 2.2 (GeneStudio, Inc., USA). Sequences newly obtained in this study were combined with sequences available in GenBank listed in Table 5. For 16S, 18S, and 28S, data sets were aligned by MAFFT ver. 7 (Kato and Standley 2013) using the automatically selected strategy L-INS-i. After being aligned with ClustalW algorithm (Thompson et al. 1994) implemented in MEGA ver. 7 (Kumar et al. 2016), COI and H3 sequences were checked by translating into amino acids to see if they did not contain stop codons. Ambiguous sites were removed by Gblocks ver. 0.91b (Castresana 2000) under a less stringent option; a 657-bp sequence for COI, a 458-bp for 16S, a 1740-bp for 18S, a 1221-bp for 28S, and a 330-bp for H3 finally remained. After tree topology is checked for each marker and consistent with each other, partial sequences of COI, 16S, 18S, 28S, and H3 were concatenated using MEGA ver. 7.

To infer phylogenetic relationships among cratenemertids examined in the present study, molecular phylogenetic trees were reconstructed based on maximum-likelihood (ML) and Bayesian inference (BI) analyses. The ML analysis was performed with RaxML ver. 8.0.0 (Stamatakis 2014) under GTR + G + I selected as an optimum nucleotide substitution model by PartitionFinder ver. 2.1.1 (Lanfear et al. 2016) employing the greedy algorithm. Nodal support values were obtained from 1000 bootstrap pseudoreplicates. The BI tree was obtained using MrBayes ver. 3.2.3 (Ronquist et al. 2012) under GTR + G + I model selected by PartitionFinder ver. 2.1.1, launching two independent Metropolis-coupled analyses with four Markov chains for 10^7 generations and sampling every 100 generations from the chains. Run convergence was assessed by Tracer ver. 1.7 (Rambaut et al. 2018).

Uncorrected pairwise genetic distances were calculated based on 657-bp COI by MEGA ver. 7.

Species delimitation analyses

Using 658 bp of COI dataset, species delimitation analyses for Nipponnemertes species with cephalic patches were performed employing three methods: Automatic Barcode

Gap Discovery (ABGD) (Puillandre et al. 2012), Bayesian implementation of the PTP (bPTP) (Zhang et al. 2013), and statistical parsimony (Templeton et al. 1992). The ABGD analyses was performed on the online server (<https://bioinfo.mnhn.fr/abi/public/abgd/abgdweb.html>) with default settings. The bPTP analyses were performed based on a ML tree generated by RAxML ver. 8.0.0 with the COI dataset. The statistical parsimony analysis was performed using the TCS algorithm (Clement et al. 2000) implemented in PopART (Leigh & Bryant, 2015). Haplotype networks for individual loci were generated for visualizing relationships between haplotypes by using PopART.

3.1.3 Results

Phylogeny

In the present study, I performed molecular phylogenetic analyses with 59 specimens of *Nipponnemertes* (Fig. 7, 8). Of these, 10 species turned out to be new to science and described based on external (Fig. 9–11) and internal morphology (Fig. 13–21). In the resulting tree (Fig. 7), a clade constituted by 59 specimens of *Nipponnemertes* was sister to another clade formed by the two bathyal species, *Cratenemertidae* sp. IZ-45644 and *Cratenemertea* sp. 25DS (possibly pelagic juvenile of *Cratenemertidae* sp. IZ-45644) and an abyssal species, *Uniporus alisae* (Chernyshev and Polyakova 2018a). The *Nipponnemertes* clade was divided into three major clades (Fig. 7). The tree topology reconstructed by the ML analysis was almost identical with the BI tree. Below, we refer to the “*N. pulchra*–*Nipponnemertes* cf. *ogumai*” clade as Clade A, the “*N. incainca*–*N. punctatula*” clade as Clade B, and the “*N. lactea*–*N. ganahai*” clade as Clade C.

In Clade A, *Nipponnemertes* cf. *ogumai* from Sado (the Sea of Japan) (Fig. 9A, B) was a sister taxon of *N. ogumai* from Arahama (Pacific Ocean); they were nested in a clade along with *N. pulchra*, *Nipponnemertes* sp. DNA105622, *Nipponnemertes* sp. DNA105589 (Andrade et al. 2012), and *Nipponnemertes* sp. 1 (Fig. 9C, D). *Nipponnemertes* sp. 1 and *Nipponnemertes* sp. DNA105622 (Andrade et al. 2012) from California constituted a clade supported by 99% of bootstrap value (BS); between the two species, the genetic distances based on COI were 3.4% in uncorrected *p*-distance (Table 6), smaller than interspecific genetic distances previously reported in *Nipponnemertes* (Chernyshev and Polyakova 2022).

Within Clade B, *N. incainca* was first branched from the rest of the members in this clade. With full support value, *Nipponnemertes neonilae* collected from the Sea of Japan and Kuril Islands (Fig. 10L–P) was sister-related with a clade constituted by “*N. bimaculata*–*N. punctatula*” clade. *Nipponnemertes ojimaorum* collected from Koganezaki was nested in the “*N. bimaculata*–*N. punctatula*” clade, together with *Nipponnemertes jambio* and *Nipponnemertes crypta*.

Clade C was detected for the first time in this study; all members were herein newly included in molecular analyses. In this clade, *Nipponnemertes lactea* (Fig. 11K, L) was sister to the rest of the members. *Nipponnemertes ganahai* (Fig. 11A–G) formed a clade weakly supported by 60% of BS and 0.75 of PP with *N. kozaensis* (Fig. 11H–J), *N. notoensis* (Fig. 11M–O), *N. ornata* sp. nov. (Fig. 11P, Q), *N. sugashimaensis* (Fig. 11R–U), *Nipponnemertes* sp. 2 (Fig. 11V, W), and Cratenemertidae sp. Guam (Fig. 11X) (Chernyshev and Polyakova 2019).

Species delimitation for Clade B

I performed species delimitation analyses using partial sequences of COI (658 bp) to examine putative species boundaries among Clade B. Five distinct lineages (each corresponding to *N. bimaculata*, *N. crypta*, *N. jambio*, *N. ojimaorum*, and *N. punctatula*) were identified by all three methods (TCS, ABGD, and bPTP) (Fig. 8). Among them, the maximum value of interspecific genetic distance was 7.9% of uncorrected *p*-distance (*N. bimaculata*–*N. jambio* and *N. bimaculata*–*N. punctatula*); the minimum value was 1.6% between *N. crypta* and *N. ojimaorum* (Table 7).

Systematics

Family Cratenemertidae Friedrich, 1968

Genus *Nipponnemertes* Gibson & Crandall, 1989

(= *Collarenemertes* Chernyshev, 1993)

Type species: *Nipponnemertes drepanophoroides* Griffin, 1898.

Diagnosis [given by Friedrich (1968); Gibson and Crandall (1989), and Riser (1998)]: Cratenemertids with generally more than 10 eyes on each side of head in adults. Mouth and rhynchodaeum opening via common pore. Anterior cephalic furrows with secondary longitudinal grooves; latter faintly visible or invisible in small individuals. Mid-dorsal cephalic ridge present or absent. Rhynchocoel extending to posterior end of body, consisting of interwoven longitudinal and circular muscle fibres, without lateral pouches. Mid-dorsal vessel penetrating into rhynchocoel to form a single vascular plug. Apical organ and cephalic glands present. Large cerebral organs extending behind brain.

Nipponnemertes cf. ogumai

(Fig. 9A, B)

Material examined: Posterior tip used for DNA extraction, collected at depths of 8 m by SCUBA diving, Shukunegi, Sado Island, Niigata Prefecture, Japan (Figs 2, 6A), by Naoto Jimi, on 26 September 2019.

Description: Body 4.5 cm in length, 4.2–6.0 mm in width; body uniformly orange colored (Fig. 9A, B). Head conspicuously demarcated from trunk; cephalic patch absent; proboscis pore depressed; mid-dorsal cephalic ridge present but not well developed (Fig. 9B). Anterior cephalic furrows completely encircling body, meeting at midline on dorsal surface (Fig. 9B). Ocelli numerous, arranged in lateral margin of head (Fig. 9B).

Remarks: In the present study, we refer to the Sado specimen as *Nipponnemertes cf. ogumai* due to morphological similarity in the uniformly orange body. A genetic distances between *N. ogumai* collected from intertidal zone of Araiama, Pacific coast of Japan (Kajihara et al. 2015)

is 6.6% in uncorrected *p*-distance (Table 6), comparable with interspecific COI divergences among hoplonemerteans (Sundberg et al. 2016). Without samples for deposition in museum except the total DNA, we leave a species description to a future study when additional materials for further observation and museum deposition are available.

Nipponnemertes sp. 1

(Fig. 9C, D)

Material examined: ICHUM 8331, preserved in 99% ethanol; posterior tip used for DNA extraction, collected at depths of 87–89 m by dredging, off Jogashima, Kanagawa Prefecture (35°8.42'N, 139°34.74'E–335°8.22'N, 139°34.67'E), by Hiroshi Kajihara, on 19 February 2014.

Description: Body 1.2 cm in length, 0.8–1.2 mm in width; body uniformly pale-orange coloured (Fig. 9C, D). Head demarcated from trunk; cephalic patch absent; mid-dorsal cephalic ridge present (Fig. 9D). Anterior cephalic furrow completely encircling body, nearly M-shaped on dorsal surface (Fig. 9D). As many as 19 ocelli irregularly arranged on each side (Fig. 9D).

Remarks: A genetic distance between the present specimen and *Nipponnemertes* sp. DNA105622 collected from California at depths of 367–389 m (Andrade et al. 2012) is 3.2% in uncorrected *p*-distance (Table 6); these values fall within previously reported intraspecific COI divergences among hoplonemerteans (Sundberg et al. 2016). Due to the lack of museum deposition, we leave a species description to a future study.

Nipponnemertes crypta Hookabe, Kajihara & Chernyshev, 2022

[Japanese name: kakure-omen-himomushi]

(Figs. 10B–D, 12, 13)

Material examined: Holotype: ICHUM 8315, serial transverse sections of anterior tip to intestine, a posterior tip used for DNA extraction, bycaught with a gill net from 10 m depth, Sugashima, Mie Prefecture, Japan (Fig. 6A, D), collected by Natsumi Hookabe and Naoto Jimi, on 3 November 2021.

Habitat and geographic distribution: The species is only known from the type locality, off Sugashima, Mie, Japan, and collected from rock samples bycaught with a gill net. After rock samples were broken into pieces using a hammer, the specimen was found in a rock crack, probably as a result of erosion by rock boring sipunculans, which sympatrically occur around the studied area in Sugashima.

Etymology: The specific name is derived from Greek, *kryptós* (concealed, hidden), referring to cryptic habitat of the new species, among a subtidal rock crack.

Description: *External features.* Body 4.2 cm in length, 5 mm in width; body dorsally pale brown and ventrally lighter colored than dorsal surface (Fig. 10B–D). Head slightly demarcated from trunk, with a pair of pale brown cephalic patches (Fig. 10C, D). Narrow longitudinal midline present between cephalic patches but not reaching to anterior tip of head (Fig. 10C). Anterior cephalic furrow incompletely encircling body, opening at mid-dorsal line (Fig. 10C). Posterior cephalic furrow V-shaped on dorsal surface (Fig. 10C). Twenty ocelli in an irregular cluster on each side of head (Fig. 10C). Cerebral ganglia visible through body wall as a pair of red spots (Fig. 10C, D).

Internal morphology. Epidermis 30–38 μm thick, with numerous red- and yellow-stained acidophilic cells, blue-stained basophilic cells, and ciliated cells (Fig. 13A, B). Dermis up to 39 μm thick, forming cup-like structures on distal surface (Fig. 13A, B). Body wall musculature with outer circular and inner longitudinal muscle layers (Fig. 13A, B); distinct diagonal muscle layers not found. Precerebral septum present (Fig. 13F). Oesophagus with red-stained acidophilic cells, blue-stained basophilic cells, and ciliated cells (Fig. 13G, J, K). Dorso-ventral muscles present in cerebral region (Fig. 13G) and between intestinal diverticula (Fig. 13L). Cephalic glands (Fig. 13D) leading to well-developed submuscular glands (Fig. 13A, E). A single apical organ present (Fig. 13C). Precerebral vessels transversely connected above rhynchodaeum at anterior tip of head, running posteriorly in both sides of rhynchodaeum (Fig.

13E). Mid-dorsal vessel protruding into rhynchocoel to form a single vascular plug in posterior region of brain (Fig. 13G); mid-dorsal vessel containing red- to orange-colored blood cells (Fig. 13K).

Proboscis pore opening ventrally (Fig. 13D). Rhynchocoel muscular wall interwoven with circular and longitudinal musculatures (Fig. 13G, J–L). Proboscis epithelium with well-developed papilla (Fig. 13H, I). Proboscis musculature composed of outer circular, middle longitudinal, and inner circular muscle layers (Fig. 13H, I). Proboscis nerves 13 in number (Fig. 13H). Stylet basis oval, 33.6 μm in length and 22.0 μm in maximum width; central stylet smooth, 72.5 μm in length; stylet-to-basis-length (S/B) ratio 2.16 (Fig. 12). Two accessory stylet pouches present, each containing 9 stylets (Fig. 12).

Pigment-cup ocelli embedded in body-wall longitudinal muscle layer (Fig. 13A, D, E). Cerebral organs unbranched, extending to posterior region of brain (Fig. 13F, J) and posteriorly followed by developed gland cells (Fig. 13G). Brain separated from body wall by outer neurilemma; inner neurilemma absent (Fig. 13G, J); dorsal lobe with glomerular structures (Fig. 13J). Lateral nerves without accessory nerves and neurochords (Fig. 13L).

Nephridial tubules anteriorly extending to brain region, forming a bundle of tubules behind brain region (Fig. 13K). Testes between intestinal diverticula (Fig. 13L).

Remarks: *Nipponnemertes crypta* is distinguished from all the congeners in the body coloration and cephalic patches (Fig. 10C, D). Body coloration of *N. pacifica* (Coe, 1905) described based on preserved specimens is most similar to, but distinguished from *N. crypta* in lacking cephalic patches. The same is true for the Antarctic species *N. variabilis* (Korotkevitch, 1983); in addition, *N. crypta* differs from *N. variabilis* in the absence of accessory stylet in a central stylet basis (Fig. 12).

Nipponnemertes jambio Hookabe, Kajihara & Chernyshev, 2022

[Japanese name: jyambio-omen-himomushi]

(Fig. 10E–H, 12)

Cf. *Cosmocephala japonica*: Kajihara, 2017, p. 436, Fig. 16F, G.

Material examined: Holotype: ICHUM 8319, preserved in 99% ethanol, posterior tip used for DNA extraction, collected by dredging from 87–89 m depth, off Jogashima, Sagami Bay, Kanagawa Prefecture, Japan (Fig. 6A, C), by Hiroshi Kajihara, on 19 February 2014. Paratype: ICHUM 8318, preserved in 99% ethanol, posterior tip used for DNA extraction, collected by dredging from 80 m depth, off Jogashima, Sagami Bay, Kanagawa Prefecture, Japan (Fig. 6A, C), by Hisanori Kohtsuka, on 25 April 2012.

Habitat and geographic distribution: The species is only known from the type locality, off Jogashima, Miura (Kanagawa), Japan.

Description: *External features.* Body 2.0–2.2 cm in length, 1.0–1.4 mm in width; in trunk region, dorsal body surface uniformly brownish orange; ventral surface slightly lighter colored than dorsal surface (Fig. 10E–H). Head with a pair of cephalic patches and a narrow longitudinal midline between cephalic patches on dorsal surface (Fig. 10E, F); mid-longitudinal line without reaching to anterior end of cephalic patches (Fig. 10E, F); ventral surface pale colored (Fig. 10G). Anterior cephalic furrows incompletely encircling body, dorsally discontinuous at midline (Fig. 10F), ventrally extending anteriorly and meeting at midline (Fig. 10G). Posterior cephalic furrows V-shaped on dorsal side (Fig. 10H). Eyes 14–17 in number, present on each side of lateral margin of head (Fig. 5F, H).

Internal morphology. Stylet basis oval, 45.0–60.0 μm in length and 35.0–48.0 μm in maximum width; central stylet smooth, 95.0–120.0 μm in length; S/B ratio 1.86–2.15 (Fig. 12). Two accessory stylet pouches present, each containing 4–5 stylets (Fig. 12).

Remarks: In external morphology, *N. jambio* is extremely similar to *N. ojimaorum*. However, it is distinguished from the latter in the following features: *i*) trunk region ventrally lighter coloured than dorsal surface (Fig. 10G), *ii*) a narrow longitudinal midline (mid-dorsal cephalic ridge) between cephalic patches reaching more anteriorly than *N. ojimaorum*, without fusing with cephalic patches (Fig. 10F).

Intraspecific COI divergences in this species are 0.0–0.3% in uncorrected *p*-distance (Table 7).

Nipponnemertes neonilae Hookabe, Kajihara & Chernyshev, 2022

[Japanese name: rishiri-himomushi]

(Figs. 10L–P, 12, 14)

Amphiporus bimaculatus: Iwata, 1954

Collarenemertes bimaculatus: Chernyshev, 1993

Collarenemertes cf. bimaculata: Chernyshev & Polyakova, 2019

Material examined: Holotype: MIMB 43013, preserved specimen, a proboscis used for DNA extraction, collected by Alexei V. Chernyshev from 5 m depth, among *Modiolus kurilensis*, Vostok Bay, Peter the Great Bay (Fig.6A), Sea of Japan, Russia, 11 August 2015. Paratypes: two specimens, MIMB 43014 and 43015, transverse and sagittal sections of anterior tips, collected by Alexei V. Chernyshev from topotype. Additional specimens: (GenBank specimen ID NipbimIt15), collected by V.G. Kuznetsov from 5–15 m depth, Iturup Island, 45°22.397'N, 148°27.296'E, Sea of Okhotsk (Fig. 6A), 19 August 2019; 16 specimens from Sea of Japan and Kuril Islands (Zoological Museum of Far East Federal University and personal collection of AVC).

Habitat and geographic distribution: The species is known from the type locality (Vostok Bay) and Kuril Islands (Iturup Island). Externally identical specimens were collected in Posyet, Ussuri, and Vladimir Bays, Valentin Bight (Sea of Japan), Aniwa Bay (South Sakhalin), Shikotan Island (Kuril Island), and Rausu, Hokkaido (Sea of Okhotsk) (Hookabe and Jimi, personal observation) from the intertidal to a depth of 14 m. As well, the species was reported from Rishiri Island, Hokkaido (Sea of Japan) (Iwata 1954). The present species is extracted from substrate samples, in which sedentary mytilids (*Modiolus kurilensis* and *Crenomytilus grayanus*) and calcareous algae (*Bossiella* sp.) were included.

Etymology: The specific name is a noun in the genitive case, after Dr. Neonila Polyakova, to whom we acknowledge her significant contribution to molecular systematics of nemerteans.

Description: *External features.* Body 5–16 cm in length (12 cm in holotype), 5–6 mm in width (up to 12–15 mm in contracted specimens); body dorsally brownish-red or brick-red and ventrally beige or whitish (Fig. 10L). Head triangular from above, with a pair of darker triangular cephalic patches and a longitudinal midline between cephalic patches on dorsal surface; mid-longitudinal line not reaching to anterior tip of head and not fused with cephalic patches (Fig. 10M, N). Narrow longitudinal midline between cephalic patches not reaching to anterior tip of head (Fig. 10M, N, P). When contracted, head can be retracted into body, forming a collar (Fig. 14D). Anterior cephalic furrow incompletely encircling body, opening at mid-dorsal line, with secondary grooves (Fig. 10O). Posterior cephalic groove V-shaped on dorsal surface (Fig. 10M). Eyes 12–23 in number, present on each side of head (Fig. 10P).

Internal morphology. Epidermis with numerous ciliated and orange-stained glandular cells. Dermis up to 60–80 μm thick in contracted specimens. Body-wall musculature as in other species in the genus, diagonal muscles detected by confocal laser scanning microscopy (Chernyshev 2010: fig. 3). Pylorus sporadically with glandular cells; intestinal caecum anteriorly extending beneath pylorus (Fig. 10P), with 5–6 pairs of lateral pouches. Dorso-ventral muscles present between intestinal diverticula (Fig. 14F). Cephalic glands (Fig. 14B) and the subsequent submuscular glands as in other species in the genus. Precerebral vessel extending above rhynchodaeum at anterior tip of head and posteriorly bifurcated (Fig. 14A). Mid-dorsal vessel protruding into rhynchocoel to form a single vascular plug in posterior region of brain (Fig. 14D, E); blood cells colorless.

Proboscis pore sub-terminal, opening ventrally. Rhynchocoel musculature consisting of interwoven longitudinal and circular muscle fibres (Fig. 14E, F). Proboscis nerves 16–17 in number. Stylet basis oval, 118–156 (156 in holotype) μm in length and 80–97 (94 in holotype) μm in maximum width; central stylet smooth, 152–222 (218 in holotype) μm in length; S/B ratio 1.14–1.67 (1.39 in holotype) (Fig. 12). Two (rarely four or five) accessory stylet pouches present, each containing 3–10 (7 in holotype) stylets (Fig. 12).

Pigment-cup ocelli embedded in body-wall muscle layer (Fig. 14A). Cerebral organs extending to posterior region of brain, posteriorly bifurcated (Fig. 14C); posterior glandular

part located between lateral blood vessel and lateral nerve (Fig. 14E). Brain and lateral nerves as in other species of the genus.

Nephridial tubules branching near lateral blood vessels. Gonads, gonoducts, and gonopores not found in the material examined.

Remarks: In external morphology, *N. neonilae* is very similar to *N. bimaculata*. However, it is distinguishable from the latter by the presence of a narrow longitudinal midline (mid-dorsal cephalic ridge) between cephalic patches (Fig. 10M, N, P). In the original description of *N. bimaculata*, Coe (1901) wrote that the head had two oval, black or very dark brown patches in material from Victoria (British Columbia, Canada), Sitka (Alaska, USA), and Puget Sound (Washington, USA). Later, Coe (1905: pl. 2, fig. 21) illustrated a specimen with two narrow triangular cephalic patches. Photographs of the similar specimens are provided in BOLD (http://v3.boldsystems.org/index.php/Taxbrowser_Taxonpage?taxid=939114). Dr. Frank Crandall (personal communication) regarded that specimens of *N. bimaculata* with oval and triangular cephalic patches belong to two different species. If this is the case, specimens with oval head spots should be assigned to *N. bimaculata*. *Nipponnemertes neonilae* is distinguished from *N. bimaculata* in the Northeast Pacific in having triangular patches by wider and darker cephalic patches.

From Japanese waters, Iwata (1954) reported a similar form with the present species from sublittoral among laminarian holdfasts, Rishiri Island (Hokkaido), the Sea of Japan. As noted in Crandall et al. (2002), the form differs from Coe's (1901) description of *N. bimaculata* in having triangular cephalic patches, and Crandall et al. (2002) noted that the form identified by Iwata (1954) differs from Coe's (1901) taxon in having a pair of quadrangular head markings, and rather resembles *N. neonilae*. Therefore, we herein redescribe Iwata's (1954) taxon as *N. neonilae*.

Nipponnemertes ojimaorum Hookabe, Kajihara & Chernyshev, 2022

[Japanese name: benishouga-omen-himomushi]

(Figs. 10I–K, 12, 15)

Material examined: Holotype: ICHUM 8324, serial transverse sections of anterior tip to intestine, posterior tip used for DNA extraction, collected by SCUBA diving from 12 m depth, Koganezaki, Shizuoka Prefecture, Japan (Fig. 6A, C), by Masanori Okanishi, on 21 October 2020. Paratypes: ICHUM 8325, fixed in Bouin's fluid and later preserved in 70% ethanol, collected by SCUBA diving from 8 m depth, Koganezaki, Shizuoka Prefecture, Japan (Fig. 6A, C), by Natsumi Hookabe, on 6 January 2021; ICHUM 8326, fixed in Bouin's fluid and later preserved in 70% ethanol, collected by SCUBA diving from 7 m depth, Koganezaki, Shizuoka Prefecture, Japan (Fig. 6A, C), by Naoto Jimi, on 6 January 2021.

Habitat and geographic distribution: The species has been currently known only from Koganezaki (Shizuoka), Japan, inhabiting subtidally under rocks on sandy substrates (Fig. 6E).

Etymology: The specific name is a noun in the genitive plural, named after Tomohito Ojima and Masako Ojima, who organized a field survey on subtidal invertebrate fauna in Koganezaki; all the specimens of the new species were collected during the survey.

Description: *External features.* Body 2.2 cm in length, 1.2 mm in width; in trunk region, dorsal body surface uniformly bright red colored; ventral surface the same color as dorsal surface (Fig. 10I–K). Head with a pair of cephalic patches on dorsal surface (Fig. 10J); ventral surface pale colored (Fig. 10K). Narrow longitudinal midline between cephalic patches not reaching to anterior tip of head (Fig. 10J). Anterior cephalic furrows incompletely encircling body, dorsally discontinuous at midline (Fig. 10J), ventrally extending anteriorly and meeting at midline (Fig. 10K). Posterior cephalic furrows V-shaped on dorsal side (Fig. 10J). Eyes arranged on each side of lateral margin (Fig. 10J); number of eyes not counted for the present species.

Internal morphology. Epidermis with numerous red-stained acidophilic cells and ciliated cells (Fig. 15A, B). Dermis up to 30 μm thick, forming cup-like structures on distal surface in precerebral region (Fig. 15A); in foregut region more flattened and without forming conspicuous cup-like structure (Fig. 10B). Body-wall musculature (Fig. 15A, B), esophagus (Fig. 15G), pylorus, intestinal caecum (Fig. 15H, K) as in other species in the genus. Dorso-ventral muscles present in cerebral region (Fig. 15J) and in intestinal region between intestinal diverticula (Fig. 15L). Cephalic glands and the subsequent submuscular glands as in other species in the genus (Fig. 15A). A single apical organ present (Fig. 15C). Precerebral vessels

transversely connected above rhynchodaeum at anterior tip of head (Fig. 15E). A single vascular plug present in posterior region of brain (Fig. 15G); mid-dorsal vessel not well distinguished thorough histology in the present species.

Proboscis pore positioned sub-terminally (Fig. 10K). Rhynchocoel musculature (Fig. 15G), proboscis epithelium (Fig. 15H), and proboscis musculature (Fig. 15H, I) as in other species in the genus. Proboscis nerves 13 in number (Fig. 15H). Stylet basis oval, 53.0 μm in length and 32.0 μm in maximum width; central stylet smooth, 98.0 μm in length; S/B ratio 1.85 (Fig. 12). Two accessory stylet pouches present, each containing 7–8 stylets (Fig. 12).

Pigment-cup ocelli embedded in body-wall muscle layer (Fig. 15A). Cerebral organs unbranched (Fig. 15F, J) and followed by glands (Fig. 15G) as in other species in the genus. Lateral nerves lacking accessory nerves and neurochords (Fig. 15K, L).

Nephridial tubules forming a bundle of tubules behind brain region (Fig. 15K). Gonads, gonoducts, and gonopores not found in the specimen examined.

Remarks: *Nipponnemertes ojimaorum* is similar to *N. arenaria* (Fig. 10A), *N. bimaculata*, *N. crypta* (Fig. 10B–D), *N. jambio* (Fig. 10E–H), *N. neonilae* (Fig. 10L–P), and *N. punctatula* (Fig. 10Q–T) because these species possess cephalic patches in common. However, *N. ojimaorum* can be differentiated from these species by the coloration of the dorsal and ventral body surfaces, which is the same in *N. ojimaorum* (Fig. 10I–K), whereas the dorsal surface is darker than the ventral in other species. In terms of the color contrast between the dorsal and ventral body surfaces, *N. ojimaorum* most closely resembles *N. jambio* because the ventral surface in the latter is only slightly lighter than the dorsal surface; in other species (*N. arenaria*, *N. bimaculata*, *N. crypta*, *N. neonilae*, and *N. punctatula*), the contrast is more prominent with a darker dorsal surface against paler ventral one. Still, *N. ojimaorum* is distinguishable from *N. jambio* by the mid-dorsal cephalic line, which is *i*) not reaching to the anterior end of the cephalic patches, *ii*) laterally fusing to the cephalic patches, and *iii*) posteriorly fusing to the dorsal dark color (Fig. 10J); in *N. jambio*, the mid-dorsal cephalic line *i*) extends anteriorly almost reaching to the level of the anterior end of the cephalic patches, *ii*) is laterally separated from the cephalic patches, and *iii*) is not continuous with the dorsal dark color (Fig. 10F).

Cosmocephala japonica Stimpson, 1857—currently *Amphiporus japonica* (Stimpson, 1857)—is anticipated to be a species in *Nipponnemertes* (Kajihara et al. 2015; Kajihara 2017a); it was originally described based on specimens intertidally collected from Shimoda (Shizuoka),

geographically close to Koganezaki, the type locality of *N. ojimaorum*. However, by having ventrally pale-colored body surface, *Cosmocephala japonica* is distinguished from *N. ojimaorum*.

Nipponnemertes punctatula (Coe, 1905)

[Japanese name: madara-himomushi]

(Fig. 10Q–T)

Material examined: ICHUM 8328, preserved in formalin, posterior tip used for DNA extraction, collected at intertidal, Tateyama, Mie Prefecture, Japan (Fig. 6A, C), by Ryuta Yoshida, on 13 June 2018. ICHUM 8329, preserved in formalin, posterior tip used for DNA extraction, collected at intertidal, Sugashima, Mie Prefecture, Japan (Fig. 6A, D), by Naoto Jimi, on 24 June 2021.

Habitat and geographic distribution: The species is widely known from Japanese coastal waters from Hokkaido to Nagasaki.

Description: External features. Body 4.0–5.2 cm in length, 8–12 mm in width; in trunk region, dorsal body surface with a dorsal brown blotch pattern (Fig. 10Q, R) or uniformly brownish (Fig. 10S, T); ventral surface pale-colored (Fig. 10T). Head with a pair of cephalic patches and a narrow longitudinal midline between cephalic patches on dorsal surface; mid-longitudinal line without reaching to anterior end of cephalic patches (Fig. 10Q–T)). Posterior cephalic furrows V-shaped on dorsal side (Fig. 10Q).

Remarks: Body coloration pattern of the present species varies from previously known brown blotch pattern (Fig. 10Q, R) to uniformly brown color without blotched (Fig. 10S, T). Intraspecific COI divergences in this species are 0.0–0.2% in uncorrected *p*-distance (Table 7), suggesting intraspecific variation of body coloration pattern in *N. punctatula*.

Nipponnemertes ganahai Hookabe, Kajihara & Chernyshev, 2022

[Japanese ganaha-omen-himomushi]

(Figs. 11A–G, 12, 16)

?*Nipponnemertes* sp. 1: Yamaoka, 2005, p. 151–152.

Material examined: Holotype: ICHUM 8317, serial transverse sections of anterior tip to intestine, a posterior tip used for DNA extraction, collected by dredging from 116–211 m depth, off Jogashima, Miura, Kanagawa Prefecture, Japan (Fig. 6A, D), by Natsumi Hookabe, on 31 July 2020. Paratypes: ICHUM 8316, serial transverse sections of anterior tip to intestine, a posterior tip used for DNA extraction, collected by dredging from 71.2–79.2 m depth, off Noto, Ishikawa Prefecture, Japan (Fig. 6A, B), by Natsumi Hookabe, on 20 November 2019.

Habitat and geographic distribution: The species has been collected off Jogashima (Kanagawa Prefecture), Noto (Ishikawa Prefecture), Ofunato (Iwate Prefecture), and Tosa (Kochi Prefecture), Japan, likely preferring muddy to sandy sediments (Fig. 6F, G).

Etymology: The specific name is a noun in the genitive case, after Ikumasa Ganaha, to whom we acknowledge his significant contribution to molecular experiments including DNA extraction, PCR amplification, sequencing, and organizing the obtained molecular data for several specimens used in the present study.

Description: *External features.* Body 1.2–1.6 cm in length, 1.5–2.0 mm in width; body uniformly pale pink to yellow colored (Fig. 11A–E). Head not conspicuously demarcated from trunk; cephalic patch absent (Fig. 11A–D). Anterior cephalic furrows completely encircling body, extending anteriorly and meeting at midline on ventral side (Fig. 11F). Posterior cephalic furrows V-shaped on dorsal surface, completely encircling body (Fig. 11B). As many as 15–21 ocelli irregularly arranged on each side, varying in sizes; 2 to 3 ocelli fused in several specimens (Fig. 11C, F, G).

Internal morphology. Epidermis with numerous red-, yellow-, and blue-stained cells and ciliated cells (Fig. 16A, B). Dermis up to 25 μm thick, forming cup-like structures on distal surface (Fig. 11A, B). Body-wall musculature (Fig. 16A, B), oesophagus (Fig. 16H, I), pylorus, and intestinal caecum (Fig. 16F) as in other species in the genus. Dorso-ventral muscles present between intestinal diverticula (Fig. 16K). Cephalic glands (Fig. 16C) leading to well-developed submuscular glands (Fig. 16A, D). A single apical organ present (Fig. 16C). Precerebral vessels transversely connected above rhynchodaeum at anterior tip of head (Fig. 16D). Mid-dorsal vessel present between rhynchocoel and pylorus (Fig. 16F); vascular plug not detected in the present species.

Proboscis pore sub-terminal, opening ventrally (Fig. 11F). Rhynchocoel musculature, proboscis epithelium, and proboscis musculature (Fig. 16F, G) as in other species in the genus. Proboscis nerves 10 in number (Fig. 16F). Stylet basis oval, 57.0–71.0 μm in length 45.0–47.0 μm in maximum width; central stylet vertically sculptured, 89.0–92.0 μm in length; S/B ratio 1.29–1.56 (Fig. 12). Two accessory stylet pouches present, each containing four stylets (Fig. 12).

Pigment-cup ocelli embedded sub-dermally (Fig. 16A, C, D). Cerebral organs ventrolaterally opening in precerebral region (Fig. 16E) and extending to posterior region of brain (Fig. 16H, I), followed by glandular cells (Fig. 16J). Brain (Fig. 16H–J) and lateral nerves (Fig. 16F, K) as in other species of the genus.

Nephridial tubules branching near lateral nerves (Fig. 16F). Ovaries between intestinal diverticula; each ovary containing 10 or more oocytes; oocytes 100–150 μm in diameter (Fig. 16K).

Remarks: *Nipponnemertes ganahai* resembles *N. africana* (Wheeler, 1940), *N. danae* (Friedrich, 1957) (probably synonym of *N. pulchra* in morphological basis (Berg 1985)), *N. magna* (Punnett, 1903), *N. pulchra* (Johnston, 1837), and *N. sanguinea* Riser, 1998 in having pale pink to yellow body (Fig. 11A–E) and intestinal caecum extending to pyloric region (Fig. 16F) and anteriorly extending in posterior region of esophagus. However, it is distinguished from *N. pulchra* (also probably from *N. danae*) in lacking an accessory stylet in the basis of the central stylet armature (Fig. 12). Between the remaining three species, the number of the proboscis nerves can be useful for species distinction; it is 10 in *N. ganahai*, 20 in *N. magna* (Punnett 1903), and 12 in *N. sanguinea* (Riser 1998). Furthermore, *N. sanguinea* is

distinguished from *N. ganahai* in the red-colored fluid contained in blood vessels due to red blood corpuscles (Riser 1998); red blood was not visible in a live squeezed specimen of *N. ganahai* sp. nov. through a light microscope (Fig. 11G).

The present species externally resembles *Nipponnemertes* sp. 1 reported by Yamaoka (2005) from a beach in Shitaru (near Shimoda (Shizuoka)) in having a pale body. Further studies with DNA barcoding are needed for Yamaoka (2005)'s unidentified forms.

Intraspecific COI divergences in this species are 0.0–0.8% in uncorrected *p*-distance (Table 8).

Nipponnemertes kozaensis Hookabe, Kajihara & Chernyshev, 2022

[Japanese name: koza-sujinashi-himomushi]

(Figs. 11H–J, 12, 17)

?*Nipponnemertes* sp. 2: Yamaoka, 2005, p. 152–153.

Material examined: Holotype: ICHUM 8320, serial transverse sections of anterior tip to intestine, posterior tip used for DNA extraction, found during anaesthetization for ascidians in menthol-floated seawater; the ascidians were collected by Naohiro Hasegawa from 12 m depth by SCUBA diving, Kaminoshima, Koza, Wakayama Prefecture (Fig. 6A), Japan, on 11 May 2021.

Habitat and geographic distribution: The present specimen was found among rocky substrate samples. In the substrate samples, several sessile invertebrates such as sponges, ascidians, and bryozoans were colonized. Currently known from the type locality, Koza (Wakayama Prefecture), Japan.

Etymology: The specific name is derived from the type locality, Koza, Wakayama Prefecture, Japan.

Description: *External features.* Body 1.5 cm in length, 1.0 mm in width; background generally pale-colored, mottled with red-brown and white dots; red-brown dots more densely distributed in dorso-anterior region of body; white dots uniformly distributed on both dorsal and ventral surfaces (Fig. 11H–J). Head devoid of red-brown pigments and slightly demarcated from body (Fig. 11I). Anterior cephalic furrows not conspicuous, probably discontinuous on dorsal midline, but ventrally meeting at midline (Fig. 11I, J). Posterior cephalic furrows completely encircling body and V-shaped on dorsal side (Fig. 11I, J). As many as 14 eyes arranged in two irregular rows at lateral margin on each side of head (Fig. 11I). Internal organs visible through body wall; proboscis pure white, extending three-fourths the body; foregut and intestine yellow colored (Fig. 11H).

Internal morphology. Epidermis with numerous red-, yellow-, blue-stained cells and ciliated cells (Fig. 17A, B). Dermis thin, up to 20 μm thick, forming cup-like structures on distal surface (Fig. 17A, B). Body-wall musculature (Fig. 17A, B), oesophagus (Fig. 17I), pylorus, and intestinal caecum (Fig. 17E) as in other species in the genus. Intestinal caecum anteriorly extending beneath pylorus (Fig. 17E). Dorso-ventral muscles present between intestinal diverticula (Fig. 17K). Cephalic glands (Fig. 17D) followed by subsequent submuscular glands (Fig. 17H). A single apical organ present (Fig. 17C). Precerebral vessels as in other species in the genus (Fig. 17D). Mid-dorsal vessel passing above pylorus and containing red to orange blood cells (Fig. 17E); vascular plug indistinguishable in the specimen examined.

Proboscis pore sub-terminal, opening ventrally (Fig. 11J). Rhynchocoel musculature (Fig. 17E, J), proboscis epithelium (Fig. 17E, F, J), and proboscis musculature (Fig. 17E–G, J) as in other species in the genus. Proboscis nerves 10 in number (Fig. 17F). Stylet basis oval, 49.6 μm in length and 32.0 μm in maximum width; central stylet vertically sculptured, 75.0 μm in length; S/B ratio 1.51 (Fig. 12). Two accessory stylet pouches present, each containing 8 stylets (Fig. 12).

Pigment-cup- ocelli embedded sub-dermally (Fig. 17A, H). Cerebral organs extending to posterior region of brain (Fig. 17I). Brain (Fig. 17I) and lateral nerves (Fig. 17E, J, K) as in other species of the genus.

Nephridial tubules branching near lateral blood vessels, latter containing red to orange blood cells (Fig. 17J). Gonads, gonoducts, and gonopores not found in the specimen examined.

Remarks: Currently five species have been known to possess pale colored body mottled with red to brown dots in the genus: *N. occidentalis* (Coe, 1905), *N. madagascarensis* (Kirsteuer, 1965), *N. kozaensis*, *N. ornata*, and *N. sugashimaensis* (Fig. 11). *Nipponnemertes kozaensis* is distinguished from the two previously known species, *N. occidentalis* and *N. madagascarensis*, by having submuscular glands originated from cephalic glands, although it could not be traced because of poor fixation or staining in *N. occidentalis* and *N. madagascarensis* and its validity as a diagnostic character needs to be reexamined in future study. In addition, *N. kozaensis* differs from *N. madagascarensis* in having 10 proboscis nerves (Fig. 17F) and 8 accessory stylets in each pouch (Fig. 12), whereas *N. madagascarensis* possesses 9 proboscis nerves and 2–3 accessory stylets per pouch (Kirsteuer, 1965). No information as to the proboscis in *N. occidentalis* is available, because the proboscis was lost in the type material (Coe, 1905). According to the original description, one of the notable anatomical features found in *N. occidentalis* is the complex alimentary canal with oesophageal and cardiac caeca (Coe, 1905, pl. XX, fig. 121). In the species herein described, including *N. kozaensis*, complex alimental canals as depicted by Coe (1905) were not observed. This would also allow the distinction of *N. occidentalis* from *N. kozaensis*.

In contrast to *N. madagascarensis* and *N. occidentalis*, no conspicuous difference is found in the internal morphology between *N. kozaensis*, *N. ornata*, and *N. sugashimaensis*. *Nipponnemertes ornata* differs from *N. kozaensis* in the lower density of brown dots on the body surface (Fig. 11P, Q). Although the species distinction is molecularly supported by the resulting phylogenetic tree, where the two species were not in a sister relation (Fig. 7), further observation based on additional materials is required to examine if any subtle differences in the spotting pattern might represent interspecific variation and thus could be used for species discrimination. A brown color pattern similar to the one found in *N. kozaensis* is also present in *N. sugashimaensis* but the pattern is irregular in the latter species (Fig. 11R) rather than the uniformly distributed dots in *N. kozaensis* (Fig. 11H).

The present species externally resembles *Nipponnemertes* sp. 2 reported by Yamaoka (2005) from Susaki and Shirahama in having a pale body with brown dots. Further studies with DNA barcoding are needed for Yamaoka (2005)'s unidentified forms.

In the resulting molecular phylogenetic tree (Fig. 7), *N. kozaensis* formed a clade with *N. sugashimaensis* with high support values. Although we could not obtain COI sequences for

them even with specific primers for their delimitation, the nucleotide differences between the two species were comparable with interspecific divergences observed in *Nipponnemertes*.

Nipponnemertes lactea Hookabe, Kajihara & Chernyshev, 2022

[Japanese name: irojiro-sujinashi-himomushi]

(Figs. 11K, L, 12, 18)

?*Nipponnemertes* sp. 1: Yamaoka, 2005, p. 151–152

Material examined: Holotype: ICHUM 8321, serial transverse sections of anterior tip to intestine, a posterior tip used for DNA extraction, collected at depths of 98–120 m by dredging, Takase, off Shimoda, Shizuoka Prefecture, Japan (Fig. 6A, C), by Natsumi Hookabe, on 25 March 2021.

Habitat and geographic distribution: The species is only known from the type locality, off Shimoda (Shizuoka), Japan.

Etymology: The specific name is derived from the Latin *lacteus*, meaning “milky”, after the uniformly pale body coloration in this species.

Description: *External features.* Body 0.7 cm in length, 0.8 mm in width; body uniformly pale; intestinal region pale-orange due to internal organs (Fig. 11K). Head demarcated from trunk and without cephalic patches. Anterior cephalic furrows completely encircling body. Posterior cephalic furrows V-shaped on dorsal surface, completely encircling body (Fig. 11K). As many as 12 eyes arranged on each side of lateral margin (Fig. 11L).

Internal morphology. Epidermis with numerous red-stained cells and ciliated cells (Fig. 18A, B). Dermis up to 25 µm thick, forming cup-like structures on distal surface in precerebral region (Fig. 18A); in foregut to intestine regions, dermis becoming thicker and more flattened without

forming cup-like structure (Fig. 18B). Body-wall musculature (Fig. 18A, B); oesophagus, pylorus, and intestinal caecum (Fig. 18I) as in other species of the genus. Dorso-ventral muscles present between intestinal diverticula (Fig. 18I). Cephalic glands (Fig. 18D) leading to well-developed submuscular glands. A single apical organ present (Fig. 18C). Precerebral vessels, mid-dorsal vessel, and vascular plug difficult to be confirmed in type material.

Proboscis pore opening ventrally (Fig. 11L). Rhynchocoel musculature (Fig. 18G–I), proboscis epithelium (Fig. 18E), and proboscis musculature (Fig. 18F) as in other species in the genus. Proboscis nerves 10 in number (Fig. 18E). Stylet basis oval, 72.0 µm in length and 40.0 µm in maximum width; central stylet smooth, 90.0 µm in length; S/B ratio 1.25 (Fig. 12). Two accessory stylet pouches present, each containing 7 stylets (Fig. 12).

Pigment-cup ocelli embedded sub-dermally (Fig. 18A). Cerebral organs unbranched, extending to posterior region of brain (Fig. 18G, H). Brain (Fig. 18G, H) and lateral nerves (Fig. 18I, J) as in other species in the genus.

Nephridial tubules difficult to be confirmed in type material. Ovaries between intestinal lateral diverticula; each ovary containing 20 or more oocytes; oocytes 42 µm in diameter (Fig. 18J).

Remarks: *Nipponnemertes lactea* resembles *N. africana* (Wheeler, 1940), *N. notoensis* (present study), and *N. variabilis* (Korotkevitch, 1983) in having uniformly pale colored body, posterior cephalic furrows completely encircling body, and the circulatory system lacking red blood corpuscles. *Nipponnemertes lactea* is characterized by the ocelli arranged in a single row at lateral margin of the head (Fig. 11L). As depicted in the original illustration based on a preserved specimen (Wheeler, 1940: fig. 9B, C), the ocelli in *N. africana* are arranged irregularly, and the similar arrangement is found in *N. notoensis* (Fig. 11N). *Nipponnemertes lactea* differs from *N. variabilis* in the absence of accessory stylet in the central stylet basis (Fig. 12).

Nipponnemertes schollaerti (Wheeler, 1934), for which no information on posterior cephalic furrows is available from Wheeler (1934)'s original description, is distinguished from *N. lactea* in the larger numbers of the ocelli on each side of head—55 in *N. schollaerti* (Berg, 1985) vs 12 in *N. lactea* (Fig. 11L)—and more than ten times larger body size—97 mm in *N. schollaerti* (preserved, mature male) vs 7 mm in *N. lactea* (living, mature female).

The present species externally resembles *Nipponnemertes* sp. 1 reported by Yamaoka (2005) from a beach in Shitaru (near Shimoda (Shizuoka)) in having a pale body. Further studies with DNA barcoding are needed for Yamaoka (2005)'s unidentified forms.

Nipponnemertes notoensis Hookabe, Kajihara & Chernyshev, 2022

[Japanese name: noto-sujinashi-himomushi]

(Figs. 11M–O, 19)

Material examined: Holotype: ICHUM 8322, serial transverse sections of anterior tip to intestine, a posterior tip used for DNA extraction, collected by dredging from 70–91 m depth, off Noto, Ishikawa Prefecture, Japan (Fig. 6A, B), by Natsumi Hookabe, on 20 November 2019. Paratype: ICHUM 8323, fixed in 10% formalin and later preserved in 70% ethanol, a posterior tip used for DNA extraction, collected by dredging from 70–91 m depth, off Noto, Ishikawa Prefecture, Japan (Fig. 6A, B), by Natsumi Hookabe, on 20 November 2019.

Habitat and geographic distribution: The species has been only known from the type locality, off Noto (Ishikawa Prefecture), Japan.

Etymology: The specific name is derived from the type locality, Noto, Ishikawa Prefecture, Japan.

Description: *External features.* Body 1.2 cm in length, 1.2 mm in width; body uniformly pale to pale-yellow colored (Fig. 11M). Head not demarcated from trunk, mottled with white spots along dorsal midline (Fig. 11N, O); cephalic patch absent (Fig. 11N, O). Anterior cephalic furrows not well developed. Posterior cephalic furrow V-shaped on dorsal surface (Fig. 11M), completely encircling body. As many as 19–20 reddish brown ocelli irregularly arranged on each side; each ocelli irregular in size and shape; 2 to 3 ocelli fusing with each other (Fig. 11N).

Internal morphology. Epidermis with numerous red-, orange-, and blue-stained cells and ciliated cells (Fig. 19A, B). Dermis up to 21 μ m thick, forming cup-like structures on distal

surface in both precerebral (Fig. 19A) and foregut regions (Fig. 19B). Body-wall musculature as in other species in the genus (Fig. 19A, B). Oesophagus (Fig. 19G) and pylorus (Fig. 19K) as in other species in the genus. Dorso-ventral muscles present between intestinal diverticula (Fig. 19L). Cephalic (Fig. 19D) and submuscular glands present (Fig. 19G). A single apical organ present (Fig. 19C). Precerebral and mid-dorsal vessel not observed.

Rhynchocoel musculature (Fig. 19G–K), proboscis epithelium (Fig. 19E), and proboscis musculature (Fig. 19E, F) as in other species in the genus. Proboscis nerves 10 in number (Fig. 19E). Stylet armature not observed in the specimen examined. Two accessory stylet pouches present, each containing more than two stylets (Fig. 19E).

Pigment-cup ocelli embedded in body-wall longitudinal muscle layer (Fig. 19D). Cerebral organs extending to posterior region of brain (Fig. 19G–I), followed by glandular cells (Fig. 19J). Brain (Fig. 19H, I) and lateral nerves (Fig. 19K, L) as in other species in the genus.

Nephridial tubules branching near lateral blood vessel (Fig. 19K). Gonads and gonoducts not found in the specimen examined.

Remarks: Among 19 species previously described and 13 species herein reported, *N. sanguinea* and *N. schollaerti* are most similar to *N. notoensis* due to the uniformly pale-colored body (Fig. 11M) and the irregular arrangement of ocelli (Fig. 11N, O) (see the Remarks section of *N. lactea*). However, *N. notoensis* can be differentiated from the two species in the number of proboscis nerves: 10 in *N. notoensis* (Fig. 19E) vs 11 in *N. sanguinea* vs 14 in *N. schollaerti*.

Nipponnemertes ornata Hookabe, Kajihara & Chernyshev, 2022

[Japanese name: kazari-sujinashi-himomushi]

(Figs. 11P, Q, 20)

?*Nipponnemertes* sp. 2: Yamaoka, 2005, p. 152–153.

Material examined: Holotype: ICHUM 8327, serial transverse sections of anterior tip to intestine, a posterior tip used for DNA extraction, collected at depths of 70–91 m by dredging, off Noto, Ishikawa Prefecture, Japan (Fig. 6A, B), by Natsumi Hookabe, on 20 November 2019.

Habitat and geographic distribution: The species is only known from the type locality, off Noto (Ishikawa Prefecture), Japan.

Etymology: The specific name is a feminine form of Latin *ornatus*, meaning “adorned or decorated”, derived from the body coloration mottled with white and brown dots in the present species.

