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A review of Asian *Hediste* species complex (Nereididae, Polychaeta) with descriptions of two new species and a redescription of *Hediste japonica* (Izuka, 1908)

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Three Asian species of the genus *Hediste* (Nereididae, Polychaeta, Annelida), including two new species, are reviewed. *Hediste japonica* (Izuka, 1908) was redescribed based on recently discovered type material and on additional specimens newly collected from the Ariake Sea in Japan and the west coast of Korea. Two new species, ***H. diadroma*** and ***H. atoka***, were described based on specimens collected from Japan. These three species are morphologically similar, but distinguishable by some differences in parapodial and chaetal morphology, at least at the sexually mature stage. The presence of homogomph falcigers and the absence of heterogomph spinigers are unique characteristics of *H. japonica* among this genus. Inconspicuous epitokous metamorphosis occurred as a slight enlargement of the eyes and parapodial ligules in mature adults of *H. diadroma* sp. nov. and *H. japonica* in combination with their reproductive swarming and pelagic larval development. The epitoke-specific sesquigomph spinigers were added in mature adults of *H. diadroma* sp. nov., while the number of ordinary homogomph spinigers increased in some adults of *H. japonica.* Epitokous metamorphosis in the *Hediste* species was compared with that in other nereidid genera. No epitokous metamorphosis was observed in *H. atoka* sp. nov., corresponding to its reproduction without swarming and to its development without a planktonic larval stage. Significant differences in paragnath numbers in the maxillary ring of the proboscis were found among the three species, suggesting different feeding habits. In Japan, the present distribution of *H. japonica* is restricted to muddy tidal flats in the inner part of the Ariake Sea, probably due to recent human impact, while the other two species are widespread. *Zoological Journal of the Linnean Society,* 2003, **137**, 403–445

ADDITIONAL KEYWORDS: epitoky – lectotype – paragnath – species diversity – taxonomy.

INTRODUCTION

Three allopatric species of *Hediste* Malmgren, 1867 have been described from shallow brackish or fresh waters in the North Temperate Zone: *H. diversicolor* (O. F. Muller, 1776) from both the European and the North American coast of the Atlantic (Smith, 1977), *H. limnicola* (Johnson, 1903) from the North American Pacific coast (Smith, 1958), and *H. japonica* (Izuka, 1908) from East Asia (Ushakov, 1955; Imajima, 1972; Wu, Sun & Yang, 1985; Khlebovich, 1996). The most important diagnostic characteristic of this genus is the presence of simple chaetae derived from falcigers in the posterior neuropodia (Fong & Garthwaite, 1994; Khlebovich, 1996). The three brackish-water *Hediste* species are morphologically very similar to one another (Smith, 1958). Recently, Kirkegaard (1995) described *H. kermadeca* from a depth of 4410 m in the Kermadec Trench. However, it is doubtful whether this species belongs to the genus *Hediste*, because there is no reference to simple chaeta.

In Asia, the distributional range of *Hediste* species extends north to Sakhalin in Russia and south to Amani-Oshima Island in Japan (Izuka, 1908; Ushakov, 1955; Imajima, 1972; Wu *et al*., 1985; Khlebovich, 1996; Sato, 1999). Previously, the Asian *Hediste* worms were regarded as belonging to a single species, *H. japonica*. However, our previous studies on the reproduction, early development, karyotype, gamete ultrastructure and electrophoretic analysis of the allozymes suggested that '*H. japonica*' consists of

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three distinct species, namely, the small-egg, large-egg and Ariake forms (the latter named after the Ariake Sea where it occurs) (Sato & Tsuchiya, 1987, 1991; Sato & Ikeda, 1992; Sato & Masuda, 1997; Sato, 1999). The type material of *H. japonica* appeared to have been lost. Judging from its original description (Izuka, 1908), we considered that the small-egg form corresponded to *H. japonica sensu stricto*, and that the large-egg and Ariake forms were undescribed species (Sato, 1999, 2000, 2001). Fortunately, the syntype of *H. japonica* was recently discovered, which has led us to revise our earlier conclusion; we now consider that the Ariake form corresponds to *H. japonica sensu stricto*.

In the present paper, we redescribe *H. japonica* based on the newly designated lectotype and newly collected specimens, and describe two new species, reviewing both our original data and the previously published findings on these species.

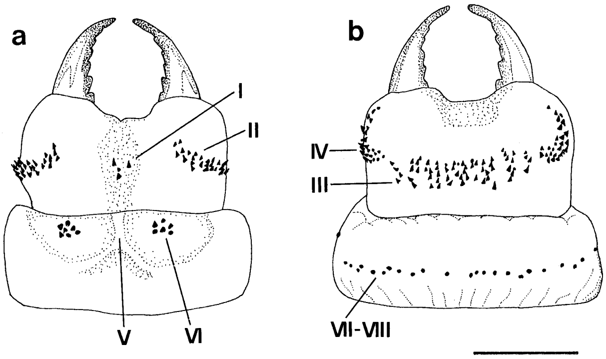


Figure 1. Grouping of paragnaths appearing in different areas on the dorsal (a) and ventral (b) surfaces of an everted proboscis. Groups I to IV on the maxillary ring form the posterior belt, while groups V to VII–VIII on the oral ring form the anterior belt on the retracted proboscis. ***Hediste atoka* sp. nov.** (nontype) collected from Kotsukigawa River. Scale bar = 1 mm.

MATERIAL AND METHODS

Sexually mature and immature specimens were collected from shallow brackish waters in Japan and Korea. Most were collected from sediment samples dug from intertidal flats within estuaries. Swarming mature adults were collected by scoop net as they were swimming around the mouths of several rivers in winter or early spring. The specimens were fixed in 10% formalin or 80% ethanol, and transferred to 80% ethanol. Some living animals were transported to the laboratory, put into small vinyl bags individually, and deep-frozen at −80°C for electrophoretic analysis.

Some mature adults were obtained by rearing immature worms for 1–6 months after collection in aquaria with sand and 50% seawater (salinity: *c.* 15‰). The aquaria were maintained at room temperature and aerated; the animals were fed commercial dry fish food (Tetramin). In a few cases, mature adults were obtained by rearing immature worms individually in a glass dish in an incubator (15–17°C). Some immature worms were obtained by rearing embryos produced by artificial fertilization (Sato & Tsuchiya, 1991), for 1–2 years in aquaria as mentioned above.

For the fixed specimens, body length (BL), anterior maximum body width excluding the parapodia (BW), and maximum eye width of the anterior pair (EW) were measured. Measurements and drawings were carried out with a camera lucida. The paragnaths in each of the groups on the proboscis (Fig. 1) were counted under a binocular microscope. Type material of *Hediste japonica*, which was recently found in Izuka's collection preserved in the University of Tokyo, was also examined. For the frozen materials, horizontal starch gel electrophoresis was carried out to compare the allozyme patterns of lactate dehydrogenase (LDH) among the three *Hediste* species, according to the method of Sato & Masuda (1997).

The type material is deposited in the University Museum, the University of Tokyo (UMUT), the National Science Museum, Tokyo (NSMT), the Zoological Institute, Hokkaido University, Sapporo (ZIHU), Coastal Branch of Natural History Museum and Institute, Chiba (CMNH), the Osaka Museum of Natural History, Osaka (OMNH), Muséum National d'Histoire Naturelle, Paris (MNHN), the Senckenberg Museum, Frankfurt (SMF), Zoological Museum, University of Copenhagen (ZMUC), and the United States National Museum of Natural History (Smithsonian Institution), Washington, D. C. (USNM).

EXAMINATION OF THE SYNTYPE OF *HEDISTE JAPONICA*

Five bottles containing *Hediste* specimens preserved in UMUT were recently discovered in Izuka's collection (Table 1). The specimens in three of the bottles were documented as having been collected from 1896 to 1906 by their labels, and were probably used as material for the original description of *H. japonica* (Izuka, 1908). Therefore, they were judged as the syntype of this species. In fact, Izuka (1908) described 11 localities, including the Sumida River in Tokyo and Kojima Bay in Okayama Prefecture, as sampling sites for this species. The specimens found in the two remaining bottles were judged as non-type, because they were collected on an unknown date or after the original description.