Description: *External features.* Body 1.8 cm in length, 0.9 mm in width; background uniformly pale colored, mottled with white and brown dots; white dots conspicuous in dorsal surface of head region; brown dots more densely distributed in dorso-anterior region of body than ventral and posterior region (Fig. 11P). Head not well demarcated from trunk; cephalic patch absent (Fig. 11Q). Anterior cephalic furrows not well developed (Fig. 11Q). Posterior cephalic groove V-shaped on dorsal surface, completely encircling body (Fig. 11Q). Ocelli 16 or 17 in number, not regularly arranged in rows, varying in size (Fig. 11Q).

Internal morphology. Epidermis with numerous red-and yellow-stained cells and ciliated cells (Fig. 20A, B). Dermis thin, up to 20 μ m thick, forming cup-like structures on distal surface in precerebral (Fig. 20A) and foregut regions (Fig. 20B). Body-wall musculature (Fig. 20A, B) and oesophagus (Fig. 20E, F) as in other species in the genus. Intestinal caecum anteriorly extending beneath pylorus (Fig. 20I). Dorso-ventral muscles present between intestinal diverticula (Fig. 19J). Cephalic glands (Fig. 20A) and the subsequent submuscular glands (Fig. 20E) as in other species in the genus. A single apical organ present (Fig. 20C). Precerebral and mid-dorsal vessel not well observed in my histological sections.

Proboscis pore opening ventrally (Fig. 20P). Rhynchocoel musculature (Fig. 20F, I), proboscis epithelium (Fig. 20D), and proboscis musculature (Fig. 20D) as in other species in the genus. Proboscis nerves 10 in number (Fig. 20D). Stylet not observed in the specimen examined.

Pigment-cup ocelli embedded sub-dermally (Fig. 20E). Cerebral organs extending to posterior region of brain (Fig. 20E–G), followed by glandular cells (Fig. 20H). Brain (Fig. 20F, G) and lateral nerves (Fig. 20I, J) as in other species of the genus.

Nephridial tubules not observed in the present specimen. Ovaries developing between intestinal diverticula (Fig. 20J).

Remarks: The present species is extremely similar to *N. kozaensis* in having white and brown dots on uniformly pale body; however, it can be differentiated from the latter in brown dots more densely scattering on body surface (Fig. 11H) (see also the Remarks section of *N. kozaensis*). Internal morphology of the two species is not remarkably different, although there might be other useful characters which are not obtained for *N. ornata* in this study (e.g., number of accessory stylets in proboscis).

The present species externally resembles *Nipponnemertes* sp. 2 reported by Yamaoka (2005) from Shirahama and Susaki in having a pale body with brown dots. Further studies with DNA barcoding are needed for Yamaoka (2005)'s unidentified forms.

The molecular phylogenetic tree shows that *N. ornata* is sister-related with *N. notoensis* (Fig. 7). *Nipponnemertes notoensis* differs from *N. ornata* in lacking dotted pattern (Fig. 11M). Moreover, uncorrected *p*-distance between the two species was 9%, higher than the threshold value (5%) used for species separation in nemertean systematics (e.g., Sundberg et al. 2016).

Nipponnemertes sugashimaensis Hookabe, Kajihara & Chernyshev, 2022

[Japanese name: sugashima-sujinashi-himomushi]

(Figs. 11R–U, 12, 21)

Material examined: Holotype: ICHUM 8330, serial transverse sections of anterior tip to intestine, a posterior tip used for DNA extraction, bycaught with a gill net from 10 m depth, Sugashima, Mie Prefecture, Japan (Fig. 6A, D), collected by Natsumi Hookabe and Naoto Jimi, on 3 November 2021.

Habitat and geographic distribution: The species is only known from the type locality. Worms were picked up from rocky substrate samples bycaught with a gill net.

Etymology: The specific name is derived from the type locality, Sugashima (Mie Prefecture), Japan.

Description: *External features.* Body 0.9 cm in length, 1.1 mm in width; background uniformly pale colored, with irregular, leopard-like brown mottling (Fig. 11R); mottling pattern ventrally faded. Head not well demarcated from trunk; cephalic patch lacking (Fig. 11S). Anterior cephalic furrows not well developed (Fig. 11S). Posterior cephalic furrows not found in the specimen examined. 12 or 13 ocelli irregularly arranged (Fig. 11T). Proboscis white in color when everted.

Internal morphology. Epidermis with numerous red- and yellow-stained cells and ciliated cells (Fig. 21A, B). Dermis up to 24 μm thick, forming cup-like structures on distal surface in precerebral (Fig. 21A) and foregut regions (Fig. 21B). Body-wall musculature (Fig. 21A, B), oesophagus (Fig. 21E, H), pylorus, and intestinal caecum (Fig. 21J) as in other species in the genus. Dorso-ventral muscles present between intestinal diverticula (Fig. 21K). Cephalic glands (Fig. 21D) and the subsequent submuscular glands as in other species in the genus. A single apical organ present (Fig. 21C). Mid-dorsal vessel (Fig. 21E, J) protruding into rhynchocoel to form a single vascular plug in posterior region of brain (Fig. 21H); mid-dorsal vessel (Fig. 21E, J) as well as lateral vessels (Fig. 21I, J) containing red to orange colored blood cells.

Rhynchocoel musculature (Fig. 21E, H, J, K), proboscis epithelium (Fig. 21F), and proboscis musculature (Fig. 21F) as in other species in the genus. Proboscis nerves 10 in number (Fig. 21F). Stylet basis oval, 75.0 μm in length and 48.0 μm in maximum width; central stylet vertically sculptured, 115.0 μm in length; S/B ratio 1.38 (Fig. 12). Two accessory stylet pouches present, each containing 7 stylets (Fig. 12).

Pigment-cup ocelli embedded sub-dermally (Fig. 21I). Cerebral organs extending to posterior region of brain (Fig. 21G–I), followed by glandular cells (Fig. 21I). Brain (Fig. 21H) and lateral nerves (Fig. 21I, J) as in other species of the genus.

Nephridial tubules branching near lateral blood vessels (Fig. 21K). Gonads, gonoducts, and gonopores not found in the specimen examined.

Remarks: Apart from *N. sugashimaensis* sp. nov., irregular brown mottling on uniformly pale body is known in *N. madagascarensis* (see the Remarks section of *N. ornata*). However, *N. sugashimaensis* is discriminated from *N. madagascarensis* by having submuscular glands (Fig. 21A) and the larger number of accessory stylets per pouch: 7 in *N. sugashimaensis* (Fig. 11T) and 2–3 in *N. madagascarensis* (Kirsteuer 1965).

Nipponnemertes sp. 2

(Figs. 11V, W, 12)

Material examined: ICHUM 8332, preserved in 70% ethanol, a posterior tip used for DNA extraction, collected at depths of 116–211 m by dredging, off Jogashima, Miura, Kanagawa Prefecture, Japan (Fig. 6A, C), by Natsumi Hookabe, on 30 July 2021.

Habitat and geographic distribution: The species is only known from Jogashima, Kanagawa Prefecture, Japan.

Description: *External features.* Body 0.9 cm in length, 0.8 mm in width; body uniformly pale; intestinal region visible through body wall as pinkish (Fig. 11V). Head not demarcated from trunk, without cephalic patches (Fig. 11V). Anterior cephalic furrows not well developed. Posterior cephalic furrow not visible. As many as 13 ocelli present (Fig. 11W). Precerebral and lateral vessels filled with red corpuscles (Fig. 11W).

Proboscis pore sub-terminal. Stylet basis oval, 76.0 μm in length and 42.0 μm in maximum width; central stylet smooth, 96.0 μm in length; S/B ratio 1.38 (Fig. 12). Two accessory stylet pouches present, each containing six stylets (Fig. 12).

Remarks: The species is characterized by pinkish intestine visible through body wall (Fig. 11V). Due to the absence of morphological voucher specimen (specimen lost during histological sectioning), I leave the species as an unidentified form in the present study.

3.1.4 Discussion

Molecular systematics of Nipponnemertes

In this study, species diversity of *Nipponnemertes* in Japanese and the Far Eastern Russian waters was significantly updated from 4 to 14 species, in which newly discovered 9 species as well as herein redescribed *N. bimaculata* sensu Iwata (1954) (= *N. neonilae*) are included (Table 2, 9). A molecular phylogenetic analyses recovered three major lineages in *Nipponnemertes* (Fig. 2). In a previous study based on six congeners (Kajihara et al. 2015), two well-supported clades each corresponding to Clade A and Clade B (Fig. 7) have been detected. With increased taxon sampling especially in subtidal to deep-sea species, Clade C was newly confirmed in the present study.

Apart from relatively small body size in Clade C (see the discussion below), each clade is not characterized by any apparent synapomorphy, but is mutually distinguishable by a combination of three external characters—demarcation between the head and body, mid-dorsal ridge on head, and cephalic patches (Fig. 11). The former two characters are likely plesiomorphic and retained in Clade A (Fig. 9) and Clade B (Fig. 10) while the last one is an apomorphy for Clade B (Fig. 10). In members in Clade C, the head is not demarcated from the body and does not have a mid-dorsal ridge and cephalic patches (Fig. 11). Although it remains unclear in Clade A due to a lack of data, stylet-to-basis-length ratio is also different between Clade B (1.85–2.16) and Clade C (1.25–1.64) (Fig. 12). Neither clade-specific characters nor combinations of characters are found in internal morphology in the species examined in the present study.

As mentioned above, species in each clade are differentiated by external morphological characters of the head: species possessing demarcated head without cephalic patches (Clade A), demarcated head with cephalic patches (Clade B), and non-demarcated head lacking cephalic patches (Clade C) (Fig. 7). Although the three lineages are presumably characterized by the combination of morphological characters of the head and could be regarded

as separate genera, we place all the species in *Nipponnemertes* since molecular data for the type species, *N. drepanophoroides* is currently unavailable. In possessing heads without cephalic patches, *N. drepanophoroides* is probably included in Clade A rather than Clade C; however, no information is available as to the head–trunk demarcation and the mid-dorsal ridge in the original description by Griffin (1898) and redescription by Coe (1905). Molecular confirmation using topotype specimens is required for future taxonomic reappraisal.

In the present study, greater external morphological disparity between specimens (body coloration and spotting pattern) generally turned out to be a manifestation of species difference with greater phylogenetic/genetic distances (Fig. 7). However, this was not the case in the *N. kozaensis*–*N. ornata* species pair. Despite having extremely similar body coloration (pale body with reddish to brownish ornamentation; Fig. 10H–J, P, Q), the two did not form a single clade (Fig. 7). On the contrary, the converse was not true for the “*N. ojimaorum*–*N. punctatula*” subclade in Clade B, consisting of *N. crypta*, *N. jambio*, *N. ojimaorum*, and *N. punctatula*. Their interspecific genetic distances based on COI were 1.6–4.4% in uncorrected *p*-distance and (Table 7), lower than interspecific COI divergences previously reported in *Nipponnemertes* (3.8–4.3% between *N. pulchra* and *N. cf. rubella*) (Chernyshev and Polyakova, 2022) and other nemertean species (Sundberg et al. 2016). The most conspicuous case was observed between *N. crypta* and *N. ojimaorum* showing 1.6% in *p*-distance, albeit apparently different body coloration (Fig. 10C, D, I–K) (similar phenomena have been found in other invertebrates such as bobtail squids (Fernández-Álvarez et al. 2021) and mites (García-Jiménez et al. 2017). Moreover, the two species were not sister taxa in the tree based on the concatenated dataset (Fig. 7). One thing we can deduce from the result is that the use of COI-based genetic distances might underestimate the true intraspecific divergences due to mutational saturation in COI region among the “*N. ojimaorum*–*N. punctatula*” subclade. As another speculation, it might be due to the relatively rapid evolutionary rate in external morphology including body coloration and pattern in the subclade.

Species delimitation analyses for Clade B show the consistent result, where five lineages are identified, preferring to separate *N. crypta* and *N. ojimaorum* as different species in all three methods (TCS, ABGD, and bPTP) (Fig. 8). The threshold used to separate nemertean species based on COI is mostly around 5% (e.g., Chen et al. 2010; Kang et al. 2015; Krämer et al. 2017; Chernyshev et al. 2018). Although the values we obtained were lower than 5%, we describe *N. crypta* and *N. ojimaorum* as separate species based on the differences in

body coloration and internal morphology, as well as the result of species delimitation analyses (Fig. 8).

Reduction in body size and blood-circulatory system in Clade C

The newly confirmed Clade C is formed by relatively smaller species (7–20 mm in the maximum body lengths) than the other two clades (12–110 mm in Clade A and 11.7–160 mm in Clade B) although we could not obtain mature adults for all species in Clade C. The smallest species in Clade C is *N. lactea* with developed gonads. Members in Clade C were collected from subtidal (8 m~) to deep sea bottoms (~985 m). In the deep-sea environment, body-size trends across environmental gradients (hydrostatic pressure, light, water temperature, and amount of food resources) have been demonstrated in invertebrates and demersal fish (Collins et al. 2005). It is difficult to determine which variable(s) most critically influences body size; still, food availability is known as one of the major factors in these animals (Collins et al. 2005). The body size reduction in Clade C in contrast with Clade A and B might reflect an adaptation to food-limitation in deep-water bottoms (herein referring to water depths deeper than 70 m) as inferred in non-scavenging fish (Collins et al. 2005) or predatory invertebrates (Van der Grient and Rogers 2015). If species in Clade C are suctorial predators on small crustaceans as reported in *Nipponnemertes* cf. *africana* (McDermott 1997), *N. pulchra* (Brunberg 1964; Berg 1972a; McDermott 1984, 1993), and *N. ogumai* (Kajihara et al. 2015), smaller body sizes might allow them to require less food demand for survival and reproduction. The phylogenetic positions among Clade C also indicate that *N. kozaensis* collected at a depth of 12 m, *N. sugashimaensis* collected at a depth of 10 m, and Cratenemertidae sp. Guam collected at a depth of 1 m (Chernyshev and Polyakova 2019) might have secondarily colonized from deep (>70 m) to shallow waters (<12 m), although I cannot exclude that it can be artificially resulted from my lack of sampling effort (e.g., sample collection from only a part of the bathymetric range they live). Since ecological factors other than amount of potential food resources can be also related to the body size evolution, the speculation above should be accompanied with further examination of ecological data on species collected from various water depths.

The vascular plug, by which nutrients and other substances are supposedly delivered from the blood vascular system to the rhynchocoel (Crandall 1993), is considered to be important to maintain the function of proboscis apparatus principally used to capture prey items. In small-bodied hoplonemertean, in contrast, this structure seems to have been often lost,

suggesting that the proboscis could function without a vascular plug by diffusion through the rhynchocoel wall (Kajihara 2021b). In Clade C, a vascular plug is not observed except for *N. sugashimaensis*. This might be actually resulted from a small body size; however, the other possibility is that a vascular plug is easily affected by body contraction during fixation and/or difficult to find through histological sectioning in small-sized species. Additional morphological examination with broader taxon sampling as well as alternative methods for observation of the fine muscular structure (e.g., phalloidin labelling) is expected in future studies.

3.2 Amphiporidae Örsted, 1843 (EUMONOSTILIFERA)

In this chapter, I describe two species in Amphiporidae Örsted, 1843 based on external/internal morphological examination and molecular phylogenetic analyses. Based on the data herein provided, *Tortus tokmakovae* Chernyshev, 1991b is redescribed as *Amphiporus* Ehrenberg, 1831. These studies were partially published in Hookabe et al. (2020).

3.2.1. Introduction

Amphiporidae Örsted, 1843 is one of the taxon-rich traditional families in Eumonostilifera (Thollesson and Norenburg 2003). The members are usually marine benthic, inhabiting intertidal to hadal zones (Chernyshev and Polyakova 2019). In the present study, I deal with three genera in the family: *Amphiporus* Ehrenberg, 1831, *Proamphiporus* Chernyshev and Polyakova, 2019, and *Tortus* Korotkevitsch, 1971.

Amphiporus is one of the largest, old-established genera in eumonostiliferans (Gibson 1989). Species in *Amphiporus* are generally characterized by possessing many eyes without extending posterior to the brain region and rhynchocoel nearly reaching to posterior end of body (Coe 1905); however, there are no definitive diagnostic characteristics so far (Gibson 1985, 1989; Norenburg 1986; Strand et al. 2014). In this situation, molecular approaches are no longer evitable for species identification and taxonomic reappraisal in the genus. As to *Amphiporus*, sequences obtained from the Welsh specimen of *A. lactifloreus* (Johnston, 1828), which can be practically regarded as a topotype (Andrade et al. 2012), are useful for systematic studies.

Tortus was established by Korotkevitsch (1971); the presence of precerebral septum and a caudal sac is main characters differentiating the genus from other eumonostiliferan genera. A close relationship between *Tortus* and *Amphiporus* were inferred based on morphology (Chaban and Chernyshev 2008), which was later supported by molecular phylogeny (Kvist et al. 2015). In the present study, I studied phylogenetic relationships among specimens of *Tortus* newly obtained from Hokkaido, Japan and several species in *Amphiporus* including the type species *A. lactifloreus* to test if *Tortus* species are nested in *Amphiporus*.

The genus *Proamphiporus* was monospecific before my study in Hookabe et al. (2020). In contrast with the two genera introduced above, species in genus have more unique morphological characteristics including anatomical features. One of the main features that characterize *Proamphiporus* among other Amphiporidae is the position of the cerebral organs,

being entirely situated alongside the brain like in cratenemertids and reptantians (Chernyshev and Polyakova 2019), instead of anteriorly to the brain in most members in the Eumonostilifera.

The present subchapter includes descriptions of two *Amphiporus* and one *Proamphiporus* species. Based on the result of phylogenetic analyses using the species herein examined, I propose to transfer *Tortus tokmakovae* to *Amphiporus*.

3.2.2 Materials and Methods

Taxon sampling and morphological examination

Specimens were collected as follows (also see Table 1): *Amphiporus* sp. Saroma collected from intertidal zones of Saroma lake on July 11, 2020 and *Proamphiporus kaimeiae* collected from soft sediment at a depth of 262 m, off Ofunato (39°06.70'N, 142°06.44'E), Iwate Prefecture, Tohoku, on 19 July 2019 by use of a remotely operated vehicle (ROV) Crambon dive #35 during KM19-05C cruise of R/V Kaimei organized by Tohoku Ecosystem-Associated Marine Sciences (TEAMS) project. Living specimens were anaesthetized with a MgCl₂ solution isotonic to seawater and photographed with a digital still camera OM-D E-M1II (Olympus, Tokyo, Japan). Internal morphology was observed following the methods used in the previous chapter.

DNA extraction, PCR amplification, and sequencing

DNA extraction, amplification, and sequencing were performed based on the same methods in Subchapters 2.2. and 3.1.1.2. Genetic distances were calculated using the same primers and the software used in Subchapter 3.2.

Sequence alignment and phylogenetic analyses

Methods used in the present study follow Subchapters 2.2 and 3.2.

3.2.3 Results

Phylogeny and genetic distances

In the resulting tree (Fig. 2), *Tortus* sp. Saroma, together with *Tortus tokmakovae* Chernyshev, 1991b and *Tortus* sp. IZ-45645 (Kvist et al. 2015), was nested in *Amphiporus*. *Proamphiporus kaimeiae* Hookabe, Tsuchida, Fujiwara & Kajihara, 2020 formed a clade along with *P. crandalli* and *P. rectangulus* (Strand, Herrera-Bachiller, Nygren & Kånneby, 2014) with full support values, corroborating the generic placement of the new species as well as the nominal species *Amphiporus rectangulus*. The interspecific genetic distances based on COI partial sequences (Table 10) were 14.1% in *p*-distance between *P. kaimeiae* and *P. crandalli*, while 13.3–13.5% in *p*-distance between *P. kaimeiae* and *P. rectangulus*. These values are comparable with 10.4–10.7% in *p*-distance between *P. crandalli* and *P. rectangulus*, and the interspecific COI divergences for Eumonostilifera suggested in Sundberg et al. (2016).

Systematics

Family Amphiporidae Örsted, 1843

Genus *Amphiporus* Ehrenberg, 1831

Type species: *Amphiporus lactifloreus* (Johnston, 1828)

Diagnosis based on Bürger (1904), Gibson and Crandall (1989), Maslakova et al. (2005), and Strand et al. 2014): Eumonostiliferous nemerteans with rhynchocoel extending almost to posterior end of body. Body wall musculature generally well developed without diagonal musculature between outer circular and inner longitudinal muscle layers. Blood vascular system with three longitudinal vessels. Lateral nerve cords without accessory nerves. Intestinal caecum with a pair of anterior diverticula and lateral pouches. Cerebral organs anterior to cerebral ganglia. Sex separate.

Amphiporus tokmakovae (Chernyshev, 1991b) comb. nov.

Tortus tokmakovae Chernyshev, 1991: Chernyshev, 1991b, p. 129–132, fig. 1, 2

Remarks: *Tortus* was established by Korotkevitch (1971) for *T. curilensis* Korotkevitch, 1971 on the following morphological characters: (i) body with epidermal constrictions becoming twisted when fixed in ethanol or formalin and (ii) rhynchocoel posteriorly narrow, forming “caudal sac”. The two characters are likely to be caused by an artifact during fixation without anesthetization. A phylogenetic position of the present species nested in the *Amphiporus* clade, in which the type species, *A. lactiflorens* is included, supports that *T. tokmakovae* should be transferred from *Tortus* to *Amphiporus*. As to the other species in the genus, *T. curilensis*, *T. incongruens* (Korotkevich, 1977), *T. iturupensis* Korotkevich, 1977, and *T. obtusorostris* (Korotkevich, 1977), I refrain from redescribing them as members in *Amphiporus* without molecular data in the present study.

Amphiporus sp. Saroma

Material examined: Museum deposition numbers not assigned, unsectioned complete specimen except for the posterior tip, fixed in Bouin’s fluid and later preserved in 70% ethanol, posterior tip preserved in 99% ethanol, collected at low tide in Saroma lake, Hokkaido, Japan, by Natsumi Hookabe, on July 11 2020. Museum deposition numbers not assigned, serial transverse sections from anterior tip to intestinal region (3 slides), everted proboscis (3 slides), and serial frontal sections of intestinal region (3 slides), posterior tip used for DNA extraction, female, collected in the same date and locality as the above specimen.

Habitat and geographic distribution: The present species was found in Saroma lake only.

Description: *External features.* Body 4.0 cm in length and 1.7 mm in width; body dorsoventrally dark brown (Fig. 22A); anterior tip of head whitish (Fig. 22B). Eyes arranged in anterior and posterior pairs of groups (Fig. 22E); anterior pairs up to 6 eyes, arranged in antero-lateral margins of head; posterior pairs up to 8 eyes (Fig. 22E). Head rounded, slightly demarcated from body (Fig. 22B). Anterior cephalic furrows not meeting at mid-line on dorsal and ventral surface (Fig. 22B–D). Posterior cephalic furrows dorsally V-shaped (Fig. 22B–D) and ventrally transverse, without meeting at mid-line (Fig. 22C). Blood uncolored. Cerebral ganglia and lateral nerves red colored (Fig. 22E).

Stylet basis oval, 24.0 μm in length and 12.0 μm in maximum width; central stylet smooth, 26.2 μm in length; stylet-to-basis-length ratio 1.09 (Fig. 22E). Two accessory stylet pouches present, each containing 2 stylets.

Remarks: The present species externally resembles *T. tokmakovae* described from the Peter the Great bay (Chernyshev 1991b) and reported from Akkeshi (Hokkaido) on the external morphological basis (Kajihara 2017). However, genetic distance based on COI between *Tortus* sp. Saroma and *T. tokmakovae* is 5.0 % in *p*-distance, which is comparable with interspecific divergences observed in eumonostiliferans (e.g., Sundberg et al. 2016; Hookabe et al. 2022b).

Amphiporus antifuscus Iwata, 1954 is also extremely similar to the present species in eye arrangement and body coloration but differs from the latter in having a pear-shaped stylet basis (Iwata 1954). The stylet basis of *Amphiporus* sp. Saroma is oval, without a constriction in the middle part.

Amphiporus sp. Saroma and *Tortus* sp. IZ-45645 seem to be conspecific in terms of COI divergence (1.0 %) lower than interspecific threshold value; both two were collected from the Sea of Okhotsk, about 650 km away from Saroma lake to Kuril Island (Kvist et al. 2015). On the basis of morphology and molecular data, *Amphiporus* sp. Saroma and *Tortus* sp. IZ-45645 would be undescribed species.

Genus *Proamphiporus* Chernyshev & Polyakova, 2019

Type species: *Proamphiporus rectangulus* (Strand, Herrera-Bachiller, Nygren & Kånneby, 2014)

Diagnosis: Eumonostiliferous nemerteans with rhynchocoel extending almost to posterior end of body. General internal characters as in *Amphiporus* except for a position of cerebral organs. Cerebral organs situated alongside the brain.

Proamphiporus rectangulus (Strand, Herrera-Bachiller, Nygren & Kånneby, 2014)

Amphiporus rectangulus Strand et al., 2014: 2–6, Fig. 2–5.

Remarks: In *Amphiporus rectangulus*, the cerebral organs are situated alongside the brain (Strand et al. 2014: fig. 5F) just as in *Proamphiporus crandalli* (type species of *Proamphiporus*) (Chernyshev and Polyakova 2019), instead of being anterior to the brain as in most of the Eumonostilifera, including *Amphiporus lactifloreus* (type species of *Amphiporus*) (Berg 1972b). The angle of the V-shape formed by the cephalic furrows on the mid-dorsal body surface in *A. rectangulus* is as acute as that in *P. crandalli*, and much more acute than that in *A. lactifloreus* (Gibson 1994; Strand et al. 2014; Chernyshev and Polyakova 2019). Based on these unique similarities, we consider it reasonable to transfer *A. rectangulus* from *Amphiporus* to *Proamphiporus*.

Proamphiporus kaimeiae Hookabe, Tsuchida, Fujiwara & Kajihara, 2020

[Japanese name: kaimei-fukamizo-himomushi]

(Figs. 23, 24)

Material examined: Holotype: ICHUM 6058, serial transverse sections of anterior body fragment (6 slides), a posterior tip used for DNA extraction, collected at a depth of 262 m, off Ofunato, Iwate, Japan (Fig. 1A, D), collected by Natsumi Hookabe, July 19, 2019.

Habitat and geographic distribution: The species is only known from the type locality.

Etymology: The specific name is after the R/V *Kaimei*, during a research cruise of which the holotype was sampled.

Description: *External features.* Body 3.6 mm in length and 0.7 mm in maximum width; background body color generally whitish (Fig. 23), epidermis semitransparent ventrally; internal organs (intestine, rhynchocoel, proboscis) visible through bodywall (Fig. 23B). Dorsal surface with brownish pigments randomly distributed and aggregated to form single middorsal longitudinal stripe of about 5/6 to 2/3 of body width (Fig. 23A); ventral surface without pigments (Fig. 23B). Anterior tip of head devoid of pigment. Head slightly narrower than middle part of body. Single pair of cephalic furrows extending posteriorly on dorsal side, meeting at mid-line and visible through body wall as orange spots (Fig. 23B). No eyes evident.

Internal morphology. Epidermis 20–40 µm in thickness, containing numerous red and yellow staining ciliated cells (Fig. 24A). Dermis up to 10–15 µm thick. Outer circular muscle layer of body wall 10–20 µm in thickness. Inner longitudinal muscle layer of body wall 50 µm in maximum thickness. Esophagus with ciliated epithelium. Stomach epithelium with glands and cilia. Intestinal diverticula deeply branched (Fig. 23B). Mid-dorsal vessel penetrating rhynchocoel wall behind ventral cerebral commissure, forming single vascular plug (Fig. 24D).

Proboscis pore terminal. Rhynchocoel long, more than two-thirds of body length, with thin wall comprised of few muscle layers (Fig. 24B, D). Proboscis with outer circular, middle longitudinal, and inner circular muscle layers (Fig. 24E); proboscis epithelium with

developed papillae containing basophilic glandular cells (Fig. 24B, D, E); ten proboscis nerves (Fig. 24E). Lateral nerves with neither accessory nerves nor neurocords (Fig. 24G). Cerebral organs entirely situated alongside brain region, with forked chamber; chamber bifurcating into an anterior radial and a posterior lateral canal, both ciliated (Fig. 24B, C); acidophilic glands developed in posterior region of cerebral organs (Fig. 24C). Eyes absent.

Testes between intestinal diverticula (Fig. 24F). Gonoducts and gonopores not found. Excretory collecting tubules near lateral nerve in pyloric region (Fig. 24G).

Remarks: The cerebral organs in *P. kaimeiae* are situated alongside the brain region behind the proboscis insertion, which accords with the generic diagnosis for *Proamphiporus* (Chernyshev and Polyakova 2019). The external appearance of *P. kaimeiae* is quite similar to that of *P. rectangulus*; however, they can be differentiated by a depigmented mid-dorsal stripe on the dorsal body surface in *P. rectangulus* but absent in *P. kaimeiae*. *Proamphiporus kaimeiae* differs from *P. crandalli* by having brown pigments on the dorsal surface.

3.2.4 Discussion

The molecular phylogenetic analyses showed that species in Amphiporidae are divided into two major lineages, brackish to shallow-water (*Amphiporus*, *Nessia* Girard, 1893, and *Paranemertes* Coe, 1901) and deep-sea lineages (*Nemertovema* Chernyshev & Polyakova, 2018b and *Proamphiporus*). Within the former lineages, *Nessia*, *Tortus*, and *Paranemertes* were nested in *Amphiporus*; thereby, I herein proposed to transfer *T. tokmakovae* to *Amphiporus* and identified specimens from Saroma lake as an undescribed species in *Amphiporus* instead of *Tortus* based on morphological examination and phylogenetic analyses. *Neesia* and *Paranemertes*—as well as *Tortus*—were earlier transferred to Neesiidae by Chernyshev (2005) based on radial precerebral septum. The result of eumonostiliferan phylogeny (Fig. 2) suggests that the feature is polyphyletic, likely a less informative character to circumscribe the family as inferred in Kvist et al. (2015). Taking into account the current tree topology (Fig. 3), where the type genus *Neesia* is nested in Amphiporidae, species in Neesiidae should be transferred to Amphiporidae. In the present study, I leave taxonomic reappraisal for Neesiidae to the future study until specimens allowing morphological examination are available.

3.3 Carcinonemertidae Sumner, Osburn & Cole, 1913 (EUMONOSTILIFERA)

In this chapter, I report a new host of *Ovicides paralithodis* Kajihara & Kuris, 2013, the snow crab *Chionoecetes opilio* (O. Fabricius, 1788) and discuss their host specificity by comparing COI sequences between the specimens obtained from Dungeness crabs.

3.3.1. Introduction

Most members in Nemertea are marine benthic and free-living, while a few are known to be parasitic. The most species-rich group of parasitic nemerteans is the family Carcinonemertidae Sumner, Osburn & Cole, 1913, members of which are predacious on decapod-crustacean eggs. Host specificity varies within the family; *Carcinonemertes epialti* Coe, 1902, for example, is known from the originally reported host *Pugettia producta* (Randall, 1839) and—on a morphological basis—10 other decapod species (Humes 1942; Wickham 1978) although worms from different hosts might represent a complex of multiple, cryptic species (Wickham and Kuris 1988). The family harbors two genera, *Carcinonemertes* Coe, 1902 (17 or 18 species) and *Ovicides* Shields, 2001 (5 species), both of which were previously reported from Japanese waters. The former is represented by *Carcinonemertes mitsukurii* Takakura, 1910 from the mitten crab *Eriocheir japonica* (De Haan, 1835) along the Pacific coast of Honshu. In the latter genus, *Ovicides paralithodis* Kajihara & Kuris, 2013 was described from the red king crab *Paralithodes camtschaticus* (Tilesius, 1815), collected off Abashiri in the Sea of Okhotsk. To date, no records of Carcinonemertidae have been documented from the Sea of Japan except an unidentified (and probably undescribed) species of *Carcinonemertes* from Peter the Great Bay (Chernyshev 2014).

Apart from Japanese waters, *O. paralithodis* has been reported (as unidentified “Form 4”) from the Bering Sea along the Aleutian Islands (Adak, Dutch Harbor, Morshovoi Bay, Pavlof Bay) and the Gulf of Alaska (Kodiak Island, Resurrection Bay, Cook Inlet, Seward, Barlow Cove, Deadman’s Reach, Gambier Cove, and Pybus Cove) by Kuris et al. (1991). In addition, unidentified carcinonemertids that lack eyes and possess accessory stylet pouches (and thus resemble *O. paralithodis*) were reported from the tanner crab *Chionoecetes bairdi* Rathbun, 1924 and the Dungeness crab *Cancer magister* Dana, 1852 in Alaskan waters (Kajihara and Kuris 2013). However, none of these records has been confirmed based on molecular barcode data, which are indispensable for accurate identification of nemertean species, especially ones

that lack obvious external characters such as body coloration, including most species of Carcinonemertidae. Indeed, no molecular study has ever been performed that investigated host range of any carcinonemertids.

During a series of population surveys on the snow crab *Chionoecetes opilio* (O. Fabricius, 1788) (Fig. 25A), off Fukui in the Sea of Japan from November to December in 2020, a large number of ribbon worms were found from egg masses of female crabs (Fig. 25B). In this chapter, I report a new host of *O. paralithodis*, the snow crab *C. opilio* and discuss their host specificity by comparing COI sequences between the specimens obtained from Dungeness crabs.

3.3.2 Materials and Methods

Taxon sampling and morphological examination

During a series of population surveys on the snow crab *C. opilio* (O. Fabricius, 1788) (Fig. 25A), off Fukui in the Sea of Japan from November to December in 2020, a large number of ribbon worms were found from egg masses of female crabs (Fig. 25B). Living worms were photographed with a digital camera for external morphological documentation and fixed in 100% ethanol for DNA extraction.

DNA extraction, PCR amplification, and sequencing

DNA extraction, amplification, and sequencing were performed based on the same methods in Subchapters 2.2 and 3.2. Genetic distances were calculated using the same primers and the software used in Subchapter 3.2.

Sequence alignment and phylogenetic analyses

Methods used in the present study follow Subchapters 2.2 and 3.2.

3.3.3 Results

Systematics

Family Carcinonemertidae, Sumner, Osburn & Cole, 1913

Genus *Ovicides* Shields, 2001

Type species: *Ovicides julieae* Shields, 2001.

Diagnosis: Carcinonemertids with two accessory stylet pouches containing zero to many accessory stylets. Proboscis reduced in size. Takakura's duct present (or may be present but not observed in *O. julieae*, *O. davidi* Shields & Segonzac, 2007, *O. jasoni* Shields & Segonzac, 2007, and *O. jonesi* Shields & Segonzac, 2007 because no adult specimens were examined for the species). Sex separate or simultaneously hermaphroditic. Living in mucus tubes on pleopods, sterna, and eggs of decapod crustaceans.

Ovicides paralithodis Kajihara & Kuris, 2013

(Fig. 25A, B)

Carcinonemertidae Form 4: Wickham and Kuris, 1988

Material examined: Preserved in 99% ethanol, collected off Fukui, the Sea of Japan, Fukui Prefecture, Japan (Fig. 25A), by Hirohito Motobayashi, on December 28, 2020.

Description: Body 0.7–1.1 cm in length, 0.9–1.1 mm in width; body uniformly pale orange (Fig. 25A, B). Eyes absent. Judging from the pale-orange body coloration, the specimens photographed in life were both female (Fig. 25D).

Remarks: The partial COI sequences (OM988200 (587 bp) and OM988201 (595 bp)) from the two worms obtained from the egg mass of the snow crab *C. opilio* totally overlapped with AB704417, a COI sequence (658 bp) derived from the holotype of *O. paralithodis*. As to the overlapped region, the three sequences were completely identical with each other (p -distance = 0).

3.3.4 Discussion

This is the first record of *O. paralithodis* from the snow crab *C. opilio*, suggesting that the species is polyxenous. The results also represent the first molecular confirmation of carcinonemertids infecting more than a single host species, ranging from anomuran to brachyuran crabs.

What ecological factors allow *O. paralithodis* to infect multiple host species? Based on the findings in the present study and previous ecological/behavioral data on other carcinonemertids, we hypothesize two processes of *O. paralithodis* infecting multiple hosts. One hypothesis is that planktonic larvae of *O. paralithodis* would have a potential to settle on both host crab species, *C. opilio* and *P. camtschaticus*. The horizontal distribution areas of the two host crab species overlap around the studied area (e.g., Osawa and Ota 2020), although the vertical distributions generally differ between the two (Kon et al. 2003; Gabaev 2007). This hypothesis is likely if the planktonic duration of *O. paralithodis* is comparable to that of other carcinonemertids. Planktonic periods in carcinonemertid larvae so far studied are mostly long (Stricker and Reed 1981; Dunn and Young 2014). In *C. epialti* Coe, 1902, it has been reported to reach 51 days (Roe 1979).

The other hypothesized infection process is that mature and/or juvenile worms would transfer from one host to another while the latter two copulate. Such behavior has been observed in *Carcinonemertes errans* Wickham, 1978, where worm individuals synchronously transfer during copulation of hosts from male to female crabs; thus *C. errans* is most likely to have an ability to accumulate on ovigerous crabs (Wickham et al. 1984).

We also hypothesize that the unidentified, eye-less carcinonemertids earlier reported from the tanner crab *C. bairdi* and possibly the Dungeness crab *C. magister* as well in Alaskan

waters (Wickham et al. 1984) actually represented *O. paralithodis*. Given the interhost adult/juvenile-worm transfer delineated in the previous paragraph, this hypothesis is likely because interspecific hybridization between *C. opilio* (distributed in the Bering Sea, the Sea of Okhotsk, and the Sea of Japan) and *C. bairdi* (distributed in the Bering Sea and Alaskan water) occurs under natural condition in the southeastern Bering Sea (Johnson 1976; Smith et al. 2005; Murphy 2020). Future molecular data, as well as behavioral study, should test the hypotheses above.

3.4 Oerstediiidae Chernyshev, 1993 (EUMONOSTILIFERA)

In this chapter, I established two new genera in Oerstediiidae based on external/internal morphological examination and molecular phylogenetic analyses. These studies were published in Hookabe et al. (2022c, d).

3.4.1. Introduction

Eumonostiliferans in the family Oerstediiidae Chernyshev, 1993 occur worldwide, in various marine ecosystems from tropical to polar regions. Recent faunistic studies have revealed unexpected species diversity in upper bathyal to hadal zones, but nemertean taxonomic discoveries remain scarce there. For example, environmental DNA (eDNA) metabarcoding analysis of sediment samples have uncovered deep-sea nemertean fauna from geographic areas and/or water depths where any nemertean species have never been documented in taxonomic literature (Guardiola et al. 2016; Sinniger et al. 2016; Klunder et al. 2020; Atienza et al. 2020). eDNA metabarcoding is widely used as a powerful tool accelerating monitoring of biodiversity even for large-scale studies but still requires baseline data reference sequence data associated with an appropriate species identification prior to ecological assessment of deep-sea biodiversity.

During a research expedition under the project ‘Development of Biodiversity Monitoring Methods for the Management of Deep-sea Marine Protected Areas’ in 2021, we obtained a single specimen of eumonostiliferous hoplonemertean from the southern part of Genroku Seamount Chain. Although this area was known to harbor diverse megafaunal populations comprising sponges, octocorals, hydrozoans, and fish (Morgan and Baco 2021; Calder and Watling 2021; Fujiwara et al. 2022), macrobenthic fauna inhabiting the bottom substrates were poorly studied; in fact, any nemertean species have not been reported from the area to date. Also, I collected oerstediid monostiliferans from sunken wood collected by a commercial trawl net at depths of 150–200 m in the Sea of Kumano. In the present study, phylogenetic positions of the two oerstediid species among Oerstediiidae are inferred with multilocus phylogenetic analyses.

3.4.2 Materials and methods

Taxon sampling and morphological examination

A single specimen was collected from the southern part of Genroku Seamount (30°39.600N, 139°02.410E), on the Nishi-Shichito Ridge, Japan (Fig. 2), during a cruise of R/V Kaimei (cruise ID: KM21-E04C Leg1) under the research project ‘Development of Biodiversity Monitoring Methods for the Management of Deep-sea Marine Protected Areas’ in 2021. The specimen was obtained with a baited trap (handmade fish-trap: hexagonal column, 40 cm in diameter, 17 cm in height). The trap containing pieces of saury was deployed on the seafloor at a depth of 2084 m on October 13, 2021 during a remotely operated vehicle (ROV) KM-ROV dive #153 and was retrieved on October 15, 2021 during KM-ROV dive #154 by use of robotic manipulators equipped on KM-ROV. Specimens from the Sea of Kumano were collected by trawling during operations of the offshore fishing trawler Jinsho-maru XVIII in the Sea of Kumano, Japan, Northwestern (NW) Pacific, at depths of 100–200 m (Table 1), on January 10 and February 7, 2022; at both sampling points, the specimens were found in crevices or burrows inside sunken wood logs. Photographs of the living specimen on board were taken with a digital still camera (NIKON D5600, Japan) before anaesthetization with a MgCl₂ solution isotonic to seawater. The anaesthetized specimen was cut into two fragments by using a razor; a posterior piece (approximately 3 mm in length) was preserved in 99% ethanol for DNA extraction, while the remaining body for histological observation was fixed in Bouin's fluid for 24 h, and later preserved in 70% EtOH. Serial sections were made at 7-mm thickness and stained with Mallory's trichrome method (Gibson 1994). Holotype has been deposited in the National Museum of Nature and Science, Tsukuba (NSMT), Japan.

DNA extraction, PCR amplification, and sequencing

DNA extraction, amplification, and sequencing were performed based on the same methods in Subchapters 2.2 and 3.2. Genetic distances were calculated using the same primers and the software used in Subchapter 3.2.

Sequence alignment and phylogenetic analyses

Methods used in the present study follow Subchapters 2.2 and 3.2.

3.4.3 Results

Phylogeny

In the resulting tree (Fig. 3), species in Oerstediiidae was sister to Plectonemertidae Gibson, 1990a. Within the clade of Oerstediiidae, monophyletic clades were confirmed with full support values for the following genera: *Antarctonemertes* (Friedrich, 1955), *Diplomma* (Stimpson, 1857), *Kurilonemertes* (Chernyshev, 1993), and *Oerstedtia* (Quatrefages, 1846).

Genrokunemertes obesa was sister-related with *Rhombonemertes rublinea* with 52% in BS; 95.0% in SH-aLRT, 97% in UFboot, and 0.77 in PP (Fig. 3). Although *R. rublinea* appeared to be relatively close-related to *Eumonostilifera* sp. Vema2B (Vema Fracture Zone), *Genrokunemertes obesa* (Japan), *Kurilonemertes dilutebasisae* (Kulikova, 1987), *K. papilliformis* (Korotkevitch, 1977) (Russia), *K. phyllospadicola* (Stricker, 1985) (Canada), *Pseudotetrastemma* sp. (Russia), and *Tetrastemma bilineatum* Coe, 1904 (USA), phylogenetic relationships within the clade were not well resolved except for monophyly of *Kurilonemertes* Chernyshev, 1993.

An uncorrected *p*-distance based on 635 bp of COI was 11.9% between *G. obesa* and *R. rublinea*.

Systematics

Family Oerstediiidae Chernyshev, 1993

Genus *Genrokunemertes* Hookabe, Koeda, Fujiwara, Tsuchida & Ueshima, 2022

Type species: *Genrokunemertes obesa* Hookabe, Koeda, Fujiwara, Tsuchida & Ueshima, 2022

Diagnosis: Eumonostiliferous nemertean lacking eyes with weakly flattened, stout body. Head with anterior and posterior cephalic furrows. Body-wall longitudinal musculature anteriorly not divided. Rhynchocoel reaching to more than 3/4 of body length; rhynchocoel musculature with

inner longitudinal and outer circular layers. Mid-dorsal vessel without vascular plug. Precerebral septum present. Dorso-ventral muscles well developed. Cephalic glands well developed. Submuscular glands not developed. Cerebral organs opening around precerebral septum, posteriorly running without branching, and replaced with yellow to green glands just anterior to the brain region. Lateral nerve cords with myofibrillae but no accessory nerves.

Etymology: The generic name is a compound word, *Genroku-* (after the type locality, the southern part of Genroku Seamount Chain) and the Greek name *Nemertes*.

Genrokunemertes obesa Hookabe, Koeda, Fujiwara, Tsuchida & Ueshima, 2022

(Figs. 26–28)

Material examined: Holotype: NSMT-Ne-002 (female), serial transverse sections of anterior body fragment, 5 slides, collected at a depth of 2084 m, south of Genroku Seamount Chain (30°39.600N, 139°02.410E), Nishi-Shichito Ridge, Japan. KM-ROV dive #154, by Natsumi Hookabe, on 15 October 2021.

Habitat and geographic distribution: The species is only known from the type locality, off south of Genroku Seamount Chain, Nishi-Shichito Ridge, Japan, at a depth of 2,084 m.

Etymology: The specific name is derived from the Latin adjective “*obesus*” (stout, plump), referring to stout body of the new species.

Description: *External features.* Body 32.0 mm in length and 2.3 mm in maximum width; body ground color uniformly pale orange slightly with reddish tone both dorsal and ventral surfaces (Fig. 26A, C); internal organs (intestine, gonads) visible through body wall as pale-colored region. Cerebral ganglia visible through body wall as reddish spots (Fig. 26B, C). Cerebral organs becoming visible through body wall as black spots after cleared in xylene (Fig. 26D). Cephalic furrows hardly distinguished in living specimen, but merely visible in cleared

specimen in xylene (Fig. 26D, E); anterior cephalic furrow incompletely encircling body, opening at mid-dorsal line (Fig. 26D, F); a single pair of posterior cephalic furrows extending posteriorly on dorsal surface and meeting each other at mid-line, ventrally forming transverse line (Fig. 26F). Eyes absent (Fig. 26B, D).

Internal morphology. Epithelium 30–42 mm in thickness, with numerous red, yellow, and blue-staining gland cells and ciliated cells (Fig. 27A, B). Dermis up to 33 mm thick, more developed in intestinal (Fig. 27B) than precerebral region (Fig. 27A). Dorsoventral muscles between intestinal diverticula present (Fig. 27Q). Acidophilic and basophilic glands well developed in precerebral region (Fig. 27D). Submuscular glands not developed throughout the body. Cephalic lacuna posteriorly bifurcated and laterally situated at both sides of rhynchodaeum (Fig. 27E, G). Mid-dorsal vessel without protruding into rhynchocoel (Fig. 27I, O). Oesophagus short, opening just posterior to precerebral septum (Fig. 27J) and leading to stomach; stomach wall gradually developed with ciliated cells and red and yellow-stained acidophilic cells at ventral commissure of brain (Fig. 27K). Intestinal caecum anteriorly branched beneath pylorus without reaching to brain region (Fig. 27P); four pairs of branched lateral diverticula present. Proboscis lost in the specimen examined.

Rhynchocoel musculature bilayered with outer circular and inner longitudinal muscle walls (Fig. 27I).

A single frontal organ present (Fig. 27C). Cerebral organ laterally opening around precerebral septum (Fig. 27H), posteriorly running without branching (Fig. 26K, L), and leading to yellow- to green staining glands just anterior to brain region (Fig. 27M); cerebral organ up to 105 mm in diameter. Brain with outer neurilemma (Fig. 27J); dorsal cerebral ganglia with glomerular structures (Fig. 27J). Lateral nerves with myofibrillae (Fig. 27N); an accessory nerve not found.

Nephridial tubules convoluted in posterior region of brain (Fig. 27O, P). Ovary-like sacs situated between inner longitudinal muscle layer and intestine (Fig. 27Q); dorsoventral processes supported by dorso-ventral muscles between ovary-like sacs (Fig. 27Q). Each ovary-like sacs containing numerous numbers of small oocyte-like cells (Fig. 28A); cells 10–13 μm in diameter; each cell containing a distinct vesicle (Fig. 28A–C).

Remarks: The present species is morphologically most similar to species in *Kurilonemertes* (Chernyshev, 1993) due to the following characters: (i) cylindrical and weakly flattened body,

(ii) well-developed anterior cephalic furrows, (iii) body-wall longitudinal musculature anteriorly without divided, (iv) a single vascular plug originated from mid-dorsal vessel lacking. However, it is differentiated from *Kurilonemertes* in the lack of eyes as well as the absence of accessory nerves in lateral nerve cords; currently known three species in the genus, *Kurilonemertes papilliformis* (Korotkevitch, 1977), *K. phyllospadicola* (Stricker, 1985), and *K. dilutebasisae* (Kulikova, 1987), possess four irregular shaped eyes (Chernyshev 1999). Having a weakly flattened body and lacking eyes, the present species resembles *Typhloerstedtia*, harboring a single species *T. anadonae* (Chernyshev 1999) (originally reported as *Oerstedtia vittata* Hubrecht, 1879 from intertidal zone in Morocco (Anadon and Bitar 1992)); however, *G. obesa* is differentiated from *Typhloerstedtia* in having well-developed anterior cephalic furrows, cephalic glands, and cerebral organs anterior to the brain region.

Genus *Rhombonemertes* Hookabe, Moritaki, Jimi & Ueshima, 2022

Type species. *Rhombonemertes rublinea* Hookabe, Moritaki, Jimi & Ueshima, 2022

Diagnosis: Eumonostiliferan with four eyes. Head with anterior and posterior cephalic furrows. Rhynchocoel short, less than half of body length; rhynchocoel musculature bilayered (outer circular and inner longitudinal muscles). Body-wall musculature not anteriorly divided. Some diagonal muscle fibers arranged as a lattice between outer longitudinal and inner circular muscle layers. Precerebral septum present. Dorsoventral muscles present. Oesophagus short, replaced with stomach in precerebral region. Intestinal caecum with lateral diverticula, not reaching brain anteriorly. Vascular plug lacking. Lateral nerves without accessory nerves.

Etymology: The generic name refers to the diamond-shaped head of the present species, a composite of the Greek feminine *rhómbos* (rhombus, diamond- or lozenge-shaped) + *Nemertes* (Nemertea).

(Figs. 29–31)

Material examined: Holotype: NMST-Ne-003 (male), unsectioned complete specimen except for the posterior tip, fixed in Bouin's fluid and later preserved in 70% ethanol, posterior tip preserved in 99% ethanol, male, collected at depths of 150–200 m, off Owase (approx. 34°06'49.0"N, 136°30'35.0"E), the Sea of Kumano, Japan, by Naoto Jimi, on January 10. Paratype: NMST-Ne-004 (female), serial transverse sections from anterior tip to intestinal region (6 slides), everted proboscis (5 slides), and serial frontal sections of intestinal region (5 slides), posterior tip used for DNA extraction, rest of body fixed in Bouin's fluid and later preserved in 70% ethanol, collected at depths of 150 m, off Kumano (33°52'18.4"N, 136°11'54.8"E), the Sea of Kumano, Japan, by Takeya Moritaki, on February 7, 2022.