The syntype series apparently included more than one species of *Hediste.* We designated one of the eight epitokous specimens (Fig. 2a), which were collected

Table 1. Recently discovered *Hediste* specimens in Izuka's collection. Collections before the original February 1908 description of *H. japonica* are judged as the syntype series

		Label information[1]			
Lot number	No. of specimens (state of maturity)	Locality	Sampling date	Species name	Corresponding to three forms of Sato (1999)
Syntype of *H. japonica*					
UMUTZ-PLC-IZ-113	15 (immature)	–	24 Dec. 1896	–	Large- or small-egg
UMUTZ-PLC-IZ-114	22 (immature)	Sumida	5 June 1900	–	Large- or small-egg
UMUTZ-PLC-IZ-55	8 (mature)	Hachihama, Kojima Bay, Okayama Prefecture	17 Dec. 1906	*Nereis japonica* Izuka	Ariake
Non-type					
UMUTZ-PLC-IZ-84	7[2] (immature)	Kojima Bay	13 Oct. 1908	*Nereis japonica* Izuka	Ariake
UMUTZ-PLC-IZ-115	248 (immature)	–	–	–	Large- or small-egg

[1] All label descriptions pencilled in Japanese.
[2] Specimens dried and badly damaged.

from Kojima Bay in 1906 and preserved in a bottle, as the lectotype of *H. japonica*, for the following reasons: (1) of the three bottles containing the syntypes, only this particular bottle included a label with a description of the species name '*Nereis japonica* Izuka' (Fig. 2b) (2) Izuka (1908) noted that sexually mature adults had been obtained from only one locality, Kojima Bay, and described details of his own observations of the reproductive swarming there on 17 December, 1906. This is exactly the date written on the label of this bottle (Fig. 2c).

The morphological characteristics of the lectotype and seven other paralectotype specimens in the same bottle were apparently consistent with those of the Ariake form, which Sato (1999, 2000, 2001) tentatively reported, as mentioned below.

SYSTEMATICS

FAMILY NEREIDIDAE JOHNSTON, 1865

SUBFAMILY NEREIDINAE JOHNSTON, 1865

GENUS *HEDISTE* MALMGREN, 1867

Type species: Hediste diversicolor (O. F. Müller, 1776)

Amended generic diagnosis

Eversible proboscis with conical paragnaths on both maxillary and oral rings. Four pairs of tentacular cirri; parapodia biramous except for the first 2 setigers. Notochaetae homogomph spinigers. Neurochaetae homogomph spinigers with or without heterogomph spinigers, and heterogomph falcigers with or without homogomph falcigers. One or a few thick simple chaetae present in supra-acicular fascicle in posterior neuropodia.

Remarks

The presence of simple straight chaetae in the posterior neuropodia is the most important diagnostic characteristic separating *Hediste* from *Nereis* and *Neanthes*. The simple chaetae were previously referred to as fused homogomph falcigers (Fauvel, 1923; Fauchald, 1977; Hartmann-Schröder, 1996). However, in the type species, *H. diversicolor*, nonfused homogomph falcigers are absent, while heterogomph falcigers are present. Our observations of the Japanese *Hediste* species suggest that a simple chaeta originates not from a homogomph but from a heterogomph falciger as pointed out by Smith (1958), and that the simple chaeta may be produced not only by fusion between a blade and a stalk of a falciger, but also by deformation of a stalk after detachment of a blade (see below).

HEDISTE JAPONICA (IZUKA, 1908) (FIGS 2–15, 38–41, 43)

Nereis japonica Izuka, 1908: 295–305, 4 text figs.; Izuka, 1912: 163–169, pl. 17, figs 14–16, 18 and 4 text figs.