Etymology: The specific name is derived from a composite of the Latin *ruber* (red colored) + *linea* (=line), referring to the mid-dorsal stripe of the present species.

Habitat and geographic distribution: The present species was found only from the type locality, off Owase to Kumano, the Sea of Kumano, Japan.

Description: *External features.* Body 0.8 cm in length and 0.1 cm in width in male (holotype); 3.2 cm in length and 0.7 cm in width in female (paratype) (Fig. 29A); single median brownish-red stripe extending from transverse band in head region to posterior tip of body (Fig. 29A, D). Four eyes present (Fig. 29B, D). Head demarcated from body, anteriorly pointed, and diamond shaped (Fig. 29A); brownish-red-colored transverse band present between head and trunk (= dorsally between anterior and posterior cephalic furrows (Fig. 29B) while ventrally overlapped with posterior cephalic furrows (Fig. 29C)). Anterior cephalic furrows not meeting mid-dorsally, each slightly curving posteriorly in lateral margin (Fig. 29B); ventrally transverse, meeting at mid-line without reaching to proboscis pore (Fig. 29C). Posterior cephalic furrows meeting at mid-line both dorsally (Fig. 2B) and ventrally (Fig. 29C); gently V-shaped in dorsal surface (Fig. 29B) and steeply A-shaped in ventral surface (Fig. 29C). Blood not red, probably uncolored. Cerebral ganglia red colored in squeezed preparation (Fig. 29D), laterally leading

from cerebral organs via cerebral canals (Fig. 2E). Intestinal caecum with lateral diverticula, anteriorly not reaching cerebral ganglia (Fig. 29D); lateral diverticula of intestinal caecum and intestinal diverticula deeply branched (Fig. 29D). Pinkish ovary or pale testes visible through body wall (Fig. 29A).

Internal morphology. Epithelium 45–60 μm thick, with red-stained acidophilic gland cells in precerebral region (Fig. 30A) and with yellow- and red-stained gland cells in foregut region (Fig. 30B). Dermis up to 20 μm , cup-shaped in precerebral region (Fig. 30A) and flattened in foregut region (Fig. 3B). Cephalic glands anteriorly with red-stained acidophilic cells (Fig. 30D, E) and posteriorly replaced with blue-stained basophilic cells (Fig. 30G). Body-wall musculature with outer longitudinal and inner circular muscles (Fig. 30A, B). Some diagonal muscle fibers present as a lattice between outer circular and inner longitudinal muscle layers (Fig. 30C). Dorsoventral muscles present between intestinal diverticula (Fig. 30L). Precerebral vessel bifurcated (Fig. 30E, F). Mid-dorsal vessel without protruding into rhynchocoel (Fig. 30H). Oesophagus short, replaced with stomach in precerebral region (around cerebral organ opening) (Fig. 31); stomach with ciliated cells, well-developed basophilic glands, and acidophilic glands (Fig. 30G). Intestinal caecum developed beneath pylorus, 2 or 3 pairs of lateral diverticula deeply branched, not reaching brain region anteriorly (Fig. 30G).

Proboscis pore subterminal, opening ventrally (Fig. 29B). Rhynchocoel short, less than half of body length (Fig. 29D). Rhynchocoel musculature with outer circular and inner longitudinal muscles (Fig. 30L). Proboscis epithelium with developed papillae up to 70 μm thick, basophilic, and acidophilic glands (Fig. 30I); outer circular, middle longitudinal, and inner circular muscles (Fig. 30I); 8 proboscis nerves (Fig. 30I). Stylet basis oval, 22 μm in length and 13.0 μm in maximum width; central stylet smooth, 26.4 μm in length; stylet-to-basis-length ratio 1.20 (Fig. 29E). Two accessory stylet pouches present, each containing 2–3 stylets (Fig. 29E).

A single frontal organ present (Fig. 30D). Cerebral organs opening ventrolaterally; cerebral canals without branching and replaced with acidophilic glands anterior to precerebral septum (Fig. 31). Brain with outer neurilemma (Fig. 30J); glomerular structures present in dorsal cerebral ganglia (Fig. 30J). Lateral nerves without accessory nerves (Fig. 30K).

Nephridial tubules convoluted and opening dorsolaterally at intestinal caecum (Fig. 30K). Each ovary containing as many as 10 oocytes; each oocyte containing a germinal vesicle; oocyte 150–200 μm in diameter (Fig. 30L, M).

Remarks: Members of Oerstediiidae Chernyshev, 1993 are morphologically circumscribed by a bilayered rhynchocoel-wall musculature, the cephalic vessels protruding into the rhynchocoel in cerebral ring, and the mid-dorsal vessel not penetrating the rhynchocoel (= lacking vascular plug) (Kajihara 2021). Although the cephalic vessels could not be traced in the cerebral region, the other two features were confirmed in *Rhombonemertes rublinea*. (Fig. 30H, L), leading to my conclusion that the species belongs to Oerstediiidae. This is further supported by the molecular phylogenetic analyses, where *R. rublinea* is nested in the Oerstediiidae clade and more closely related to the monophyletic *Oerstedtia* clade than to *Argonemertes australiensis* (Dendy, 1892) in Plectonemertidae (Fig. 3).

Among 127 monostiliferan genera (Kajihara 2021b), comparatively few are known to possess a short rhynchocoel (Gibson 1990b). In Table 11, free-living marine oerstediid genera with a bilayered rhynchocoel wall, rhynchocoel extending half the body length or shorter, and without a vascular plug are listed with selected morphological characters. *Rhombonemertes rublinea* is dissimilar to any known oerstediid species except *Ischyronemertes* Gibson, 1990b. This is especially due to: *i*) the presence of body-wall diagonal musculature, *ii*) the longitudinal muscle layer of body wall not divided anteriorly, *iii*) the presence of a precerebral septum, *iv*) 4 eyes, and *v*) free-living, not parasitic or commensal lifestyle. *Ischyronemertes* was originally described from mid- to lower shore in Vancouver Peninsula (Western Australia) for three species, *I. albanensis* Gibson, 1990b, *I. erythrophleps* Gibson, 1990b, and *I. tetraphthalma* Gibson, 1990b. Most conspicuous difference between *R. rublinea* and *Ischyronemertes* is the shape of head; the former possesses a lozenge-shaped head while the latter is characterized by a more rectangular, somewhat anteriorly bilobed head (Gibson 1990b, fig. 17, 19, 21). The new species is also differentiated from *I. albanensis* and *I. erythrophleps* in the number and arrangement of eyes—four large eyes in the new species (Fig. 29A, B, D) and 2–6 small eyes more or less irregularly arranged in four groups in *I. albanensis* and *I. erythrophleps* (Gibson 1990b). Furthermore, accessory nerves are present in lateral nerve cords of *Ischyronemertes* (Gibson 1990b), which is another difference between *R. rublinea*. and *Ischyronemertes*. Based on these morphological differences between the existing genera in Oerstediiidae, we establish a new genus, *Rhombonemertes*, for *R. rublinea*.

In addition to the oerstediid genera in Table 11, *Nemertopsis tetraclitophila* Gibson, 1990a resembles *R. rublinea* gen. et sp. nov. in terms of the internal morphology; Hong Kong specimens of *N. tetraclitophila* exceptionally lack a mid-dorsal vascular plug in the genus (Gibson 1990a), and thereby display a character state that fits morphological circumscription

of Oerstediiidae (Kajihara 2021). As was the case in *N. quadripunctata* (Quoy & Gaimard, 1833) (Kajihara 2007b), however, the vascular plug might be hard to recognize in the Hong Kong specimens due to the small-sized body. Thus, the internal morphology of *N. tetraclitophila* needs to be revisited with additional materials. Apart from the internal morphology, *R. rublinea* is clearly distinguished from *N. tetraclitophila* in the external appearance—two longitudinal dark-green stripes on the dorsal surface and the rounded head in *N. tetraclitophila*.

3.4.4 Discussion

Systematic discussion for Genrokunemertes and Rhombonemertes

In the resulting tree, *R. rublinea* was a sister clade of *G. obesa* (Fig. 3); uncorrected *p*-distance based on COI between the two species was higher than interspecific COI divergences (e.g., Chen et al. 2010; Sundberg et al. 2016; Hookabe et al. 2022b). Morphological differences between the two species also warrant the recognition of *R. rublinea* as a species distinct from *G. obesa*—the latter species lacking eyes and diagonal muscles of the body wall, and having rhynchocoel extending 3/4 of the body length. The sister-taxon relationships with the clade formed by *R. rublinea* and *G. obesa* were not well resolved in the ML and BI analyses (Fig. 3). More extensive taxon sampling including apparently polyphyletic, and yet-to-be-discovered deep-sea oerstediid lineages will be required to obtain a more robust phylogeny of these taxa.

Ovary-like structures in Genrokunemertes

Apart from the absence of eyes, *Genrokunemertes obesa* is also characterized by having sacs packed with numerous oocyte-like cells (Fig. 27Q, R, 28A, B), each of which contains a single distinct vesicle (Fig. 28C). I considered this structure to be oocytes in ovaries of this species due to: *i*) sacs situated between intestinal caecum or diverticula as in nemertean gonads (Fig. 3Q, 3R and 4A), *ii*) each sac surrounded by thin wall (Fig. 28A, B) as in hoplonemertean ovaries in the peak breeding season (Stricker et al. 2001). On the other hand, atypical features are also found in the organs unusually small-sized oocytes compared to other hoplonemerteans (Stricker et al. 2001) and dorsoventral tube-like processes connecting dorsal and ventral ovary-like sacs (Fig. 28D). Supposedly, the two features are due to the immature state of the present specimen; immature ovaries between intestinal caecum and diverticula might appear as

dorsoventral tube-like processes. With the currently available data, I cannot exclude another possibility for the structure body of female orthonectids interiorly packed with irregularly arranged oocytes (e.g., plasmodium of several species in *Rhopalura* (Caullery and Lavalley 1912; Atkins 1933)) parasitizing in nemertean ovaries although any contaminated DNA sequences were detected from total DNA extracted from the posterior body fragment containing ovary-like sacs. Further morphological studies especially on gonads and gametes with additional specimens are needed for a firm conclusion.

Ecological notes of Rhombonemertes

Sunken wood has been increasingly recognized as important oases in the deep-sea environment, providing organic resources, shelter, and substrata (Turner 1977; Wolff 1979; Bernardino et al. 2010; Bienhold et al. 2013) for various marine invertebrates recently discovered (e.g., Hasegawa 1997; Sigwart and Sirenko 2012; Magalhães et al. 2017; Barroso et al. 2018; Voight et al. 2019; Ariyama and Moritaki 2020). Nemerteans were also reported from wood falls or experimental wood deployments in previous ecological studies; in most cases, however, they were listed as *Nemertea* sp. (Judge and Barry 2016; Amon et al. 2017; Saeedi et al. 2019). Thus, species diversity of nemerteans associated with organic falls or prey invertebrates such as annelids and amphipods predominantly occurred there is not well understood. Current knowledge and data are not enough to tell us whether *R. rublinea* actually prefers sunken wood pieces or porous hard structures because their colonization preference is not well investigated in the present study (e.g., ecological comparisons of specimen occurrence rates between bottom sediments and sunken woods). If some deep-water inhabiting nemerteans prefer cryptic environments as do shallow-water species, the species diversity may become even clearer by focusing on structures that sink on the sea floor.

3.5 Plectonemertidae Gibson, 1990a (EUMONOSTILIFERA)

This chapter deals with a description of new species in *Leptonemertes* Girard, 1893, which is the first member of Plectonemertidae Gibson, 1990a in Japan.

第3章-5については、5年以内に雑誌等で刊行予定のため、非公開とする。

3.6 Prosorhochmidae Bürger, 1895 (EUMONOSTILIFERA)

This chapter deals with a description of an unidentified species of *Geonemertes* Semper, 1863 from Ogasawara Island, Japan, which had long been recognized as widespread species *G. pelaensis* Semper, 1863. I also provide a redescription of *G. pelaensis* and *Prosadenoporus spectaculum* (Yamaoka, 1940) based on specimens collected from Okinawa, Japan with DNA sequences.

第3章-6については、5年以内に雑誌等で刊行予定のため、非公開とする。

3.7 Sacconemertidae Chernyshev, 2005 (EUMONOSTILIFERA)

This chapter deals with redescriptions of Japanese species in Sacconemertidae Chernyshev, 2005 including *Sacconemertopsis olivifera* Iwata, 1970 and *Potamostomma shizunaiense* Kajihara, Gibson & Mawatari, 2003, as well as a description of an undescribed species in Sacconemertidae collected from Hokkaido, Japan. Furthermore, I herein provide a new record of *Gurjanovella littoralis* Uschakov, 1926 from Japanese waters, which was published in Hookabe and Kajihara (2020).

第3章-7については、5年以内に雑誌等で刊行予定のため、非公開とする。

3.8 Tetrastemmatidae Hubrecht, 1879 (EUMONOSTILIFERA)

This chapter deals with descriptions of five new species in *Tetrastemma* Ehrenberg, 1828. These studies were partially published in Hookabe et al. (2021).

3.8.1. Introduction

A histology-free description has been progressively introduced to nemertean systematics in the past decade (e.g., Kajihara 2015; Gonzalez-Cueto et al. 2017; Simpson et al. 2017; Kajihara et al. 2018, 2022b; Chernyshev et al. 2020; Hookabe et al. 2021a, b; Leiva et al. 2021). This approach has been applied to two cases, one of which is a description of species with internal characters interspecifically differentiated and observable without histology (e.g., number of proboscis branches in *Gorgonorhynchus* Dakin & Fordham, 1931 [Kajihara 2015; Hookabe et al. 2021a]). In the other case, especially when internal morphology is uniform between most species in a genus, a species description has been performed solely based on characters examined *in-vivo* (shape of head, body coloration and markings, number of eyes, blood color, and stylet apparatus) [e.g., *Baseodiscus* Diesing, 1850 (Kajihara et al. 2022) and *Ototyphlonemertes* Diesing, 1863 (Kajihara et al. 2018)]. Recent descriptions of species in the genus *Tetrastemma* Ehrenberg, 1828, fitting the latter case, have been performed based on characters of living specimens without histological observations (Chernyshev et al. 2020; Hookabe et al. 2021b).

Tetrastemma is a species-rich genus in Monostilifera (Kajihara 2021b), currently harboring about 110 species from tropical to polar areas (Chernyshev et al. 2021). As the generic name suggests—a composite of the Latin feminine “tetra” (=four) + “stemma” (=simple eyes), members in the genus are generally characterized by having four eyes. Several species in *Tetrastemma* were described based on internal morphology; however, the internal characters were inferred to be almost homogenous within the genus by taxonomic reappraisal based on molecular phylogeny (Chernyshev et al. 2021). Recent examples of a histology-free approach based on characteristics studied *in-vivo* and molecular data are descriptions of *T. freyae* Chernyshev et al., 2020 and *T. cupido* Hookabe, Kohtsuka & Kajihara, 2021.

During faunal surveys in Sagami Bay (2006–2019) with support by Misaki Marine Biological Station, the University of Tokyo and Nishi-Shichito ridge in 2020 under the project ‘Development of Biodiversity Monitoring Methods for the Management of Deep-sea Marine Protected Areas’ in 2021, I obtained several specimens of *Tetrastemma*. The descriptions are

histology-free, based on characters of living specimens examined with a light microscope. To test phylogenetic relationships with the congeners, I performed molecular phylogenetic reconstruction using partial sequences of 16S, COI, 18S, 28S and H3.

3.8.2 Materials and Methods

Taxon sampling and morphological examination

Specimens were collected in 2019–2021 by use of a biological dredge in Sagami Bay or a remotely operated vehicle (ROV) on Shoho Seamount of the Nishi-Shichito Ridge, northwestern Pacific Ocean. External morphology of the living specimens was documented on the vessel or the laboratory with a Nikon D5600 Digital SLR Camera equipped with an AF-S DX Micro-NIKKOR 40mm f/2.8G macro lens (Nikon, Japan). A single specimen collected from Shoho Seamount was further observed under a compound light microscope by preparing a squeezed specimen with a cover slip and a slide glass. Specimens were anaesthetized with few drops of bitterns Tenpi Nigari (Amashio, Japan); after the worms were relaxed, the posterior tips were preserved in 99% ethanol for DNA extraction and the rest of the body was fixed in Bouin's fluid for 24–48 hours and later transferred to 70% ethanol. Type specimens have been deposited in the National Museum of Nature and Science, Tsukuba (NSMT), Japan.

DNA extraction, PCR amplification, and sequencing

DNA extraction, amplification, and sequencing were performed based on the same methods in Subchapters 2.2 and 3.2. Genetic distances were calculated using the same primers and the software used in Subchapter 3.2.

Sequence alignment and phylogenetic analyses

Methods used in the present study follow Subchapters 2.2. and 3.2. OTUs used in the present study are listed in Table 13.

3.8.3 Results

Phylogeny

The sequence data set for molecular phylogenetic analyses in the present study is primarily based on Chernyshev et al. (2021), and the monophyly for *Tetrastemma* species and phylogenetic relationships between species were tested. Since I confirmed that the new species are nested in *Tetrastemma* Clade B in Chernyshev et al. (2021) by phylogenetic analyses based on sequence dataset from other *Tetrastemma* and eumonostiliferan species in Chernyshev et al. (2021), I used three species in Clade A (*Tetrastemma* sp. GM1 Gulf of Mexico, *Tetrastemma* sp. GM2 USA FL, and *Tetrastemma* sp. GM3 Gulf of Mexico) as outgroup taxa (Fig. 42). Clade B was subdivided into two clades with a full support value, one of which was a clade formed by *T. freyae* and *T. shohoense* sp. nov. The two species are characterized by having a cylindrical stylet basis in the proboscis. In the other subclade in Clade B, *T. alba* sp. nov. and *T. persona* sp. nov. were included (Fig. 42). A clade constituted by newly described species, *T. album* sp. nov. and *T. persona* sp. nov., from Sagami Bay (Japan) with 99% of BS, was nested in the American Atlantic clade formed by *T. elegans* (Girard, 1852) (Virginia), *T. enteroplecta* (Corrêa, 1954), (Florida and Venezuela), *T. merulum* (Corrêa, 1954) (Florida), and *Tetrastemma* sp. F7 (Florida). The clade formed by *T. album* sp. nov. and *T. persona* sp. nov. was sister-related to a clade formed by *T. enteroplecta* (Florida and Venezuela) and *Tetrastemma* sp. F7 (Florida) with 77% of BS (Fig. 42).

Systematics

Family Tetrastemmatidae Hubrecht, 1879

Genus *Tetrastemma* Ehrenberg, 1878

Type species: *Tetrastemma flavidum* Ehrenberg, 1878

Diagnosis: Eumonostiliferous nemertean lacking eyes with weakly flattened, stout body. Head with anterior and posterior cephalic furrows. Body-wall longitudinal musculature anteriorly not

divided. Rhynchocoel reaching to more than 3/4 of body length; rhynchocoel musculature with inner longitudinal and outer circular layers. Mid-dorsal vessel without vascular plug. Precerebral septum present. Dorso-ventral muscles well developed. Cephalic glands well developed. Submuscular glands not developed. Cerebral organs opening around precerebral septum, posteriorly running without branching, and replaced with yellow to green glands just anterior to the brain region. Lateral nerve cords with myofibrillae but no accessory nerves.

Tetrastemma album sp. nov.

[New Japanese name: misaki-oshiroi-himomushi]

(Fig. 43A–C)

Material examined: Holotype: NMST-Ne-H-06, unsectioned complete specimen except for the posterior tip, fixed in Bouin's fluid and later preserved in 70% ethanol, posterior tip preserved in 99% ethanol, collected with biological dredge (R/V *Rinkai-maru*) at depths of 144–200 m, off Jogshima (35°07'41"N, 139°34'11"E–35°07'32"N, 139°33'57"E), Miura, Kanagawa, Japan, on March 12 2021 by Natsumi Hookabe.

Etymology: The species name is derived from the Latin *album* (white), referring to pure-white colored body of the new species.

Habitat and geographic distribution: The present species was found from the type locality, off Owase to Kumano, the Sea of Kumano, Japan.

Description: Head spatulate to circular shaped (Fig. 43A–C), demarcated from body (Fig. 43A). A live specimen 17 mm in body length and 1.0–1.2 mm in body width. Body uniformly pale colored without longitudinal or transverse stripe markings (Fig. 43A). Pure white cephalic patch present between anterior and posterior pairs of eyes (Fig. 43B). Head not wider than maximum body width (Fig. 43A–C). A pair of cephalic furrows present; anterior pair without meeting mid-dorsally and ventrally curving anteriorly but not reaching to proboscis pore; posterior pair

V-shaped and barely meeting mid-dorsally (Fig. 43B) and running transversely on ventral surface (Fig. 43C). Cerebral ganglia and blood not red and probably uncolored. Internal organs (proboscis, foregut, and intestine) visible as pale regions. Four reddish brown eyes present (Fig. 43B).

Remarks: Having a pure white cephalic patch on a uniformly pale body, *T. album* sp. nov. differs from all the congeners. *Tetrastemma diadema* Hubrecht, 1879 has two white cephalic patches but is distinguished from *T. album* sp. nov. in possessing a light brown transverse band on the head (Hubrecht 1879). *Tetrastemma albomaculatum* Chernyshev, 2016 also possesses a white cephalic patch but differs from the new species in pale-ochre body dorsally spotted with small white dots (Chernyshev 2016).

Tetrastemma carneum sp. nov.

(Fig. 43D–F)

Material examined: Holotype: museum ID not assigned, unsectioned complete specimen except for the posterior tip, fixed in 10% formalin, posterior tip preserved in 99% ethanol, collected during KM19-05C cruise of R/V *Kaimei*, at a depth of 398 m, off Ofunato (39°03'0.17"N, 142°09'0.67"E), Iwate, Japan, by Yuki Oya on July 31, 2019. Paratypes: museum ID not assigned, unsectioned complete specimen except for the posterior tip, fixed in 10% formalin, posterior tip preserved in 99% ethanol, collected on the same date and locality as the holotype.

Etymology: The species name is derived from the Latin *carneum* (flesh-colored), referring to the body coloration of the new species.

Description: Head spatulate to circular shaped (Fig. 43E), demarcated from body (Fig. 43D, E); marginal region between anterior and posterior cephalic furrows constricted (Fig. 43D). Live specimens 11–25 mm in body length and 1.1–1.5 mm in body width. Background body

color generally pale tinged with flesh red, semitransparent either dorsally or ventrally (Fig. 43D). Head with a mid-dorsal thin dark longitudinal line and without a cephalic patch (Fig. 43E). A pair of cephalic furrows present; anterior pair without meeting mid-dorsally and ventrally curving anteriorly but not reaching to proboscis pore; posterior pair shallow V-shaped and meeting mid-dorsally (Fig. 43D, F) and slightly curving anteriorly on ventral surface (Fig. 43F). Cerebral ganglia and blood probably not red but hard to distinguish from flesh-colored internal organs. Alimentary canals visible flesh-colored organs through body wall (Fig. 43D). Proboscis pale colored, extending about 1/2 of the body length. Four large eyes present (Fig. 43E).

Habitat and geographic distribution: The species is only known from the type locality, off Ofunato, Iwate Prefecture, Japan, at a depth of 398 m, sorted from demosponge.

Remarks: The present species is extremely similar to *T. appendiculatum* Chernyshev, 1998b described from the Peter the Great Bay, the Sea of Japan and *T. cephalophorum* Bürger, 1895 from Naples in having four large eyes—it is rarely found in *Tetrastemma*—and pale-yellow to flesh colored body. However, it is distinguished from the latter two species in possessing a narrow mid-dorsal longitudinal line on the head.

Tetrastemma appendiculatum was described based on two specimens collected from demosponge (*Amphilectus digitatus* (Koltun, 1958)) at depths of 40–60 m (Chernyshev 1998b). *Tetrastemma appendiculatum* and *T. carnea* sp. nov. is similar to each other not only in the external morphology but also in the habitat preference to sponge. Several adult and juvenile specimens of *T. carnea* sp. nov. were found from one sponge, rather indicating specific association with the sponge in the species.

An uncorrected genetic distance based on 657-bp of COI was 5 % between *T. carneum* sp. nov. and *Tetrastemma* sp. IT Iturup, which was also collected from deep-sea demosponge (Alexei V. Chernyshev, personal observation) off Iturup, Russia.

Tetrastemma cupido Hookabe, Kohtsuka & Kajihara, 2021

[Japanese name: Misaki-kyūpiddo-himomushi]

(Fig. 44)

Material examined: ICHUM 6298 (holotype), preserved in 99% ethanol, collected with biological dredge (R/V *Rinkai-maru*), at depths of 90 m by dredging, off Jogashima, Kanagawa (35°8.9'N, 139°34.6'E), by Hisanori Kohtsuka, on 11 March 2014. ICHUM 6299 (paratype), preserved in 70% ethanol, collected with biological dredge (R/V *Rinkai-maru*), at depths of 97 m by dredging, off Jogashima, Kanagawa (35°8.3'N, 139°33.7'E), by Hiroshi Kajihara, on 15 February 2017. ICHUM 6300 (paratype), preserved in 70% ethanol, collected with biological dredge (R/V *Rinkai-maru*), at depths of 92 m by dredging, off Jogashima, Kanagawa (35°8.7'N, 139°34.4'E), by Hiroshi Kajihara, on 15 February 2017. ICHUM 6300 (paratype), preserved in 99% ethanol; posterior tip used for DNA extraction, collected with biological dredge (R/V *Rinkai-maru*), at depths of 112 m by dredging, off Jogashima, Kanagawa (35°8.5'N, 139°32.8'E), by Hiroshi Kajihara, on 22 February 2019.

Habitat and geographic distribution: The species is only known from the type locality, Sagami Bay, Kanagawa, Japan, at a depth of about 100 m (Table 1).

Etymology: The new specific name is a noun in the nominative case, derived from the Latin Cupīdō, a god of desire, erotic love, attraction and affection; a person, or even a deity, shot by Cupid's arrow is filled with uncontrollable desire in classical mythology. The new species is so named because its body looks like an arrow due to the triangular head, reminiscent of an arrow shot by Cupid.

Description: Head triangular to semicircular in shape on dorsal view, wider than trunk, often significantly constricted between succeeding portion of trunk; dark brownish cephalic patch present, slightly paler along mid-line (Fig. 44A–E). Body 2.0–5.3 cm in length (1.0 cm in holotype) and 1.3–2.0 mm in width (1.5–2.0 mm in holotype) (Fig. 44A–E); dorsally brownish

in color (often brighter than cephalic patch), with single median longitudinal white stripe; ventrally uniformly pale-colored. Head not wider than maximum body width (Fig. 44A–G). Single pair of cephalic furrows present, not meeting mid-dorsally, commencing on dorsal surface from slightly lateral to midline, running laterally along posterior edge of cephalic patch (Fig. 44F), curving anteriorly on ventral surface to meet mid-ventrally but not reaching to proboscis pore (Fig. 44G). Ocelli arranged in two, anterior and posterior, pairs of groups; anterior group consisting of up to eight ocelli, arranged just below antero-lateral edges of cephalic patch (and thus difficult to be discerned unless otherwise observed in squeezed preparation); posterior group consisting of up to four ocelli, situated slightly posterior to, and arranged along, posterior margins of cephalic patch (Fig. 44H). Cerebral sensory organs situated underneath paired clusters of posterior eye spots (Fig. 44I). Paired cerebral ganglia are not visible through body wall. Blood not red, probably uncolored. Intestinal caecum with lateral diverticula; anterior-most of latter seemingly reaching dorsal ganglia (Fig. 44I); lateral diverticula of intestinal caecum and main intestine often deeply branched (Fig. 44I).

Remarks: The generic placement of *T. cupido* is unquestionable in reference to its phylogenetic position in the resulting tree, being nested in a clade comprised of the other congeners including the type species *T. flavidum* (Fig. 3). Among ~570 species presently known in Eumonostilifera, the one that is morphologically most similar to *T. cupido* is perhaps *Amphiporus suberbus* (Girard in Stimpson, 1854). Having morphological allies in different genera is not surprising in this case if I consider the peculiarity of *T. cupido* (with >4 eyes) as well as the historical taxonomic entanglement between *Amphiporus* and *Tetrastemma* (Coe 1901; Friedrich 1955; Berg 1973; Gibson 1985, 1989; Norenburg 1986; Gibson and Crandall 1989; Chernyshev 2005; Strand and Sundberg 2005a).

Amphiporus suberbus was originally described as a new species in a new genus *Nareda superba* based on material dredged at a depth of 35 fathoms (64 m) deep in Hake Bay, Grand Manan, on the Atlantic coast of Canada (Stimpson 1854, p. 28, fig. 17); because Stimpson (1854) ascribed the species and genus to Charles Girard, the author(s) of these taxa should be cited as ‘Girard in Stimpson, 1854’ in accordance with Recommendation 51E of the International Code of Zoological Nomenclature (International Commission on Zoological Nomenclature 1999). Verrill (1892, p. 403) transferred *Nareda superba* to *Amphiporus* with some reservation, practically synonymizing *Nareda* Girard in Stimpson, 1854 with *Amphiporus* Ehrenberg, 1831. Subsequently, Berg (1973) reported 10 specimens of monostiliferans

collected from a depth of 45 m on a soft bottom with bivalve shells in Balsfjord, Tromsø, Norway, as *Amphiporus superbis*. At the same time, Berg (1973) considered *Tetrastemma albicollis* Uschakow, 1928 from Kola Bay in the Barents Sea, Russia, as conspecific with *A. superbis*. These worms from Canada (Stimpson 1854), Norway (Berg 1973), Russia (Uschakow 1928) and Japan (present study) commonly possess *i*) pale-colored body, dorsally brownish; *ii*) head with brownish cephalic patch, often conspicuously demarcated from the trunk; *iii*) eyes in four groups (not obvious in Stimpson (1854)); and *iv*) body not as long as the width. Assessing if the Canadian, Norwegian and Russian specimens constitute a single species is beyond the scope of this study, but the present Japanese material is clearly different from the others by having a narrow, mid-dorsal, white stripe (Fig. 44A–E). Although *Nareda* was later restored in Chernyshev and Maslakova (2011), taxonomic reappraisals for the genus would be needed if the Canadian form is included in a *Tetrastemma* clade in future molecular phylogenetic studies.

Tetrastemma persona sp. nov.

[New Japanese name: misaki-kamen-himomushi]

(Fig. 43G)

Material examined: Holotype: NMST-NE-H-07, unsectioned complete specimen except for the posterior tip, fixed in Bouin's fluid and later preserved in 70% ethanol, posterior tip preserved in 99% ethanol, collected with biological dredge (R/V *Rinkai-maru*) at depths of 116–211 m, off Jogshima (35°08.32'N, 139°32.857'E–35°08.40'N, 139°32.504'E), Miura, Kanagawa, Japan, on July 31 2020 by Natsumi Hookabe. Paratype: NMST-NE-P-08, unsectioned complete specimen fixed in Bouin's fluid and later preserved in 70% ethanol, collected on the same date and locality as the holotype.

Etymology: The species name is derived from the Latin *persōna* (mask), referring to a broad cephalic patch of the new species masking eyes and internal organs in head region.

Habitat and geographic distribution: The species is only known from the type locality, Sagami Bay, Kanagawa, Japan, at depths of 116–211 m (Fig. 2, Table 1).

Description: Head slightly narrower than middle part of body and weakly demarcated from trunk (Fig. 43D). A live specimen 7.0–10 mm in body length and 0.8–1.0 mm in body width. Body uniformly pale to yellow colored without longitudinal or transverse stripe markings (Fig. 43D). Vermilion-red cephalic patch spade-shaped (Fig. 43D), covering both anterior and posterior pairs of eyes (Fig. 43D) but not posteriorly reaching to anterior pair of cephalic furrows. A posterior pair of cephalic furrows not well distinguished probably due to the small body size. Cerebral ganglia and blood not red and probably uncolored. Internal organs (proboscis, foregut, and intestine) not well visible through body wall. Rhynchocoel visible as whitish region through body wall, extending about 1/2–2/3 of the body length.

Remarks: *Tetrastemma persona* sp. nov. most resembles *T. roseocephalum* (Yamaoka, 1947) and *T. yamaokai* Iwata, 1954 in having a pale body without any markings and a red cephalic patch. Pattern variation of a cephalic patch (shield shape or horse-shoe shape) was reported in both *T. roseocephalum* and *T. yamaokai*; referring to the original description of *T. yamaokai*, the name may be a junior synonym of *T. roseocephalum*. The external morphology of *T. persona* sp. nov. is similar to a form with a shield-shaped cephalic patch of *T. roseocephalum*. The subtle difference in the shape of cephalic patch between *T. persona* sp. nov. (spade-shaped) and *T. roseocephalum* (shield-shaped) was supported by my molecular analysis. The new species did not constitute a clade with *T. roseocephalum* but with *T. album* sp. nov. (Fig. 42).

An uncorrected genetic distance based on 657 bp of COI was 16 % between *T. album* sp. nov. and *T. persona* sp. nov., comparable with interspecific values observed among Monostilifera (e.g., Sundberg et al. 2016; Hookabe et al. 2022b).

Tetrastemma shohoense sp. nov.

[New Japanese name: shoho-kakubari-himomushi]

(Fig. 43H–L)

Material examined: Holotype: NMST-Nem-H-05, unsectioned complete specimen except for the posterior tip, fixed in Bouin's fluid, posterior tip preserved in 99% ethanol, collected by use of ROV KM-ROV (dive #123) during KM20-10C cruise of R/V *Kaimei*, at a depth of 455 m, near the summit of Shoho Seamount of the Nishi-Shichito Ridge (32°19.39'N, 138°44.48'E), Japan, on November 29 2020 by Natsumi Hookabe.

Etymology: The species name is named after the type locality, Shoho Seamount of the Nishi-Shichito Ridge, Japan.

Description: Head spatulate shaped (Fig. 43I, J), not well demarcated from body (Fig. 43H, K). Live specimen 5.5 mm in body length and 0.3 mm in body width. Background body color generally white tinged with bright yellow to orange and almost transparent (Fig. 43H, I). Head with a red cephalic patch (Fig. 43H, I). Anterior pair of cephalic furrows present (Fig. 43I) but posterior one not well distinguished. Cerebral ganglia and blood uncolored (Fig. 43K). Alimentary canals visible as bright-yellow-colored organs through body wall (Fig. 43H). Proboscis pale colored, extending about 3/4 of the body length (Fig. 43H). Four eyes present.

Stylet basis cylindrical, 55.0 μm in length and 25.0 μm in maximum width; central stylet smooth, 47.0 μm in length; (stylet length)/ (basis length) ratio 0.85 (Fig. 43L). Two accessory stylet pouches present, each containing 2 stylets (Fig. 43L).

Habitat and geographic distribution: The species is only known from the type locality, Shoho Seamount of the Nishi-Shichito Ridge, Japan, at a depth of 455 m (Fig. 2, Table 1), among the sandy sediments on rocky substrates.

Remarks: Having a cephalic patch and cylindrical stylet basis and lacking a longitudinal line on the dorsal surface of the body, *T. shohoense* sp. nov. resembles *T. freyae* Chernyshev et al.,

2020 originally described based on Hawaiian and Indian specimens. The new species is differentiated from *T. freyae* in the color of the cephalic patch as well as the non-flared posterior margin of the cylindrical stylet basis.

A genetic distance based on COI between *T. shohoense* sp. nov. and *T. freyae* was 13%; the value is comparable with interspecific values observed among Monostilifera (e.g., Sundberg et al. 2016 and Hookabe et al. 2022a).

3.8.4 Discussion

Number of eyes in Tetrastemma

Tetrastemma cupido is atypical among *Tetrastemma* in possessing more than four eyes (Fig. 44H). Considering the character distribution, the most recent common ancestor of all the *Tetrastemma* species likely possessed four eyes. If true, an evolutionary increase of eye number must have happened somewhere between the *Tetrastemma* common ancestor and *T. cupido*. In this context, it is interesting to see *T. stimpsoni* be closely related to *T. cupido* in the phylogenetic tree (Fig. 3, 42). In *T. stimpsoni*, the number of eyes decreases from six to four after free-swimming larvae settle to a benthic stage, with the eyes in the second pair fusing with those in the first pair (Chernyshev 2008). The extent of the generality of this phenomenon among other *Tetrastemma* species is currently uncertain, but the number of the eyes is ontogenetically/phylogenetically plastic at least in the vicinity of this subclade consisting of *T. cupido* and *T. stimpsoni* within the entire *Tetrastemma* clade.

Geographic distribution of Tetrastemma species

In the resulting tree (Fig. 42), *T. album* sp. nov. and *T. persona* sp. nov. are nested in a clade formed by several American Atlantic species [*T. enteroplecta*, *T. elegans*, *T. merulum*, and *Tetrastemma* sp. F7 (Chernyshev et al. 2021)] (Fig. 42). Previous molecular analysis inferred that the *Tetrastema* clade B is subdivided into geographically distinct structures, North Pacific and American Atlantic subclades (Chernyshev et al. 2021), but it is less likely based on the new data herein obtained. Speciation in this lineage seems not to be explained solely by geographic distribution. To obtain a more accurate picture of *Tetrastemma* phylogeny and speciation, again, further taxon sampling without biased toward shallow-water species is needed for future phylogenetic analyses.

Chapter 4.

Concluding Remarks

4.1 Species diversity of Monostilifera in Japan

Monostiliferous nemerteans have been reported in various habitats, including marine, freshwater, and terrestrial environments, and some of them are known as commensal/parasitic species associated with other marine invertebrates (Gibson 1994). It is one promising taxon that uncovers the evolutionary process of morphological and physiological adaptation, where the ancestral marine benthic forms evolved into novel habitats, such as freshwater and terrestrial environments, and lifestyles, including commensal and parasitic relationships. Despite the potential, a paucity of taxonomic and phylogenetic studies on monostiliferans has hindered progress in understanding their evolution. To facilitate the baseline of evolutionary research, I worked on systematic studies of the Monostilifera, including taxonomy and phylogeny of 8 families: Cratenemertidae (Hookabe et al. 2022b), Amphiporidae (Hookabe et al. 2020), Carcinonemertidae (Hookabe et al. 2022a), Oerstediiidae (Hookabe et al. 2022c, d), Plectonemertidae (Hookabe et al. under review), Prosorhochmidae (Hookabe et al. in preparation), Sacconemertidae (Hookabe and Kajihara 2020), and Tetrastemmatidae (Hookabe et al. 2021b; Hookabe et al. under review). In these studies, I established 2 new genera and 20 new species and proposed two new combinations by species redescrptions. Moreover, the marine and semiterrestrial family Plectonemertidae was recorded for the first time from Japan (Hookabe et al. under review). Before my studies, a total of 44 species of monostiliferans were recognized in Japan (Kajihara 2017; Kajihara et al. 2018) and has been increased to 66 in this dissertation. Yet, there are still at least 5 unnamed forms (also see: Hookabe et al. 2022b) and more than 50 unreported/undescribed forms (also see: Kajihara 2017) awaiting formal description. I wish to continue to explore nemertean faunae in Northwestern Pacific to grasp and document the current species diversity of Monostilifera.

4.2 Monostiliferan molecular phylogeny

In the previous molecular phylogenetics in Monostilifera (Andrade et al. 2012; Kvist et al. 2014, 2015; Chernyshev and Polyakova 2019, 2022; Chernyshev et al 2021), phylogenetic

relationships between families were not well recovered. In Chapter 2, I performed phylogenetic analyses with increased taxon sampling for monostiliferans, which is one method to obtain a robust phylogenetic tree (Zwicki and Hillis 2002). In the result, the monophyly of Amphiporina, Oerstedina, and some monostiliferan families (Amphiporidae, Carcinonemertidae, Oerstedidae, Plectonemertidae, Sacconemertidae, Tetrastemmatidae *sensu stricto* (Chernyshev et al. 2021) was confirmed (Fig. 3). However, the inter-familial relationships within Amphiporina were not well resolved. Apart from one study employing transcriptome approaches with limited taxon sampling (Andrade et al. 2014), nemertean phylogeny has been conventionally made based on two mitochondrial (COI, 16S) and three nuclear gene markers (18S, 28S, and H3). In future phylogenetic studies for Monostilifera, it would be well worth trying to include a greater number of phylogenetic markers sampled from transcriptome datasets, in addition to extensive and dense taxon sampling, to obtain a robust phylogeny.

Amphiporina is the most ecologically diverse taxa in Monostilifera but diversification patterns within the lineage were not well understood. Our tree topology infers that ecological adaptation to parasitic lifestyle to crustacean hosts (Carcinonemertidae) and to interstitial habitat (Ototyphlonemertidae) might have basally happened in Amphiporina (Fig. 45) although it must be further tested with an increased number of OTUs and phylogenetic markers, especially for Carcinonemertidae exhibiting long branches (Fig. 3) probably due to those relatively rapid substitutional rates.

4.3 Future challenges in monostiliferan systematics

Deep sea or supratidal zones, especially in Japanese waters, are habitats where nemertean diversity has been scarcely understood. Specimens collected during my Ph. D course from these habitats revealed that their morphological characters sometimes fall outside of the morphological taxonomic circumscriptions such as the deep-sea lineage of *Nipponnemertes* without a vascular plug (Subchapter 3.1) (Hookabe et al. 2022b) and *Tetrastemma cupido* Hookabe, Kohtsuka & Kajihara 2021 with multiple eyes (Subchapter 3.8) (Hookabe et al. 2021). In many metazoan lineages putatively with a shallow-water origin, colonization and diversification in deep-sea waters occurred independently multiple times accompanied by morphological evolution (Lee et al. 2019; Miller et al. 2022; Rodríguez-Flores et al. 2022). Further faunistic studies in various ecosystems without biased toward intertidal to shallow-

water species should be made for monostiliferan systematics. In my future study, I wish to advance monostiliferan systematics by prioritizing the comprehension of the species diversity and then uncover the evolutionary background that drives ecological diversification in Monostilifera.

References

- Abe, T., Makino, S. I., Okochi, I. (2010). Why have endemic pollinators declined on the Ogasawara Islands?, in Restoring the Oceanic Island Ecosystem, eds. K. Kawakami, I. Okochi (Tokyo, Japan: Springer), 75–83.
- Abe, H., Kan, K., Matsumasa, M., Suzuki, T., Kajihara, H. (2022). Northernmost record of *Nipponnemertes ogumai* (Nemertea: Monostilifera) in Otomo-ura, Iwate Prefecture, Japan. *Michi. Ben.* 6, 21–27 [in Japanese].
- Ali, H., Deo, R.N., Rutherford, M.G. (2021). First records of the terrestrial nemertean *Geonemertes pelaensis* (Hoploneuridae; Prosorhochmidae) in Trinidad, WI. *Living World 2021*, J. T. T. Field Naturalists' Club, 1–2.
- Amon, D.J., Copley, J.T., Dahlgren, T.G., Horton, T., Kemp, K.M., Rogers, A.D., Glover, A. G. (2017). Observations of fauna attending wood and bone deployments from two seamounts on the Southwest Indian Ridge. *Deep Sea Res. Part II Top. Stud. Oceanogr.* 136, 122–132.
- Anadon, N., Bitar, G. (1992). On the anatomy of *Oerstedia vittata* Hubrecht, 1879 (Nemertes, Enopla, Monostiliferoidea). *Bull. Soc. Zool. France* 117, 383–391.
- Andrade, S.C.S., Strand, M., Schwartz, M., Chen, H., Kajihara, H., von Döhren, J., Sun, S., Junoy, J., Thiel, M., Norenburg, J.L., Turbeville, J.M., Giribet, G., Sundberg, P. (2012). Disentangling ribbon worm relationships: multi-locus analysis supports traditional classification on the phylum Nemertea. *Cladistics* 28, 141–159.
- Ariyama, H., Moritaki, T. (2020). A new species of the genus *Bathyceradocus* from the Kumano-nada, central Japan (Crustacea: Amphipoda: Maeridae). *Crustac. Res.* 49, 61–71.
- Atienza, S., Guardiola, M., Præbel, K., Antich, A., Turon, X., Wangensteen, O.S. 2020. DNA metabarcoding of deep-sea sediment communities using COI: community assessment, spatio-temporal patterns and comparison with 18S rDNA. *Diversity* 12, 123.
- Atkins, D. (1933). *Rhopalura granosa* sp. nov., an orthonectid parasite of a lamellibranch *Heteranomia squamula* L., with a note on its swimming behaviour. *J. Mar. Biol. Assoc. U.K.* 19, 233–252.
- Băcescu, M.C., Müller, G.J., Gomoiu, M.T. (1971). *Ecologie Marină. Cercetări de ecologie bentală în Marea Neagră; analiza cantitativă, calitativă și comparată a faunei bentale pontice. Vol. 4. Academiei Republicii Socialiste Româna, Bucuresti [Bucharest], 357 pp.*
- Barroso, R., Kudenov, J.D., Halanych, K.M., Saeedi, H., Sumida, P.Y., Bernardino, A.F. 2018. A new species of xylophilic fireworm (Annelida: Amphinomidae: Cryptonome) from deep-sea wood falls in the SW Atlantic. *Deep Sea Res. Part II Top. Stud. Oceanogr.* 137, 66–75.

- Beckers, P., Krämer, D., Bartolomaeus, T. (2018). The nervous systems of Hoplonemertea (Nemertea). *Zoomorphology* 137, 473–500.
- Berg, G. (1972a). Studies on *Nipponnemertes* Friedrich, 1968 (Nemertini, Hoplonemertini) I. Redescription of *Nipponnemertes pulcher* (Johnston, 1837) with special reference to intraspecific variation of the taxonomical characters. *Zool. Scr.* 1, 211–225.
- Berg, G. (1972b). Taxonomy of *Amphiporus lactifloreus* (Johnston, 1828) and *Amphiporus dissimulans* Riches, 1893 (Nemertini, Hoplonemertini). *Astarte* 5, 19–26.
- Berg, G. (1973). On morphology and distribution of some hoplonemertean species along Scandinavian coasts (Nemertini). *Zool. Scr.* 2, 63–70.
- Berg, G. (1985). Studies on *Nipponnemertes* Friedrich (Nemertini, Hoplonemertini). II. Taxonomy of *Nipponnemertes pulcher* (Johnston) and some other species. *Zool. Scr.* 14, 239–246.
- Bergendal, D. (1891). Kurzer Bericht über eine im Sommer d. J. 1890 unternommene zoologische Reise nach Nord-Grönland. *Bihang till Kongl. Sve. Vet. Handl.* 17, 1–20.
- Bernardino, A.F., Smith, C. R., Baco, A., Altamira, I., Sumida, P.Y. (2010). Macrofaunal succession in sediments around kelp and wood falls in the deep NE Pacific and community overlap with other reducing habitats. *Deep Sea Res. Part II Top. Stud. Oceanogr.* 57, 708–723.
- Bernt, M., Donath, A., Jühling, F., Externbrink, F., Florentz, C., Fritsch, G., Pütz, J., Middendorf, M., Stadler, P.F. (2013). MITOS: improved de novo metazoan mitochondrial genome annotation. *Mol. Phylogenet. Evol.* 69, 313–319.
- Bienhold, C., Pop Ristova, P., Wenzhöfer, F., Dittmar, T., Boetius, A. (2013). How deep-sea wood falls sustain chemosynthetic life. *PLOS ONE* 8, e53590.
- Blainville, H.M. de (1827). "Espèces molles, sans articulations distinctes (Genre Malacobdella, de Bl.) ", in Dictionnaire des sciences naturelles. Tome quarante-septième. Vol. 47, ed. F.G. Levrault (Paris, France: Normant), 270–271.
- Bömig, L. (1898). Beiträge zur Anatomie und Histologie der Nemertinen (*Stichosteninia graecense* (Bömig), *Geonemertes chalicophora* (Graff)). *Z. Wiss. Zool.* 64, 479–564.
- Brinkmann, A. (1914–1915). *Uniporus*, ein neues Genus der Familie Drepanophoridae Verrill. *Ber. Mus. Aar.* 6, 1–29.
- Brunberg, L. (1964). On the nemertean fauna of Danish waters. *Ophelia* 1, 77–111.
- Bürger, O. (1890). Untersuchungen über die Anatomie und Histologie der Nemertinen nebst Beiträgen zur Systematik. *Z. Wiss. Zool.* 50, 1–277.
- Bürger, O. (1895) Die Nemertinen des Golfes von Neapel und der angrenzenden Meeres-Abschnitte. *F. F. Golf Neapel* 22, 1–743.

- Bürger, O. (1904). Nemertini. Das Tierreich. Eine Zusammenstellung und Kennzeichnung der rezenten Tierformen. 20. Lieferung. Platyhelminthes. R. Friedländer und Sohn, Berlin, xviii + 151 pp.
- Calder, D.R., Watling, L. (2021). Report on hydrozoans (Cnidaria), excluding Stylasteridae, from the Emperor Seamounts, western North Pacific Ocean. *Zootaxa* 4950, 201–247.
- Castresana, J. (2000). Selection of conserved blocks from multiple alignments for their use in phylogenetic analysis. *Mol. Biol. Evol.* 17, 540–552.
- Caullery, M., Lavalley, A. (1912). Recherches sur le cycle évolutif des Orthonectides: les phases initiales dans l'infection expérimentale de l'ophiure, *Amphipholis squamata*, par *Rhopalura ophiocomae* Giard. *Bull. Sci. France Belgique* 46, 139–171.
- Chaban, E.M., Chernyshev, A.V. (2008). Type specimens of benthic nemerteans (Nemertea, Enopla) in the Zoological Institute (St. Petersburg). *Zoosyst. Ross.* 17, 53–60.
- Chen, H., Strand, M., Norenburg, J.L., Sun, S., Kajihara, H., Chernyshev, A.V., Maslakova, S.A., Sundberg, P. (2010). Statistical parsimony networks and species assemblages in cephalotrichid nemerteans (Nemertea). *PLOS ONE* 5, e12885.
- Cherneva, I.A., Chernyshev, A.V., Ekimova, I.A., Polyakova, N.E., Schepetov, D.M., Turanov, N.E., Neretina, T.V., Chaban, E.M., Malakhov, V.V. (2019). Species identity and genetic structure of nemerteans of the “*Lineus ruber–viridis*” complex (Müller, 1774) from Arctic waters. *Pol. Biol.* 42, 497–506.
- Chernyshev, A.V. (1991a) *Sacconemertopsis belogurovi* sp. nov. (Hoploneimertini, Tetrastemmatidae)—novaya solonovatovodnaya nemertina iz Yuzhnogo Primorya. *Biol. Nauki* 5, 45–49.
- Chernyshev, A.V. (1991b) *Tortus tokmakovae* sp. n. (Hoploneimertini; Emplectonematidae) from the Sea of Japan. *Zool. Zhur.* 70, 129–132.
- Chernyshev, A.V. (1992). On the names of some nemertines. *Zool. Zhur.* 71, 134–136.
- Chernyshev, A.V. (1993). A review of the genera of nemerteans allied to *Oerstedia* (Monostilifera, Tetrastemmatidae) with description of four new species. *Zool. Zhur.* 72, 11–20.
- Chernyshev, A.V. (1998a). On the genus *Gurjanovella* Uschakov, 1926 (Nemertea: Enopla). *Zoosystem. Ross.* 7, 9–11.
- Chernyshev, A.V. (1998b). Nemerteans of the genus *Tetrastemma* (Enopla, Monostilifera) from the Far East seas of Russia. *Zool. Zhur.* 77, 995–1002.
- Chernyshev, A.V. (1999). Nemerteans of the genus *Antarctonemertes* (Enopla, Monostilifera). *Zool. Zhur.* 78, 939–948.
- Chernyshev, A.V. (2005). System of families of enoplous nemerteans of the order Eumonostilifera (Nemertea: Enopla). *Russ. J. Mar. Biol.* 31, 27–33.
- Chernyshev, A.V. (2008). Larval development of nemerteans of the genus *Quasitetrastemma* (Nemertea: Monostilifera). *Russ. J. Mar. Biol.* 34, 258–262.