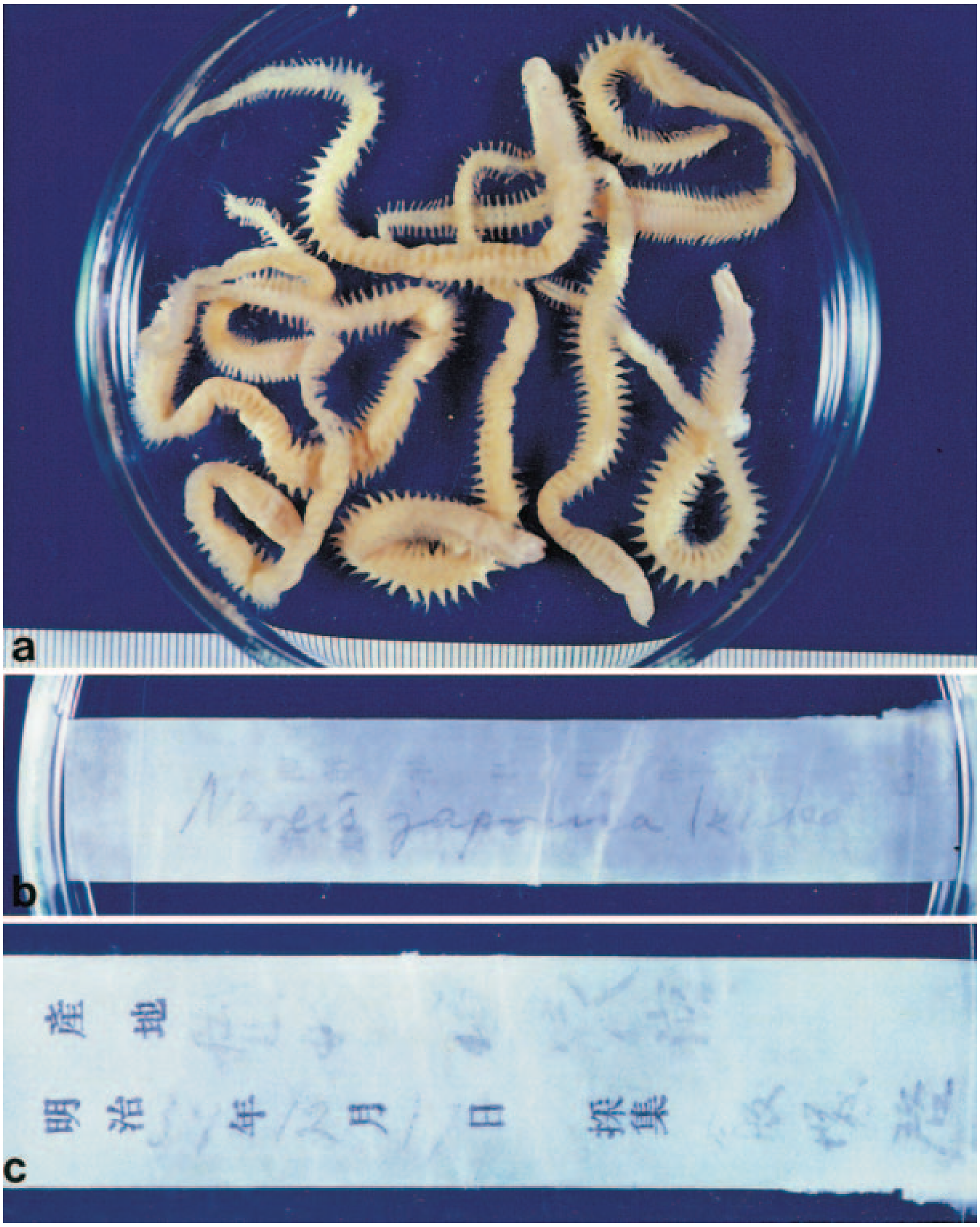


Figure 2. Part of the syntype series of *Hediste japonica* (Izuka, 1908). (a) Eight mature females in a bottle of Izuka's collection recently discovered in UMUT. One of them is designated as the lectotype. (b) Detail of label: the original species name of '*Nereis japonica* Izuka' can be seen. (c) Detail of label, in Japanese: the locality 'Hachihama, Kojima Bay, Okayama Prefecture', the sampling date '17 December, Meiji 39 (= 1906)', and the collector's name 'Akira Izuka'.