- Chernyshev, A.V. (2014). "Nemertean biodiversity in the Sea of Japan and adjacent areas", in Marine biodiversity and ecosystem dynamics of the Northwest Pacific Ocean, eds. S. Sun, A.V. Adrianov, K.A. Lutaenko, X.X. Sun (Beijing, China: Science Press), 119–135.
- Chernyshev, A.V. (2016). "Nemerteans of the coastal waters of Vietnam", in Biodiversity of the western part of the South China Sea, eds. A.V. Adrianov, K.A. Lutaenko (Vladivostok, Russia: Dalnauka), 279–314.
- Chernyshev, A.V. (2020). Nemerteans from the Far Eastern Seas of Russia. *Russ. J. Mar. Biol.* 46, 141–153.
- Chernyshev, A.V. (2021). An updated classification of the phylum Nemertea. *Invert. Zool.* 18, 188–196.
- Chernyshev, A.V., Maslakova, S.A. (2011). "Phylum Nemertea", in Illustrated keys to free-living invertebrates of Eurasian Arctic Seas and adjacent deep waters, Vol. 2., ed. B.I. Shirenko (Fairbanks, USA: Alaska Sea Grant, University of Alaska Fairbanks), 3–34.
- Chernyshev, A.V., Polyakova, N.E. (2018a). Nemerteans of the Vema-TRANSIT expedition: first data on diversity with description of two new genera and species. *Deep-Sea Res. II: Top. Stud. Oceanogr.* 148, 64–73.
- Chernyshev, A.V., Polyakova, N.E. (2018b). Nemerteans from deep-sea expedition SokhoBio with description of *Uniporus alisae* sp. nov. (Hoploneurata: Reptantia s.l.) from the Sea of Okhotsk. *Deep Sea Res. Part II Top. Stud. Oceanogr.* 154, 121–139.
- Chernyshev, A.V., Polyakova, N.E. (2019). Nemerteans from the deep-sea expedition KuramBio II with descriptions of three new hoplonemerteans from the Kuril-Kamchatka Trench. *Prog. Oceanogr.* 178, 102–148.
- Chernyshev, A.V., Polyakova, N.E. (2022). Nemerteans collected in the Bering Sea during the research cruises aboard the R/V Akademik MA Lavrentyev in 2016, 2018, and 2021 with an analysis of deep-sea heteronemertean and hoplonemertean species. *Deep Sea Res. Part II Top. Stud. Oceanogr.* 199, 105081.
- Chernyshev, A.V., Polyakova, N.E., Turanov, S.V., Kajihara, H. (2018). Taxonomy and phylogeny of *Lineus torquatus* and allies (Nemertea, Lineidae) with descriptions of a new genus and a new cryptic species. *Syst. Biodiv.* 16, 55–68.
- Chernyshev, A.V., Polyakova, N.E., Vignesh, M.S., Jain, R.P., Sanjeevi, P., Norenburg, J.L., Rajesh, R.P. (2020). A histology-free description of a new species of the genus *Tetrastemma* (Nemertea: Hoplonemertea: Monostilifera) from Hawaii and India. *Zootaxa* 4808, 379–383.
- Chernyshev, A.V., Polyakova, N.E., Norenburg, J.L., Kajihara, H. (2021). A molecular phylogeny of *Tetrastemma* and its allies (Nemertea, Monostilifera). *Zool. Scr.* 50, 824–836.

- Clement, M., Posada, D., Crandall, K.A. (2000). TCS: a computer program to estimate gene genealogies. *Mol. Ecol.* 9, 1657–1659.
- Coe, W.R. (1901). Papers from the Harriman Alaska Expedition. XX. The nemerteans. *Proc. Wash. Acad. Sci.* 3, 1–110.
- Coe, W.R. (1902). The nemertean parasites of crabs. *Am. Nat.* 36, 431–450.
- Coe, W.R. (1904). Nemerteans of the Pacific coast of North America. Part II. *Harriman Alaska Ser. 11* 111–220.
- Coe, W.R. (1905). Nemerteans of the west and northwest coasts of America. *Bull. Mus. Comp. Zool.* 47, 1–318.
- Coe, W.R. (1938). A new genus and species of Hoplonemertea having differential bipolar sexuality. *Zool. Anz.* 124, 220–224.
- Coe, W.R. (1939). Sexual phases in terrestrial nemerteans. *Biol. Bull.* 76, 416–427.
- Coe, W.R. (1940). Revision of the nemertean fauna of the Pacific coasts of North, Central, and northern South America. *Allan Hancock Pac. Exped.* 2, 247–322.
- Coe, W.R. (1943). Biology of the nemerteans of the Atlantic coast of North America. *Trans. Conn. Acad. Arts Sci.* 35, 129–328.
- Coe, W.R. (1945). *Malacobdella minuta*, a new commensal nemertean. *J. Wash. Acad. Sci.* 35, 65–67.
- Cognetti, G., Maltagliati, F. (2000). Biodiversity and adaptive mechanisms in brackish water fauna. *Mar. Poll. Bull.* 40, 7–14.
- Colgan, D.J., McLauchlan, A., Wilson, G.D.F., Livingston, S.P., Edgecombe, G.D., Macaranas, J., Cassis, G., Gray, M.R. (1998). Histone H3 and U2 snRNA DNA sequences and arthropod molecular evolution. *Aust. J. Zool.* 46, 419–437.
- Collins, M.A., Bailey, D.M., Ruxton, G.D., Priede, I.G. (2005). Trends in body size across an environmental gradient: a differential response in scavenging and non-scavenging demersal deep-sea fish. *Proc. R. Soc. B: Biol. Sci.* 272, 2051–2057.
- Corrêa, D.D. (1954). Nemertinos do litoral Brasileiro. *Bol. Facul. Uni. São Paulo* 19, 1–90.
- Crandall, F.B. (1993). Major characters and enoplan systematics. *Hydrobiologia* 266, 115–140.
- Dakin, W.J. (1915). Fauna of West Australia—III. A new nemertean, *Geonemertes dendyi*, sp. n., being the first recorded land nemertean from Western Australia. *Proc. Gen. Meet. Sci. Bus. Zool. Soc. London* 1915, 567–570.
- Dakin, W.J., Fordham, M.G.C. (1931). A new and peculiar marine nemertean from the Australian coast. *Nature* 128, 796.
- Dendy, A. (1892). On an Australian land nemertine (*Geonemertes australiensis*, n. sp.). *Proc. R. Soc. Vic.* 4, 85–122.
- Dendy, A. (1895). Notes on a New Zealand land nemertine. *Trans. Proc. N.Z.* 27, 191–194.
- Diesing, C.M. (1850). *Systema Helminthum* Vol I. W Braumuller, Vindobonae.

- Diesing, K.M. (1863). Nachträge zur Revision der Turbellarien. *Sitzung Kaiser Akade. Wissen. Math Class* 46, 173–188.
- Dunn, P. H., Young, C.M. (2014). Larval settlement of the nemertean egg predator *Carcinonemertes errans* on the Dungeness crab, *Metacarcinus magister*. *Invert. Biol.* 133, 201–212.
- Durham, M.J. (1947). Occurrence of the land nemertine, *Geonemertes dendyi* Dakin in Wirral, Cheshire. *N.W. Naturalist* 22, 182–183.
- Ehrenberg, C.G. (1828–1831). Phytozoa Turbellaria Africana et Asiatica in Phytozoorum Tabula IV. et V. delineata. In: Hemprich, F.G. & Ehrenberg, C.G. (Eds.), *Symbolae physicae, seu icones et descriptiones corporum naturalium novorum aut minus cognitorum quae ex itineribus per Libyam, Aegyptium, Nubiam, Dongalam, Syriam, Arabiam et Habessiniam, pars Zoologica II, animalia evertebrata exclusis insectis*. Officina Academica: Berlin, pp. 53–67 (without pagination), pls. IV–V (plates published in 1828; text in 1831).
- Envall, M., Sundberg, P. (1998). Phylogenetic relationships and genetic distances between some monostiliferous interstitial nemerteans (*Ototyphlonemertes*, Hoplonemertea, Nemertea) indicated from the 16S rRNA gene. *Zool. J. Linn. Soc.* 123, 105–115.
- Fernández-Álvarez, F.Á., Machordom, A. (2013). DNA barcoding reveals a cryptic nemertean invasion in Atlantic and Mediterranean waters. *Helgol. Mar. Res.* 67, 599–605.
- Fernández-Álvarez, F.Á., Sánchez, P., Villanueva, R. (2021). Morphological and molecular assessments of bobtail squids (cephalopoda: Sepiolidae) reveal a hidden history of biodiversity. *Front. Mar. Sci.* 7, 632261.
- 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. *Mol. Mar. Biol. Biotechnol.* 3, 294–299.
- Friedrich, H. (1933). Morphologische Studien an Nemertinen der Kieler Bucht, I und II. *Z. Wiss. Zool.* 144, 496–509.
- Friedrich, H. (1935). Studien zur Morphologie, Systematik und Ökologie der Nemertinen der Kieler Bucht. *Arc. Nat.* 4, 293–375.
- Friedrich, H. (1955). Beiträge zu einer Synopsis der Gattungen der Nemertini monostilifera nebst Bestimmungsschlüssel. *Z. Wiss. Zool.* 158, 133–192.
- Friedrich, H. (1957). Beiträge zur Kenntnis der Arktischen Hoplonemertinen. *Vidensk. Meddel. Natuirist. Foren. Kjobenhavn.* 119, 129–154.
- Friedrich, H. (1968). *Sagaminemertes*, eine bemerkenswerte neue Gattung der Hoplonemertinen und ihre systematische Stellung. *Zool. Anz.* 180, 33–36.
- Fujiwara, Y., Tsuchida, S., Kawato, M., Masuda, K., Sakaguchi, S.O., Sado, T., Miya, M., Yoshida, T. (2022). Detection of the largest deep-sea-endemic teleost fish at depths of over 2,000 m through a combination of eDNA metabarcoding and baited camera observations. *Front. Mar. Sci.* 9, 945758

- Gabaev, D.D. (2007). Ecology of reproduction of the red king crab. *Russ. J. Ecol.* 38, 112–118.
- García-Jiménez, R., Horreo, J.L., Valdecasas, A.G. (2017). Minimal barcode distance between two water mite species from Madeira Island: a cautionary tale. *Exp. Appl. Acarol.* 72, 133–143.
- Gering, G. (1912). Neue Nemertinen der schwedischen Westküste. *Zool. Anz.* 39, 520–523.
- Gerlach, J. (1998). The behaviour and captive maintenance of the terrestrial nemertine (*Geonemertes pelaensis*). *J. Zool.* 246, 233–237.
- Gibson, R. (1968). Studies on the biology of the entocommensal rhynchocoelan *Malacobdella grossa*. *J. Mar. Biol. Assoc. U. K.* 48, 637–656.
- Gibson, R. (1972). Nemerteans. (London, UK: Hutchinson and Co Ltd)
- Gibson, R. (1985) The need for a standard approach to taxonomic descriptions of Nemertea. *Am. Zool.* 25, 5–14.
- Gibson, R. (1989) Redescription and taxonomic reappraisal of *Amphiporus elongatus* Stephenson (Nemertea, Enopla) from the Firth of Clyde. *Zool. Scr.* 18, 357–364.
- Gibson, R. (1990a). "The macrobenthic nemertean fauna of Hong Kong". in Proceedings of the Second International Marine Biological Workshop: The Marine Flora and Fauna of Hong Kong and Southern China. Vol. 1, ed. B.S., Morton (Hong Kong, China: Hong Kong University Press), 33–212.
- Gibson, R. (1990b). "The macrobenthic nemertean fauna of the Albany region, Western Australia". in roceedings of the Third International Marine Biological Workshop: The Marine Flora and Fauna of Albany, Western Australia, Vol. 1, eds. F.E. Wells, D.I. Walker, H. Kirkman, R. Lethbridge (Perth, Australia: Western Australian Museum), 89–94.
- Gibson, R. (1994). Nemerteans. (Shrewsbury, UK: Field Studies Council), 224 p.
- Gibson, R. (1995). Nemertean genera and species of the world: an annotated checklist of original names and description citations, synonyms, current taxonomic status, habitats and recorded zoogeographic distribution. *J. Nat. Hist.* 29, 271–562.
- Gibson, R., Crandall, F.B. (1989). The genus *Amphiporus* Ehrenberg (Nemertea, Enopla, Monostiliferoidea). *Zool. Scr.* 18, 453–470.
- Gibson, R., Crandall, F.B. (1991). Case 2707. *Amphiporus* Ehrenberg, 1831 (Nemertea): proposed designation of *Planaria lactiflorea* Johnston, 1828 as the type species. *Bull. Zool. Nomencl.* 48, 22–24.
- Gibson, R., Moore, J. (1998). Further observations on the genus *Geonemertes* with a description of a new species from the Philippine Islands. *Hydrobiologia* 365, 157–171.
- Gibson, R., Strand, M. (2002). A new genus and species of monostiliferous hoplonemertean (Enopla: Hoplonemertea: Monostilifera) from New Zealand. *Zootaxa* 50, 1–20.
- Girard, C., (1852). Descriptions of two new genera and two species of Nemertes. *Proc. Boston Soc. Nat. Hist.* 4, 185–186.

- Girard, C. (1893). Recherches sur les Planariés et les Némertiens de l'Amérique du Nord. *Ann. Sci. Nat., Zool. Sér.* 7 15, 145–310.
- Giribet, G., Carranza, S., Baguña, J., Riutort, M., Ribera, C. (1996). First molecular evidence for the existence of a Tardigrada + Arthropoda clade. *Mol. Biol. Evol.* 13, 76–84.
- Gonzalez-Cueto, J., Castro, L.R., Quiroga, S. (2017). *Nipponnemertes incaïna* sp. n. Adoption of the new taxonomic proposal for nemerteans (Nemertea, Cratenemertidae). *ZooKeys* 693, 1–15.
- Graff, L. von (1879). *Geonemertes chalicophora*, eine neue Landnemertine. *Morpho. Jah.* 5, 430–449.
- Griffin, B.B. (1898). Description of some marine nemerteans of Puget Sound and Alaska. *Ann. N. Y. Acad. Sci.* 11, 193–217.
- Guardiola, M., Wangensteen, O.S., Taberlet, P., Coissac, E., Uriz, M.J., Turon, X. (2016). Spatio-temporal monitoring of deep-sea communities using metabarcoding of sediment DNA and RNA. *PeerJ* 4, e2807.
- Gulliver, G., (1879). Turbellaria. *Phil. Trans. R. Soc. London* 168, 557–563.
- Habe, T. (1961). Descriptions of fifteen new species of Japanese shells. *Venus* 21, 416–431.
- Hao, Y., Kajihara, H., Chernyshev, A.V., Okazaki, R.K., Sun, S.C. (2015). DNA taxonomy of *Paranemertes* (Nemertea: Hoplonemertea) with spirally fluted stylets. *Zool. Sci.* 32, 571–578.
- Hasegawa, K. (1997). Sunken wood-associated gastropods collected from Suruga Bay, Pacific side of central Honshu, Japan, with descriptions of 12 new species. *Nat. Sci. Mus. Monogr.* 12, 59–123.
- Hickman, V.V. (1963). The occurrence in Tasmania of the land nemertine, *Geonemertes australiensis* Dendy, with some account of its distribution, habits, variations and development. *Pap. proc. R. Soc. Tasmania* 97, 63–75.
- Hiebert, T.C., Maslakova, S. (2015). Integrative taxonomy of the *Micrura alaskensis* Coe, 1901 species complex (Nemertea: Heteronemertea), with descriptions of a new genus *Maculaura* gen. nov. and four new species from the NE Pacific. *Zool. Sci.* 32, 615–637.
- Hiebert, L.S., Gavelis, G., von Dassow, G., Maslakova, S.A. (2010). Five invaginations and shedding of the larval epidermis during development of the hoplonemertean *Pantinonemertes californiensis* (Nemertea: Hoplonemertea). *J. Nat. Hist.* 44, 2331–2347.
- Hillis, D.M., and Dixon, M.T. (1991). Ribosomal DNA: molecular evolution and phylogenetic inference. *Q. Rev. Biol.* 66, 411–453.
- Hookabe, N., Kajihara, H. (2020). New record of the monostiliferous hoplonemertean *Gurjanovella littoralis* Uschakov, 1926 from Japan (Nemertea, Hoplonemertea, Monostilifera). *Check List*, 16, 265–269.

- Hookabe, N., Tsuchida, S., Fujiwara, Y., Kajihara, H. (2020). A new species of bathyal nemertean, *Proamphiporus kaimeiae* sp. nov., off Tohoku, Japan, and molecular systematics of the genus (Nemertea: Monostilifera). *Spec. Div.* 25, 183–188.
- Hookabe, N., Xu, C.M., Tsuyuki, A., Jimi, N., Sun, S.C., Kajihara, H. (2021a). A new nemertean with a branched proboscis, *Gorgonorhynchus citrinus* sp. nov. (Nemertea: Pilidiophora), with molecular systematics of the genus. *Invert. Syst.* 35, 350–359.
- Hookabe, N., Kohtsuka, H., Kajihara, H. (2021b). A histology-free description of *Tetrastemma cupido* sp. nov. (Nemertea: Eumonostilifera) from Sagami Bay, Japan. *Mar. Biol. Res.* 17, 467–474.
- Hookabe, N., Motobayashi, H., Jimi, N., Kajihara, H., Ueshima, R. (2022a). First record of the decapod-egg predator *Ovicides paralithodis* (Nemertea, Carcinonemertidae) from the snow crab *Chionoecetes opilio* (Decapoda, Brachyura). *Parasitol. Int.* 89, 102567.
- Hookabe, N., Kajihara, H., Chernyshev, A.V., Jimi, N., Hasegawa N., Kohtsuka H., Okanishi M., Tani, K., Fujiwara Y., Tsuchida, S., Ueshima, R. (2022b). Molecular phylogeny of the genus *Nipponnemertes* (Nemertea: Monostilifera: Cratenemertidae) and descriptions of 10 new species, with notes on small body size in a newly discovered clade. *Front. Mar. Sci.* 9, 906383.
- Hookabe, N., Koeda, K., Fujiwara, Y., Tsuchida, S., Ueshima, R. (2022c). First eumonostiliferous nemertean from the Nishi-Shichito Ridge, *Genrokunemertes obesa* gen. et sp. nov. (Eumonostilifera, Nemertea). *PeerJ* 10, e13857.
- Hookabe, N., Moritaki, T., Jimi, N., Ueshima, R. (2022d). A new oerstediid discovered from wood falls in the Sea of Kumano, Japan: Description of *Rhombonemertes rublinea* gen. et sp. nov. (Nemertea: Eumonostilifera). *Zool. Anz.* 301, 154–162.
- Hubrecht, A.A.W. (1879). The genera of European nemerteans critically revised, with description of several new species. *Note Leyden Mus.* 1, 193–232.
- Hubrecht, A.A.W. (1887). Report on the Nemertea collected by H.M.S. Challenger during the years 1873–76. *Rep. Sci. Res. Chall.* 19, 1–150.
- Humes, A.G. (1942). The morphology, taxonomy, and bionomics of the nemertean genus *Carcinonemertes*. *Biol. Monogr.* 18, 1–105.
- International Commission on Zoological Nomenclature (1992). Opinion 1675. *Amphiporus* Ehrenberg, 1831 (Nemertea): *Planaria lactiflorea* Johnston, 1828 designated as the type species. *Bull. Zool. Nomencl.* 49, 157.
- International Commission on Zoological Nomenclature. (1999). International Code of Zoological Nomenclature, 4th Edition. London: International Trust for Zoological Nomenclature.
- Iwata, F. (1954). The fauna of Akkeshi Bay. XX. Nemertini in Hokkaido (revised report). *J. Fac. Sci. Hokkaido Univ. 6 Zool.* 12, 1–39.

- Iwata, F. (1970). On the brackish water nemerteans from Japan, provided with special circulatory and nephridial organs useful for osmoregulation. *Zool. Anz.* 184, 133–154.
- Iwata, F. (2001). *Nipponnemertes fernaldi*, a new species of swimming monostiliferous hoplonemertean from the San Juan Archipelago, Washington, USA. *Proc. Biol. Soc. Wash.* 114, 833–857.
- Jensen, O.S. (1878). Turbellarier ved Norges Vestkust. J.W. Eides, Bergen, 97 pp., pls I–VIII.
- Johnston, G. (1828). Contributions to the British fauna. *Zool. J.* 3, 486–491.
- Johnston, G. (1837). Miscellanea zoologica. II. A description of some planarian worms. *Mag. Zool. Bot.* 1, 529–538.
- Johnson, A.G. (1976). Electrophoretic evidence of hybrid snow crab, *Chionoecetes bairdi* × *opilio*. *Fish. Bull.* 74, 693–694.
- Jones, H.D., Sterrer, W. (2005). Terrestrial planarians (Platyhelminthes, with three new species) and nemertines of Bermuda. *Zootaxa* 1001, 31–58.
- Judge, J., Barry, J.P. (2016). Macroinvertebrate community assembly on deep-sea wood falls in Monterey Bay is strongly influenced by wood type. *Ecology* 97, 3033–3043.
- Kajihara, H. (2007a). A taxonomic catalogue of Japanese nemerteans (phylum Nemertea). *Zoo. Sci.* 24, 287–326.
- Kajihara, H. (2007b). Two species of *Nemertopsis* (Nemertea: Hoplonemertea: Monostilifera) living in association with *Capitulum mitella* (Crustacea: Cirripedia: Thoracica: Lepadomorpha). *Zootaxa* 1446, 43–58.
- Kajihara, H. (2015). A histology-free description of the branched-proboscis ribbonworm *Gorgonorhynchus albocinctus* sp. nov. (Nemertea: Heteronemertea). *Publ. Seto Mar. Biol. Lab.* 43, 92–102.
- Kajihara, H. (2021a). "Phylum Nemertea", in *Invertebrate Zoology. A Tree of Life Approach*, eds. B. Schierwater, R. DeSalle (Boca Raton: CRC Press), 357–368.
- Kajihara, H. (2021b). Higher classification of the Monostilifera (Nemertea: Hoplonemertea). *Zootaxa* 4920, 151–199.
- Kajihara, H., Kuris, A.M. (2013). *Ovicides paralithodis* (Nemertea, Carcinonemertidae), a new species of symbiotic egg predator of the red king crab *Paralithodes camtschaticus* (Tilesius, 1815) (Decapoda, Anomura). *ZooKeys* 258, 1–15.
- Kajihara, H., Gibson, R., Mawatari, S.F. (2003). *Potamostoma shizunaiense* gen. et sp. nov. (Nemertea: Hoplonemertea: Monostilifera): a new brackish-water nemertean from Japan. *Zool. Sci.* 20, 491–500.
- Kajihara, H., Olympia, M., Kobayashi, N., Katoh, T., Chen, H.X., Strand, M., Sundberg, P. (2011). Systematics and phylogeny of the hoplonemertean genus *Diplomma* (Nemertea) based on molecular and morphological evidence. *Zool. J. Linn. Soc.* 161, 695–722.

- Kajihara, H., Nishi, E., Kawabata, M., Kohtsuka, H., Uyeno, D. (2015). Records of the poorly known ribbon worm *Nipponnemertes ogumai* (Nemertea: Monostilifera) and its phylogenetic position. *Mar. Biodiv.* 45, 175–182.
- Kajihara, H., Tamura, K., Tomioka, S. (2018). Histology-free descriptions for seven species of interstitial ribbon worms in the genus *Ototyphlonemertes* (Nemertea: Monostilifera) from Vietnam. *Spec. Div.* 23, 13–37.
- Kajihara, H., Abukawa, S., Chernyshev, A.V. (2022). Exploring the basal topology of the heteronemertean tree of life: establishment of a new family, along with turbotaxonomy of Valenciniidae (Nemertea: Pilidiophora: Heteronemertea). *Zool. J. Linn. Soc.* 196, 503–548.
- Kang, X.X., Fernández-Álvarez, F.Á., Alfaya, J.E., Machordom, A., Strand, M., Sundberg, P., Sun, S.C. (2015). Species diversity of *Ramphogordius sanguineus*/*Lineus ruber*-like nemerteans (Nemertea: Heteronemertea) and geographic distribution of *R. sanguineus*. *Zool. Sci.* 32, 579–589.
- Katoh, K., Standley, D.M. (2013). MAFFT multiple sequence alignment software version 7: improvements in performance and usability. *Mol. Biol. Evol.* 30, 772–780.
- Kawakatsu, M. (1991). "Nemertinea", in Pictorial Keys to Soil Animals of Japan, ed. J. Aoki (Tokyo, Japan: Tōkaidai-gaku Shuppan Kai), 2 [In Japanese].
- Kawakatsu, M. (1999). "Nemertinea", in Pictorial Keys to Soil Animals of Japan. ed. J. Aoki (Tokyo, Japan: Tōkaidai-gaku Shuppan Kai), 11 [In Japanese].
- Keferstein, W. (1862). Untersuchungen über niedere Seethiere. *Z. Wiss. Zool.* 12, 1–147.
- Kem, W.R., Abbott, B.C., Coates, R.M. (1971). Isolation and structure of a hoplonemertine toxin. *Toxicon* 9, 15–22.
- Kern, E.M., Kim, T., Park, J.K. (2020). The mitochondrial genome in nematode phylogenetics. *Front. Ecol. Evol.* 8, 250.
- Kirsteuer, E. (1965). Über das Vorkommen von Nemertinen in einem tropischen Korallenriff. 4. Hoplonemertini monostilifera. *Zool. Jahrb. Abt. Syst. Geog. Biol. Tiere* 92, 289–326.
- Klunder, L., De Stigter, H., Lavaleye, M.S., Van Bleijswijk, J.D., Van der Veer, H.W., Reichart, G.J., Duineveld, G.C. (2020). A molecular approach to explore the background benthic fauna around a hydrothermal vent and their larvae: implications for future mining of deep-sea SMS deposits. *Front. Mar. Sci.* 7, 1–12.
- Kon, T., Adachi, T., Suzuki, Y. (2003). Distribution of snow crab, *Chionoecetes* spp., larvae off Wakasa Bay in the Sea of Japan. *Fish. Sci.* 69, 1109–1115.
- Korotkevitch, V.S. (1961). A new nemertean species and its position in the system. *Zool. Zhur.* 40, 1416–1420.

- Korotkevitsch, V.S. (1971). "On the ecology and systematics of nemerteans from Posyet Bay, Sea of Japan", in *Explorations of the Fauna of the Seas*, vol. 8, Flora and Fauna of Posyet Bay, Sea of Japan (Leningrad, Russia: Nauka), 29–41.
- Korotkevitsch, V.S. (1977). "Nemertini pribrezhnuikh rod kurilskikh ostrovov", in *Fauna Pribrezhnuikh zon Kurilskikh Ostrovov*, ed. V.V. Gulbin (Moscow, Russia: Izadatelstvo Nauka), 49–124.
- Korotkevitsch, V.S. (1983). O stiletakh nemertin i novom vide Cratenemertes (Hoplonemertini, Amphiporidae) iz Antarktiki. *Antark. Dok. Kom.* 22, 137–143. [in Russian]
- Kozlov, A.M., Darriba, D., Flouri, T., Morel, B., Stamatakis, A. (2019). RAxML-NG: a fast, scalable and user-friendly tool for maximum likelihood phylogenetic inference. *Bioinformatics* 35, 4453–4455.
- Krämer, D., von Döhren, J. (2015). *Arenogigas armoricus*, a new genus and species of a monostiliferous hoplonemertean (Nemertea) from the north-west coast of France. *Zool. Sci.* 32, 605–614.
- Krämer, D., Schmidt, C., Podsiadlowski, L., Beckers, P., Horn, L., von Döhren, J. (2017). Unravelling the *Lineus ruber/viridis* species complex (Nemertea, Heteronemertea). *Zool. Scr.* 46, 111–126.
- Kruse, I., Buhs, F. (2000). Preying at the edge of the sea: the nemertine *Tetrastemma melanocephalum* and its amphipod prey on high intertidal sandflats. *Hydrobiologia* 426, 43–55.
- Kulikova, V.I. (1987). New species of the genus *Oerstedrella* (Nemertini, Hoplonemertini) from the Vostok Bay, Sea of Japan. *Izv. Akad. Nauk SSSR* 6, 3828–3836.
- Kumar, S., Stecher, G., Tamura, K. (2016). MEGA7: molecular evolutionary genetics analysis version 7.0 for bigger datasets. *Mol. Biol. Evol.* 33, 1870–1874.
- Kuris, A.M., Blau, S.F., Paul, A.J., Shields, J.D., Wickham, D.E. (1991). Infestation by brood symbionts and their impact on egg mortality of the red king crab, *Paralithodes camtschatica*, in Alaska: geographic and temporal variation. *Can. J. Fish. Aquat. Sci.* 48, 559–568.
- Kuroda, T. (1949). "Mollusca (Gastropoda, Scaphopoda and Pelecypoda)", in Revised Edition Illustrated Encyclopaedia of the Fauna of Japan, ed. S. Uchida (Tokyo, Japan: The Hokuryukan Co. Ltd.) [In Japanese].
- Kvist, S., Laumer, C.E., Junoy, J., Giribet, G. (2014). New insights into the phylogeny, systematics and DNA barcoding of Nemertea. *Invert. Syst.* 28, 287–308.
- Kvist, S., Chernyshev, A.V., Giribet, G. (2015). Phylogeny of Nemertea with special interest in the placement of diversity from Far East Russia and northeast Asia. *Hydrobiologia* 760, 105–119.
- Lanfear, R., Frandsen, P.B., Wright, A.M., Senfeld, T., Calcott, B. (2016). PartitionFinder 2: new methods for selecting partitioned models of evolution for molecular and morphological phylogenetic analyses. *Mol. Biol. Evol.* 34, 772–773.

- Leasi, F., Norenburg, J.L. (2014). The necessity of DNA taxonomy to reveal cryptic diversity and spatial distribution of meiofauna, with a focus on Nemertea. *PLOS ONE* 9, e104385.
- Leasi, F., Andrade, S.D.S, Norenburg, J. (2016). At least some meiofaunal species are not everywhere. Indication of geographic, ecological and geological barriers affecting the dispersion of species of *Otocyphlonemertes* (Nemertea, Hoplonemertea). *Mol. Ecol.* 25, 1381–1397.
- Lee, H., Chen, W.J., Puillandre, N., Aznar-Cormano, L., Tsai, M.H., Samadi, S. (2019). Incorporation of deep-sea and small-sized species provides new insights into gastropods phylogeny. *Mol. Phylogenet. Evol.* 135, 136–147.
- Leidy, J., (1851). Helminthological contributions—No. 3. *Proc. Acad. Nat. Sci. Phil.* 5, 239–244.
- Leigh, J.W., Bryant D. (2015). Popart: full-feature software for haplotype network construction. *Methods Ecol. Evol.* 6, 1110–1116.
- Leiva, N.V., Ñacari, L., Baeza, J.A., González, M.T. (2021). First report of an egg-predator nemertean worm in crabs from the south-eastern Pacific coast: *Carcinonemertes camanchaco* sp. nov. *Sci. Rep.* 11, 1–13.
- Levinsen, G.M.R. (1879–1880). Bidrag til Kundskab om Grønlands Turbellariefauna. *Vid. Medd. Foren.* 41–42, 165–204, pl. III.
- Littlewood, D.T.J. (1994). Molecular phylogenetics of cupped oysters based on partial 28S rRNA gene sequences. *Mol. Phylogenet. Evol.* 3, 221–229.
- Magarlamov, T.Y., Dyachuk, V., Chernyshev, A.V. (2020). Does the frontal sensory organ in adults of the hoplonemertean *QuasitetraSTEMMA stimpsoni* originate from the larval apical organ? *Front. Zool.* 17, 1–19.
- Martens, E. von (1861). Die Japanesischen Binnenschnecken im Leidner Museum. *Malakozool. Blätt.* 7, 32–61.
- Maslakova, S.A., Norenburg, J.L. (2008). Revision of the smiling worms, genus *Prosorhochmus* Kefenstein, and description of a new species, *Prosorhochmus belizeanus* sp. nov. (Prosorhochmidae, Hoplonemertea, Nemertea) from Florida and Belize. *J. Nat. Hist.* 42, 1219–1260.
- Maslakova, S.A., von Döhren, J. (2009). Larval development with transitory epidermis in *Paranemertes peregrina* and other hoplonemerteans. *Biol. Bull.* 216, 273–292.
- Maslakova, S.A., Thiel, M., Vásquez, N., Norenburg, J.L. (2005). The smile of *Amphiporus nelsoni* Sanchez, 1973 (Nemertea: Hoplonemertea: Monostilifera: Amphiporidae) leads to a redescription and a change in family. *Proc. Biol. Soc. Wash.* 118, 483–498.
- Mateos, E., Giribet, G. (2008). Exploring the molecular diversity of terrestrial nemerteans (Hoplonemertea, Monostilifera, Acteonemertidae) in a continental landmass. *Zool. Scr.* 37, 235–243.

- McDermott, J.J. (1984). The feeding biology of *Nipponnemertes pulcher* (Johnston) (Hoploneurtea), with some ecological implications. *Ophelia* 23, 1–21.
- McDermott, J.J. (1993). Nemertea inhabiting the *Haploops* (Amphipoda) community of the northern Øresund with special reference to the biology of *Nipponnemertes pulcher* (Hoploneurtea). *Hydrobiologia* 266, 15–28.
- McDermott, J.J. (1997). Observations on feeding in a South African suctorial hoploneurtean, *Nipponnemertes* sp. (Family Cratenemertidae). *Hydrobiologia* 365, 251–256.
- McDermott, J.J., Roe, P. (1985). Food, feeding behavior and feeding ecology of nemerteans. *Am. Zool.* 25, 113–125.
- Montgomery, T.H. (1894). Kritische Uebersicht der Süßwasser-Nemertinen. Inaugural Dissertation, University of Berlin, Berlin.
- Montgomery, T.H. (1897). Descriptions of new metanemerteans, with notes on other species. *Zool. Jb. Abt. Syst.* 10, 1–14.
- Moore, J., Gibson, R. (1972). On a new genus of freshwater hoploneurtean from Campbell Island. *Freshw. Biol.* 2, 187–202.
- Moore, J., Gibson, R. (1973). A new genus of freshwater hoploneurtean from New Zealand. *Zool. J. Linn. Soc.* 52, 141–157.
- Moore, J., Gibson, R. (1981). The *Geonemertes* problem (Nemertea). *J. Zool.* 194, 175–201.
- Moore, J., Gibson, R. (1985). The evolution and comparative physiology of terrestrial and freshwater nemerteans. *Biol. Rev.* 60, 257–312.
- Moore, J., Gibson, R. (1988). Further studies on the evolution of land and freshwater nemerteans: generic relationships among the paramonostiliferous taxa. *J. Zool.* 216, 1–20.
- Moore, J., Moore, N.M. (1972). Land nemertines of Madeira and the Azores. *Bol. Mus. Mun. Fun.* 26, 31–44.
- Moore, J., Moore, R.N.W., Gibson, R., (1995). Land nemertines of Rodrigues. *J. Zool., Lond.* 237, 241–257.
- Moore, J., Gibson, R., Jones, H.D. (2001). Terrestrial nemerteans thirty years on. *Hydrobiologia* 456, 1–6.
- Morffe, J., García, N., Breugelmans, K. (2020). First record of the terrestrial nemertean *Geonemertes pelaensis* Semper, 1863 (Hoploneurtea: Prosorhochmidae) for Cuba. *Bioinvasions Rec.* 9, 399–407.
- Morgan, N.B., Baco, A.R. (2021). Recent fishing footprint of the high-seas bottom trawl fisheries on the Northwestern Hawaiian Ridge and Emperor Seamount Chain: A finer-scale approach to a large-scale issue. *Ecol. Indic.* 121, 107051.
- Murphy, J.T. (2020). Climate change, interspecific competition, and poleward vs. depth distribution shifts: spatial analyses of the eastern Bering Sea snow and tanner crab (*Chionoecetes opilio* and *C. bairdi*). *Fish. Res.* 223, 105417.

- Norenburg, J. (1986). Redescription of a brooding nemertine, *Cyanophthalma obscura* (Schultze) gen. et comb. n., with observations on its biology and discussion of the species of *Prostomatella* and related taxa. *Zool. Scr.* 15, 275–293.
- Nunomura, N. (1984). Studies on the terrestrial isopod crustaceans in Japan, II. Taxonomy of the family Scyphacidae. *Bull. Toyama Sci. Mus.* 6, 1–43.
- Oki, I., Tamura, S., Ogren, R.E., Kitagawa, K., Kawakatsu, M. (1987). The karyotype and a new locality for the land nemertine *Geonemertes pelaensis* Semper, 1863. *Bull. Fuji Women's Coll. Ser. 2* 25, 66–77.
- Okochi, I., Sato, H., Ohbayashi, T. (2004). The cause of mollusk decline on the Ogasawara Islands. *Biodiv. Conserv.* 13, 1465–1475.
- Örsted, A.S. (1843). Forsøg til en ny Classification af Planarierne (Planariae Dugès) grundet paa mikroskopisk-anatomiske Undersogelser. *Natur. Tidsk.* 4, 519–581.
- Osawa, M., Ota, Y. (2020). New records of species of Gebiidea and Anomura (Crustacea: Decapoda) from the Sea of Japan. *Spec. Div.* 25, 295–307.
- Page, L.R., Parries, S.C. (2000). Comparative study of the apical ganglion in planktotrophic caenogastropod larvae: ultrastructure and immunoreactivity to serotonin. *J. Comp. Neurol.* 418, 383–401.
- Palumbi, S.R., Martin, A., Romano, S., McMillan, W.O., Stice, L., Grabowski, G. (1991). The simple fool's guide to PCR, v. 2.0. Honolulu: Department Zoology, Kewalo Marine Laboratory, University of Hawaii.
- Pantin, C.F.A. (1961). *Acteonemertes bathamae*, gen. et sp. nov. An upper littoral nemertine from Portobello, New Zealand. *Proc. Linn. Soc. Lond.* 172, 153–156.
- Puillandre, N., Lambert, A., Brouillet, S., Achaz, G. (2012). ABGD, automatic barcode gap discovery for primary species delimitation. *Mol. Ecol.* 21, 1864–1877.
- Punnett, R.C. (1903). On the nemerteans of Norway. *Ber. Mus. Aarbog* 2, 3–35.
- Quatrefages, A. de. (1846). Étude sur les types inférieurs de l'embranchement des annelés. *Ann. Sci. Nat, Zoologie, Sér.* 3 6, 173–303.
- Quoy, J.R.C., Gaimard, J.P. (1833). Voyage de découvertes de l'Astrolabe exécuté par ordre du Roi, pendant les années 1826–1827–1828–1829, sous le commandement de M. J. Dumont d'Urville, Zoologie. Tome Quatrième. J. Tastu, Paris, 390 pp.
- Rambaut, A., Drummond, A.J., Xie, D., Baele, G., Suchard, M.A. (2018). Posterior summarisation in Bayesian phylogenetics using Tracer ver. 1.7. *Syst. Biol.* 67, 901–904.
- Reisinger, E. (1926). "Nemertini. Schnurwürmer.", in *Biologie der Tiere Deutschlands*. Gebruder Bornsraeger, ed. Schulze, P. (Berlin, Germany: Gebruder Bornsraeger), 7.1–7.24.
- Richter, S., Loesel, R., Purschke, G., Schmidt-Rhaesa, A., Scholtz, G., Stach, T., Vogt, L., Wanninger, A., Brenneis, G., Döring, C., Faller, S., Fritsch, M., Grobe, P., Heuer,

- C.M., Kaul, S., Møller O.S., Müller C.H.G., Rieger, V., Rothe, B.H., Stegner, M.E.J., Harzsch, S. (2010). Invertebrate neurophylogeny: suggested terms and definitions for a neuroanatomical glossary. *Front. Zool.* 7, 1–49.
- Riser, N.W. (1989). Speciation and time—relationships of the nemertines to the acoelomate metazoan Bilateria. *Bull. Mar. Sci.* 45, 531–538.
- Riser, N.W. (1998). New Zealand nemertines from kelp holdfasts: Hoplonemertinea 1. *Nipponnemertes sanguinea* sp. n. *N. Z. J. Zool.* 25, 287–294.
- Roe, P. (1979). Aspects of development and occurrence of *Carcinonemertes epialti* (Nemertea) from shore crabs in Monterey Bay, California. *Biol. Bull.* 156, 130–140.
- Rodríguez-Flores, P.C., Macpherson, E., Schnabel, K.E., Ahyong, S.T., Corbari, L., Machordom, A. (2022). Depth as a driver of evolution and diversification of ancient squat lobsters (Decapoda, Galatheaidea, *Phylladorhynchus*). *Mol. Phylogenet. Evol.* 171, 107467.
- Ronquist, F., Teslenko, M., Van der Mark, P., Ayres, D.L., Darling, A., Höhna, S., Larget, B., Liu, L., Suchard, M.A., Huelsenbeck, J.P. (2012). MrBayes 3.2: efficient Bayesian phylogenetic inference and model choice across a large model space. *Syst. Biol.* 61, 539–542.
- Saccone, C., De Giorgi, C., Gissi, C., Pesole, G., Reyes, A. (1999). Evolutionary genomics in Metazoa: the mitochondrial DNA as a model system. *Gene* 238, 195–209.
- Saeedi, H., Bernardino, A.F., Shimabukuro, M., Falchetto, G., Sumida, P.Y. (2019). Macrofaunal community structure and biodiversity patterns based on a wood-fall experiment in the deep South-west Atlantic. *Deep Sea Res. Part I: Oceanogr.* 145, 73–82.
- Sagorny, C., Wesseler, C., Krämer, D., von Döhren, J. (2019). Assessing the diversity and distribution of *Cephalothrix* species (Nemertea: Palaeonemertea) in European waters by comparing different species delimitation methods. *Zoolog. Syst. Evol. Res.* 57, 497–519.
- Schwartz, M.L., Norenburg, J.L. (2001). Can we infer heteronemertean phylogeny from available morphological data? *Hydrobiologia* 456, 165–174.
- Schwendinger, P.J., Giribet, G. (2005). The systematics of the southeast Asian genus *Fangensis* Rambla (Opiliones: Cyphophthalmi: Stylocellidae). *Invert. Syst.* 19, 297–323.
- Semper, C. (1863). Reisebericht. Briefliche Mittheilung an A. Kölliker. *Z. Wiss. Zool.* 13, 558–570.
- Shields, J.D. (2001). *Ovicides julieae* n. gen., n. sp. (Nemertea: Carcinonemertidae) on xanthid crabs from the Great Barrier Reef, Australia. *J. Crustac. Biol.* 21, 304–312.
- Shields, J.D., Segonzac, M. (2007). New nemertean worms (Carcinonemertidae) on bythograeid crabs (Decapoda: Brachyura) from Pacific hydrothermal vent sites. *J. Crustac. Biol.* 27, 681–692.

- Shinobe, S., Uchida, S., Mori, H., Okochi, I., Chiba, S. (2017). Declining soil crustacea in a world heritage site caused by land nemertean. *Sci. Rep.* 7, 1–8.
- Simpson, L.A., Ambrosio, L.J., Baeza, J.A. (2017). A new species of *Carcinonemertes*, *Carcinonemertes conanobrieni* sp. nov. (Nemertea: Carcinonemertidae), an egg predator of the Caribbean spiny lobster, *Panulirus argus*. *PLOS ONE* 12, e0177021.
- Sinniger, F., Pawlowski, J., Harii, S., Gooday, A.J., Yamamoto, H., Chevalloné, P., Cedhagen, T., Carvalho, G., Creer, S. (2016). Worldwide analysis of sedimentary DNA reveals major gaps in taxonomic knowledge of deep-sea benthos. *Front. Mar. Sci.* 3, 1–14.
- Smith, C.T., Grant, W.S., Seeb, L.W. (2005). A rapid, high-throughput technique for detecting tanner crabs *Chionoecetes bairdi* illegally taken in Alaska's snow crab fishery. *Trans. Am. Fish. Soc.* 134, 620–623.
- Stamatakis, A. (2014). RAxML version 8: a tool for phylogenetic analysis and post-analysis of large phylogenies. *Bioinformatics* 30, 1312–1313.
- Stimpson, W. (1857). Prodomus descriptionis animalium evertibratorum, quae in Expeditione ad Oceanum Pacificum Septentrionalem, a Republica Federata missa, Cadwaladaro Ringgold et Johanne Rodgers Ducibus, observavit et descripsit. Pars II. Turbellarieorum Nemertineorum. *Proc. Acad. Nat. Sci.* 9, 159–165.
- Strand, M., Sundberg, P. (2005a). Genus *Tetrastemma* Ehrenberg, 1831 (Phylum Nemertea)—a natural group? Phylogenetic relationships inferred from partial 18S rRNA sequences. *Mol. Phylogenetics Evol.* 37, 144–152.
- Strand, M., Sundberg, P. (2005b). Delimiting species in the hoplonemertean genus *Tetrastemma* (phylum Nemertea): morphology is not concordant with phylogeny as evidenced from mtDNA sequences. *Biol. J. Linn. Soc.* 86, 201–212.
- Strand, M., Sundberg, P. (2011). A DNA-based description of a new nemertean (phylum Nemertea) species. *Mar. Biol. Res.* 7, 63–70.
- Strand, M., Herrera-Bachiller, A., Nygren, A. & Kånneby, T. (2014). A new nemertean species: what are the useful characters for ribbon worm descriptions? *J. Mar. Biol. Assoc. U.K.* 94, 317–330.
- Stricker, S.A. (1985). A new species of *Tetrastemma* (Nemertea, Monostilifera) from San Juan Island, Washington, U.S.A. *Can. J. Zool.* 63, 682–690.
- Stricker, S.A., Reed, C.G. (1981). Larval morphology of the nemertean *Carcinonemertes epialti* (Nemertea: Hoplonemertea). *J. Morphol.* 169, 61–70.
- Stricker, S.A., Smythe, T.L., Miller, L., Norenburg, J.L. (2001). Comparative biology of oogenesis in nemertean worms. *Acta Zool.* 82, 213–230.
- Sumner, F.B., Osburn, R.C., Cole, L.J. (1913). A biological survey of the waters of Woods Hole and vicinity. Section III.—A catalogue of the marine fauna of Woods Hole and vicinity. *Bull. Bur. Fish.* 31, 549–794.

- Sun, S. (2001). A new mangrove-dwelling nemertean from China. *Hydrobiologia* 456, 199–209.
- Sun, W.Y., Shen, C.Y., Sun, S.C. (2016). The complete mitochondrial genome of *Tetrastemma olgarum* (Nemertea: Hoplonemertea). *Mitochondrial DNA A* 27, 1086–1087.
- Sundberg, P. (1989a). Phylogeny and cladistic classification of the paramonostiliferous family Plectonemertidae (phylum Nemertea). *Cladistics* 5, 87–100.
- Sundberg, P. (1989b). Classification of the family Plectonemertidae (Nemertea): a phenetic comparison. *Zool. J. Linn. Soc.* 97, 57–68.
- Sundberg, P., Gibson, R. (1995). The nemerteans (Nemertea) of Rottnest Island, Western Australia. *Zool. Scr.* 24, 101–141.
- Sundberg, P., Saur, M. (1998). Molecular phylogeny of some European heteronemertean (Nemertea) species and the monophyletic status of *Riseriellus*, *Lineus*, and *Micrura*. *Mol. Phylogenet. Evol.* 10, 271–280.
- Sundberg, P., Turbeville, J.M., Lindh, S. (2001). Phylogenetic relationships among higher nemertean (Nemertea) taxa inferred from 18S rDNA sequences. *Mol. Phylogenet. Evol.* 20, 327–334.
- Sundberg P, Gibson R, Strand M. (2007). Swedish nemerteans (phylum Nemertea), with description of a new hoplonemertean genus and species. *J. Nat. Hist.* 41, 2287–2299.
- Sundberg, P., Chernyshev, A. V., Kajihara, H., Kånneby, T., Strand, M. (2009). Character-matrix based descriptions of two new nemertean (Nemertea) species. *Zool. J. Linn. Soc.* 157, 264–294.
- Sundberg, P., Kvist, S., Strand, M. (2016). Evaluating the utility of single-locus DNA barcoding for the identification of ribbon worms (Phylum Nemertea). *PLOS ONE* 11, e0155541.
- Taboada, S., Junoy, J., Andrade, S.C.S., Giribet, G., Cristobo, J., Avila, C. (2013). On the identity of two Antarctic brooding nemerteans: redescription of *Antarctonemertes valida* (Bürger, 1893) and description of a new species in the genus *Antarctonemertes* Friedrich, 1955 (Nemertea, Hoplonemertea). *Pol. Biol.* 36, 1415–1430.
- Taboada, S., Leiva, C., Junoy, J., Alexander, F., Riesgo, A. (2018). A new member of the genus *Antarctonemertes* (Hoplonemertea, Nemertea) from Antarctic waters. *Pol. Biol.* 41, 1463–1473.
- Takakura, U. (1897). On a new species of *Malacobdella* (*M. japonica*). *Ann. Zool. Jpn.* 1, 105–112.
- Takakura, U. (1910). Kisei-himomushi-no-ichi-shinshu [A new species of parasitic nemertean]. *Zool. Mag.* 22, 111–116 [In Japanese].
- Templeton, A.R., Crandall, K.A., Sing, C.F. (1992). A cladistic analysis of phenotypic associations with haplotypes inferred from restriction endonuclease mapping and DNA sequence data. III. Cladogram estimation. *Genetics* 132, 619–633.