?*Nereis japonica* (populations of Yellow Sea): Khlebovich *et al*., 1982: 1110–1113.

Neanthes japonica (epitokes): Smith, 1958: 60–73.

Hediste sp. 2 (Ariake form): Sato, 1999: 129–143; Sato, 2000: 187–191, figs 8-1 and 8-3; Sato, 2001: 66–86.

Not *Hediste japonica*: Imajima, 1972: 102–105, figs 30 a–n, 37 (= *H. diadroma* sp. nov. or *H. atoka* sp. nov.); Imajima, 1996: 139, fig. 111 (= *H. diadroma* sp. nov. or *H. atoka* sp. nov.); Sato, 1999: 129–143 (= *H. diadroma* sp. nov.); Sato, 2000: 187–191, figs 8-2 and 8-3 (= *H. diadroma* sp. nov.); Sato, 2001: 66–86, figs 4-1–4-10 (= *H. diadroma* sp. nov.); Rouse & Pleijel, 2001: pl. 5c (= *H. diadroma* sp. nov.).

Type material (Fig. 2)

Lectotype (UMUTZ-PLC-IZ-55A): complete ovigerous female (BL: 100 mm, BW: 4.0 mm, 98 setigers), Hachihama, Kojima Bay, Okayama Prefecture, the Seto

Inland Sea, 17 December 1906, coll. A. Izuka. A part of paralectotype (UMUTZ-PLC-IZ-55B): 7 ovigerous females (BL: 70–100 mm, BW: 3.0–4.1 mm, 87–102 setigers), data as for lectotype.

Other material examined

Mature specimens (epitokes): collected during reproductive swarming in Ariake Sea, Japan. Nanaura, Kashima-shi, Saga Prefecture, 29 December 2001, coll. A. Nakashima, 6 specimens (OMNH-Iv-4223, 4224).

Epitokes obtained after rearing in laboratory: Daijugarami, Higashiyoka-cho, Saga Prefecture, Ariake Sea, Japan, 1 female fixed 24 February 1997 after rearing for 4 months (ZIHU-2102), 1 male fixed 20 January 1997 after rearing for 3 months (ZIHU-2103), coll. A. Nakashima.

Specimens collected from sediment samples: Seto Inland Sea, Japan: Kojima Bay, Okayama Prefecture, 13 October 1908, coll. Eitaro Iwaki, 7 specimens (damaged by drying, UMUTZ-PLC-IZ-0000). Ariake Sea, Japan: Around mouth of Honmyogawa River, Isahaya Bay, Takaki-cho, Nagasaki Prefecture, 21 August 1994, coll. H. Yamashita, 5 specimens; 19 November 1994, coll. H. Yamashita, 11 specimens; 28 April 1995, coll. T. Sakai, 17 specimens (juveniles); 29 May 1995, coll. T. Sakai, 17 specimens; 21 January 1996, coll. T. Sakai, 2 specimens (juveniles); 28 November 1996, coll. A. Nakashima, 31 specimens. Onojima, Isahaya Bay, Isahaya-shi, Nagasaki Prefecture, 30 March 1994, coll. M. Sato, 52 specimens (juveniles); 1 July 1994, coll. H. Yamashita, 13 specimens; 20 September 1994, coll. H. Yamashita, 9 specimens; 28 October 1995, coll. T. Sakai, 1 specimen; 10 November 1997, coll. M. Sato, 1 specimen. Around mouth of Nitandagawa River, Isahaya Bay, Moriyama-cho, Nagasaki Prefecture, 20 August 1994, coll. M. Sato, 12 specimens. Ariakegawa River, Isahaya Bay, Moriyama-cho, Nagasaki Prefecture, 20 August 1994, coll. M. Sato, 36 specimens. Yamadagawa River, Isahaya Bay, Azuma-cho, Nagasaki Prefecture, 24 April 1995, coll. H. Yamashita, 22 specimens (juveniles). Kojirogawa River, Isahaya Bay, Nagasaki Prefecture, 3 November 1994, coll. H. Yamashita, 9 specimens. Nanaura, Kashima-shi, Saga Prefecture, 27 September 1996, coll. H. Yamashita, 1 specimen; 29 April 2001, coll. M. Sato, 11 specimens (ZIHU-2104). Rokkakugawa River, Ashikari-cho, Saga Prefecture, 10 October 1995, coll. H. Yamashita & T. Sakai, 11 specimens. Daijugarami, Higashiyoka-cho, Saga Prefecture, 20 March 1995, coll. A. Nakashima, 12 specimens (juveniles); 3 April 1995, coll. A. Nakashima, 5 specimens (juveniles); 28 July 1995, coll. A. Nakashima, 90 specimens (ZIHU-2101); 21 October 1996, coll. A. Nakashima, 21 specimens (CMNH-ZW-1888, MNHN, NSMT-Pol-107441, SMF-12064, USNM-1008435, ZMUC-Pol-1502); 8 December 1996, coll. A. Nakashima, 16 specimens. Omutagawa River, Omuta-shi, Fukuoka Prefecture, 24 November 1998, coll. K. Ichimiya, 21 specimens; 4 January 1999, coll. K. Ichimiya, 21 specimens (including mature or nearly mature adults); 17 May 1999, coll. M. Sato, S. Iwamatsu & A. Kubo, 32 specimens; 21 December 1999, coll. S. Iwamatsu, 5 specimens (including mature or nearly mature adults). Yellow Sea, Korea: Yongyudo Island, Inchon, 19 January 1999, coll. M. Sato & J.-S. Hong, 11 specimens (including nearly mature adults). Around mouth of Kumu River, Kunsan, 2, 3 May 2000, coll. A. Kubo, 17 specimens.

Diagnosis

Few (less than 10 in most cases) large paragnaths on right and left sides of proboscis in group II. Homogomph falcigers with slender blade present, and heterogomph spinigers absent in neuropodia. Neuropodial postchaetal ligule tapering to digitate lobe throughout. Epitokous stage without any addition of epitoke-specific chaetae. Full-grown oocytes 180–210 μm in diameter. Reproductive swarming in winter.

Description

Body stout anteriorly, posteriorly gradually tapering toward pygidium (Figs 3a,c, 4a,d). Dorsum convex, venter relatively flat with longitudinal midventral groove. Colour in preserved specimens whitish cream with dark brown pigmentation on the anterior dorsal surface; colour in life at sexually immature stage reddish or greenish brown with anterior dorsal surface darkened.

Prostomium pyriform, broader than long, with pair of smooth tapered antennae situated at anterior end. Pair of palps with massive palpophores about twice as long as antennae and short round palpostyles. Two pairs of round or reniform eyes almost equal in size (border of eye unclear in type material because of discoloration from long-term preservation), arranged trapezoidally (space between anterior pair 1.3–1.4 times as wide as that between posterior pair). Longitudinal mid-dorsal groove present on anterior dorsum of prostomium. Partial dark pigmentation present on dorsal anterior surface of prostomium and palpophore (Fig. 4a), though pigmentation unclear in type material (Fig. 3a).

Peristomium nearly as long as following setigers, with 4 pairs of tentacular cirri of unequal length; posterior dorsal tentacular cirri longest, reaching back to setigers 6–9; anterior dorsal tentacular cirri next longest, reaching back to setigers 3–4.