- Thiel, M., Kruse, I. (2001). Status of the Nemertea as predators in marine ecosystems. *Hydrobiologia* 456, 21–32.
- Thollessen, M., Norenburg, J.L. (2003). Ribbon worm relationships: a phylogeny of the phylum Nemertea. *Proc. R. Soc. B Biol. Sci.* 270, 407–415.
- Thompson, J.D., Higgins, D.G., Gibson, T.J. (1994). CLUSTAL W: improving the sensitivity of progressive multiple sequence alignment through sequence weighting, position-specific gap penalties and weight matrix choice. *Nucleic Acids Res.* 22, 4673–4680.
- Tilic, E., Sayyari, E., Stiller, J., Mirarab, S., Rouse, G.W. (2020). More is needed—Thousands of loci are required to elucidate the relationships of the ‘flowers of the sea’ (Sabellida, Annelida). *Mol. Phylogenet. Evol.* 151, 106892.
- Turbeville, J.M., Field, K.G., Raff, R.A. (1992). Phylogenetic position of phylum Nemertini, inferred from 18S rRNA sequences: molecular data as a test of morphological character homology. *Mol. Biol. Evol.* 9, 235–249.
- Turner, R.D. (1977). Wood, mollusks, and deep-sea food chains. *Bull. American Malaco. Uni.* 1976, 13–19.
- Uschakov, P. (1926). K faune nemertin Belogo morya. *Issl. Russ. Mor.* 3, 47–71.
- Uschakov, P. (1927). Eine neue Nemertine des japanischen Meeres. *Zool. Anz.* 72, 289–290.
- Uschakov, [‘Uschakow’ in print] P. (1928). Beschreibung einiger neuen Nemertinenarten vom Barents-Meere, Weißen Meere und Nowaja-Semlja. *Zool. J. Abteil. Syst. Ökolog. Geograp.* 54, 407–424.
- Van der Grient, J.M, Rogers, A.D. (2015). Body size versus depth: regional and taxonomical variation in deep-sea meio-and macrofaunal organisms. *Adv. Mar. Biol.* 71, 71–108.
- Verdes, A., Arias, M.B., Junoy, J., Schwartz, M.L., Kajihara, H. (2021). Species delimitation and phylogenetic analyses reveal cryptic diversity within *Cerebratulus marginatus* (Nemertea: Pilidiophora). *Syst. Biodiv.* 19, 895–905.
- Verrill, A.E. (1874a). Brief contributions to zoölogy from the Museum of Yale College. No. XXVI. Results of recent dredging expeditions on the coast of New England. No. 4. *Am. J. Sci. Arts, Ser. 3 7*, 38–46.
- Verrill, A.E. (1874b). Brief contributions to zoölogy from the Museum of Yale College. No. XXIX. Results of recent dredging expeditions on the coast of New England. No. 7. *Am. J. Sci. Arts, Ser. 3 7*, 498–505.
- Verrill, A.E. (1874c). Explorations of Casco Bay by the U. S. Fish Commission, in 1873. *Proc. Am. Assoc. Advance. Sci.* 22, 340–395.
- Verrill, A.E. (1879). Notice of recent additions to the marine Invertebrata, of the northeastern coast of America, with descriptions of new genera and species and critical remarks on others. Part I—Annelida, Gephyraea, Nemertina, Nematoda, Polyzoa, Tunicata, Mollusca, Anthozoa, Echinodermata, Porifera. *Proc. U.S. Nat. Mus.* 2, 165–205.

- Verrill, A.E. (1892). The marine nemerteans of New England and adjacent waters. *Trans. Conn. Acad. Arts Sci.* 8, 382–456.
- Voight, J.R., Marshall, B.A., Judge, J., Halanych, K.M., Li, Y., Bernardino, A.F., Grewe, F., Maddox, J.D. (2019). Life in wood: preliminary phylogeny of deep-sea wood-boring bivalves (Xylophagidae), with descriptions of three new genera and one new species. *J. Molluscan Stud.* 85, 232–243.
- von Dassow, G., Mendes, C., Robbins, K., Andrade, S., Maslakova, S. (2022). Hoplonemertean larvae are planktonic predators that capture and devour active animal prey. *Inv. Biol.* 141, e12363.
- Wang, H., Sun, S. (2006). Studies on the chemoreception of the nemertean, *Procephalothrix simulus* Iwata, 1952. *J. Exp. Mar. Biol. Ecol.* 336, 146–152.
- Wanninger, A. (2009). Shaping the things to come: ontogeny of Lophotrochozoan neuromuscular systems and the tetraneuralia concept. *Biol Bull.* 216, 293–306.
- Waterston, A.R., Quick, H.E. (1938). XXIV.—*Geonemertes dendyi* Dakin, a land nemertean, in Wales. *Proc. R. Soc. Edinb.* 57, 379–384.
- Wheeler, J.F.G. (1934). Nemerteans from the South Atlantic and southern oceans. ‘Discovery’ Rep.
- Whiting, M.F., Carpenter, J.M., Wheeler, Q.D., Wheeler, W.C. (1997). The Strepsiptera problem: phylogeny of the holometabolous insect orders inferred from 18S and 28S ribosomal DNA sequences and morphology. *Syst. Biol.* 46, 1–68.
- Wickham, D.E. (1978). A new species of *Carcinonemertes* (Nemertea, Carcinonemertidae) with notes on the genus from the Pacific coast. *Proc. Biol. Soc. Wash.* 91, 197–202.
- Wickham, D.E., Kuris, A.M. (1988). Diversity among nemertean egg predators of decapod crustaceans. *Hydrobiologia* 15, 23–30.
- Wickham, D.E., Roe, P., Kuris, A.M. (1984). Transfer of nemertean egg predators during host molting and copulation. *Biol. Bull.* 167, 331–338.
- Wolff, T. (1979). Macrofaunal utilization of plant remains in the deep sea. *Sarsia* 64, 117–143.
- Yamaoka, T. (1940). Two nemerteans from the Riukiu Islands. *Ann. Zool. Jpn.* 19, 13–18.
- Yamaoka, T. (1947). "*Amphiporus ogumai* Yamaoka", submitted by S. Okuda, in Revised edition, illustrated encyclopedia of the fauna of Japan (exclusive of insects) (Tokyo: Hokuryukan) 1468, fig. 4138 (1–3) [In Japanese].
- Yamaoka, T. (2005). On fourteen monostiliferous hoplonemerteans from the Izu Peninsula, Middle Japan. *Publ. Seto Mar. Biol. Lab.* 40, 141–158.
- Zhang, J., Kapli, P., Pavlidis, P., Stamatakis, A. (2013). A general species delimitation method with applications to phylogenetic placements. *Bioinformatics* 29, 2869–2876.
- Zaitseva, O.V., Petrov, S.A., Petrov, A.A. (2020). Sensory systems of *Lineus ruber* (Nemertea, Pilidiophora). *Zoomorphology* 139, 447–459.

Zwickl, D.J., Hillis, D.M. (2002). Increased taxon sampling greatly reduces phylogenetic error.
Syst. Biol. 514, 588–598.

Figures

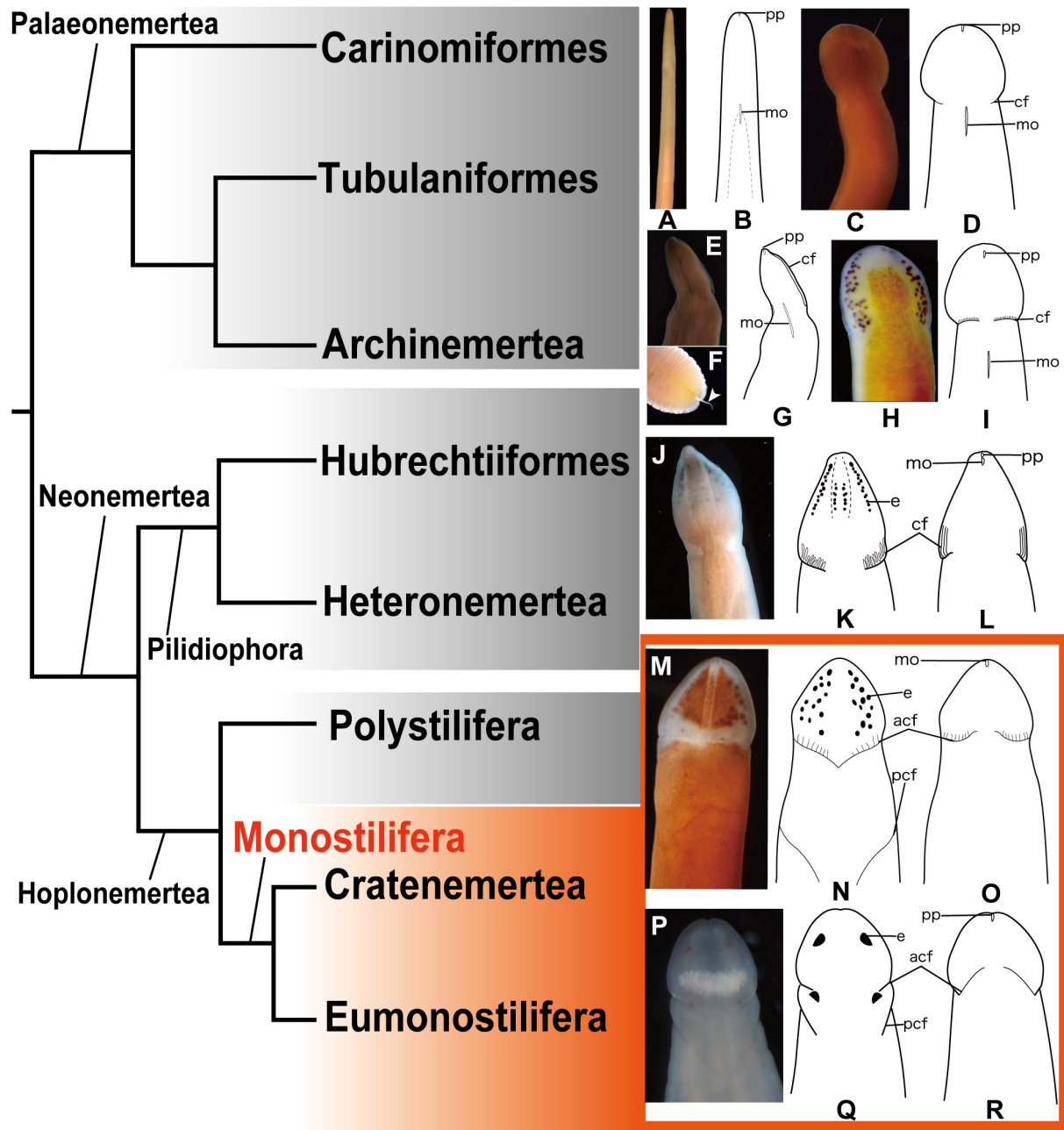


Figure 1. A diagram of phylogenetic relationships among Nemertea (modified from Chernyshev (2021)) and head characteristics of each higher taxon. (A–D) Palaeonemertea. (E–I) Pilidiophora. (J–R) Hoplonemertea. (A) *Cephalothrix* sp. (Cephalotrichidae), dorsal; (B) ventral; (C) *Tubulanus* sp. (Tubulanidae), dorsal; (D) ventral; (E) *Cerebratulus* sp. (Lineidae), dorsal; (F) caudal cirrus (arrowhead); (G) ventral; (H) *Baseodiscus* sp. (Valenciniidae), dorsal; (I) ventral; (J, K) *Reptantia* sp., dorsal; (L) ventral; (M, N) *Nipponnemertes jambio* Hookabe et al. 2022 (Cratenemertidae), dorsal; (O) ventral; (P, Q) *Tetrastemma album* sp. nov.

(Tetrastemmatidae), dorsal; (R) ventral. Abbreviations: acf, anterior cephalic furrow; cf, cephalic furrow; e, eye; mo, mouth; pcf, poseterior cephalic furrow; pp, proboscis pore.

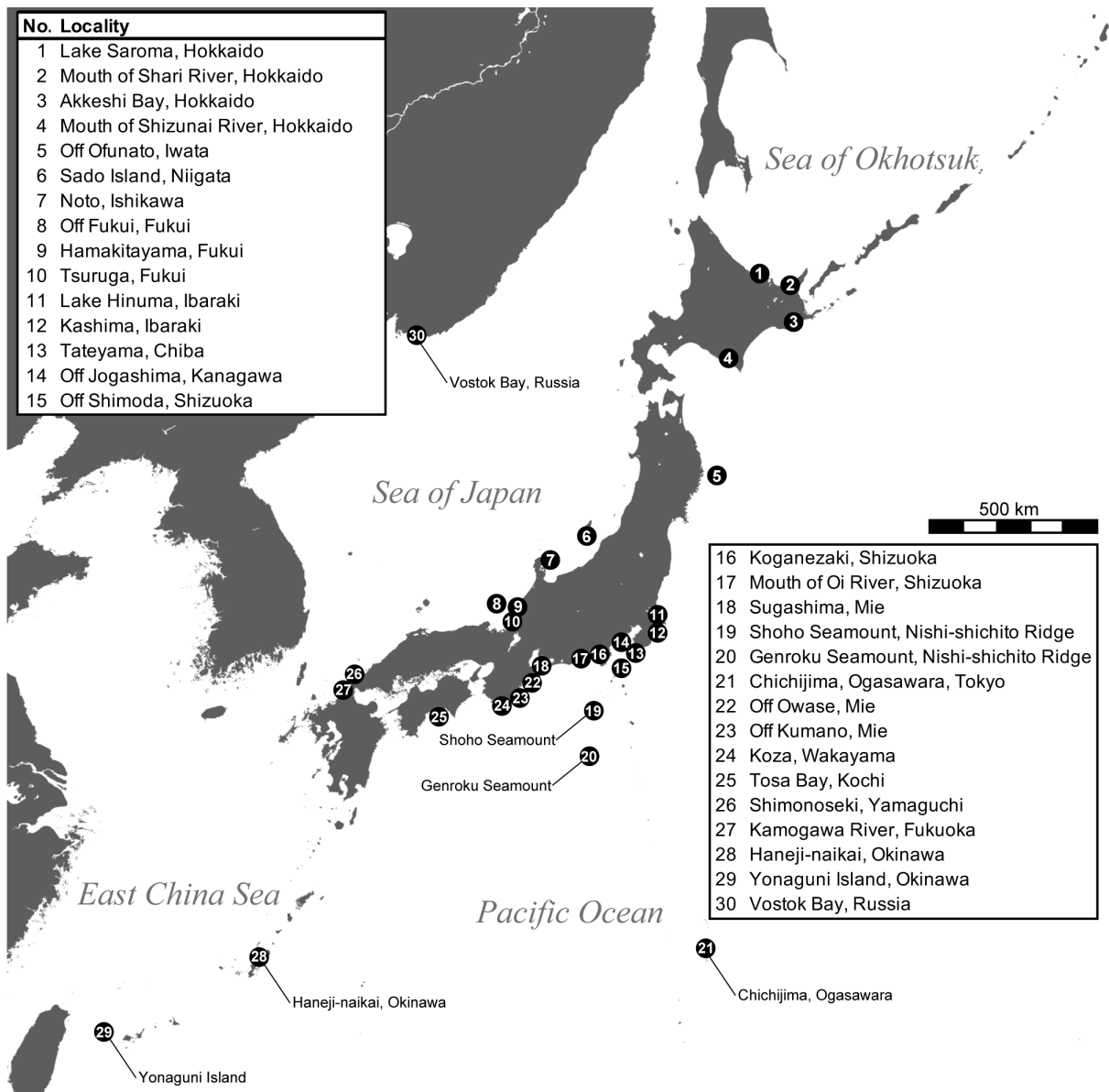


Figure 2. Collection sites for monostiliferan specimens studied in the dissertation. Detailed information on each location is listed in Tables 1 and 2.

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Figure 3. A phylogenetic tree reconstructed with ML analyses based on concatenated partial sequences of COI, 16S, 18S, 28S, and H3. Numbers near nodes indicate IQTree maximum-likelihood ultrafast bootstrap value (UFboot)/SH-aLRT/RAxML bootstrap

support values (BS) generated by maximum likelihood analysis with 1000 replicates/posterior probability (PP) of a separate partitioned Bayesian analysis (UFBoot/SH-aLRT/BS/PP). Solid circles indicate full support values (100/100.0/100/1.00). Circular circles indicate 1.00 of PP. Species names written in bold indicate the species studied in this study.

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Figure 4. A phylogenetic tree reconstructed with ML analyses based on concatenated dataset (12,841 bp) including the nucleotide sequences of 13 mitochondrial genome PCGs, 16S, and 12S. Numbers near nodes indicate IQTree maximum-likelihood ultrafast bootstrap value (UFBoot)/ RAxML bootstrap support values (BS) generated by maximum likelihood analysis with 1000 replicates/posterior probability (PP) of a separate partitioned Bayesian analysis (UFBoot/BS/PP). Solid circles indicate full support values (100/100/1.00). Species names written in bold indicate the species studied in this thesis.

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Figure 5. Serotonergic nervous system of *Malacobdella japonica* larvae. Green: 5-HT-like immunoreactivity; blue or purple: cilia, acetylated tubulin immunoreactivity; cyan: nuclei labelled with DAPI; red: muscles labelled with phalloidin. (A, A') 2-day-old larvae, a 20 μm -substack from dorsal view (A) and a 20 μm -substack lateral view (A') showing apical neurons (an) leading to lateral neurons (ln) via lateral nerve cords. (B, B', B''); 3-day-old larvae, a 20 μm -substack from dorsal view (B), a 20 μm -substack from lateral view (B'), and a 20 μm -substack from dorsal view with higher magnification of apical neurons (B''). (C, C') 5-day-old larvae, a 15 μm -substack from dorsal view (C) and a 15 μm -substack lateral view (C') showing well-developed lateral nerve cords. (D) 7-day-old larvae with higher magnification of dorsal nerve commissure (dnc), an arrowhead pointing to the mouth opening. (E, E', E'') 14-day-old larvae, a 15 μm -substack from dorsal view (E), a 15 μm -substack from dorsal view, with higher magnification of dorsal nerve commissure (dnc) and nerve

processes of 5-HT-like immunoreactive cells running from cephalic nerves (cn) into the body wall (arrowheads pointing to cerebral organs) (E'), and magnification of posterior nerve ring pointed with an arrowhead, a 10 μ m-substack from dorsal view (E''). (F, F') 21-day-old larvae, a 10 μ m-substack from lateral view (F) and a 10 μ m-substack from ventral view, mouth opening (arrowhead) leading to foregut lumen (F'). Abbreviations: an, apical neuron; ap, apical taft; dnc, dorsal nerve commissure; fg, foregut; ln, lateral neuron; lnc, lateral nerve cord; mg, midgut; pc, posterior cirrus; pnc, posterior nerve commissure. Scale bars: 30 μ m.

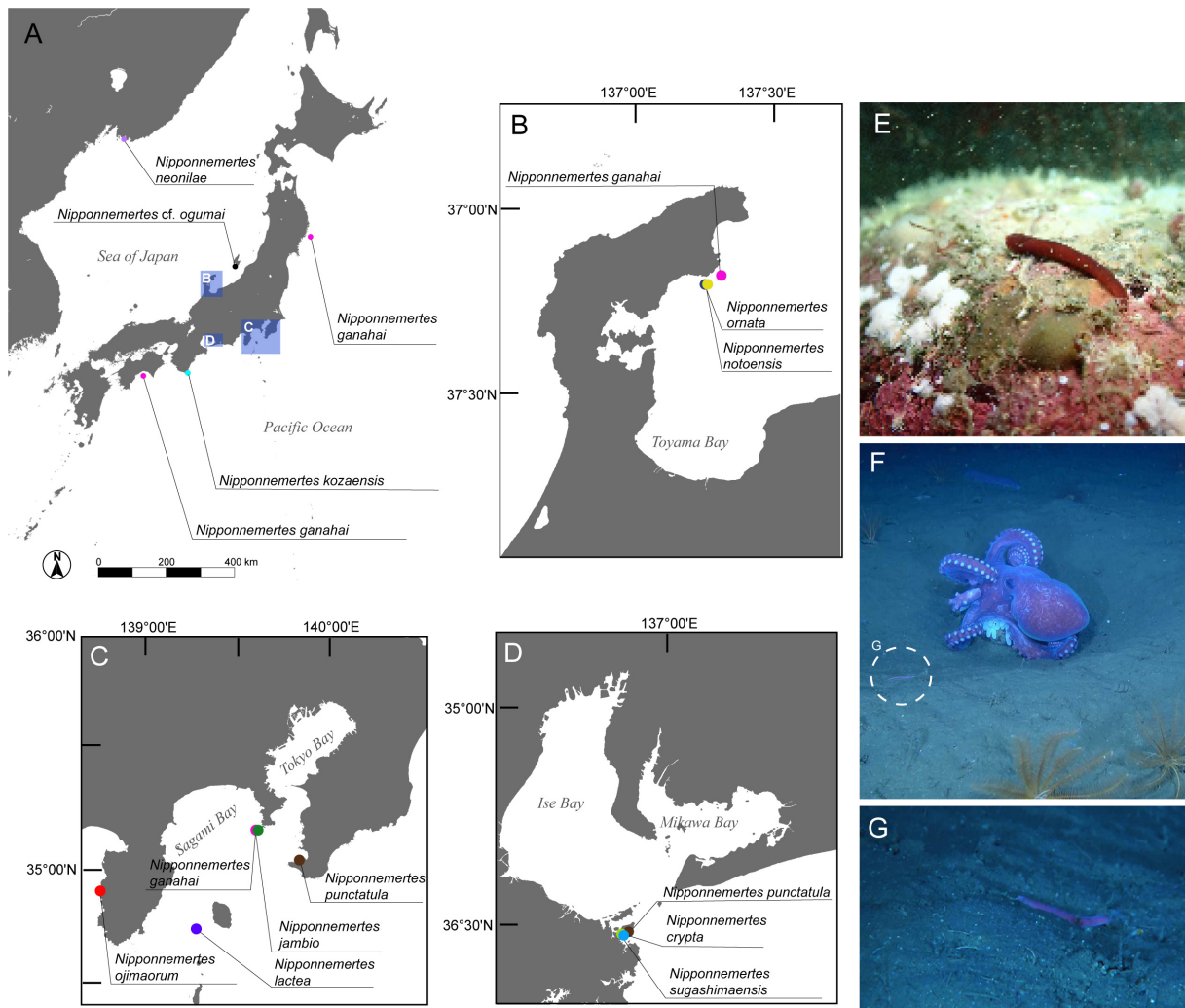


Figure 6. Sampling sites of specimens used in Chapter 3.1. (A) overview map; (B) collection sites in Noto, Japan; (C) collection sites in Kanto region and adjacent areas, Japan, including Tateyama, off Jogashima, and off Izu Peninsula; (D) collection sites around Sugashima, Japan; (E) in-situ photographs of *Nipponnemertes ojimaorum*, Koganezaki, Shizuoka, Japan; (F) in-situ photograph of *N. ganahai*, crawling near an octopus brooding eggs (circle with broken line), off Ofunato, Iwate, Japan; (G) close-up of *N. ganahai* in Fig. 6F.

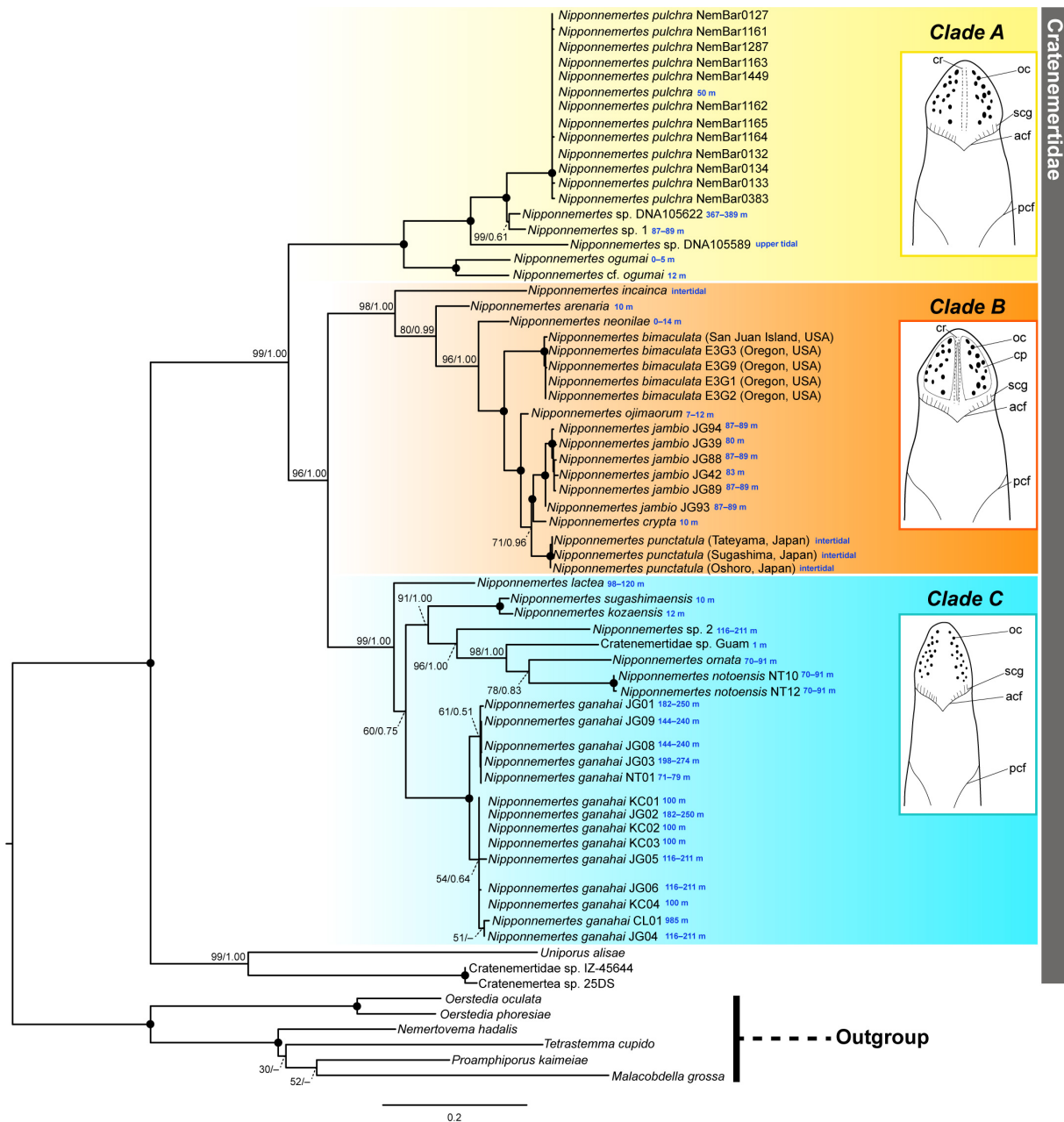


Figure 7. A phylogenetic tree of *Nipponnemertes* reconstructed with ML analyses using concatenated sequences of 16S, COI, 18S, 28S and H3, with illustrations showing general head morphology of species constituting each clade. Tree topology obtained by BI was generally identical with ML. Numbers near nodes indicate bootstrap support values (BS) generated by 1000 replicates and posterior probability (PP) of a separate partitioned BI analysis (BS/PP). Solid circles indicate full support values (100/1.00). Collection depths are provided in blue letters. Abbreviations: acf, anterior cephalic furrow; cp, cephalic patch; cr, mid-dorsal cephalic ridge; oc, ocelli; pcf, posterior cephalic furrow; scg, secondary transverse groove.

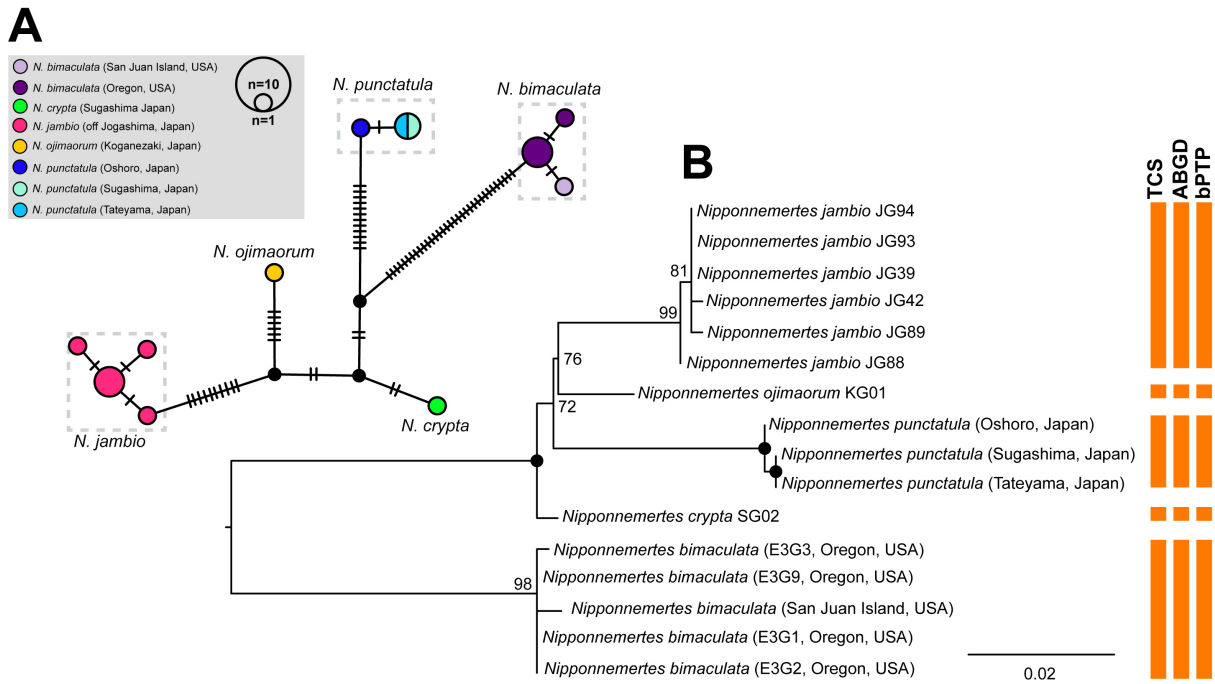


Figure 8. Results of species delimitation analyses for *Nipponnemertes* spp. in Clade B. (A) a haplotype network generated by statistical parsimony analysis based on partial sequence (658 bp) of COI. Numbers of black bars correspond to the numbers of nucleotide substitutions between the haplotypes; (B) a ML tree based on 658 bp of COI. Vertical bars indicate the putative species delineated with TCS, ABGD, and bPTP methods. Solid circles indicate 100 % of bootstrap values.

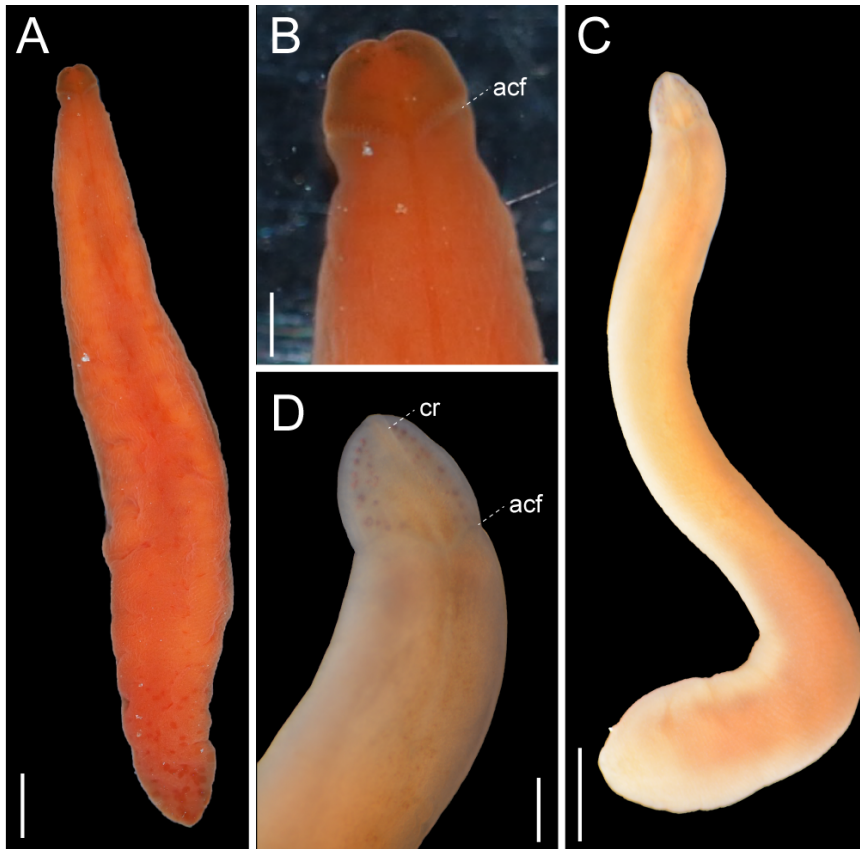


Figure 9. Images of *Nipponnemertes* spp., taken in life. (A, B) *Nipponnemertes* cf. *ogumai*, Sado. (A) whole body; (B) magnification of head; (C, D) *Nipponnemertes* sp. 1, off Jogashima. (C), whole body; (D) magnification of head. Abbreviations: acf, anterior cephalic furrow; cr, mid-dorsal cephalic ridge. Scale bars: 1 cm (A, C), 5 mm (B, D).

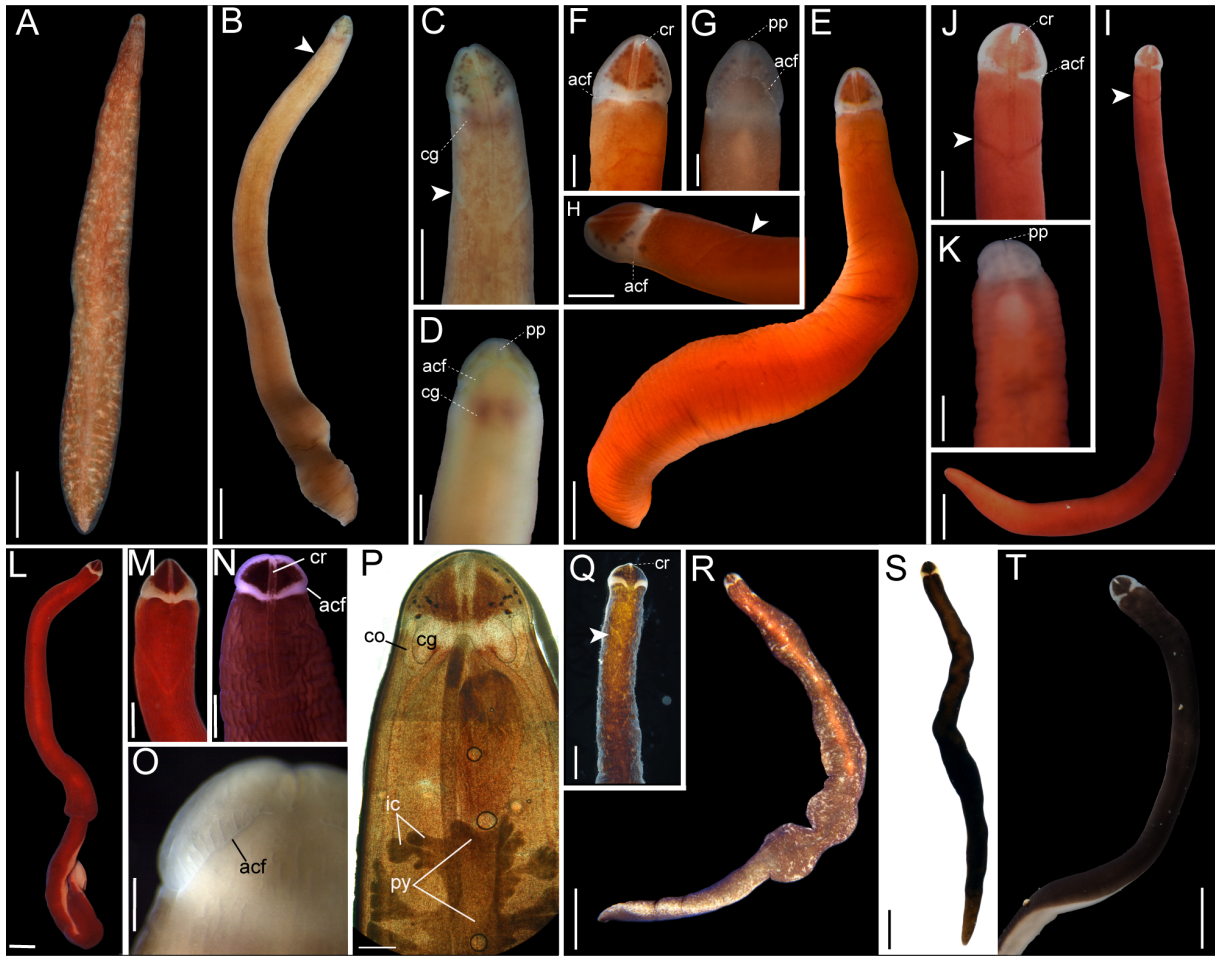


Figure 10. *Nipponnemertes* spp., taken in life. (A) *Nipponnemertes arenaria*, Peter the Great Bay. (B–D) *Nipponnemertes crypta*, Sugashima. (B) whole body; (C) magnification of head, dorsal; (D) ventral. (E–H) *Nipponnemertes jambio*, off Jogashima. (E) whole body; (F) magnification of head, dorsal; (G) ventral; (H) lateral. (I–K) *Nipponnemertes ojimaorum*, Koganezaki. (I) whole body; (J) magnification of head, dorsal; (K) ventral. (L–P) *Nipponnemertes neonilae*, Vostok Bay. (L) whole body; (M) magnification of head, dorsal, living specimen; (N) magnification of head, dorsal, fixed specimen in formalin; (O) magnification of head, ventral, living specimen; (P) microphotograph of a live squeezed specimen, dorsal. (Q–T) *Nipponnemertes punctatula*. (Q) Tateyama; (R, S) Misaki; (T) Sugashima. Each arrowhead points to posterior cephalic furrows. Abbreviations: acf, anterior cephalic furrow; cg, cerebral ganglia; co, cerebral organ; cr, mid-dorsal cephalic ridge; ic, lateral diverticulum of intestinal caecum; pp, proboscis pore; py, pylorus. Scale bars: 1 cm (A, B, L, S), 5 mm (B, C, I, M, N, Q, T); 2.5 mm (D); 1 mm (E, R); 500 μ m (F–H, J, K, O, P); 1.5 mm (I).



Figure 11. Images of *Nipponnemertes* spp., taken in life. (A–F) *Nipponnemertes ganahai*, collected off Ofunato (A–C), off Jogashima (D), Tosa Bay (E–G). (A) CL01, whole body; (B) magnification of head, dorsal; (C) lateral; (D) JG01, whole body; (E) KC01, whole body; (F) magnification of head, ventral; (G) microphotograph of a live squeezed specimen, dorsal. (H–J) *Nipponnemertes kozaensis*, Koza. (H) whole body; (I), magnification of head, ventral; (J) dorsal. (K, L) *Nipponnemertes lactea*, off Izu. (K) whole body; (L) microphotograph of a live squeezed specimen, ventral. (M) *Nipponnemertes notoensis*. (M) whole body; (N) magnification of head, dorsal; (O) ventral. (P, Q) *Nipponnemertes ornata*, off Noto. (P) whole body; (Q) magnification of head, dorsal. (R–U) *Nipponnemertes sugashimaensis*, Sugashima. (R) whole body; (S) magnification of head, dorsal; (T) microphotograph of a live squeezed specimen, dorsal; (U) lateral blood vessel. (V, W) *Nipponnemertes* sp. 2, off Jogashima. (V) whole body; (W) microphotograph of a live squeezed specimen, dorsal. (X)

Cratemertidae sp. Guam, a live squeezed specimen. Each arrowhead points to posterior cephalic furrows. Abbreviations: acf, anterior cephalic furrow; bv, blood vessel; cg, cerebral ganglia; pb, proboscis; pp, proboscis pore. Scale bars: 2 mm (A, D, E); 1.5 mm (B); 1 mm (C, H, M, P, V); 500 μm (F, K, L, N, O, U); 300 μm (G, J, Q, S); 700 μm (I, R); 100 μm (T, W, X).

	Clade B						Clade C					
	<i>N. crypta</i>	<i>N. jambio</i>		<i>N. neonilae</i>	<i>N. ojimaorum</i>	<i>N. ganahai</i>		<i>N. kozaensis</i>	<i>N. lactea</i>	<i>N. augsahmnaensis</i>	<i>Nipponnemertes</i> sp. 2	
	SG02	JG12	JG13	JG14	RU01	KG01	KC01	NT01	KZ01	TK01	SG01	JG16
Central stylet												
Stylet length (µm)	2.16	2.08	1.86	2.15	1.39	1.85	1.29	1.56	1.51	1.25	1.64	1.38
Basis length (µm)												
Accessory stylets												
Number of pouches	2	2	2	2	2	2	2	2	2	2	2	2
Number of stylets per pouch	9	4	5	5	7	7	4	4	8	7	7	6

Figure 12. Stylet apparatus of *Nipponnemertes* spp. herein described, micrographs of squeezed live specimens.

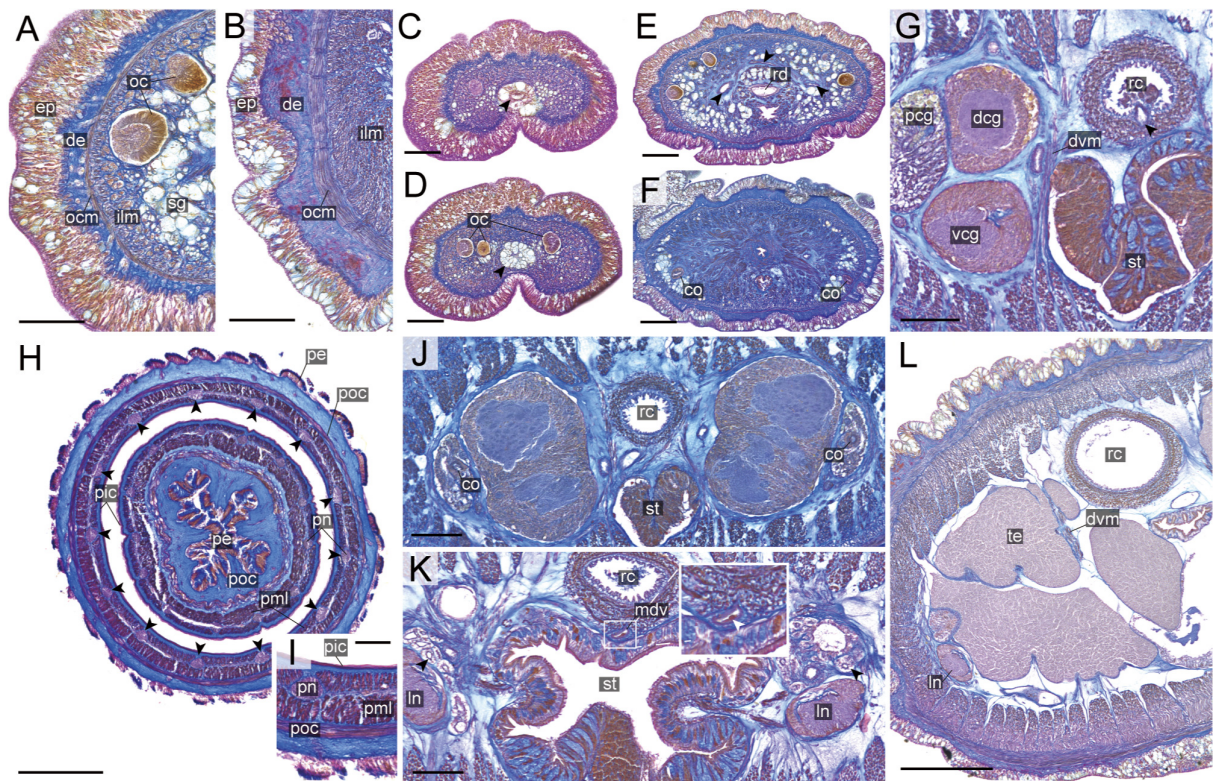


Figure 13. *Nipponnemertes crypta*, transverse sections. (A) body wall in precerebral region; (B) body wall in foregut region; (C) apical organ (arrowhead); (D) cephalic glands (arrowhead); (E) precerebral vessels (arrowheads); (F) precerebral septum; (G) vascular plug (arrowhead); (H) proboscis, arrowheads pointing to proboscis nerves; (I) magnification of proboscis musculature; (J) posterior region of cerebral ganglia; (K) nephridial canals (black arrowheads) and mid-dorsal vessel (white box, a white arrowhead pointing to a blood cell in mid-dorsal vessel); (L) testes. Abbreviations: co, cerebral organ; dcg, dorsal cerebral ganglia; de, dermis; dvm, dorso-ventral muscle in body wall; ep, epidermis; ilm, body-wall inner longitudinal muscle; mdv, mid-dorsal vessel; oc, ocelli; ocm, body-wall outer circular muscle; pcg, postcerebral glands of cerebral organ; pe, proboscis epithelium; pic, proboscis inner circular muscle; pml, proboscis middle longitudinal muscle; pn, proboscis nerve; poc, proboscis outer circular muscle; rc, rhynchocoel; rd, rhynchodaeum; st, stomach; sg, submuscular glands; te, testis; vcg, ventral cerebral ganglia. Scale bars: 50 μm (A–D, I); 100 μm (E, F, K); 200 μm (G); 150 μm (H, J); 250 μm (L)

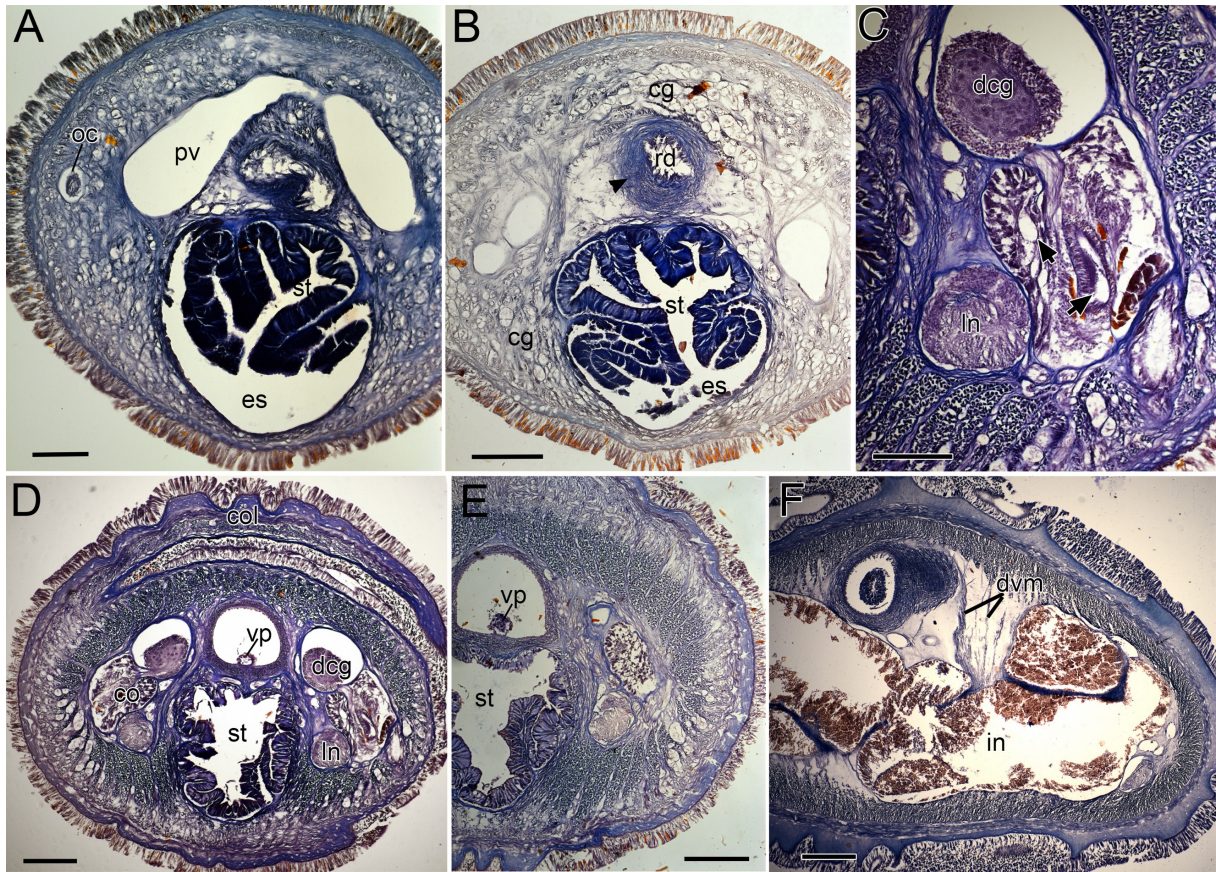


Figure 14. *Nipponnemertes neonilae*, transverse sections. (A) anterior part of cephalic blood lacuna; (B) rhynchodeal sphincter (arrowhead); (C) cerebral organ (arrows indicate bifurcate canal of cerebral organ); (D) collar; (E) stomach; (F) intestine. Abbreviations: cg, cephalic glands; co, cerebral organ; col, collar; dcg, dorsal cerebral ganglia; dvm, dorso-ventral muscles; es, esophagus; in, intestine; ln, lateral nerve; oc, ocelli; pv, precerebral vessel; rd, rhynchodaeum, st, stomach; vp, vascular plug. Scale bars: 100 μm (A, C); 200 μm (B, D–F).

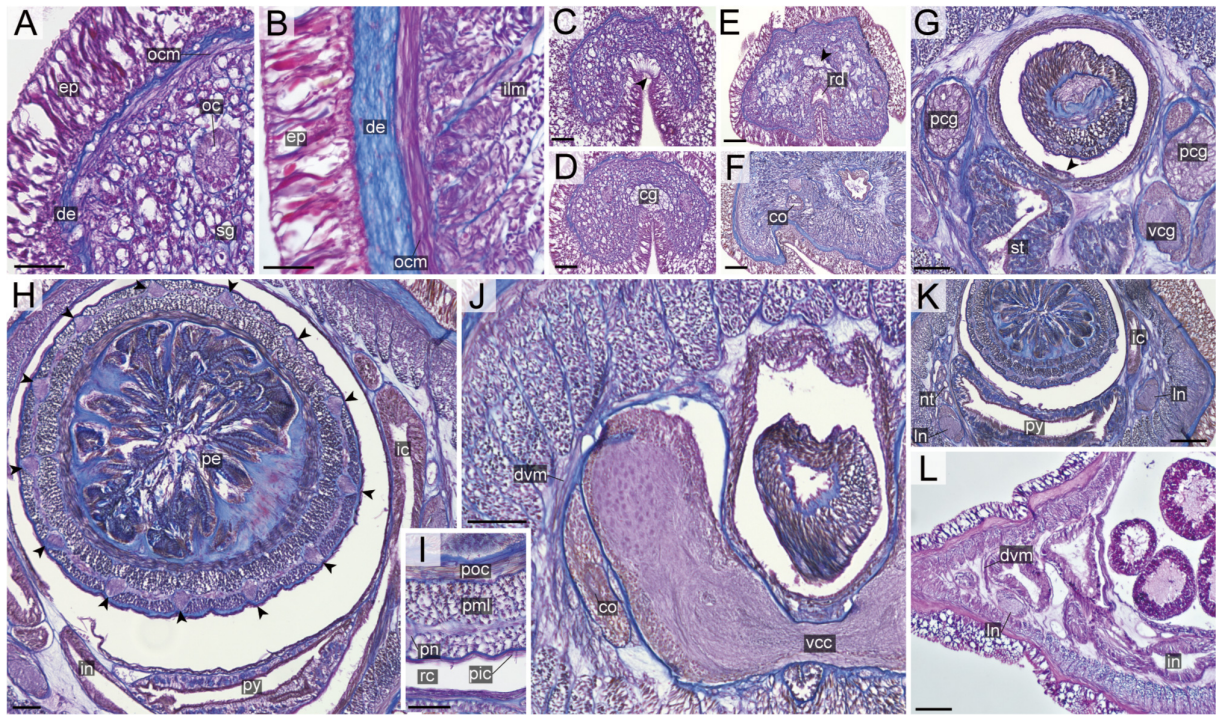


Figure 15. *Nipponnemertes ojimaorum*, transverse sections. (A) body wall in precerebral region; (B) body wall in foregut region; (C) apical organ (arrowhead); (D) cerebral glands; (E) precerebral vessel (arrowhead); (F) cerebral organ opening; (G) mid-dorsal vessel (arrowhead); (H) proboscis, arrowheads pointing to proboscis nerves; (I) magnification of proboscis musculature; (J) ventral cerebral commissure; (K) nephridial tubules; (L) intestine. Abbreviations: co, cerebral organ; dcg, dorsal cerebral ganglia; de, dermis; dvm, dorso-ventral muscle in body wall; ep, epidermis; ic, lateral diverticulum of intestinal caecum; ilm, body-wall inner longitudinal muscle; in, intestine; ln, lateral nerve; mdv, mid-dorsal vessel; nt, nephridial tubule; oc, ocelli; ocm, body-wall outer circular muscle; pcg, postcerebral glands of cerebral organ; pe, proboscis epithelium; pic, proboscis inner circular muscle; pml, proboscis middle longitudinal muscle; pn, proboscis nerve; poc, proboscis outer circular muscle; rc, rhynchocoel; rd, rhynchodaeum; sg, submuscular glands; st, stomach; vcg, ventral cerebral ganglia. Scale bars: 50 μm (A, C, D, E); 25 μm (B, F); 100 μm (G, J–L); 75 μm (H); 30 μm (I)

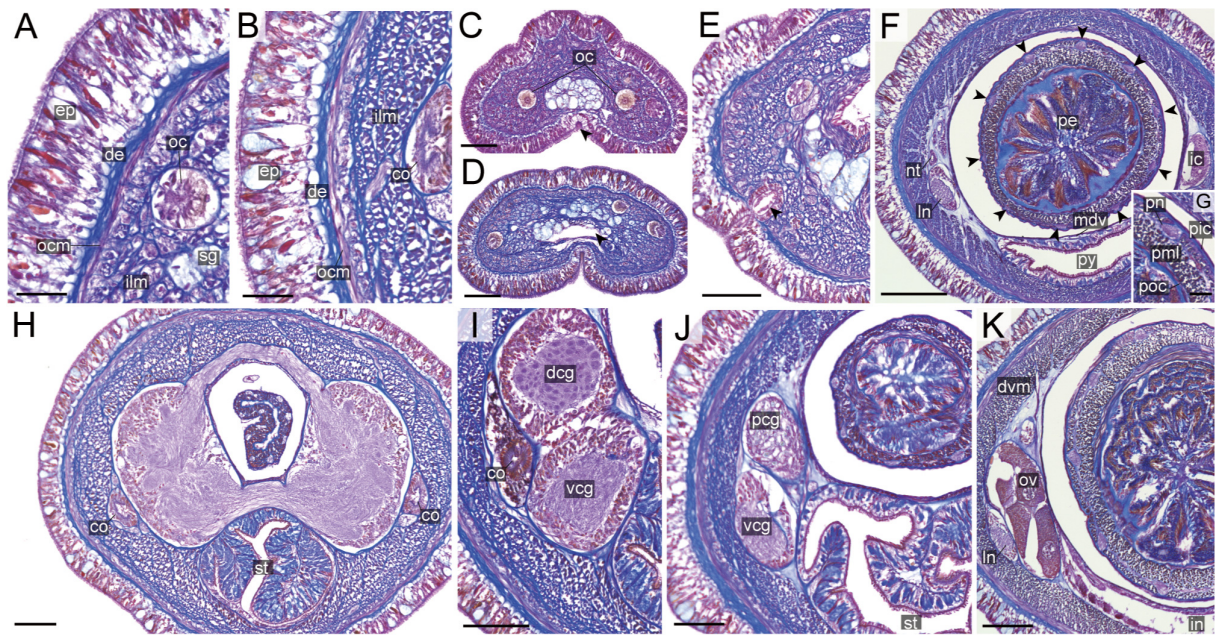


Figure 16. *Nipponnemertes ganahai*, transverse sections. (A) body wall in precerebral region; (B) body wall in foregut region; (C) apical organ (white arrowhead) and cephalic glands (black arrowhead); (D) precerebral vessels (arrowhead); (E) cerebral organ opening (arrowhead); (F) proboscis, arrowheads pointing to proboscis nerves; (G) magnification of proboscis musculature; (H) dorsal and ventral cerebral commissure; (I) cerebral organ; (J) posterior region of cerebral ganglia; (K) ovaries. Abbreviations: co, cerebral organ; dcg, dorsal cerebral ganglia; de, dermis; dvm, dorso-ventral muscle in body wall; ep, epidermis; ic, lateral diverticulum of intestinal caecum; ilm, body-wall inner longitudinal muscle; in, intestine; ln, lateral nerve; mdv, mid-dorsal vessel; nt, nephridial tubule; oc, ocelli, ocm; body-wall outer circular muscle; ov, ovaries; pcg, postcerebral glands of cerebral organ; pe, proboscis epithelium; pic, proboscis inner circular muscle; pml, proboscis middle longitudinal muscle; pn, proboscis nerve; poc, proboscis outer circular muscle; py, pylorus; sg, submuscular glands; st, stomach; vcg, ventral cerebral ganglia. Scale bars: 30 μm (A, B, G); 50 μm (C–E, J, K); 150 μm (F); 75 μm (H, I)

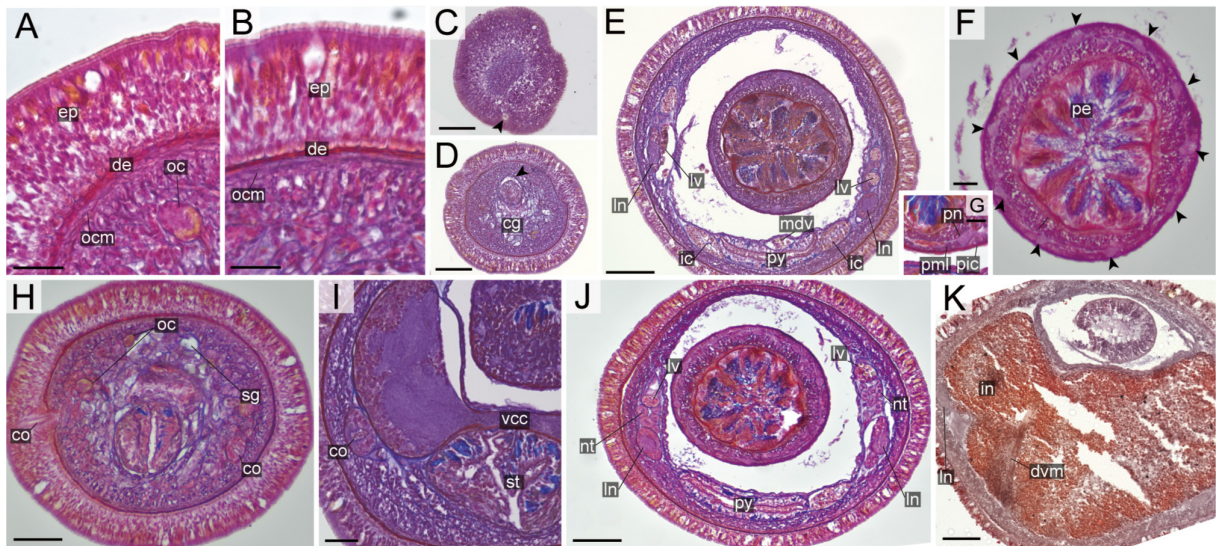


Figure 17. *Nipponnemertes kozaensis*, transverse sections. (A) body wall in precerebral region; (B) body wall in foregut region; (C) apical organ (arrowhead); (D) precerebral vessel (arrowhead) running above cerebral glands; (E) mid-dorsal vessel; (F) proboscis, arrowheads pointing to proboscis nerves; (G) magnification of proboscis musculature; (H) cerebral organ opening; (I) cerebral organ; (J) nephridial tubules; (K) intestine. Abbreviations: cg, cephalic glands; co, cerebral organ; de, dermis; dvm, dorso-ventral muscle in body wall; ep, epidermis; ic, lateral diverticulum of intestinal caecum; in, intestine; ln, lateral nerve; lv, lateral vessel; mdv, mid-dorsal vessel; nt, nephridial tubule; oc, ocelli; ocm, body-wall outer circular muscle; pcg, postcerebral glands of cerebral organ; pe, proboscis epithelium; pic, proboscis inner circular muscle; pml, proboscis middle longitudinal muscle; pn, proboscis nerve; py, pylorus; st, stomach; vcc, ventral cerebral commissure. Scale bars: 15 μm (A, B); 50 μm (C, D, F); 75 μm (E, H, J, K); 25 μm (G); 30 μm (I)

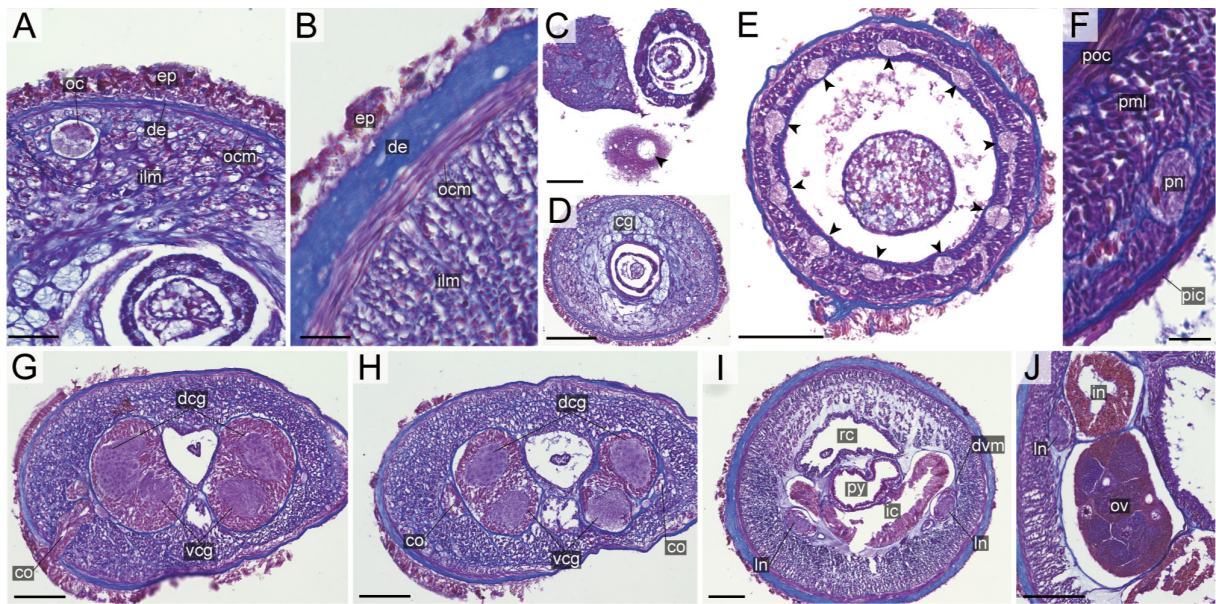


Figure 18. *Nipponnemertes lactea*, transverse sections. (A) body wall in precerebral region; (B) body wall in foregut region; (C) apical organ (arrowhead); (D) cerebral gland; (E) proboscis, arrowheads pointing to proboscis nerves; (F) magnification of proboscis musculature; (G) cerebral organ opening; (H) posterior region of cerebral ganglia; (I) pylorus; (J) ovaries. Abbreviations: co, cerebral organ; dcg, dorsal cerebral ganglia; de, dermis; dvm, dorso-ventral muscle in body wall; ep, epidermis; ic, lateral diverticulum of intestinal caecum; ilm, body-wall inner longitudinal muscle; in, intestine; ln, lateral nerve; oc, ocelli; ocm, body-wall outer circular muscle; ov, ovary; pe, proboscis epithelium; pic, proboscis inner circular muscle; pml, proboscis middle longitudinal muscle; pn, proboscis nerve; poc, proboscis outer circular muscle; py, pylorus; rc, rhynchocoel; vcg, ventral cerebral ganglia. Scale bars: 30 μm (A, F); 25 μm (B, C); 100 μm (D, E); 75 μm (G–I); 50 μm (J)

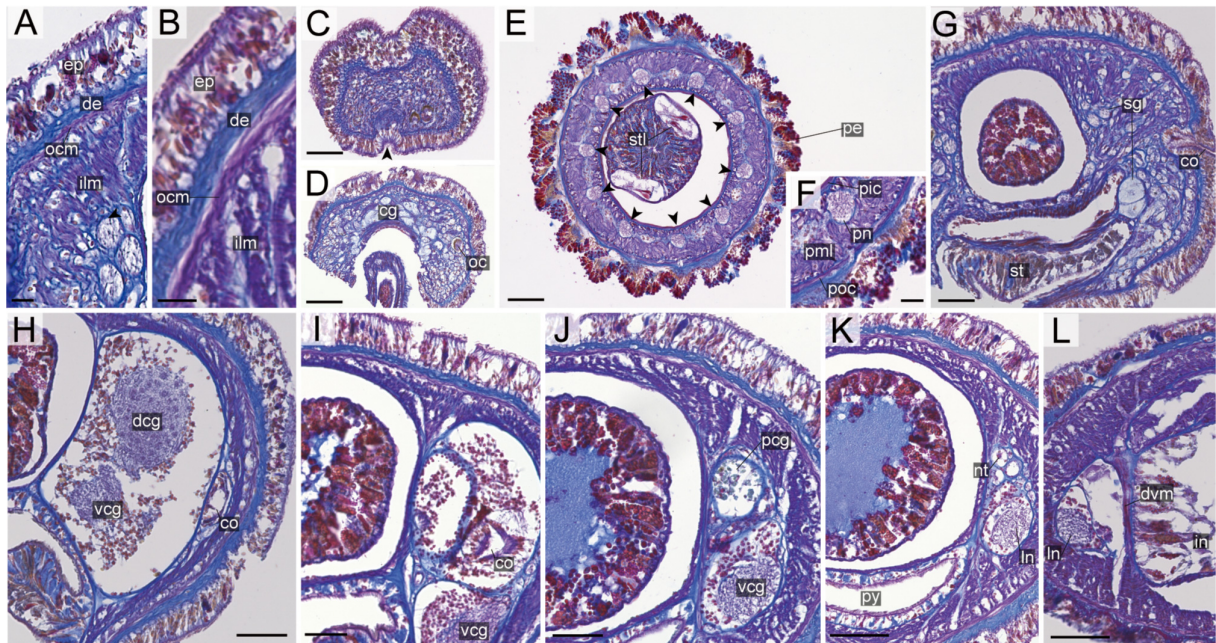


Figure 19. *Nipponnemertes notoensis*, transverse sections. (A) body wall in precerebral region; (B) body wall in foregut region; (C) apical organ (arrowhead); (D) cerebral glands; (E) proboscis, arrowheads pointing to proboscis nerves; (F) magnification of proboscis musculature; (G) cerebral organ opening; (H) middle region of cerebral organ; (I) posterior region of cerebral organ; (J) postcerebral glands of cerebral organ; (K) nephridial tubules; (L) intestine. Abbreviations: cg, cephalic glands; co, cerebral organ; dcg, dorsal cerebral ganglia; de, dermis; dvm, dorso-ventral muscle in body wall; ep, epidermis; ilm, body-wall inner longitudinal muscle; in, intestine; ln, lateral nerve; nt, nephridial tubule; oc, ocelli; ocm, body-wall outer circular muscle; pcg, postcerebral glands of cerebral organ; pe, proboscis epithelium; pic, proboscis inner circular muscle; pml, proboscis middle longitudinal muscle; pn, proboscis nerve; poc, proboscis outer circular muscle; py, pylorus; sg, submuscular glands; st, stomach; stl, stylet; vcg, ventral cerebral ganglia. Scale bars: 15 μm (A, B); 60 μm (C, D, L); 75 μm (E); 30 μm (F, I, J); 50 μm (G, H, K)

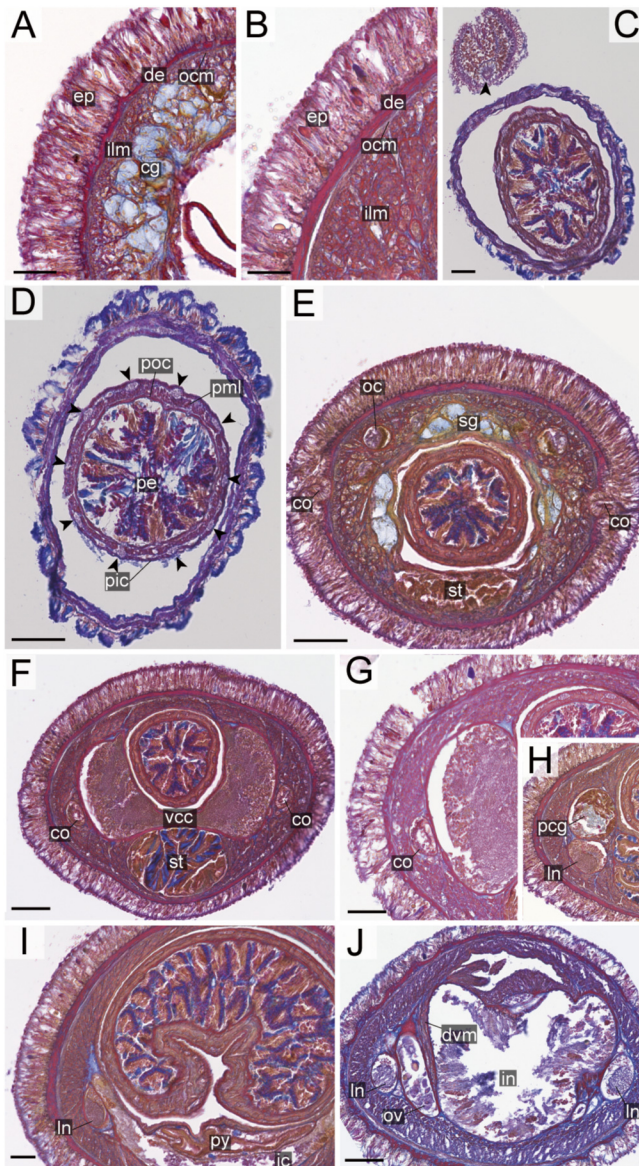


Figure 20. *Nipponnemertes ornata*, transverse sections. (A) body wall in precerebral region; (B) body wall in foregut region; (C) apical organ (arrowhead); (D) proboscis, arrowheads pointing to proboscis nerves; (E) cerebral organ opening; (F) ventral cerebral commissure; (G) cerebral organ; (H) postcerebral glands; (I) pylorus and intestinal caecum; (J) ovaries. Abbreviations: cg, cephalic glands; co, cerebral organ; de, dermis, dvm; dorso-ventral muscle in body wall; ep, epidermis; ic, lateral diverticulum of intestinal caecum; ilm, body-wall inner longitudinal muscle; in, intestine; ln, lateral nerve; nt, nephridial tubule; ocm, body-wall outer circular muscle; ov, ovaries; pcg, postcerebral glands of cerebral organ; pe, proboscis epithelium; pic, proboscis inner circular muscle; pml, proboscis middle longitudinal muscle; pn, proboscis nerve; pcc, proboscis outer circular muscle; py, pylorus; st, stomach; sg,

submuscular glands; vcc, ventral cerebral commissure. Scale bars: 20 μm (A, B, H, I); 50 μm (C, E, F, J); 75 μm (D); 30 μm (G)

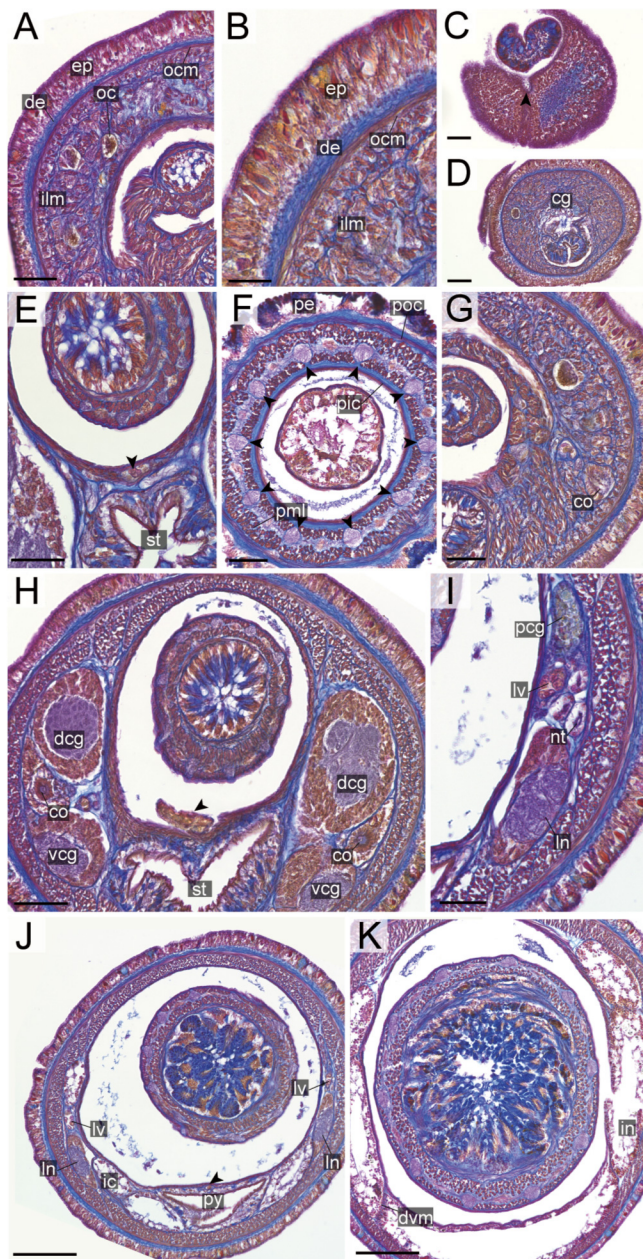


Figure 21. *Nipponnemertes sugashimaensis*, transverse sections. (A) body wall in precerebral region; (B) body wall in foregut region; (C) apical organ (arrowhead); (D) cerebral glands; (E) mid-dorsal vessel (arrowhead); (F) proboscis, arrowheads pointing to proboscis nerves; (G) cerebral organ opening; (H) posterior region of cerebral ganglia, arrowhead pointing to vascular plug; (I) nephridial tubules; (J) pylorus, arrowhead pointing to mid-dorsal vessel; (K) intestine. Abbreviations: cg, cephalic glands; co, cerebral organ; dcg, dorsal cerebral ganglia; de, dermis; dvm, dorso-ventral muscle in body wall; ep, epidermis; ic, lateral diverticulum of intestinal caecum; ilm, body-wall inner longitudinal muscle; in, intestine; ln, lateral nerve; lv, lateral vessel; nt, nephridial tubule; oc, ocelli; ocm, body-wall outer circular muscle; pcg, postcerebral

glands of cerebral organ; pe, proboscis epithelium; pic, proboscis inner circular muscle; pml, proboscis middle longitudinal muscle; pn, proboscis nerve; poc, proboscis outer circular muscle; py, pylorus; vcg, ventral cerebral ganglia. Scale bars: 30 μm (A); 20 μm (B); 50 μm (C–F, G–I); 150 μm (J); 100 μm (K)

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Figure 22. *Amphiporus* sp. Saroma. (A) whole body, dorsal; (B) magnification of head, dorsal; (C) ventral; (D) lateral; (E) magnification of head, squeezed specimen; (F) stylet apparatus. Abbreviations: acf, anterior cephalic furrow; cg, cerebral ganglia; pcf, posterior cephalic furrow; pp, proboscis pore. Scale bars: 2 cm (A); 2.5 mm (B–E); 100 μ m (F)

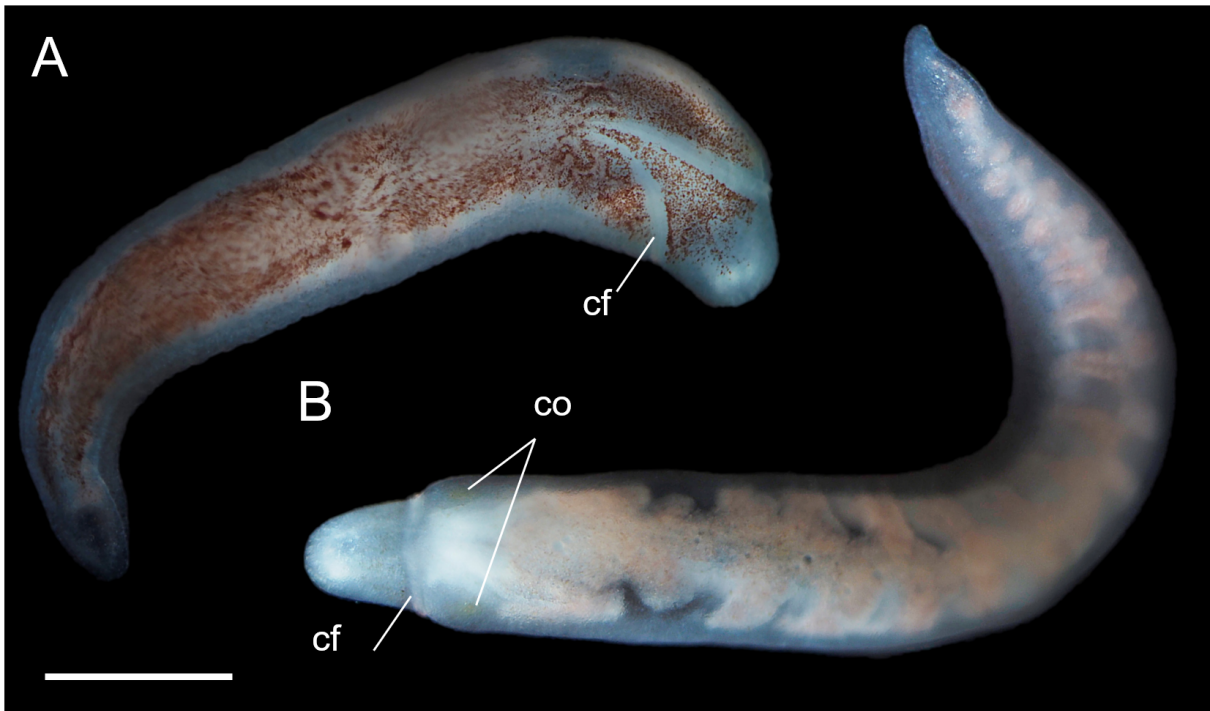


Figure 23. *Proamphiporus kaimeiae*. (A) whole body, dorsal; (B) ventral. Abbreviations: cf, cephalic furrow; cg, cerebral ganglia; in, intestine. Scale bar: 500 μm .

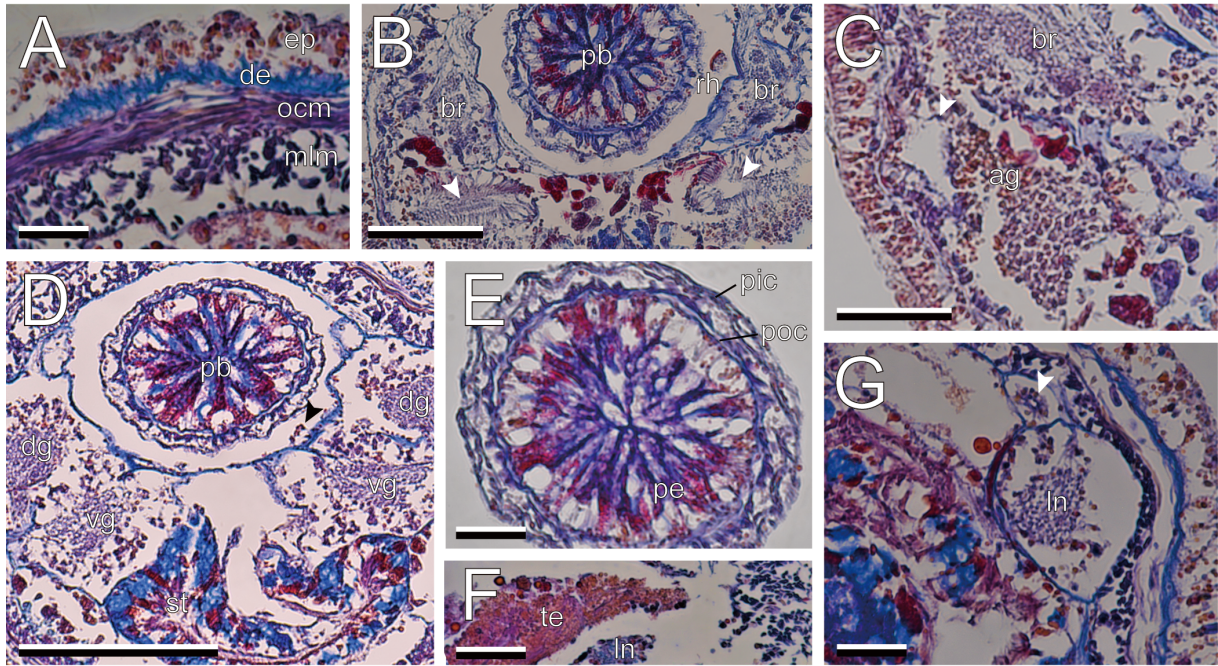


Figure 24. *Proamphiporus kaimeiae*, photomicrographs of transverse sections. (A) body wall in brain region; (B) anterior part of cerebral organs; arrowheads pointing to the radial canal; (C) posterior part of cerebral organs; arrowhead pointing to the lateral canal; (D) brain; arrowhead pointing to vascular plug; (E) proboscis; (F) testis; (G) excretory collecting tubule (arrowhead). Abbreviations: ag, acidophilic glands; br, brain; de, dermis; dg, dorsal ganglion; ep, epidermis; ln, lateral nerve cord; mlm, middle longitudinal muscle layer of body wall; ocm, outer circular muscle layer of body wall; pb, proboscis; pe, proboscis epithelium; pic, proboscis inner circular muscle layer; poc, proboscis outer circular muscle layer; rh, rhynchocoel; st, stomach; te, testis; vg, ventral ganglion. Scale bars: 20 μm (A); 75 μm (B); 50 μm (C, G); 100 μm (D); 40 μm (E, F)

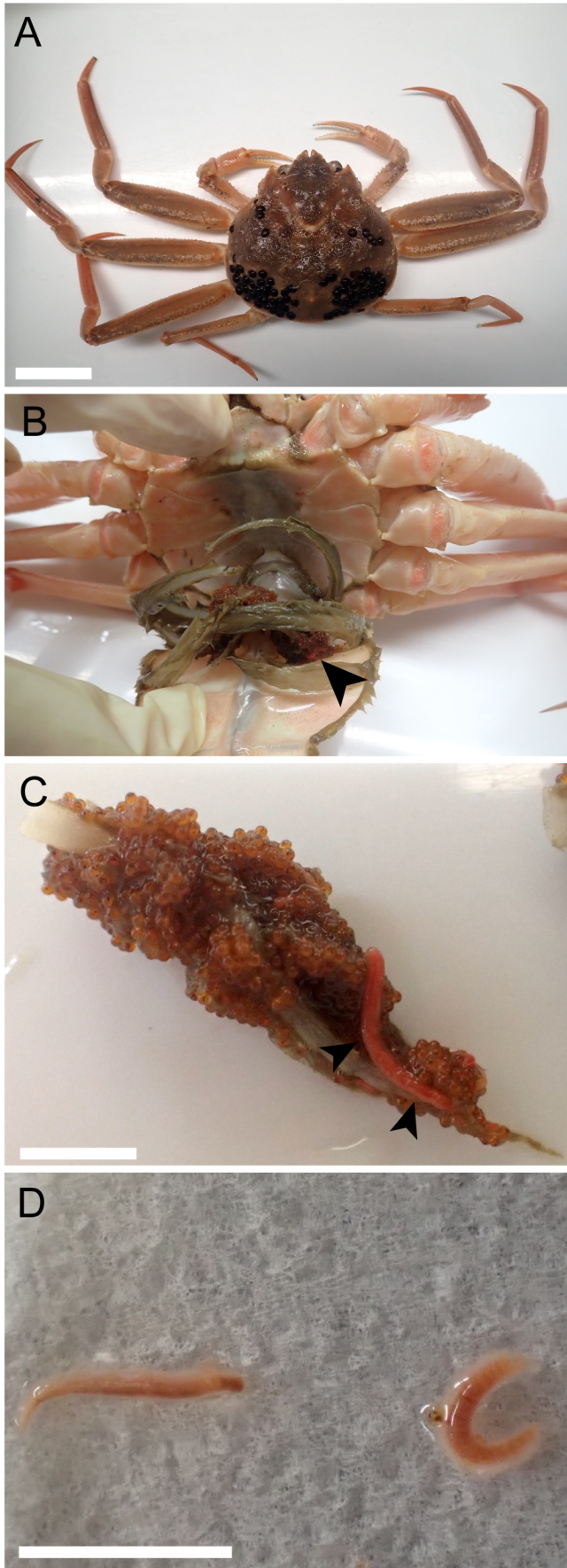


Figure 25. *Ovicides paralithodes* and its host *Chinocetes ophilio*. (A) a newly reported host in the present study, *Chinocetes ophilio*; (B) a female crab carrying its egg masses in the abdomen, an arrowhead indicating a specimen of *O. paralithodes*; (C) egg masses extracted from a female crab with a single specimen of *O. paralithodes*; (D) living specimens of *O. paralithodes*. Scale bars: 4 cm (A); 5mm (C); 1 cm (D)

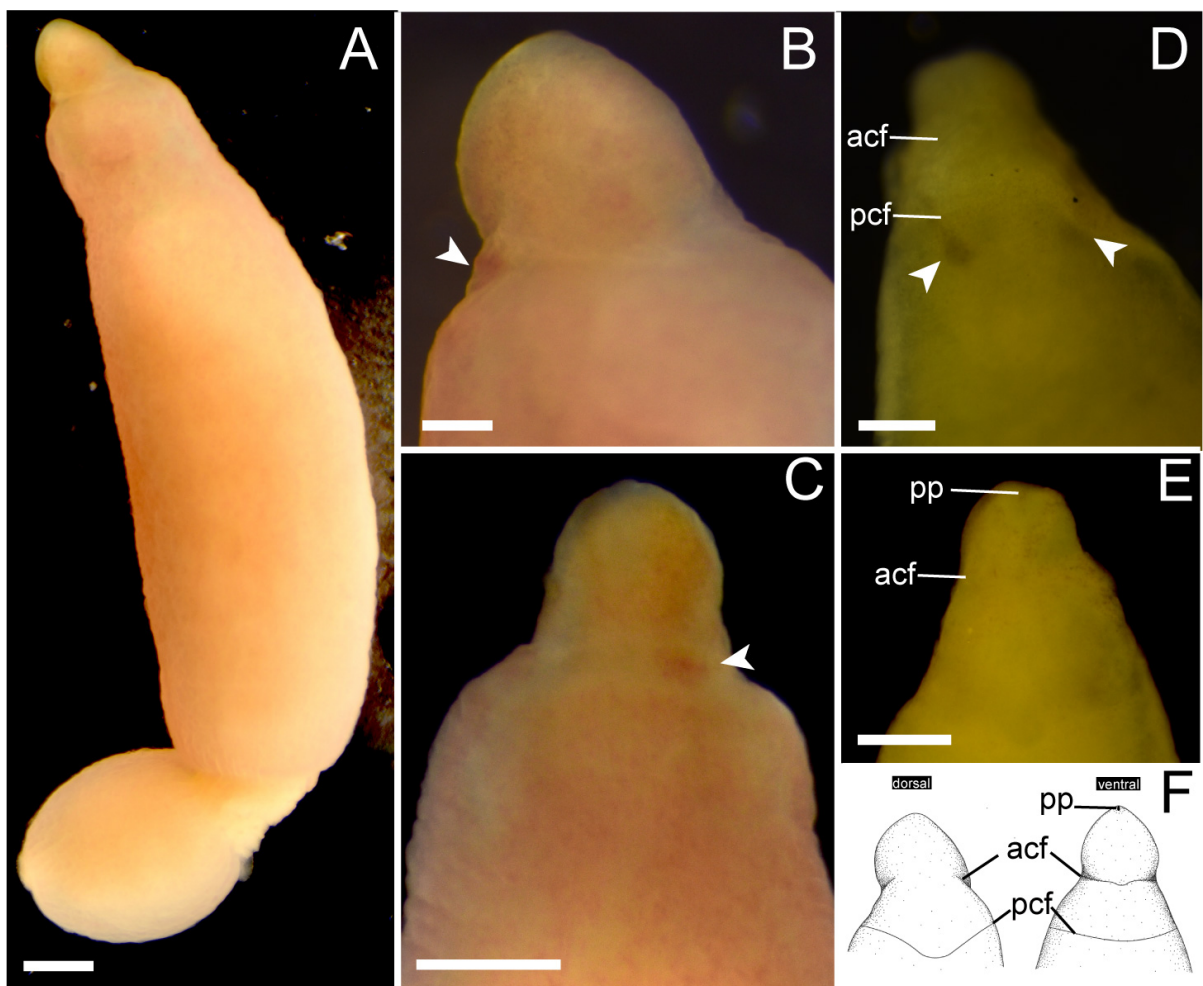


Figure 26. *Genrokunemertes obesa*. (A) Whole body, dorsal view; (B) magnification of head, living state, dorsal view, an arrowhead pointing to cerebral ganglia; (C) magnification of head, living state, ventral view, an arrowhead pointing to cerebral ganglia; (D) magnification of head, cleared in xylene, dorsal view, arrows pointing to cerebral organs; (E) magnification of head, cleared in xylene, ventral view; (F) illustrations of head, dorsal and ventral side. Abbreviations: acf, anterior cephalic furrow; pcf, posterior cephalic furrow; pp, proboscis pore. Scale bars: 500 μm (A, C); 250 μm (B–D)

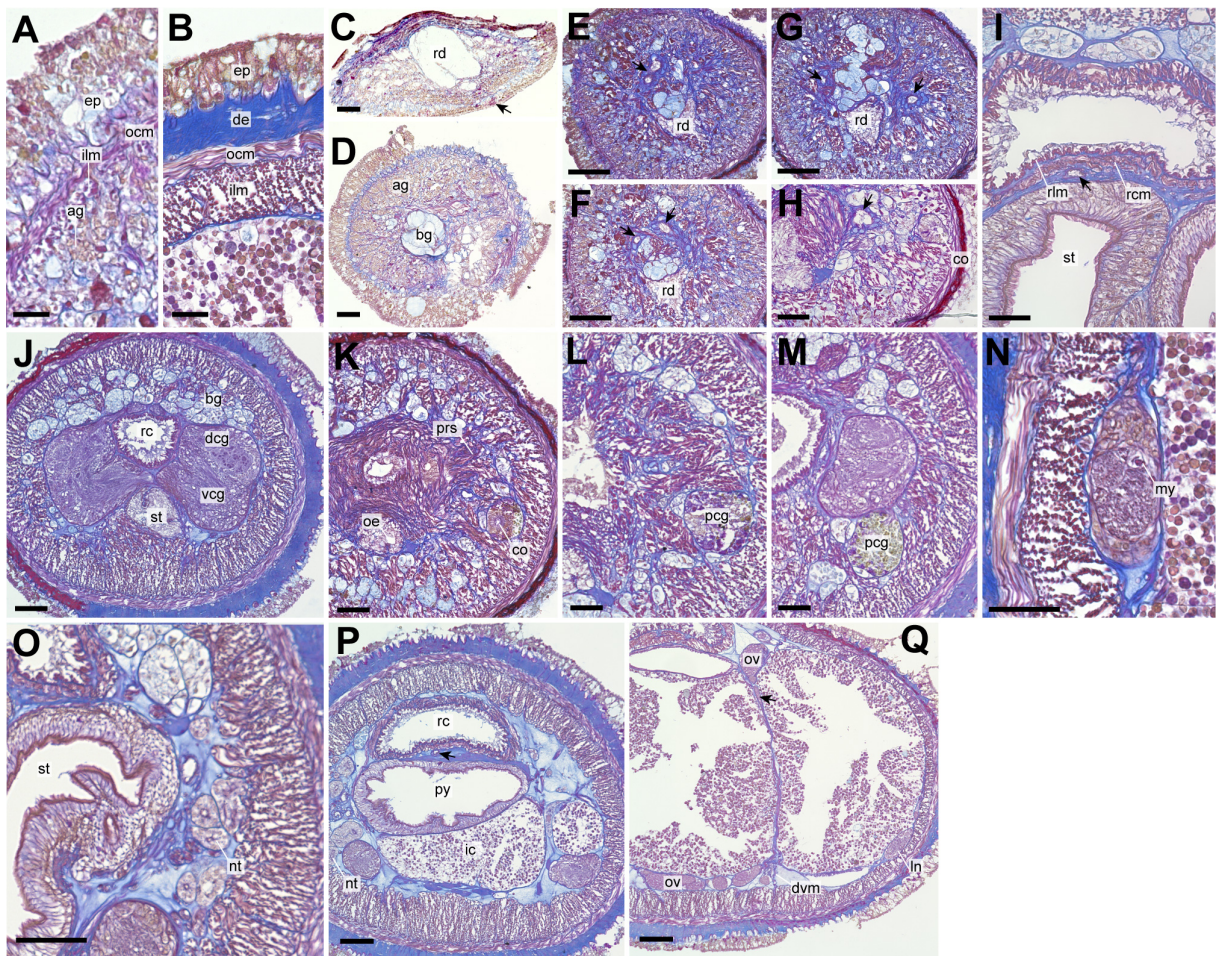


Figure 27. *Genrokunemertes obesa*, photomicrographs of transverse sections. (A) Body wall in precerebral region; (B) body wall in intestine region; (C) frontal organ; (D) precerebral cephalic glands; (E–H) precerebral vessels posteriorly branching above rhynchodaeum (arrow); (I) rhynchocoel wall, an arrow pointing to mid-dorsal vessel; (J) brain; (K, L) cerebral organ located anterior to brain; (M) posterior glands of cerebral organ; (N) lateral nerve cord; (O) nephridial tubules; (P) pylorus, an arrow pointing to mid-dorsal vessel; (Q) intestine, an arrow pointing to a dorsoventral process between ovary-like sacs. Abbreviations: ag, acidophilic glands; bc, basophilic glands; co, cerebral organ; de, dermis; dcg, dorsal cerebral ganglia; dvm, dorsoventral muscle; ep, epithelium; in, intestine; my, myofibril; ocm, outer circular muscle layer of body wall; oe, oesophagus; ov, ovary-like sac; pcg, posterior glands of cerebral organ; prs, precerebral septum; py, pylorus; rc, rhynchocoel; rcm, rhynchocoel outer circular muscle layer; rd, rhynchodaeum; rlm, rhynchocoel inner longitudinal muscle layer; vcg, ventral cerebral ganglia. Scale bars: 25 μm (A, B); 30 μm (C, D, N, O); 100 μm (E–H); 50 μm (I–M, P, Q)

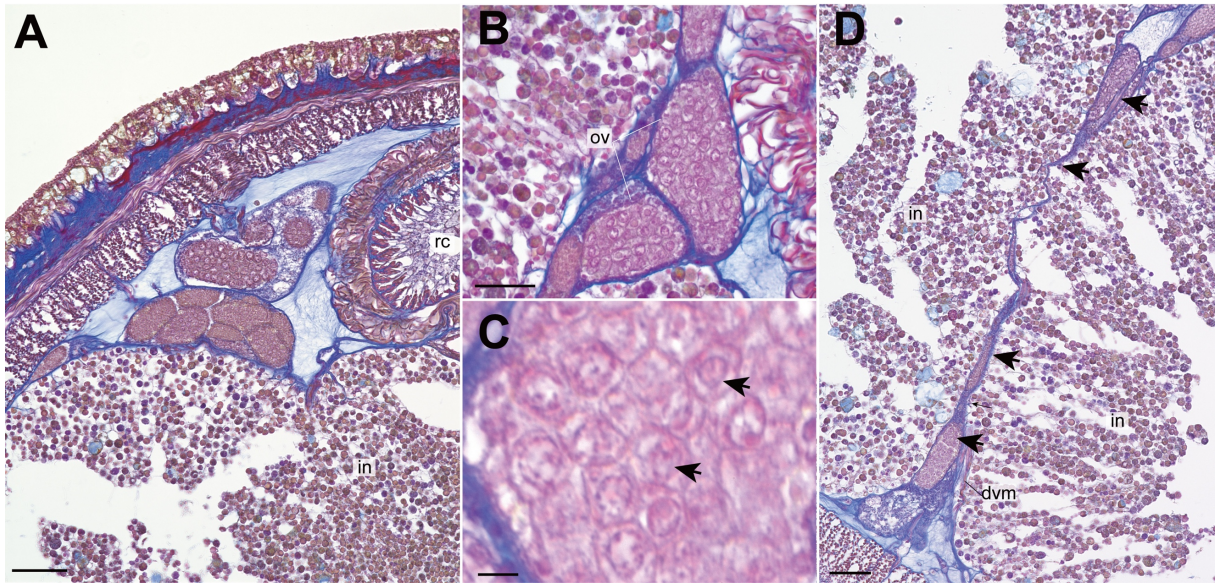


Figure 28. *Genrokunemertes obesa*, photomicrographs of transverse sections. (A) ovary-like sacs; (B) magnification of ovary-like sacs; (C) vesicles contained in each oocyte-like cell; (D) dorsoventral process (arrows) between ovary-like sacs. Abbreviations: dvm, dorsoventral muscle, intestine; ov, ovary-like sac; rc, rhyngocoel. Scale bars: 50 μm (A, D); 25 μm (B); 5 μm (C)

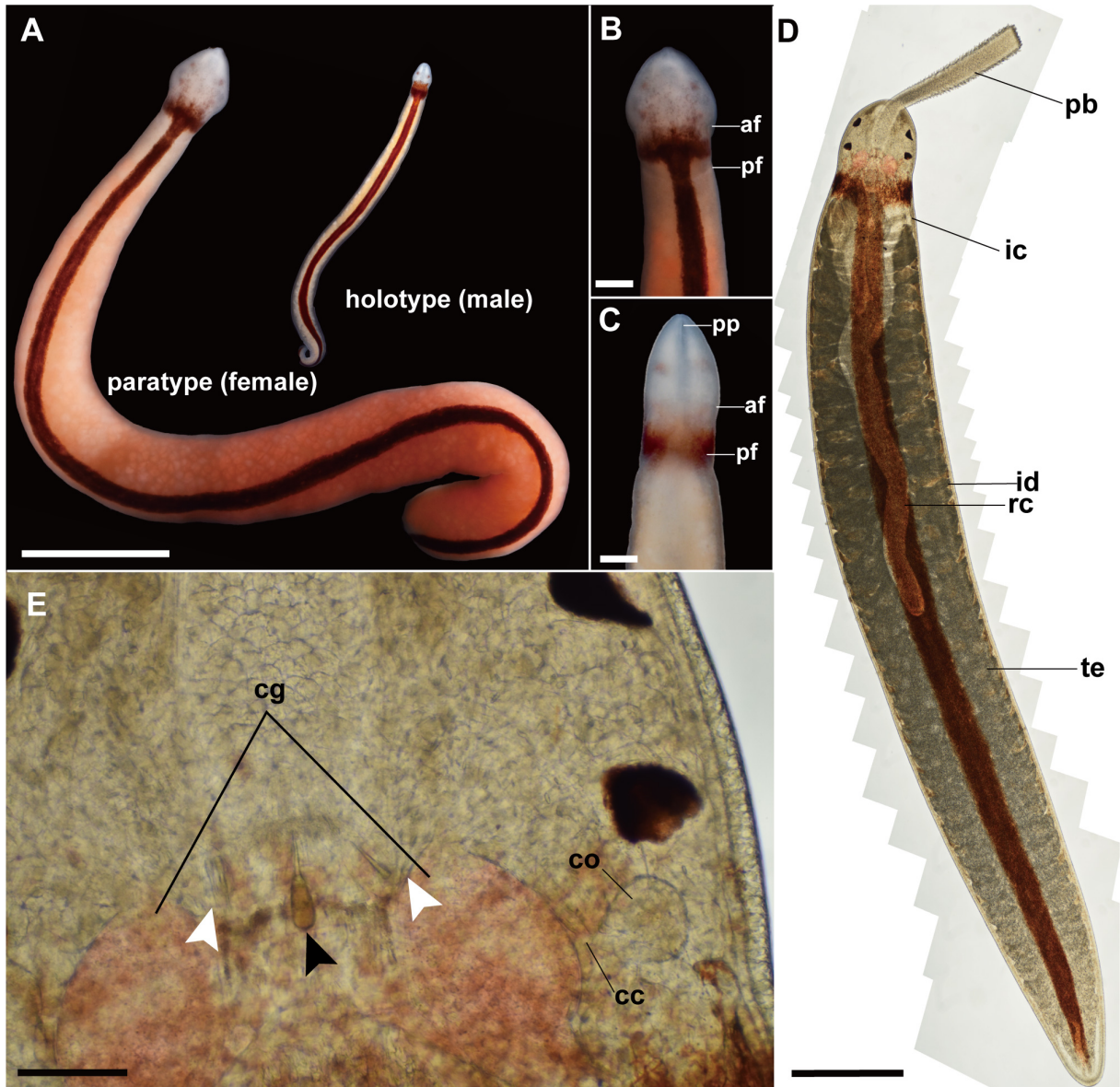


Figure 29. *Rhombonemertes rublinea*. (A) whole body, dorsal view, holotype (male) and paratype (female); (B) magnification of head, dorsal view, holotype; (C) magnification of head, ventral view, paratype; (D) Microphotograph of a living specimen, squeezed with a coverslip after anesthetization, holotype; (E) magnification of stylet apparatus in everted proboscis, a black arrowhead pointing to a central stylet and white arrowheads to accessory stylet pouches. Abbreviations: af, anterior cephalic furrows; cc, cerebral canal, cg, cerebral ganglia; co, cerebral organ; ic, intestinal caecum; id, intestinal diverticulum; pb, proboscis; pf, posterior cephalic furrows; pp, proboscis pore; rc, rhynchocoel; te, testis. Scale bars: 5 mm (A); 500 μ m (B, C); 750 μ m (D); 100 μ m (E)

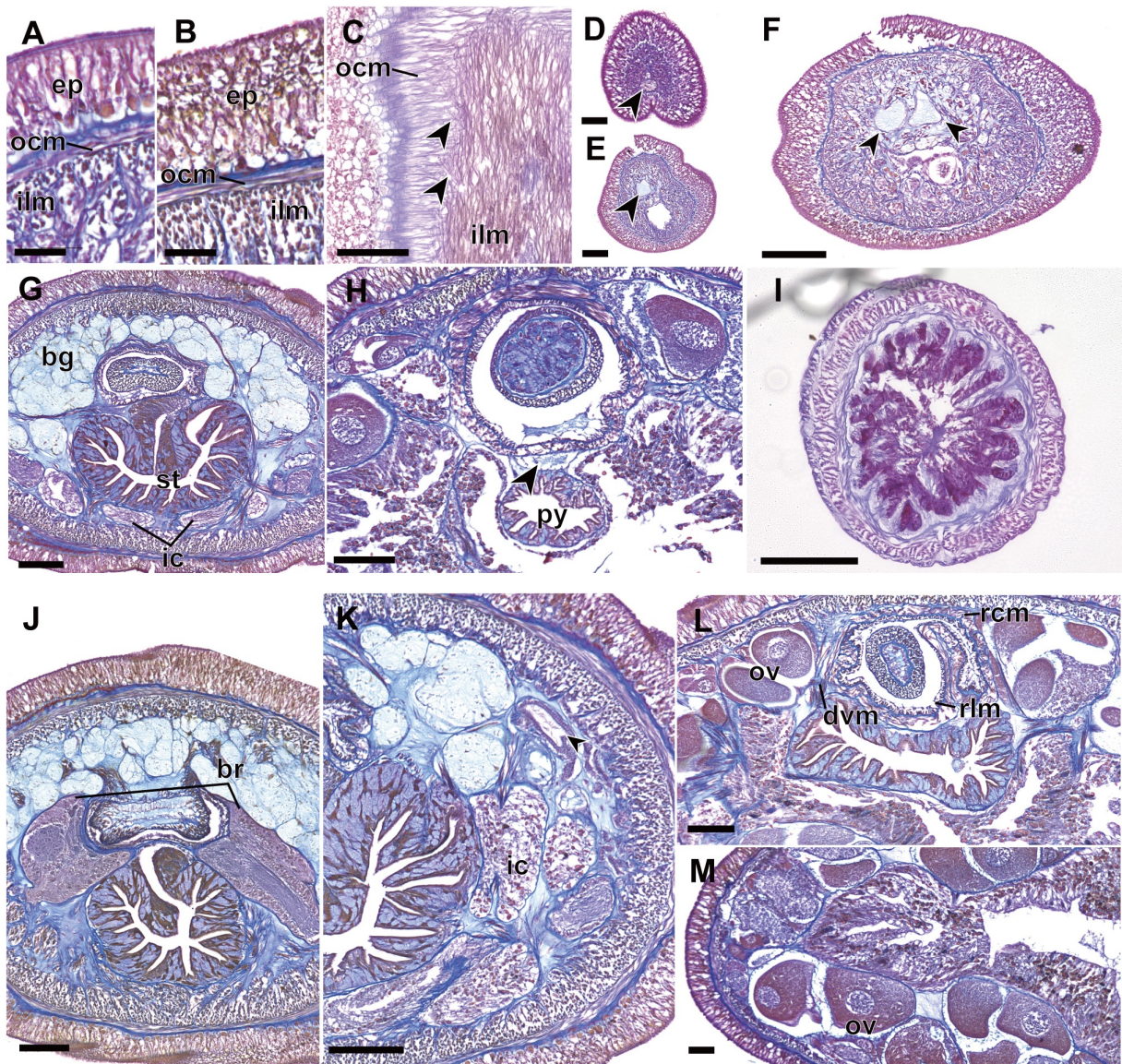


Figure 30. *Rhombonemertes rublinea*, photomicrographs of transverse (A, B, D–M) and frontal (C) sections. (A) Body wall in precerebral region; (B) Body wall in foregut region; (C) diagonal muscle fibers (arrowheads) between outer circular and inner longitudinal muscle layers of body wall, intestinal region; (D) frontal organ; (E, F) precerebral vessels (arrowheads); (G) cephalic glands; (H) mid-dorsal vessel (arrowhead); (I) proboscis, arrowheads pointing to proboscis nerves; (J) brain; (K) nephridial canals (arrowhead); (L) rhyngocoel musculature and dorsoventral muscles; (M) ovaries. Abbreviations: br, brain; co, cerebral organ; dvm, dorso-ventral muscle of body wall; ep, epidermis; ic, intestinal caecum; ilm, body-wall inner longitudinal muscle; ocm, body-wall outer circular muscle; pe, proboscis epithelium; pic, proboscis inner circular muscle; pml, proboscis middle longitudinal muscle; poc, proboscis outer circular muscle; py, pylorus, rcm, rhyngocoel circular muscle; rlm, rhyngocoel

longitudinal muscle; ov, ovary. Scale bars: 45 μm (A, B); 50 μm (C–E); 100 μm (F–K), 75 μm (L, M)

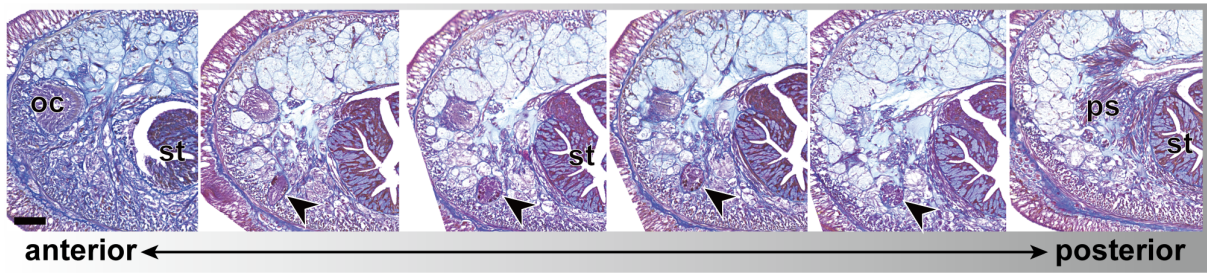


Figure 31. *Rhombonemertes rublinea*, photomicrographs of serial transverse sections, showing cerebral organ ventrolaterally opening and replaced with acidophilic glands anterior to precerebral septum. Abbreviations: oc, ocelli; ps, precerebral septum; st, stomach. Scale bar: 75 μ m

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Figure 32. A phylogenetic tree reconstructed with ML analyses. Numbers near each node indicates BS values. Solid circles represent 100% of BS.