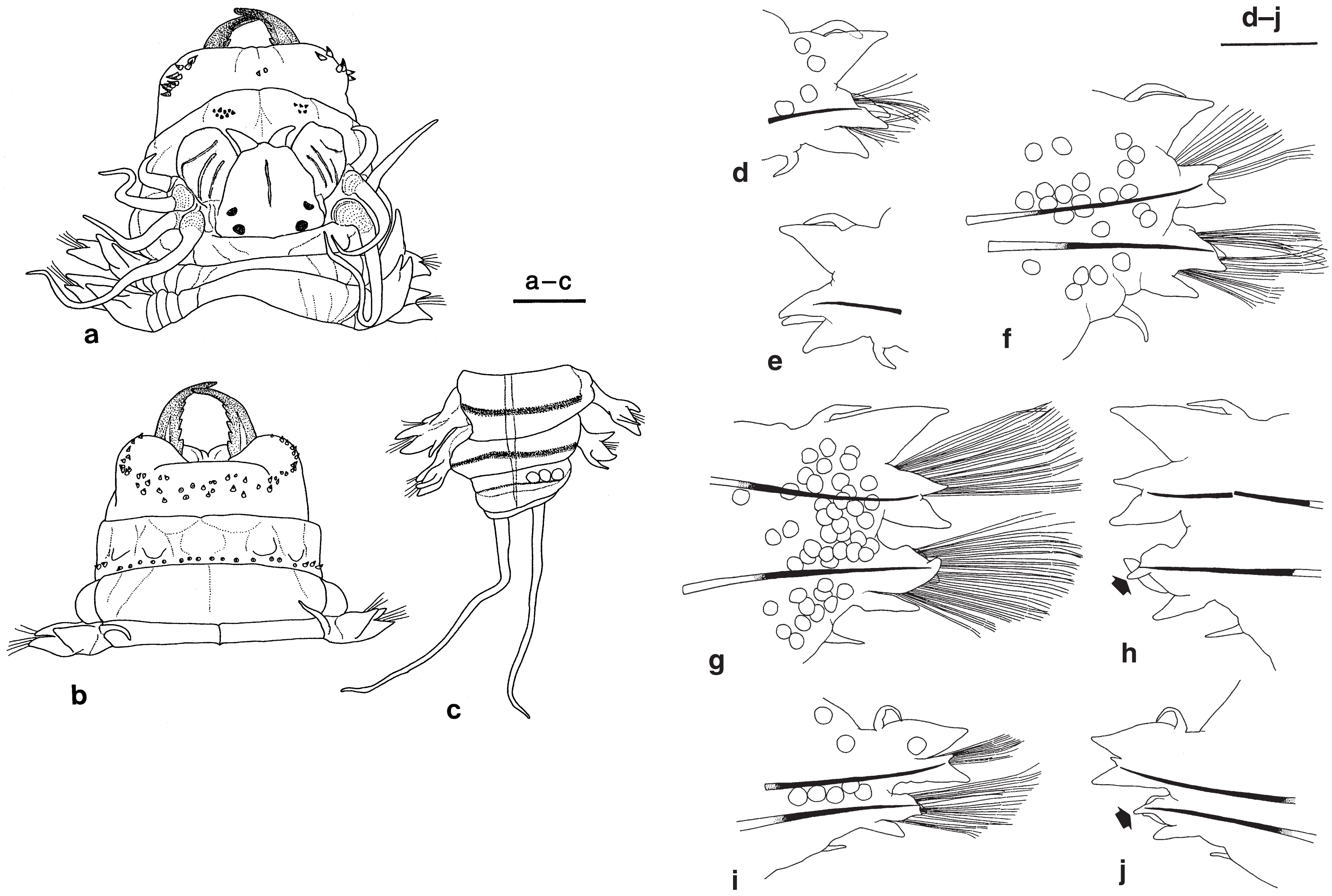


Figure 3. Mature female of *Hediste japonica* (Lectotype, UMUTZ-PLC-IZ-55A). (a) Dorsal view of anterior end with an everted proboscis. (b) Ventral view of proboscis. (c) Dorsal view of posterior end. (d–j) Parapodia of the left side (chaetae not shown in e, h, j). (d & e) Anterior and posterior views of parapodium 2. (f) Anterior view of parapodium 10. (g & h) Anterior and posterior views of parapodium 30. (i & j) Anterior and posterior views of parapodium 71. Arrows in h and j indicate a diagnostic characteristic, presence of a digitate lobe at tip of the postchaetal ligule in posterior neuropodia. Scale bar = 1 mm.