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Figure 33. *Leptonemertes* sp. Japan, holotype specimen collected from Tsuruga (A, D) and paratype specimens collected from Shimonoseki (B, E–G) and Hagi (C). (A) holotype, whole body, dorsal; (B) paratype, whole body, dorsal; (C) paratype, whole body, dorsal; (D) magnification of head, dorsal, holotype; (E) paratype, dorsal; (F) paratype, ventral; (G) paratype, lateral; (H) a squeezed specimen, an arrowhead pointing to a gregarine parasitizing between intestinal diverticula; (I) stylet apparatus. Abbreviations: acf, anterior cephalic furrow; cg, cerebral ganglia; pb, proboscis; pp, proboscis pore. Scale bars: 5 mm (A, B); 2 mm (C–E); 1 mm (F, G); 200 μ m (H); 25 μ m (I)

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Figure 34. *Leptonemertes* sp. Japan, photomicrographs of transverse sections. (A) Body wall in precerebral region; (B) body wall in intestine region; (C) rhynchocoel wall, an arrowhead pointing to mid-dorsal vessel; (D) cerebral ganglia; (E) proboscis nerves (arrowheads); (F) vascular plugs (arrowheads); (G, H) cerebral organ; (I) lateral nerve cord; (J) dorsoventral muscles; (K) a gregarine parasitizing between intestinal diverticula (arrowhead). Abbreviations: ag, acidophilic glands; co, cerebral organ; dcg, dorsal cerebral ganglia; dvm, dorsoventral muscle; ep, epithelium; ln, lateral nerve cord; in, intestine; ocm, outer circular muscle layer of body wall; plc, proboscis middle longitudinal muscle layer; pic, proboscis inner circular muscle layer; poc, proboscis outer circular muscle layer; rcm, rhynchocoel muscle layer; vcg, ventral cerebral ganglia. Scale bars: 25 μm (A, B); 50 μm (C, D, F–I, K); 75 μm (E, J)

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Figure 35. *Geonemertes* spp. herein described. (A) *Geonemertes pelaensis*, whole body; (B) magnification of head, anterodorsal; (C) ventral; (D) squeezed specimen; (E) stylet apparatus; (F) *Geonemertes* sp. Ogasawara, specimens photographed *in-situ*; (G) whole body; (H) magnification of head, dorsal; (I) stylet apparatus. Abbreviations: cf, cephalic furrow; fo, frontal organ; pp, proboscis pore. Scale bars: 1 mm (A, G); 500 μ m (B, C, H); 100 μ m (D, E, I)

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Figure 36. *Prosadenoporus spectaculum*. (A) a specimen photographed *in-situ*; (B, C) whole body; (D) magnification of head, dorsal; (E) ventral; (F) anterolateral; (G) stylet apparatus. Abbreviations: cf, cephalic furrow; pb, proboscis; pp, proboscis pore; ps, prosorhochmid smile. Scale bars: 5 mm (A, B); 2 mm (D–F); 100 μ m (G)

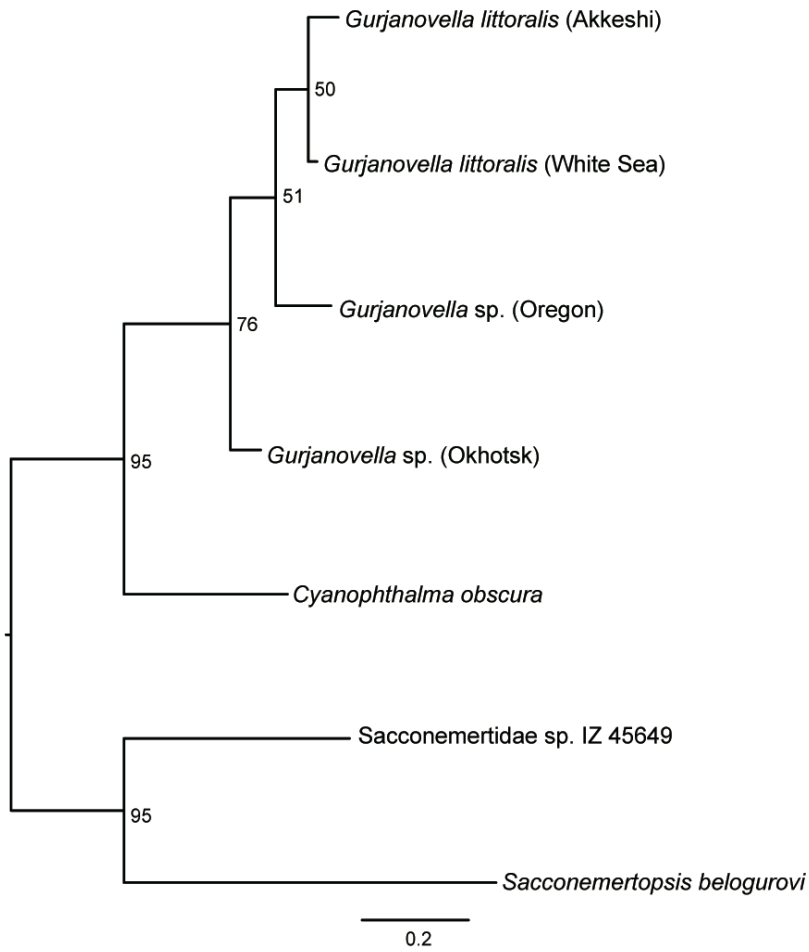


Figure 37. A phylogenetic tree reconstructed with ML analysis based on COI (658 bp) with support values generated by a separate partitioned ML bootstrap analysis based on 1000 replicates.

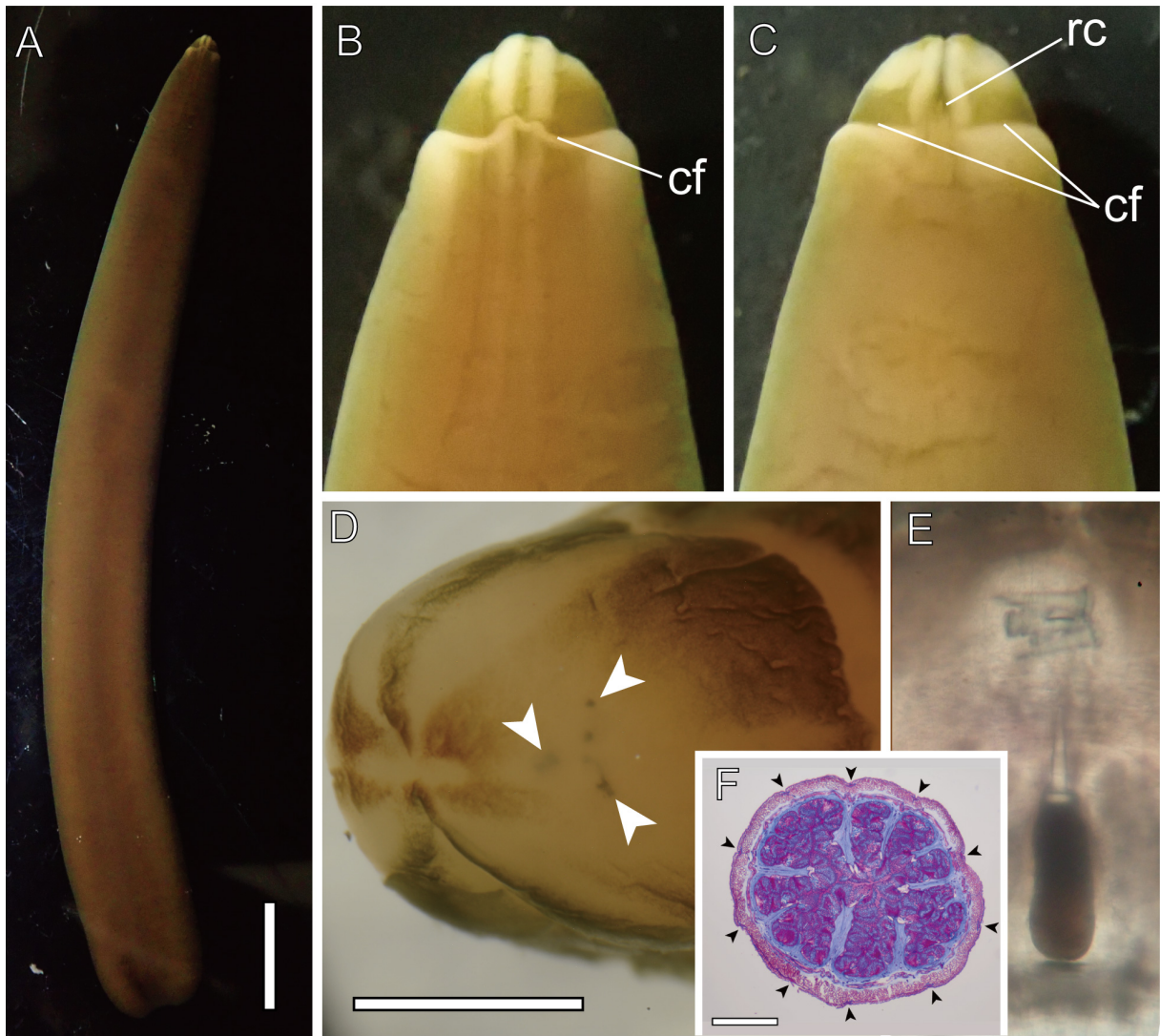


Figure 38. *Gurjanovella littoralis*. (A) whole body; (B) magnification of head, dorsal; (C) ventral; (D) cephalic tip, anterolateral view, fixed specimen, cleared in xylene; arrowheads indicating eyes (arrowheads); (E) stylet apparatus; (F) proboscis nerves. 1 cm (A); 1 mm (D); 100 μ m (F)

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Figure 39. *Potamostomma shizunaiense*. (A) immature specimen (Shizunai); (B) male specimen (Tsuruga); (C) female specimen (Tsuruga); (D) magnification of head, female (Fig. 39C), ventral; (E) squeezed specimen; (F) stylet apparatus, stylet basis slightly distorted; (G) central stylet on the basis without distorted. Abbreviations: acf, anterior cephalic furrow; cg, cerebral ganglia; ic, intestinal caecum; ov, ovary; pcf, posterior cephalic furrow; pp, proboscis pore; te, testis. Scale bars: 1 mm (A, D); 2 mm (B, C); 500 μm (E); 100 μm (F); 30 μm (G)

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Figure 40. *Sacsonemertopsis olivifera*. (A) whole body; (B) magnification of head, dorsal; (C) ventral; (D) epidermal rings (arrowheads); (E–G) squeezed specimen; (H) stylet apparatus. Abbreviations: acf, anterior cephalic furrow; pb, proboscis. Scale bars: 2 mm (A); 500 μm (B, C); 1 mm (D, E); 200 μm (F, G); 100 μm (H)

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Figure 41. *Sacsonemertidae* sp. Shari. (A) whole body; (B) magnification of head, dorsal; (C) ventral; (D) squeezed specimen, arrowheads pointing to eyes; (E) stylet apparatus. Abbreviations: cg, cerebral ganglia; pb, proboscis; pp, proboscis pore. Scale bars: 1 cm (A); 500 μ m (B, C)

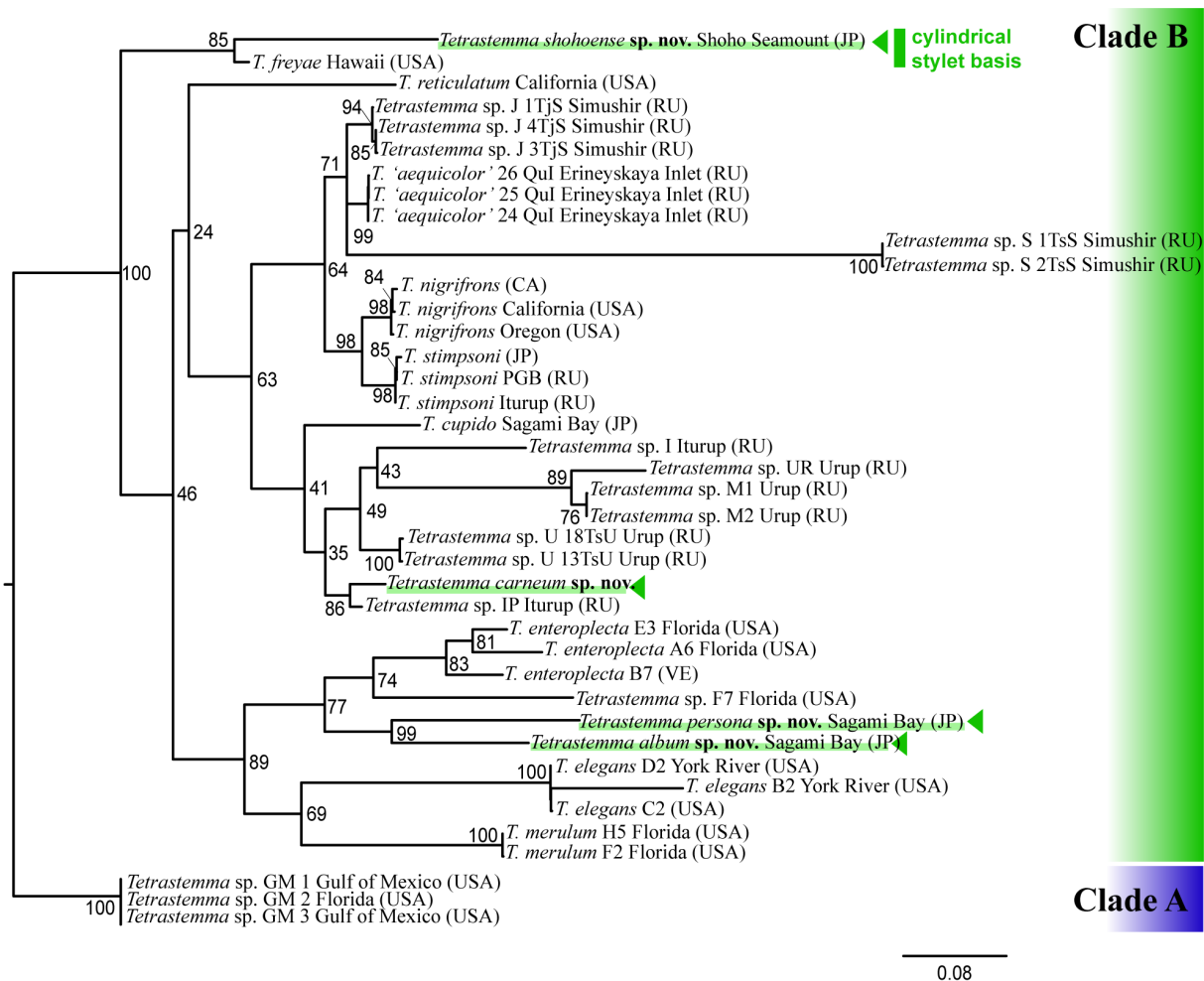


Figure 42. A phylogenetic tree reconstructed with ML analyses for *Tetrastemma*. Numbers near each node are support values generated by a separate partitioned ML bootstrap analysis with 1000 replicates. Country names of each species sampling location are abbreviated as follows: CA = Canada, JP = Japan, RU = Russia, USA = United States of America, and VE = Venezuela.

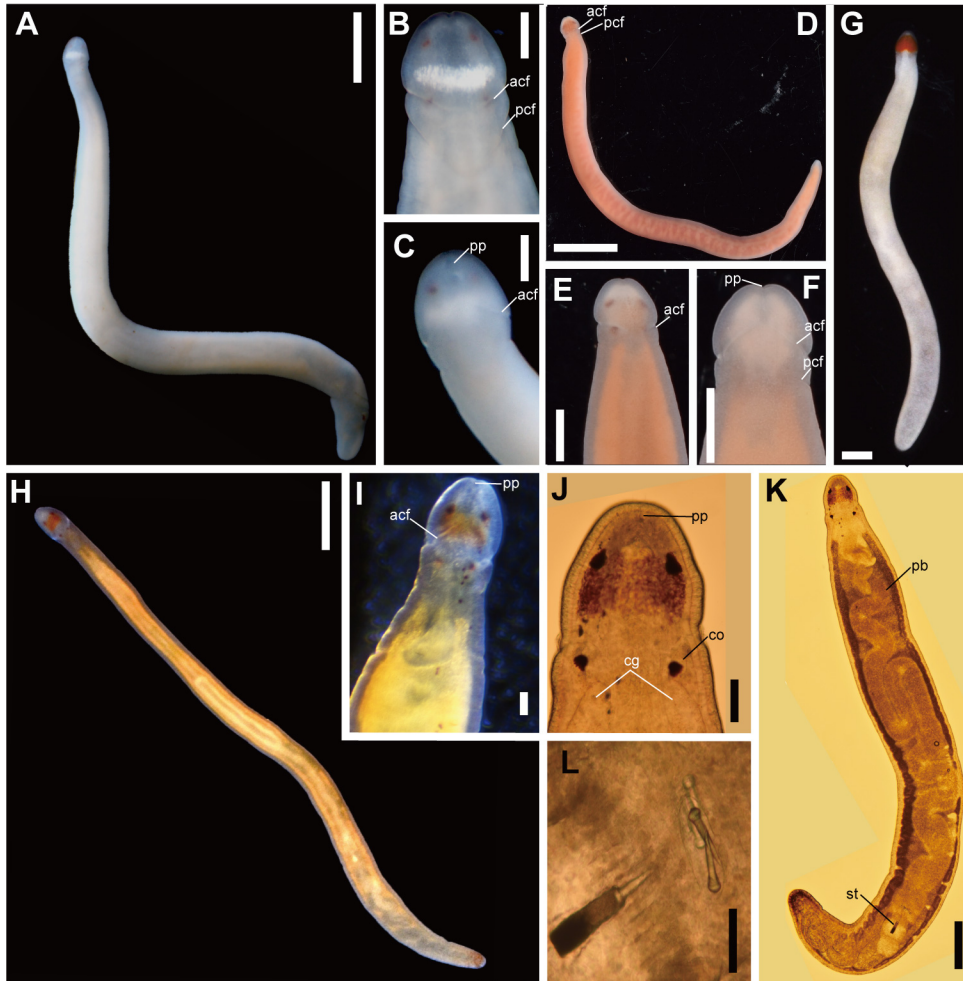


Figure 43. *Tetrastemma* spp. herein described. (A–C) *T. album* sp. nov.; (A) whole body, dorsal; (B) magnification of head, dorsal; (C) ventral; (D–F) *T. carneum* sp. nov.; (D) whole body; (E) magnification of head, dorsal; (F) ventral; (G) *T. persona* sp. nov.; whole body, dorsal; (H–L) *T. shohoense* sp. nov.; (H) whole body; (I) magnification of head, ventral; (J) squeezed specimen; (K) magnification of head; (L) stylet apparatus. Abbreviations: acf, anterior cephalic furrow; pcf, posterior cephalic furrow; cg, cerebral ganglia; co, cerebral organ; pb, proboscis; pp, proboscis pore. Scale bars: 2 mm (A, D); 500 μ m (B, C, F, G); 1 mm (E, G, H); 100 μ m (I, K); 50 μ m (L)

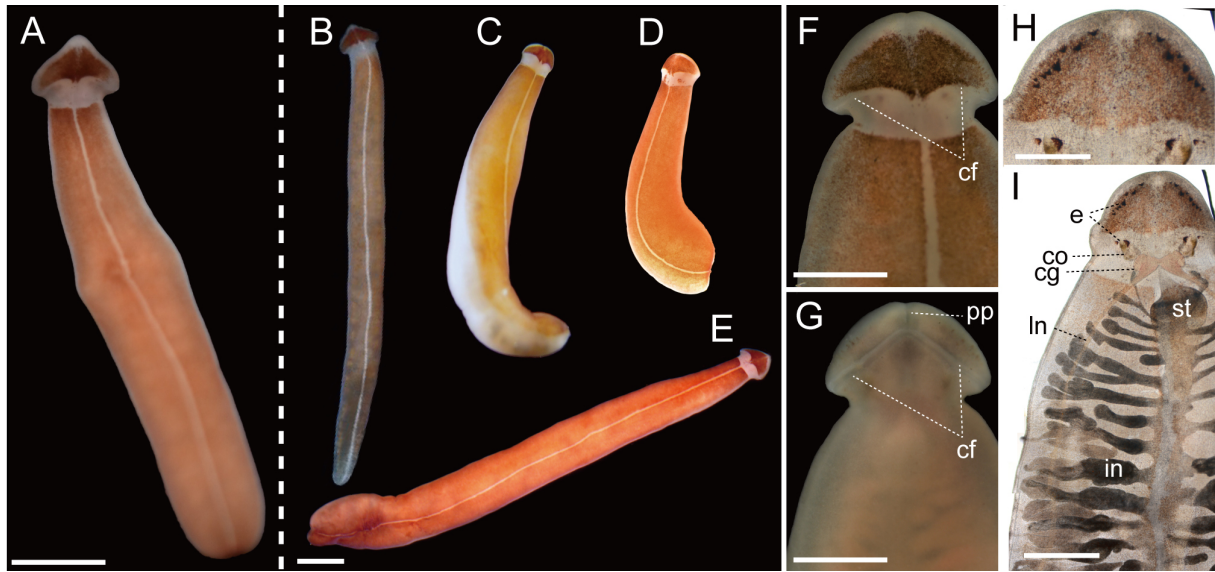


Figure 44. Photographs of *Tetrastemma cupido*. (A) holotype, ICHUM 6298; (B) MMBS-NEM 005; (C–E) paratypes. (C) ICHUM 6299; (D) ICHUM 6300; (E) ICHUM 6301; (F, G) magnification of head. (F) dorsal view; (G) ventral view; (H, I) squeezed specimen under a cover slip. (H) magnification of head; (I) anterior part of body. Abbreviations: cf, cephalic furrow; cg, cerebral ganglia; co, cerebral organ; e, eye; in, intestine; ln, lateral nerve cord; st, stomach; pp, proboscis pore. Scale bars: 1.5 mm (A–E); 500 μ m (F–I)



Figure 45. A diagram of phylogenetic relationships within Monostilifera newly recovered in the present study.

Tables

Table 1. Specimen collection information for eumonostiliferans obtained in the present study with museum depositon numbers and GenBank accession numbers.

Family	Species	Collection location	Coordinate	Location No. in Fig. 2	Depth	Collection method	Collector (s)	Collection date	Deposition number	GenBank accession number				
										COI	16S	18S	28S	H3
Amphiporidae	<i>Amphiporus</i> sp.	Saroma lake, Hokkaido, JP	–	1	–	By hand	N. Hookabe	11 Jul. 2020		In prep.	In prep.	In prep.	In prep.	In prep.
	<i>Proamphiporus kaimeiae</i>	Off Ofunato, Iwate, JP	39°06.70'N, 142°06.44'E	5	262 m	Slurp gun, ROV <i>Crambon</i> , R/V <i>Kaimei</i>	N. Hookabe	19 Jul. 2019		LC505451	LC505452	LC520132	LC505453	LC520133
Carcinonemertidae	<i>Ovicides paralithodis</i>	Off Fukui, JP	–	8	–	–	H. Motobayashi	28 Dec. 2021		OM988200	–	–	–	–
Oerstediiidae	<i>Genrokunemertes obesa</i>	Genroku Seamount, Nishishichito Ridge, JP	30°39.600N, 139°02.410E	20	2084 m	Slurp gun, ROV <i>KM-ROV</i> , R/V <i>Kaimei</i>	N. Hookabe	15 Oct. 2021		ON255709	ON244700	ON244699	ON244696	–
	<i>Rhombonemertes rublinea</i>	The Sea of Kumano, Mie, JP	34°06'49.0"N, 136°30'35.0"E	22	150–200 m	Trawling, <i>Jinsho-maru</i>	N. Jimi	10 Jan. 2022		OP34708	OP028962	OP028974	OP028967	–
	<i>Rhombonemertes rublinea</i>	The Sea of Kumano, Mie, JP	33°52'18.4"N, 136°11'54.8"E	23	150 m	Trawling, <i>Jinsho-maru</i>	N. Hookabe	7 Feb. 2022		–	–	–	–	–
Plectonemertidae	<i>Leptonemertes</i> sp.	Kinukake-nosaki, Tsuruga, Fukui, JP	35°40'3.08"N, 136°4'23.83"E	10	–	By hand	Y. Fujino	11 Jan. 2021		In prep.	In prep.	In prep.	In prep.	In prep.

	<i>Leptonemertes</i> sp.	Kinukake-no-saki, Tsuruga, Fukui, JP	35°40'3.08"N, 136°4'23.83"E	9	–	By hand	Y. Fujino, N. Hookabe, N. Jimi	25 Feb. 2021		–	–	–	–	–
	<i>Leptonemertes</i> sp.	Toyoura, Shimonoseki, Yamaguchi, JP	34°09'07.8"N, 130°54'01.8"E	26	–	By hand	M. Matsuda, Y. Fujino, N. Hookabe, N. Jimi	18 Jul. 2022		–	–	–	–	–
	<i>Leptonemertes</i> sp.	Toyokita, Shimonoseki, Yamaguchi, JP	34°19'59.2"N, 130°53'32.2"E	26	–	By hand	M. Matsuda, Y. Fujino, N. Hookabe, N. Jimi	18 Jul. 2022		–	–	–	–	–
	<i>Leptonemertes</i> sp.	Myojin-ike pond, Hagi, Yamaguchi, JP	34°26'60.0"N, 131°24'29.8"E	26	–	By hand	Y. Fujino	20 Jul. 2022		–	–	–	–	–
Prosorhochmidae	<i>Geonemertes pelaensis</i>	Yonaguni, Okinawa, JP	24°27.387N, 123°00.153E	29	–	By hand	N. Sawada	16 Apr. 2022		In prep.	In prep.	In prep.	In prep.	In prep.
	<i>Geonemertes</i> sp.	Ogasawara, Ogasawara	–	21	–	By hand	N. Hookabe	12 Dec. 2020		In prep.	In prep.	In prep.	In prep.	In prep.
	<i>Prosadenoporus spectaculum</i>	Makiya, Okinawa, JP	–	28	intertida 1	By hand	N. Hookabe	14 Mar. 2020	ICHUM831 6	In prep.	In prep.	In prep.	In prep.	In prep.
Sacconemertidae	<i>Gurjanovella littoralis</i>	Akkeshi, Hokkaido, JP	42°57.625'N, 144°50.961'E	3	25 m	Smith–McIntire grab sampler, TR/V <i>Misago-maru</i>	K. Kakui	21 Jun. 2019	ICHUM831 7	LC50545 0		–	–	–

	<i>Potamostomma shizunaiense</i>	Mouth of Shizunai River, Hokkaido, JP	–	4	–	By hand	N. Hookabe	5 Jul. 2020		In prep.	In prep.	In prep.	In prep.	In prep.
	<i>Potamostomma shizunaiense</i>	Tsuruga, Fukui, JP	–	10	–	By hand	Y. Fujino	13 May. 2020		In prep.	In prep.	In prep.	In prep.	In prep.
	<i>Potamostomma shizunaiense</i>	Mouth of Oi River, Shizuoka, JP	–	17	–	By hand	Y. Fujino	6 Mar. 2022		In prep.	In prep.	In prep.	In prep.	In prep.
	<i>Potamostomma shizunaiense</i>	Fukuoka, JP	–	27	–	By hand	H. Nagano	9 Mar. 2022		In prep.	In prep.	In prep.	In prep.	In prep.
	<i>Sacconemertopsis olivifera</i>	Lake Hinuma, Ibaraki, JP	–	11	–	By hand	N. Hookabe, N. Jimi	21 Jun. 2020		In prep.	In prep.	In prep.	In prep.	In prep.
	Sacconemertidae sp. Shari	Mouth of Shari River, Hokkaido, JP	–	2	–	By hand	H. Abe	16 Oct. 2019		In prep.	In prep.	In prep.	In prep.	In prep.
Tetrastemmatidae	<i>Tetrastemma album</i>	Off Jogashima, Kanagawa, JP	35°07.41'N, 139°34.11'E–35°07.32'N, 139°33.572'E	14	144–200 m	Biological dredge, R/V <i>Rinkai-maru</i>	N. Hookabe	12 Mar. 2021	NSMT Ne-	In prep.	In prep.	In prep.	In prep.	In prep.
	<i>Tetrastemma carneum</i>	Off Ofunato, Iwate, JP	39°3.017'N, 142°9.067'E	5	398 m	Slurp gun, ROV <i>KM-ROV</i> , R/V <i>Kaimei</i>	Y. Oya	31 Jul. 2019		In prep.	In prep.	In prep.	In prep.	In prep.
	<i>Tetrastemma carneum</i>	Off Ofunato, Iwate, JP	39°3.017'N, 142°9.067'E	5	398 m	Slurp gun, ROV <i>KM-ROV</i> , R/V <i>Kaimei</i>	Y. Oya	31 Jul. 2019		In prep.	In prep.	In prep.	In prep.	In prep.

<i>Tetrastemma cupido</i>	Off Jogashima, Kanagawa, JP	35°7.7'N, 139°34.4'E	14	97 m	Biological dredge, R/V <i>Rinkai-maru</i>	Y. Ise, N. Miyamoto	25 Oct. 2006	MMBS-Nem005	In prep.	In prep.	In prep.	In prep.	In prep.
<i>Tetrastemma cupido</i>	Off Jogashima, Kanagawa, JP	35°8.9'N, 139°34.6'E	14	90 m	Biological dredge, R/V <i>Rinkai-maru</i>	H. Kohtsuka	11 Mar. 2014	ICHUM629 8	-	-	-	-	-
<i>Tetrastemma cupido</i>	Off Jogashima, Kanagawa, JP	35°8.3'N, 139°33.7'E	14	97 m	Biological dredge, R/V <i>Rinkai-maru</i>	H. Kajihara	15 Feb. 2017	ICHUM629 9	-	-	-	-	-
<i>Tetrastemma cupido</i>	Off Jogashima, Kanagawa, JP	35°8.7'N, 139°34.4'E	14	92 m	Biological dredge, R/V <i>Rinkai-maru</i>	H. Kohtsuka	18 Apr. 2018	ICHUM630 0	-	-	-	-	-
<i>Tetrastemma cupido</i>	Off Jogashima, Kanagawa, JP	35°8.5'N, 139°32.8'E	14	112 m	Biological dredge, R/V <i>Rinkai-maru</i>	N. Hookabe	22 Feb. 2019	ICHUM630 1	OK4140 13	OK428649	OK428689	OK428648	-

<i>Tetrastemma persona</i>	Off Jogashima, Kanagawa, JP	35°08.32'N, 139°32.857'E– 35°08.40'N, 139°32.504'E	14	116– 211 m	Biological dredge, R/V <i>Rinkai- maru</i>	N. Hookabe	31 Jul. 2020	NSMT Ne-	In prep.	In prep.	In prep.	In prep.	In prep.
<i>Tetrastemma shohoense</i>	Shoho Seamount, Nishi-shichito Ridge, JO	32°19.39'N, 138°44.48'E	19	455 m	Slurp gun, ROV <i>KM- ROV</i> , R/V <i>Kaimei</i>	N. Hookabe	29 Nov. 2020	NSMT Ne-	In prep.	In prep.	In prep.	In prep.	In prep.

Table 2. Specimen collection information for *Nipponnemertes* with ICHUM numbers and Genbank accession numbers.

Species	Sample ID	Collection location	Coordinate	Location No. in Fig. 2	Depth	Collection method	Collector (s)	Collection date	Deposition number	GenBank accession number				
										COI	16S	18S	28S	H3
<i>Nipponnemertes crypta</i>	SG02	Sugashima, Mie, JP	–	24	10 m	Gill net	N. Hookabe, N. Jimi	3 Nov. 2021	ICHUM8315	ON357606	ON356061	ON356033	ON356046	ON382008
<i>Nipponnemertes ganahai</i>	JG01	Off Jogashima, Kanagawa, JP	35°08.448'N, 139°32.603'E –35°8.29N, 139°32.30'E	14	182–250 m	Biological dredge, R/V <i>Rinkai-maru</i>	H. Kajihara	12 Feb. 2015		ON357620	ON356063	–	–	ON382011
<i>Nipponnemertes ganahai</i>	JG02	Off Jogashima, Kanagawa, JP	35°08.448'N, 139°32.603'E –35°8.29N, 139°32.30'E	14	182–250 m	Biological dredge, R/V <i>Rinkai-maru</i>	H. Kajihara	12 Feb. 2015		ON357598	ON356053	ON356027	ON356038	ON382001
<i>Nipponnemertes ganahai</i>	JG03	Off Jogashima, Kanagawa, JP	35°08.448'N, 139°32.603'E –35°8.29N, 139°32.30'E	14	198–274 m	Biological dredge, R/V <i>Rinkai-maru</i>	H. Kajihara	12 Feb. 2015		ON357619	ON356062	–	ON356047	ON382010
<i>Nipponnemertes ganahai</i>	KC01	Tosa Bay, Kochi, JP	33°18.452'N, 133°35.829'E	25	100-m	Sledge, R/V <i>Toyohata-maru</i>	N. Hookabe	24 Jul. 2019		ON357615	–	–	–	–
<i>Nipponnemertes ganahai</i>	KC02	Tosa Bay, Kochi, JP	33°18.452'N, 133°35.829'E	25	100-m	Sledge, R/V <i>Toyohata-maru</i>	N. Hookabe	24 Jul. 2019		ON357616	–	–	–	–
<i>Nipponnemertes ganahai</i>	KC03	Tosa Bay, Kochi, JP	33°18.452'N, 133°35.829'E	25	100-m	Sledge, R/V <i>Toyohata-maru</i>	N. Hookabe	24 Jul. 2019		ON357617	–	–	–	–

<i>Nipponnemertes ganahai</i>	KC04	Tosa Bay, Kochi, JP	33°18.452'N, 133°35.829'E	25	100-m	Sledge, R/V <i>Toyohata- maru</i>	N. Hookabe	24 Jul. 2019		ON357 618	-	-	-	-
<i>Nipponnemertes ganahai</i>	NT01	Off Noto, Ishikawa, JP	37°18.239'N, 137°16.091'E -37°1.222'N, 137°16.166'E	7	71.2- 79.2 m	Biological dredge, R/V <i>Aosagi</i>	N. Hookabe	20 Nov. 2019	ICHUM831 6	ON357 604	ON3560567	ON356030	ON356042	ON382004
<i>Nipponnemertes ganahai</i>	JG04	Off Jogashima, Kanagawa, JP	35°08.402'N, 139°32.857'E - 139°32.504'E	14	116- 211 m	Biological dredge, R/V <i>Rinkai-maru</i>	N. Hookabe	31 Jun. 2020	ICHUM831 7	ON357 608	-	-	-	-
<i>Nipponnemertes ganahai</i>	JG05	Off Jogashima, Kanagawa, JP	35°08.402'N, 139°32.857'E - 139°32.504'E	14	116- 211 m	Biological dredge, R/V <i>Rinkai-maru</i>	N. Hookabe	31 Jun. 2020		ON357 609	-	-	-	-
<i>Nipponnemertes ganahai</i>	JG06	Off Jogashima, Kanagawa, JP	35°08.402'N, 139°32.857'E - 139°32.504'E	14	116- 211 m	Biological dredge, R/V <i>Rinkai-maru</i>	N. Hookabe	31 Jun. 2020		ON357 610	-	-	-	-
<i>Nipponnemertes ganahai</i>	CL01	Off Ofunato, Iwate, JP	39°00.726'N, 142°21.971'E	5	985 m	Slurp gun, ROV <i>Crambon, R/V Kaimei</i>	N. Hookabe	19 Jul. 2019		ON391 164	-	-	-	-
<i>Nipponnemertes ganahai</i>	JG07	Off Jogashima, Kanagawa, JP	35°7.454'N, 139°33.666'E -35°7.623'N, 139°33.100'E	14	139- 222 m	Biological dredge, R/V <i>Rinkai-maru</i>	N. Hookabe	12 Mar. 2021		ON357 607	-	-	-	-

<i>Nipponnemertes ganahai</i>	JG08	Off Jogashima, Kanagawa, JP	35°7.419'N, 139°34.112'E -35°7.325N, 139°33.572'E	14	144– 240 m	Biological dredge, R/V <i>Rinkai-maru</i>	N. Hookabe	12 Mar. 2021		ON357 611	–	–	–	–
<i>Nipponnemertes ganahai</i>	JG09	Off Jogashima, Kanagawa, JP	35°7.419'N, 139°34.112'E -35°7.325N, 139°33.572'E	14	144– 240 m	Biological dredge, R/V <i>Rinkai-maru</i>	N. Hookabe	12 Mar. 2021		ON357 612	–	–	–	–
<i>Nipponnemertes jambio</i>	JG39	Off Jogashima, Kanagawa, JP	—	14	80 m	Gill net	H. Kohtsuka	10 Apr. 2012		ON357 625	ON356067	ON356035	ON356050	ON382016
<i>Nipponnemertes jambio</i>	JG42	Off Jogashima, Kanagawa, JP	35°8.456'N, 139°34.900'E	14	83 m	Biological dredge, R/V <i>Rinkai-maru</i>	H. Kohtsuka	25 Apr. 2012	ICHUM831 8	ON357 624	–	ON356034	–	ON382015
<i>Nipponnemertes jambio</i>	JG88	Off Jogashima, Kanagawa, JP	35°8.42'N, 139°34.74'E– 35°8.22N, 139°34.67'E	14	87–89 m	Biological dredge, R/V <i>Rinkai-maru</i>	H. Kajihara	19 Feb. 2014	ICHUM831 9	ON357 597	ON356052	–	ON356037	ON382000
<i>Nipponnemertes jambio</i>	JG89	Off Jogashima, Kanagawa, JP	35°8.42'N, 139°34.74'E– 35°8.22N, 139°34.67'E	14	87–89 m	Biological dredge, R/V <i>Rinkai-maru</i>	H. Kajihara	19 Feb. 2014		ON357 623	ON356066	–	ON356049	ON382014
<i>Nipponnemertes jambio</i>	JG93	Off Jogashima, Kanagawa, JP	35°8.42'N, 139°34.74'E– 35°8.22N, 139°34.67'E	14	87–89 m	Biological dredge, R/V <i>Rinkai-maru</i>	H. Kajihara	19 Feb. 2014		ON357 622	ON356065	–	–	ON382013

<i>Nipponnemertes jambio</i>	JG94	Off Jogashima, Kanagawa, JP	35°8.42'N, 139°34.74'E–35°8.22N, 139°34.67'E	14	87–89 m	Biological dredge, R/V <i>Rinkai-maru</i>	H. Kajihara	19 Feb. 2014	ICHUM832	ON357621	ON356064	–	ON356048	ON382012
<i>Nipponnemertes kozaensis</i>	KZ	Koza, Wakayama, JP	–	24	12 m	SCUBA	N. Hasegawa	11 May. 2021	ICHUM832		ON356059	ON356032	ON356044	ON382006
<i>Nipponnemertes lactea</i>	TK	Off Shimoda, Shizuoka, JP	34°28.650'N, 139°11.380'E–34°28.559'N, 139°11.474'E	15	98–120 m	Biological dredge, R/V <i>Tsukuba II</i>	N. Hookabe	25 Mar. 2021	ICHUM832	ON357603	ON356056	ON356029	ON356041	–
<i>Nipponnemertes neonilae</i>	RU1	Vostok bay, RU	–	30	5 m	Snorkel	A.V. Chernyshev	11 Aug. 2015	MIMB43013	MN211492	MN205510	MN211395	MN211445	MN205467
<i>Nipponnemertes notoensis</i>	NT12	Off Noto, Ishikawa, JP	37°18.239'N, 137°16.091'E–37°1.222'N, 137°16.166'E	7	70–91 m	Biological dredge, R/V <i>Aosagi</i>	N. Hookabe	20 Nov. 2019	ICHUM832	ON357599	–	–	–	–
<i>Nipponnemertes notoensis</i>	NT13	Off Noto, Ishikawa, JP	37°18.239'N, 137°16.091'E–37°1.222'N, 137°16.166'E	7	70–91 m	Biological dredge, R/V <i>Aosagi</i>	N. Hookabe	20 Nov. 2019	ICHUM832	ON357600	–	–	–	–
<i>Nipponnemertes ojimaorum</i>	KG01	Koganezaki, Shizuoka, JP	–	16	12 m	SCUBA	M. Okanishi	21 Oct. 2020	ICHUM832	ON357605	ON356058	ON356031	ON356043	ON382005
<i>Nipponnemertes ojimaorum</i>	KG02	Koganezaki, Shizuoka, JP	–	16	8 m	SCUBA	N. Jimi	6 Jan. 2021	ICHUM832	–	–	–	–	–

<i>Nipponnemertes ojimaorum</i>	KG03	Koganezaki, Shizuoka, JP	–	16	7 m	SCUBA	N. Hookabe	6 Jan. 2021	ICHUM832 6	–	–	–	–	–
<i>Nipponnemertes ornata</i>	NT10	Off Noto, Ishikawa, JP	37°18.239'N, 137°16.091'E –37°1.222'N, 137°16.166'E	7	70–91 m	Biological dredge, R/V <i>Aosagi</i>	N. Hookabe	20 Nov. 2019	ICHUM832 7	ON357 602	ON356055	–	ON356040	ON382003
<i>Nipponnemertes punctatula</i>	TT	Tateyama, Chiba, JP	–	13	intertida 1	By hand	R. Yoshida	13 Jun. 2018	ICHUM832 8	ON357 613	–	–	–	–
<i>Nipponnemertes punctatula</i>	SG03	Sugashima, Mie, JP	–	18	intertida 1	By hand	N. Jimi	24 Jun. 2021	ICHUM832 9	ON357 626	–	–	–	–
<i>Nipponnemertes sugashimaensis</i>	SG01	Toba bay, Mie, JP	–	18	10–15 m	Gill net	N. Hookabe, N. Jimi	3 Nov. 2021	ICHUM833 0	–	ON356060	–	ON356045	ON382007
<i>Nipponnemertes cf. ogumai</i>	SD	Sado, Niigata, JP	–	6	12 m	SCUBA	N. Jimi	26 Sep. 2019	–	ON357 614	–	–	–	–
<i>Nipponnemertes</i> sp. 1	JG15	Off Jogashima, Kanagawa, JP	35°8.42'N, 139°34.74'E– 35°8.22'N, 139°34.67'E	14	87–89 m	Biological dredge, R/V <i>Rinkai-maru</i>	H. Kajihara	19 Feb. 2014	ICHUM833 1	ON357 589	ON356051	–	ON356036	ON382009
<i>Nipponnemertes</i> sp. 2	JG16	Off Jogashima, Kanagawa, JP	5°08.402'N, 139°32.857'– 139°32.504'E	14	116– 211 m	Biological dredge, R/V <i>Rinkai-maru</i>	N. Hookabe	31 Jun. 2020	ICHUM833 2	ON357 601	ON356054	ON356028	ON356039	ON382002

Table 3. List of primers used for the phylogenetic analyses in the present study.

Locus	Primer	Sequence (5'–3')	References
16S rRNA	ar-L	CGCCTGTTTATCAAAAACAT	Palumbi et al. (1991)
16S rRNA	br-H	CCGGTCTGAACTCAGATCACGT	Palumbi et al. (1991)
COI	LCO1490	GGTCAACAAATCATAAAGATATTGG	Folmer et al. (1994)
COI	HCO2198	TAAACTTCAGGGTGACCAAAAAATCA	Folmer et al. (1994)
18S rRNA	1F	TACCTGGTTGATCCTGCCAGTAG	Giribet et al. (1996)
18S rRNA	5R	CTTGGCAAATGCTTTCGC	Giribet et al. (1996)
18S rRNA	3F	GTTTCGATTCCGGAGAGGGA	Giribet et al. (1996)
18S rRNA	18Sbi	GAGTCTCGTTCGTTATCGGA	Whiting et al. (1997)
18S rRNA	S2.0	ATGGTTGCAAAGCTGAAAC	Whiting et al. (1997)
18S rRNA	9R	GATCCTTCCGCAGGTTACCTAC	Giribet et al. (1996)
28S rRNA	LSU5	ACCCGCTGAAYTTAAGCA	Littlewood (1994)
28S rRNA	LSU3	TCCTGAGGGAAACTTCGG	Littlewood (1994)
28S rRNA	D2f	CTTTGAAGAGAGAGTTC	Littlewood (1994)
28S rRNA	28Z	CTTGGTCCGTGTTTCAAGAC	Hillis and Dixon (1991)
28S rRNA	Sa	GACCCGTCTTGAAACACGGA	Whiting et al. (1997)
28S rRNA	rd5b	CCACAGCGCCAGTTCTGCTTAC	Schwendinger and Giribet (2005)
H3	aF	ATGGCTCGTACCAAGCAGAC	Colgan et al. (1998)
H3	aR	ATATCCTTRGGCATRATRGTGAC	Colgan et al. (1998)

Table 4. List of species included in the phylogenetic analysis, DDBJ/EMBL/GenBank accession numbers for the COI, 16S, 18S, 28S, and H3, and their references.

Infraorder	Species	COI	16S	18S	28S	H3	References
Amphiporina	<i>Amphiporus formidabilis</i>	KF935547	KF935498	KF935331	KF935387	KF935443	Kvist et al. (2014)
	<i>Amphiporus imparispinosus</i>	HQ848612	JF277618	JF293029	HQ856878	JF277696	Andrade et al. (2012)
	<i>Amphiporus lactifloreus</i>	HQ848611	JF277617	JF293018	HQ856876	—	Andrade et al. (2012)
	<i>Arctostemma articum</i>	MZ231216	MZ231150	MZ231306	MZ216538	MZ216608	Chernyshev et al. (2021)
	<i>Carcinonemertes carcinophila</i>	HQ848619	JF277603	JF293007	HQ856893	JF277693	Andrade et al. (2012)
	<i>Carcinonemertes errans</i>	KU197594	KU197258	—	—	—	Hiebert and Maslakova (unpublished)
	<i>Cyanophthalma obscura</i>	EF208980	—	AY039667	—	—	Sundberg et al. (2001, 2007)
	<i>Emplectonemma gracile</i>	HQ848620	JF277621	JF293022	HQ856883	JF277751	Andrade et al. (2012)
	<i>Emplectonemma</i> sp. IZ-135333	KF935546	KF935497	KF935330	KF935386	KF935442	Kvist et al. (2014)
	<i>Geonemertes pelaensis</i>	HQ848593	JF277611	JF304779	HQ856888	JF277736	Andrade et al. (2012)
	<i>Geonemertes</i> sp. IZ-132534	KF935548	KF935499	KF935333	KF935389	KF935445	Kvist et al. (2014)
	<i>Gurjanovella littoralis</i>	AJ436904	AJ436794	—	AJ436849	JF277737	Thollesson and Norenburg (2003)
	<i>Gurjanovella</i> sp. 11DS	MF512123	MF512082	MF512082	MF512107	MF512149	Chernyshev et al. (2021)
	<i>Malacobdella japonica</i>	MZ216520	—	MZ231198	MZ231288	MZ216591	Chernyshev et al. (2021)

<i>Malacobdella grossa</i>	HQ848591	JF277614	JF293015	HQ856882	JF277700	Andrade et al. (2012)
<i>Neesia buergeri</i>	HQ848600	JF277616	JF293066	HQ856883	JF277751	Chernyshev et al. (2021)
<i>Nemertovema hadalis</i>	KY296912	KY296891	KY296895	KY296903	KY296921	Chernyshev and Polyakova (2018b)
<i>Nemertovema norenburgi</i>	MN205524	MN211507	MN211413	MN211463	MN205485	Chernyshev and Polyakova (2019)
<i>Ototyphlonemertes correae</i>	HQ848613	JF277612	JF293025	HQ856884	JF277706	Andrade et al. (2012)
<i>Ototyphlonemertes macintoshi</i>	HQ848605	JF277613	JF293024	HQ856886	JF277707	Andrade et al. (2012)
<i>Paranemertes peregrina</i>	MN205526	MN211509	MN211415	MN211465	—	Chernyshev et al. (2021)
<i>Poseidonemertes maslakovae</i>	KP270879	KP270856	KP270806	KP270831	—	Kvist et al. (2015)
<i>Proamphiporus crandalli</i>	MN205524	MN211505	MN211413	MN211463		Chernyshev and Polyakova (2019)
<i>Prosadenoporus californiensis</i>	EF157597	EF157585	—	—	—	Maslakova and Norenburg (2008)
<i>Prosadenoporus floridensis</i>	EF157596	EF157584	—	—	—	Maslakova and Norenburg (2008)
<i>Prosadenoporus winsori</i>	EF157594	EF157581	—	—	—	Maslakova and Norenburg (2008)
<i>Prosadenoporus</i> sp. 115	AJ436914	AJ436804	—	AJ436859	AJ436962	Thollesson and Norenburg (2003)
<i>Prostoma</i> cf. <i>eilhardi</i>	HQ848594	JF277620	JF293027	HQ856875	JF277695	Andrade et al. (2012)
<i>Sacconemertopsis belogurovi</i>	KP270884	KP270861	KP270811	KP270836	MN205479	Kvist et al. (2015); Chernyshev and Polyakova (2019)
<i>Sacconemertidae</i> sp. IZ-45649	KP270883	KP270860	KP270810	KP270835	—	Kvist et al. (2015)

	<i>Tetrastemma candidum</i>	OK414012	—	AY928357	AB505827	—	Strand and Sundberg (2005a); Kajihara et al. (2011); Hookabe et al. (2021)
	<i>Tetrastemma elongatum</i>	HQ848609	JF277622	JF293026	HQ856874	JF277702	Andrade et al. (2012)
	<i>Tetrastemma nigrifrons</i>	MZ216531	MZ231144	MZ231209	MZ231299	MZ216601	Chenyshev et al. (2021)
	<i>Tetrastemma olgarum</i>	KJ717948 ^e	KJ717948 ^e	—	—	—	Sun et al. (2016)
	<i>Tetrastemma stimpsoni</i>	KP270885	KP270862	KP270812	KP270837	MZ231213	Kvist et al. (2015); Chernyshev et al. (2021)
	<i>Tortus tokmakovae</i>	MN205527	MN211510	MN211416	MN211466	MN205487	Chenyshev et al. (2021)
	<i>Tortus</i> sp. IZ-45645	KP270881	KP270858	KP270808	KP270833	—	Kvist et al. (2015)
	<i>Zygonemertes virescense</i>	HQ848590	JF277615	JF293016	HQ856885	JF277694	Andrade et al. (2012)
	<i>Zygonemertes</i> sp. Russia	—	MN211503	MN211409	MN211459	MN205480	Chernyshev and Polyakova (2019)
Oerstediiina	<i>Abyssonemertes kajiharai</i>	KY296906	KY296888	KY296892	KY296897	KY296915	Chernyshev and Polyakova (2018a)
	<i>Antarctonemertes riesgoae</i>	KF935538	KF935490	KF935322	KF935378	KF935434	Kvist et al. (2014)
	<i>Antarctonemertes unilineata</i>	MG948790	MG920846	—	MG920847	—	Taboada et al. (2018)
	<i>Antarctonemertes valida</i>	KF935537	KF935489	KF935321	KF935377	KF935433	Kvist et al. (2014)
	<i>Antarctonemertes</i> sp. Urup48	OM456697	—	OM423116	OM423054	OM468148	Chernyshev and Polyakova (2022)
	<i>Antiponemertes novazealandiae</i>	—	—	AY928345	—	—	Strand and Sundberg (2005b)

<i>Argonemertes australiensis</i>	HQ848601	JF277605	JF293010	HQ856892	JF277750	Andrade et al. (2012)
<i>Asteronemertes cf. gibsoni</i>	MN205519	MN211500	MN211406	MN211456	MN205477	Chernyshev and Polyakova (2019)
<i>Diplomma bothwellae</i>	MZ216517	MZ231133	MZ231195	MZ231285	MZ216588	Chernyshev et al. (2021)
<i>Diplomma serpentina</i>	MN205514	—	MN211400	MN211450	MN205471	Chernyshev and Polyakova (2019)
Eumonostilifera sp. Vema2B	KY296910	—	KY296894	KY296901	KY296919	Chernyshe and Polyakova (2018a)
<i>Galathenemertes giribeti</i>	MN205516	MN211497	MN211402	MN211452	MN205473	Chernyshev and Polyakova (2019)
<i>Gononemertes parasita</i>	AB505821	JF277606	JF304779	HQ856889	JF277745	Andrade et al. (2012); Kajihara et al. (2011)
<i>Kurilonemertes dilutebasisae</i>	MN205511	MN211494	MN211397	MN211447	—	Chernyshev and Polyakova (2019)
<i>Kurilonemertes papilliformis</i>	MZ216515	MZ231131	MZ231194	MZ231283	—	Chernyshev et al. (2021)
<i>Kurilonemertes phyllospadicola</i>	FJ594418*	MN211493	MN211396	MN211446	MN205468	*Maslakova and von Döhren (2009); Chernyshev et al. (2021)
<i>Leptonemertes cf. chalicophora</i>	HQ848596	JF277608	JF293011	HQ856898		Kvist et al. (2014)
Monostilifera sp. 9 Iturup	MZ216521	MZ231136	MZ231199	MZ231289	MZ216592	Chernyshev et al. (2021)
Monostilifera sp. KuramBio2 82	—	—	MN211405	MN211455	MN205476	Chernyshev and Polyakova (2019)
<i>Nemertellina yamaokai</i>	AJ436907	AJ436797	AB505826	AJ436852	AJ436959	Thollesson and Norenburg (2003)
<i>Oerstedia dorsalis</i>	MZ216523	—	MZ231201	MZ231291	—	Chernyshev et al. (2021)

<i>Oerstedia oculata</i>	MN205512	MN211495	MN211398	MN211448	MN205469	Chernyshev and Polyakova (2019)
<i>Oerstedia phoresiae</i>	MN205513	MN211496	MN211399	MN211449	MN205470	Chernyshev and Polyakova (2019)
<i>Oerstedia polyorbis</i>	MZ216524	MZ231137	MZ231202	MZ231292	MZ216594	Chernyshev et al. (2021)
<i>Pseudotetrastemma</i> sp. KB2hop25	MN205520	MN211501	MN211407	MN211457	—	Chernyshev et al. (2021)
Plectonemertidae sp. IZ-25173	KF935529	KF935480	KF935313	KF935369	<i>KF935423</i>	Kvist et al. (2014)
Plectonemertidae sp. IZ-25174	KF935530	KF935481	KF935314	KF935370	<i>KF935424</i>	Kvist et al. (2014)
Plectonemertidae sp. IZ-25175	KF935531	KF935482	KF935315	KF935371	<i>KF935425</i>	Kvist et al. (2014)
Plectonemertidae sp. IZ-25169	KF935528	KF935479	KF935312	KF935368	<i>KF935422</i>	Kvist et al. (2014)
Plectonemertidae sp. IZ-25166	KF935526	KF935477	KF935310	KF935366	<i>KF935421</i>	Kvist et al. (2014)
Plectonemertidae sp. IZ-25167	KF935527	KF935478	KF935311	KF935367	—	Kvist et al. (2014)
Plectonemertidae sp. NT000046	EU255614	—	EU255585	—	—	Mateos and Giribet (2008)
Plectonemertidae sp. NT000072	EU255626	KF935486	EU255596	—	—	Mateos and Giribet (2008)
<i>cf. Potamonemertes percivali</i>	KF935532	KF935483	KF935316	KF935372	—	Kvist et al. (2014)
<i>Tetranemertes antonina</i>	KF935534	—	KF935318	KF935374	KF935430	Kvist et al. (2014)
<i>Tetrastemma bilineatum</i>	MZ216539	MZ231151	MZ231217	MZ231307	MZ216609	Chernyshev et al. (2021)
<i>Tetrastemma wilsoni</i>	AJ436921	AJ436811	—	AJ436866	—	Thollessen and Norenburg (2003)
<i>Tetrastemma</i> sp. IR Iturup	MZ216568	MZ231181	MZ231254	MZ231344	MZ216645	Chernyshev et al. (2021)

	<i>Tetrastemma</i> sp. IT Iturup	MZ216569	—	MZ231255	MZ231345	MZ216646	Chernyshev et al. (2021)
	<i>Pseudotetrastemma</i> sp. KB2hop25	MN205520	MN211501	MN211407	MN211457	—	Chernyshev et al. (2021)
	<i>Tetrastemma vittigerum</i>	MZ216585	MZ231192	MZ231272	MZ231362	MZ216663	Chernyshev et al. (2021)
	<i>Vieitezia luzmurubeae</i>	HQ443426	JF277607	HQ443428	HQ856890	JF277746	Andrade et al. (2012)
Outgroup	<i>Nipponnemertes ogumai</i>	AB920907	AB921008	AB921009	AB921010	—	Kajihara et al. (2015)
	<i>Nipponnemertes pulchra</i>	HQ848597	JF277625	JF293012	HQ856871	JF277704	Andrade et al. (2012)

*Registered as *Psammamphiporus elongatus* in DDBJ/EMBL/GenBank

Table 5. GenBank accession numbers of eumonostiliferans except species herein described, which are used in the molecular phylogenetic analyses.

Species	16S	COI	18S	28S	H3	References
<i>Nipponnemertes arenaria</i>	MN211491	MN205509	MN211394	MN211444	MN205466	Chernyshev and Polyakova (2019)
<i>Nipponnemertes bimaculata</i>	AJ436799	AJ436909	—	AJ436854	AJ436961	Thollesson and Norenburg (2003)
<i>Nipponnemertes bimaculata</i> E3F9	KU197269	KU197602	—	—	—	Hiebert and Maslakova (unpublished)
<i>Nipponnemertes bimaculata</i> E3G1	KU197270	KU197603	—	—	—	Hiebert and Maslakova (unpublished)
<i>Nipponnemertes bimaculata</i> E3G2	KU197271	KU197604	—	—	—	Hiebert and Maslakova (unpublished)
<i>Nipponnemertes bimaculata</i> E3G3	—	KU197605	—	—	—	Hiebert and Maslakova (unpublished)
<i>Nipponnemertes incainca</i>	—	KX879856	—	—	—	Gonzalez-Cueto et al. (2017)
<i>Nipponnemertes ogumai</i>	AB921008	AB920927	AB921009	AB921010	—	Kajihara et al. (2015)

<i>Nipponnemertes pulchra</i>	JF277625	HQ848597	JF293012	HQ856871	JF277704	Andrade et al. (2012)
<i>Nipponnemertes pulchra</i> NemBar0127	—	KU839800	—	—	—	Sundberg et al. (2016)
<i>Nipponnemertes pulchra</i> NemBar0132	—	KU839804	—	—	—	Sundberg et al. (2016)
<i>Nipponnemertes pulchra</i> NemBar0133	—	KU839805	—	—	—	Sundberg et al. (2016)
<i>Nipponnemertes pulchra</i> NemBar0134	—	KU839806	—	—	—	Sundberg et al. (2016)
<i>Nipponnemertes pulchra</i> NemBar0383	—	KU839981	—	—	—	Sundberg et al. (2016)
<i>Nipponnemertes pulchra</i> NemBar1161	—	KP697764	—	—	—	Sundberg et al. (2016)
<i>Nipponnemertes pulchra</i> NemBar1162	—	KP697765	—	—	—	Sundberg et al. (2016)
<i>Nipponnemertes pulchra</i> NemBar1163	—	KP697767	—	—	—	Sundberg et al. (2016)

<i>Nipponnemertes pulchra</i> NemBar1164	—	KP697761	—	—	—	Sundberg et al. (2016)
<i>Nipponnemertes pulchra</i> NemBar1165	—	KP697763	—	—	—	Sundberg et al. (2016)
<i>Nipponnemertes pulchra</i> NemBar1449	—	KP697762	—	—	—	Sundberg et al. (2016)
<i>Nipponnemertes pulchra</i> NemBar1287	—	KP697766	—	—	—	Sundberg et al. (2016)
<i>Nipponnemertes punctatula</i>	KU230292	KU230295	KU230293	KU230294	—	Leasi et al. (2016)
<i>Nipponnemertes</i> sp. DNA105622	JF277624	HQ848598	JF293020	HQ856872	JF277703	Andrade et al. (2012)
<i>Nipponnemertes</i> sp. DNA105589	JF277623	HQ848599	JF293019	HQ856870	JF277705	Andrade et al. (2012)
Cratenemertidae sp. Guam	MN211490	MN205508	MN211393	MN211443	MN205465	Chernyshev and Polyakova (2019)
<hr/>						
Outgroup						
Cratenemertidae sp. IZ-45644	KP270854	KP270877	KP270804	KP270829	—	Kvist et al. (2015)

<i>Cratenemertea</i> sp. 25DS	MF512058	MF512127	MF512086	MF512111	—	Chernyshev and Polyakova (2018)
<i>Malacobdella</i> <i>grossa</i>	HQ848591	HQ848591	JF293015	HQ856882	JF277700	Andrade et al. (2012)
<i>Nemertovema</i> <i>hadaris</i>	KY296891	KY296912	KY296895	KY296903	KY296921	Chernyshev and Polyakova (2018b)
<i>Oerstedia</i> <i>oculata</i>	MN211495	MN205512	MN211398	MN211448	MN205469	Chernyshev and Polyakova (2019)
<i>Oerstedia</i> <i>phoresiae</i>	MN211496	MN205513	MN211399	MN211449	MN205470	Chernyshev and Polyakova (2019)
<i>Proamphiporus</i> <i>kaimeiae</i>	LC505452	LC505451	LC520132	LC505453	LC520133	Hookabe et al. (2020)
<i>Tetrastemma</i> <i>cupido</i>	OK428649	OK414013	OK428689	OK428648	—	Hookabe et al. (2021)
<i>Uniporus</i> <i>alisae</i>	MF512053	MF512121	MF512080	MF512105	MF512147	Chernyshev and Polyakova (2018a)

Table 6. Inter- and intraspecific genetic distance (%) among species in Clade A based on uncorrected *p*-distance.

	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	
1. <i>N. ogumai</i>	—																		
2. <i>Nipponnemertes</i> cf. <i>ogumai</i> (Sado)	7.3	—																	
3. <i>N. pulchra</i> (Sweden)	14.6	13.4	—																
4. <i>N. pulchra</i> NemBar0127 (Sweden)	13.2	13.2	0.0	—															
5. <i>N. pulchra</i> NemBar0132 (Sweden)	13.2	13.2	0.0	0.0	—														
6. <i>N. pulchra</i> NemBar0133 (Sweden)	13.5	13.5	0.2	0.2	0.2	—													
7. <i>N. pulchra</i> NemBar0134 (Sweden)	13.2	13.2	0.0	0.0	0.0	0.2	—												
8. <i>N. pulchra</i> NemBar0383 (Sweden)	13.0	13.5	0.2	0.2	0.2	0.4	0.2	—											
9. <i>N. pulchra</i> NemBar1161 (Sweden)	13.9	13.4	0.2	0.2	0.2	0.4	0.2	0.4	—										
10. <i>N. pulchra</i> NemBar1162 (Sweden)	14.1	13.0	0.0	0.0	0.0	0.2	0.0	0.2	0.2	—									
11. <i>N. pulchra</i> NemBar1163 (Sweden)	13.8	13.3	0.0	0.0	0.0	0.2	0.0	0.2	0.2	0.0	—								
12. <i>N. pulchra</i> NemBar1164 (Sweden)	14.3	13.0	0.2	0.2	0.2	0.4	0.2	0.4	0.3	0.2	0.2	—							
13. <i>N. pulchra</i> NemBar1165 (Sweden)	14.1	13.0	0.0	0.0	0.0	0.2	0.0	0.2	0.2	0.0	0.0	0.2	—						
14. <i>N. pulchra</i> NemBar1187 (Sweden)	13.9	13.4	0.0	0.0	0.0	0.2	0.0	0.2	0.3	0.2	0.2	0.3	0.2	—					
15. <i>N. pulchra</i> NemBar1449 (Sweden)	14.1	13.0	0.0	0.0	0.0	0.2	0.0	0.2	0.2	0.0	0.0	0.2	0.0	0.2	—				
16. <i>Nipponnemertes</i> sp. DNA105589	13.1	14.7	10.3	10.6	10.6	10.9	10.6	10.6	10.4	10.3	10.3	10.5	10.3	10.2	10.3	—			
17. <i>Nipponnemertes</i> sp. DNA105622	12.7	12.2	4.4	4.1	4.1	4.4	4.1	4.4	4.6	4.2	4.4	4.4	4.2	4.4	4.2	11.0	—		
18. <i>Nipponnemertes</i> sp. 1	13.1	12.1	4.7	4.6	4.6	4.8	4.6	4.8	4.8	4.5	4.6	4.7	4.5	4.8	4.5	12.0	3.4	—	

Table 7. Inter- and intraspecific genetic distance (%) among species in Clade B based on uncorrected *p*-distance.

	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18
1. <i>N. arenaria</i>	—																	
2. <i>N. bimaculata</i> (San Juan Island)	10.2	—																
3. <i>N. bimaculata</i> E3F9 (Oregon)	10.0	0.5	—															
4. <i>N. bimaculata</i> E3G1 (Oregon)	10.1	0.3	0.2	—														
5. <i>N. bimaculata</i> E3G2 (Oregon)	10.1	0.3	0.2	0.0	—													
6. <i>N. bimaculata</i> E3G3 (Oregon)	10.0	0.3	0.2	0.0	0.0	—												
7. <i>N. neonilae</i>	10.0	9.6	9.4	9.3	9.3	9.3	—											
8. <i>N. crypta</i>	10.2	6.5	6.0	6.0	6.0	6.0	9.6	—										
9. <i>N. incainca</i>	10.7	12.5	12.7	12.6	12.6	12.6	11.9	12.0	—									
10. <i>N. jambio</i> . JG39	11.0	7.8	7.3	7.3	7.3	7.3	9.4	2.3	13.0	—								
11. <i>N. jambio</i> . JG42	10.8	7.9	7.5	7.4	7.4	7.4	9.6	2.4	13.2	0.2	—							
12. <i>N. jambio</i> . JG89	10.8	7.6	7.2	7.1	7.1	7.1	9.3	2.1	12.8	0.2	0.3	—						
13. <i>N. jambio</i> . JG93	11.0	7.9	7.5	7.4	7.4	7.4	9.3	2.4	13.2	0.2	0.3	0.3	—					
14. <i>N. jambio</i> . JG94	11.0	7.8	7.3	7.3	7.3	7.3	9.4	2.3	13.0	0.0	0.2	0.2	0.2	—				
15. <i>N. ojimaourum</i> .	11.0	7.1	6.8	6.8	6.8	6.8	9.4	1.6	11.7	2.7	2.9	2.5	2.9	2.7	—			
16. <i>N. punctatula</i> (Oshoro)	11.0	7.6	7.5	7.4	7.4	7.4	9.6	2.7	13.3	4.3	4.4	4.1	4.1	4.3	3.5	—		
17. <i>N. punctatula</i> (Sugashima)	10.8	7.9	7.6	7.6	7.6	7.6	9.7	2.9	13.5	4.4	4.3	4.3	4.3	4.4	3.6	0.2	—	
18. <i>N. punctatula</i> (Tateyama)	10.8	7.9	7.6	7.6	7.6	7.6	9.7	2.9	13.5	4.4	4.3	4.3	4.3	4.4	3.6	0.2	0.0	—

Table 8. Inter- and intraspecific genetic distance (%) among species in Clade C based on uncorrected *p*-distance.

	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16
1. <i>N. ganahai</i> CL01	—															
2. <i>N. ganahai</i> JG01	0.0	—														
3. <i>N. ganahai</i> JG02	0.2	0.3	—													
4. <i>N. ganahai</i> JG03	0.2	0.2	0.2	—												
5. <i>N. ganahai</i> JG07	0.0	0.0	0.2	0.2	—											
6. <i>N. ganahai</i> JG08	0.2	0.2	0.0	0.0	0.2	—										
7. <i>N. ganahai</i> JG09	0.2	0.2	0.0	0.0	0.2	0.0	—									
8. <i>N. ganahai</i> JG04	0.7	0.7	0.8	0.8	0.7	0.8	0.8	—								
9. <i>N. ganahai</i> JG05	0.0	0.2	0.5	0.3	0.0	0.2	0.2	0.7	—							
10. <i>N. ganahai</i> KC01	0.0	0.0	0.3	0.2	0.0	0.2	0.2	0.7	0.2	—						
11. <i>N. ganahai</i> KC02	0.0	0.0	0.2	0.2	0.0	0.2	0.2	0.7	0.0	0.0	—					
12. <i>N. ganahai</i> KC03	0.0	0.0	0.3	0.2	0.0	0.2	0.2	0.7	0.2	0.0	0.0	—				
13. <i>N. lactea</i>	12.5	12.5	12.3	12.3	12.5	12.3	12.3	12.4	12.5	12.5	12.5	12.5	—			
14. <i>N. notoensis</i>	12.6	12.6	12.3	12.3	12.6	12.3	12.3	12.6	12.6	12.6	12.6	12.6	13.4	—		
15. <i>N. ornata</i>	12.3	11.9	11.6	11.7	12.2	12.1	12.0	12.5	12.1	11.9	12.3	11.9	13.1	9.0	—	
16. Cratenemertidae sp. Guam	13.7	13.4	13.1	13.2	13.8	13.7	13.6	14.0	13.6	13.4	13.7	13.4	14.3	11.5	10.0	—

Table 9. An updated species list in *Nipponnemertes* and *Cratenemertes amboiensis* with selected morphological characters and ecological information.

Species	Type locality	Distribution	Depth	Habitat	External Coloration	Cephalic patches	Mid-dorsal cephalic ridge	Numbers of proboscis nerves	Cerebral organs	References
<i>Nipponnemertes africana</i> (Wheeler, 1940)	Shelly Beach (South Africa)	Algoa Bay, Shelly Beach, Saldanha Bay (South Africa)	Intertidal	Among algae <i>Hypnea specifica</i>	Color white, pink, pinkish yellow or buff, becoming lighter anteriorly and deeper posteriorly	Absent	Present	11	Reaching behind the brain	Wheeler (1940); Berg (1985); McDermott (1998)
<i>Nipponnemertes arenaria</i> (Uschakov, 1927)	Ussuri Bay (Russia)	Peter the Great Bay (Russia)	15–62 m	Subtidally on sandy bottom	Dorsally brown with white mottling (Fig. 4A); lateral and ventral margin paler	Present	Present	14–15	–	Uschakov (1927); Chernyshev (1993)
<i>Nipponnemertes bimaculata</i> (Coe, 1901)	Not designated; originally reported from Victoria, British Columbia (Canada) and Puget Sound (USA)	Pacific coasts of North America; specimens identified as this species reported from eastern Russia and Japan likely to be <i>N. neonilae</i> ; as far south as Ensenada in Mexico but the species identity uncertain	Intertidal to subtidal	Intertidal to sublittoral, among laminarian holdfasts, in rock crevices, beneath stones or among algae, hydroids, mussels and other growths on rocks and pier pilings	Dorsally deep brownish orange and ventrally uniformly pale	Present; narrow triangular	Present	16	Large cerebral organs extending behind the brain	Coe (1901); Gibson & Crandall (1989); Gibson (1995)
<i>Nipponnemertes crypta</i>	Sugashima (Japan)	Sugashima (Japan)	10 m	In rock cracks probably resulting from rock boring sipunculans	Dorsally pale brown and ventrally lighter colored without mottling (Fig. 10B–D)	Present	Present	13	Reaching behind the brain	Present study
<i>Nipponnemertes danae</i> (Friedrich, 1957)	Faroe Island (Denmark)	Faroe Island (Denmark); Arctic waters	Intertidal	–	Dorsally reddish and ventrally whitish; Berg (1985) indicated morphological similarities with <i>N. drepanophoroides</i>	Absent	–	–	Large	Friedrich (1957); Berg (1985); Gibson (1995)

<i>Nipponnemertes drepanophoroides</i>	Puget Sound (USA)	Puget Sound (USA)	–	–	Dorsally reddish and ventrally whitish	Absent	–	–	Cephalic organs large, anterior portion opposite ventral commissure; posteriorly between dorsal and ventral	Griffin (1898); Berg (1985); Gibson (1995)
<i>Nipponnemertes fernaldi</i> Iwata, 2001	Peavine Pass (USA)	Peavine Pass, Friday Harbor, Willow Island (USA)	4–52 m	Gravel bottom; collected by night light in water column	Dorsally pale brown and ventrally lighter in color; lateral margin transparent	Absent	Present	14	Very large and extend behind the cerebral ganglia	Iwata (2001)
<i>Nipponnemertes ganahai</i>	Off Jogashima (Japan)	Widely found in Japanese coastal waters (off Jogashima, off Ofunato, off Noto, and Tosa Bay)	71–985 m	Muddy to sandy substrates mixed with shell debris	Uniformly pale pink to yellow colored in both dorsal and ventral surface	Absent	Absent	10	Reaching behind the brain	Present study
<i>Nipponnemertes incainca</i>	Inca-Inca beach (Colombia)	Inca-Inca beach and Taganga (Colombia)	Intertidal	Under boulders	Uniformly solid bright red colored	Present	Absent	12	–	Gonzalez-Cueto <i>et al.</i> (2017)
<i>Nipponnemertes jambio</i>	Off Jogashima (Japan)	Off Jogashima (Japan)	80–89 m	Muddy to sandy substrates mixed with shell debris	Dorsal body surface uniformly brownish orange; ventral surface slightly lighter colored than dorsal surface	Present	Present	–	–	Present study
<i>Nipponnemertes kozaensis</i>	Koza (Japan)	Koza (Japan)	12 m	Among rocky substrates colonized by sponges, ascidians, and bryozoans	Background generally pale-colored, mottled with red-brown and white dots; red-brown dots more densely distributed in dorso-anterior region of body; white dots uniformly distributed on both dorsal and ventral surfaces	Absent	Absent	10	Reaching behind the brain	Present study

<i>Nipponnemertes lactea</i>	Off Izu (Japan)	Off Izu (Japan)	98–120 m	Among sandy substrates	Uniformly pale in both dorsal and ventral surface	Absent	Absent	10	Reaching behind the brain	Present study
<i>Nipponnemertes madagascarensis</i> (Kirsteuer, 1965)	Mozambique Channel (Madagascar)	Mozambique Channel (Madagascar)	–	Among dead encrusting bryozoans covering branches of a coral	Ochre colored on dorsal surface mottled with irregular reddish to brown patterns	Absent	Absent	9	Large cerebral organs extending behind the brain	Kirsteuer (1965); Gibson (1995)
<i>Nipponnemertes magna</i> (Punnet, 1903)	Pysfjord (Norway)	Pysfjord (Norway)	500 m	On coral bottom (<i>Lophohelia prolifera</i> (currently known as <i>Desmophyllum pertusum</i>))	Light orange brown	–	–	20	Well developed, extending behind the brain	Punnet (1903); Berg (1985)
<i>Nipponnemertes marioni</i> (Hubrecht, 1887)	Not designated; originally reported from Marion Island and Christmas Harbor, Kerguelen Island	Antarctic and Subantarctic waters	474 m	Among kelp holdfasts and on volcanic sands	In the original description, only a body diameter was provided: 5.5 mm in maximum; according to Wheeler (1934), body coloration resembles <i>Amphiporus moseleyi</i> Hubrecht, 1887 (i.e., dorsally blue-green, yellow-green, pale buff or light brown, and ventrally pale buff)	Absent?	–	15	Large, extending behind the brain	Hubrecht (1887); Berg (1985); Gibson (1995)
<i>Nipponnemertes neonilae</i>	Peter the Great Bay (Russia)	East Russian waters including the Sea of Okhotsk and the Sea of Japan	Intertidal to 15 m	Among mytilid beds (<i>Modiolus kurilensi</i> , <i>Crenomytilus grayanus</i>) and calcareous algae (<i>Bossiella</i> sp.)	Dorsally brownish-red or brick-red and ventrally beige or whitish (Fig. 10L)	Present; paired dark colored triangular patches	Present; mid-dorsal line without reaching to anterior tip of head and not fused with patches	16–17	Reaching behind the brain	Chernyshev (1993); present study

<i>Nipponnemertes notoensis</i>	Off Noto (Japan)	Off Noto (Japan)	70–91 m	Among muddy to sandy substrates	Uniformly pale to pale-yellow colored	Absent	Absent	10	Reaching behind the brain	Present study
<i>Nipponnemertes occidentalis</i> (Coe, 1905)	Off Washington (USA)	Off Washington (USA)	70–190 m	On subtidal pebbles or among muddy substrates mixed with pebbles	Thickly mottled and blotched with dark reddish brown on a pale ground color in dorsal surface; ventrally pale colored	Absent?	–	–	–	Coe (1905)
<i>Nipponnemertes ogumai</i> (Yamaoka, 1947)	Shimoda (Japan)	Widely reported from the Pacific coast of Honshu (Japan)	Intertidal to 5 m	Under a rock in intertidal to subtidal rocky bottom	Uniformly orange; the periphery of the ventral surface of the head is edged with a thin, blurred, black line	Absent	Present	16	Reaching behind the brain	Yamaoka (1947); Kajihara <i>et al.</i> (2015); Abe <i>et al.</i> (2022)
<i>Nipponnemertes ojimaorum</i>	Koganezaki (Japan)	Koganezaki (Japan)	7–12 m	Subtidally under a rock on sandy substrates	dorsal body surface uniformly bright red colored; ventral surface the same color as dorsal surface (Fig. 10I–K)	Present	Present; mid-longitudinal line not reaching to the anterior end of the cephalic patches, laterally fusing to the cephalic patches, and posteriorly fusing to the dorsal dark color (Fig. 4J)	13	Reaching behind the brain	Present study
<i>Nipponnemertes ornata</i>	Off Noto (Japan)	Off Noto (Japan)	70–91 m	Among muddy to sandy substrates	Background uniformly pale colored, mottled with white and brown dots; white dots conspicuous in dorsal surface of head region; brown dots more densely distributed in dorso-anterior region of body than ventral and posterior region (Fig. 11H–J)	Absent	Absent	10	Reaching behind the brain	Present study

<i>Nipponnemertes pacifica</i> (Coe, 1905)	Not designated; originally reported from the Bering Sea, off Washington, and off central California	Off central California to the Bering Sea; the coast of central Chile (Bahia San Vicente)	70–190 m	Among black sand, broken pebbles, and green muddy substrates mixed with pebbles	Reddish or brownish color on dorsal surface and pale on ventral surface in a preserved specimen	Absent	Absent?	14	Cerebral organs remarkably large, extending behind the brain	Coe (1905); Friedrich (1968)
<i>Nipponnemertes pulchra</i> (Johnston, 1837)	Not designated; originally reported from Berwickshire (UK)	Widely known from the northern hemisphere (North America, Greenland, the Faroe Is., the White Sea and northern Europe, Scandinavia, Mediterranean) as well as the southern hemisphere in Chile, South Africa, Antarctica and Subantarctica; species identities from these areas not confirmed using molecular data	Intertidal to 569 m	Under rocks, among sandy to muddy substrates, shell debris, and gravels	Fresh red colored (Johnston, 1837); Berg (1985) indicated the body coloration varies by individuals between brown, red and pink.	Absent	Present	8–14 (normally 12)	Large, extending behind the brain	Johnston (1837); Berg (1985); Gibson (1995)
<i>Nipponnemertes punctatula</i> (Coe, 1905)	Not designated; originally reported from Isthmus Cove, Catalina Island (USA)	Pacific coast of USA, eastern Russian and Japanese coastal waters (Hokkaido to Nagasaki)	Intertidal to subtidal	Under rocks and among algal holdfasts	Dorsal body surface with a dorsal brown blotch pattern (Fig. 10Q, R) or uniformly brownish (Fig. 10S, T)	Present	Present	12–16	Remarkable voluminous, extending behind the brain	Coe (1905); Gibson (1995); Iwata (2001); Kajihara (2007)
<i>Nipponnemertes rubella</i> (Coe, 1905)	Not designated; originally reported from San Diego and San Pedro (USA)	Pacific coasts of USA	35–200	Among mussels, on rocks or wharf pilings	Deep fresh color, pale orange or pale red on dorsal surface; ventrally paler colored	Absent	Absent	14	Remarkably highly developed and situated beside and behind the dorsal ganglia	Coe (1905); Gibson (1995)

<i>Nipponnemertes sanguinea</i> Riser, 1998	Kakikoura (New Zealand)	Kakikoura (New Zealand)	intertidal	Holdfast of <i>Lessonia variegata</i> washed up on beach	Varied from buffy white to pale yellow to orange, with reddish blood vessels; ventral surface paler than dorsal; brain lobes pink	Absent	Not evident (Riser, 1998)	12	Elongated, posteriorly reaching behind the brain	Riser (1998)
<i>Nipponnemertes schollaerti</i> (Wheeler, 1934)	Schollaert Channel, Palmer Archipelago, Antarctica	Antarctic waters	278–500 m	–	Pale buff in a living specimen	Absent	Absent	14	Reaching behind the brain	Wheeler (1934); Berg (1985)
<i>Nipponnemertes sugashimaensis</i>	Sugashima (Japan)	Sugashima (Japan)	10 m	Among subtidal rocky substrates	Background uniformly pale colored, with irregular, leopard-like brown mottling	Absent	Absent	10	Reaching behind the brain	Present study
<i>Nipponnemertes variabilis</i> (Korotkevich, 1983)	Kerguelen Island, Antarctic waters	Antarctic and subantarctic waters (off the southeastern coast of South America near Patagonia, north-west of the Falkland Island, off South Africa, and near South Georgia and the Kerguelen Island)	64–295 m	Sandy and muddy substrates, mixed with broken shells and gravels; attached to floating algae	Beige colored both dorsally and ventrally	Absent	Absent	12–13	Reaching behind the brain	Korotkevitch (1983); Wheeler (1934)
<i>Cratenemertes amboiensis</i> (Bürger, 1890)	Banda Sea, Ambon (Indonesia)	Banda Sea, Ambon (Indonesia)	–	–	Uniformly fresh colored in both dorsal and ventral surface	Absent	Absent	–	Small, almost fused with dorsal ganglia in posterior region	Bürger (1890); Friedrich (1955)

Table 10. Range of genetic distance within the genus *Proamphiporus* (a single specimen for *P. kaimeiae* and *P. crandalli*; seven specimens for *P. rectangulus*). Values below diagonal indicate K2P, while values above diagonal indicate uncorrected *p*-distance.

	<i>P. kaimeiae</i>	<i>P. crandalli</i>	<i>P. rectangulus</i>
<i>P. kaimeiae</i>	—	14.1	13.3–13.5
<i>P. crandalli</i>	15.8	—	10.4–10.7
<i>P. rectangulus</i>	14.9	11.2–11.6	0.00–0.05

Table 11. Comparison of selected morphological characters of free-living marine oerstediiids or monostiliferan genera presumably classified as Oerstediiidae due to having a two-layered (inner longitudinal and outer circular) muscular rhynchocoel wall and lacking a vascular plug, with rhynchocoel half-length of the body or shorter.

Genus or species	Diagonal musculature in body wall	Body-wall longitudinal musculature anteriorly divided/undivided	Precerebral septum closed/lacking	Number of eyes	References
<i>Abyssonemertes</i> Chernyshev & Polyakova, 2018	absent	undivided	closed	no eyes	Chernyshev and Polyakova (2018a)
<i>Aenigmanemertes</i> Sundberg & Gibson, 1995	absent	divided	lacking	20	Sundberg and Gibson (1995)
<i>Dichonemertes</i> <i>hartmanae</i> (Coe, 1938)*	absent	N/A	closed	4	Coe (1938)
<i>Digononemertes</i> Gibson, 1990b	absent	undivided	closed	4	Gibson (1990b)
<i>Halimanemertes</i> Gibson, 1990b	absent	undivided	closed	12	Gibson (1990b)
<i>Ischyronemertes</i> Gibson, 1990b	present	undivided	closed	4 or more eyes arranged in 4 groups	Gibson (1990b)
<i>Nemertellina</i> Friedrich, 1935	absent	undivided	closed	4	Friedrich (1935); Kajihara (2021)

<i>Paranemertopsis</i> Gibson, 1990b	present	divided	closed	1–3 eyes arranged in 4 groups	Gibson (1990b)
<i>Rhombonemertes</i>	present	undivided	closed	4	Present study
<i>Vulcanonemertes</i> Gibson & Strand, 2002	absent	divided	lacking	4	Gibson and Strand (2002)

*The rhynchocoel is illustrated as shorter than a half-length of the body in *D. hartmanae* (Coe, 1938), while it extends to the posterior tip of the body in *D. coensis* (Friedrich, 1970).

Table 12. List of species included in the phylogenetic analysis and DDBJ/EMBL/GenBank accession numbers for each gene.

Species	COI	16S	18S	28S	References
<i>Antiponemertes novazealandiae</i>	–	–	AY928345	–	Strand and Sundberg (2005)
<i>Argonemertes australiensis</i> IZ-135314	HQ848601	JF277605	JF293010	HQ856892	Andrade et al. (2012)
<i>Argonemertes australiensis</i> IZ-135315	KF935525	KF935476	KF935309	KF935365	Kvist et al. (2014)
cf. <i>Potamonemertes percivali</i> IZ-25172	KF935532	KF935483	KF935316	KF935372	Kvist et al. (2014)
<i>Leptonemertes</i> cf. <i>chalicophora</i>	HQ848596	JF277608	JF293011	HQ856898	Kvist et al. (2014)
<i>Leptonemertes</i> sp. Japan	In prep.	In prep.	In prep.	In prep.	Present study
Plectonemertidae sp. IZ-25173	KF935529	KF935480	KF935313	KF935369	Kvist et al. (2014)
Plectonemertidae sp. IZ-25174	KF935530	KF935481	KF935314	KF935370	Kvist et al. (2014)
Plectonemertidae sp. IZ-25175	KF935531	KF935482	KF935315	KF935371	Kvist et al. (2014)
Plectonemertidae sp. IZ-25169	KF935528	KF935479	KF935312	KF935368	Kvist et al. (2014)
Plectonemertidae sp. IZ-25166	KF935526	KF935477	KF935310	KF935366	Kvist et al. (2014)
Plectonemertidae sp. IZ-25167	KF935527	KF935478	KF935311	KF935367	Kvist et al. (2014)
Plectonemertidae sp. NT000046	EU255614	–	EU255585	–	Mateos and Giribet (2008)
Plectonemertidae sp. NT000072	EU255626	KF935486	EU255596	–	Mateos and Giribet (2008)
Outgroup taxa					
<i>Amphiporus lactifloreus</i>	HQ848611	JF277617	JF293018	HQ856876	Andrade et al. (2012)
<i>Malacobdella grossa</i>	HQ848591	JF277614	JF293015	HQ856882	Andrade et al. (2012)

<i>Geonemertes pelaensis</i>	HQ848593	JF277611	JF304779	HQ856888	Andrade et al. (2012)
<i>Nipponnemertes ogumai</i>	AB920907	AB921008	AB921009	AB921010	Kajihara et al. (2015)
<i>Nipponnemertes pulchra</i>	HQ848597	JF277625	JF293012	HQ856871	Andrade et al. (2012)

Table 13. List of species included in the phylogenetic analysis and DDBJ/EMBL/GenBank accession numbers for each gene. Country names of each species sampling location are abbreviated as follows: CA = Canada, JP = Japan, RU = Russia, USA = United States of America, and VE = Venezuela.

Species	Sampling location	16S	COI	18S	28S	H3	References
<i>Tetrastemma 'aequicolor'</i> 24 QuI	Erineyskaya Inlet, RU	MZ231141	MZ216528	MZ231206	MZ231296	MZ216598	Chernyshev et al. (2021)
<i>Tetrastemma 'aequicolor'</i> 25 QuI	Erineyskaya Inlet, RU	MZ231142	MZ216529	MZ231207	MZ231297	MZ216599	Chernyshev et al. (2021)
<i>Tetrastemma 'aequicolor'</i> 26 QuI	Erineyskaya Inlet, RU	MZ231143	MZ216530	MZ231208	MZ231298	MZ216600	Chernyshev et al. (2021)
<i>Tetrastemma album</i> sp. nov.	Sagami Bay, JP	OQ248525	OQ249697	OQ248517	OQ248520	OQ248166	Present study
<i>Tetrastemma carneum</i> sp. nov.	Off Ofunato, JP	In prep.	In prep.	In prep.	In prep.	In prep.	Present study
<i>Tetrastemma cupido</i>	Sagami Bay, JP	OK428649	OK414013	OK428689	OK428648	—	Hookabe et al. (2021b)
<i>Tetrastemma nigrifrons</i>	CA	MZ231144	MZ216531	MZ231209	MZ231299	MZ216601	Chernyshev et al. (2021)
<i>Tetrastemma nigrifrons</i>	Oregon, USA	MZ231145	MZ216532	MZ231210	MZ231300	MZ216602	Chernyshev et al. (2021)
<i>Tetrastemma nigrifrons</i>	California, USA	MZ231146	MZ216533	MZ231211	MZ231301	MZ216603	Chernyshev et al. (2021)
<i>Tetrastemma stimpsoni</i>	JP	MZ231147	MZ216534	MZ231212	MZ231301	MZ216604	Chernyshev et al. (2021)
<i>Tetrastemma stimpsoni</i>	RU	MZ231148	MZ216535	MZ231213	MZ231303	MZ216605	Chernyshev et al. (2021)
<i>Tetrastemma stimpsoni</i>	Iturup, RU	MZ231149	MZ216536	MZ231214	MZ231304	MZ216606	Chernyshev et al. (2021)

<i>Tetrastemma elegans</i> B2	York River, USA	MZ231156	MZ216543	MZ231222	MZ231312	MZ216614	Chernyshev et al. (2021)
<i>Tetrastemma elegans</i> C2	USA	MZ231157	MZ216544	MZ231223	MZ231313	MZ216615	Chernyshev et al. (2021)
<i>Tetrastemma elegans</i> D2	York River, USA	MZ231158	—	MZ231224	MZ231314	—	Chernyshev et al. (2021)
<i>Tetrastemma enteroplecta</i> A6	Florida, USA	MZ231159	MZ216545	MZ231225	MZ231314	MZ216616	Chernyshev et al. (2021)
<i>Tetrastemma enteroplecta</i> E3	Florida, USA	MZ231160	—	MZ231226	MZ231316	MZ216618	Chernyshev et al. (2021)
<i>Tetrastemma enteroplecta</i> B7	VE	MZ231161	—	MZ231227	MZ231317	—	Chernyshev et al. (2021)
<i>Tetrastemma freyae</i>	Hawaii, USA	—	MT247877	MZ231229	MZ231319	MT247879	Chernyshev et al. (2020)
<i>Tetrastemma merulum</i> F2	Florida, USA	MZ231163	MZ216550	MZ231231	MZ231321	MZ216622	Chernyshev et al. (2021)
<i>Tetrastemma merulum</i> H5	Florida, USA	MZ231164	MZ216551	MZ231232	MZ231322	MZ216623	Chernyshev et al. (2021)
<i>Tetrastemma persona</i> sp. nov.	Sagami Bay, JP	OQ248526	OQ249698	OQ248518	OQ248521	OQ248167	present study
<i>Tetrastemma reticulatum</i>	California, USA	MZ231168	MZ216556	MZ231238	MZ231328	MZ216629	Chernyshev et al. (2021)
<i>Tetrastemma shohoense</i> sp. nov.	Shoho Seamount, JP	OQ249700	—	—	—	—	Present study
<i>Tetrastemma</i> sp. F7	Florida, USA	MZ231173	MZ216564	MZ231246	MZ231336	MZ216637	Chernyshev et al. (2021)
<i>Tetrastemma</i> sp. GM 1	Gulf of Mexico, USA	MZ231175	—	MZ231248	MZ231338	MZ216639	Chernyshev et al. (2021)
<i>Tetrastemma</i> sp. GM 2	Florida, USA	MZ231176	MZ216565	MZ231249	MZ231339	MZ216640	Chernyshev et al. (2021)

<i>Tetrastemma</i> sp. GM 3	Gulf of Mexico, USA	MZ231177	—	MZ231250	MZ231340	MZ216641	Chernyshev et al. (2021)
<i>Tetrastemma</i> sp. I	Iturup, RU	MZ231179	—	MZ231252	MZ231342	MZ216643	Chernyshev et al. (2021)
<i>Tetrastemma</i> sp. IP	Iturup, RU	MZ231180	MZ216567	MZ231253	MZ231343	MZ216644	Chernyshev et al. (2021)
<i>Tetrastemma</i> sp. J 1TjS	Simushir, RU	MZ231182	MZ216570	MZ231256	MZ231346	MZ216647	Chernyshev et al. (2021)
<i>Tetrastemma</i> sp. J 3TjS	Simushir, RU	MZ231183	MZ216571	MZ231257	MZ231347	MZ216648	Chernyshev et al. (2021)
<i>Tetrastemma</i> sp. J 4TjS	Simushir, RU	MZ231184	MZ216572	MZ231258	MZ231348	MZ216649	Chernyshev et al. (2021)
<i>Tetrastemma</i> sp. M1	Urup, RU	—	MZ216573	MZ231259	MZ231349	MZ216650	Chernyshev et al. (2021)
<i>Tetrastemma</i> sp. M2	Urup, RU	—	MZ216574	MZ231260	MZ231350	MZ216651	Chernyshev et al. (2021)
<i>Tetrastemma</i> sp. S 1TsS	Simushir, RU	—	MZ216575	MZ231261	MZ231351	MZ216652	Chernyshev et al. (2021)
<i>Tetrastemma</i> sp. S 2TsS	Simushir, RU	—	MZ216576	MZ231262	MZ231352	MZ216653	Chernyshev et al. (2021)
<i>Tetrastemma</i> sp. U 13TsU	Urup, RU	MZ231185	MZ216577	MZ231263	MZ231353	MZ216654	Chernyshev et al. (2021)
<i>Tetrastemma</i> sp. U 18TsU	Urup, RU	MZ231186	MZ216578	MZ231264	MZ231354	MZ216655	Chernyshev et al. (2021)
<i>Tetrastemma</i> sp. UR	Urup, RU	MZ231187	—	MZ231265	MZ231355	MZ216656	Chernyshev et al. (2021)