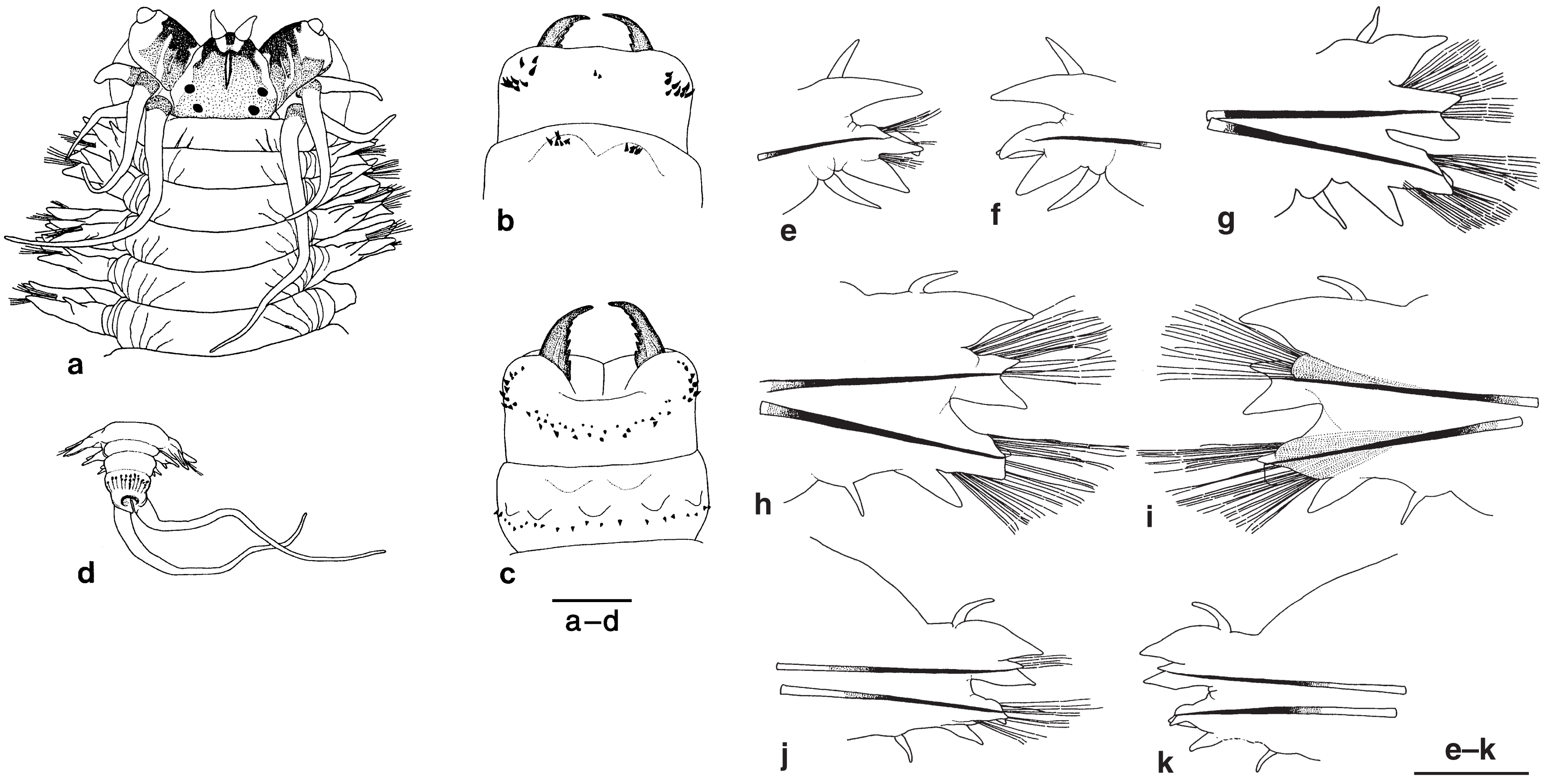


Figure 4. Immature specimen of *Hediste japonica* (nontype, a–d, NSMT-Pol-107441, e–k, CMNH-ZW-1888). (a) Dorsal view of anterior end (everted proboscis not shown). (b) Dorsal view of the everted proboscis. (c) Ventral view of the everted proboscis. (d) Dorsal view of posterior end. (e–k) Parapodia of the left side (chaetae not shown in f, k). (e & f) Anterior and posterior views of parapodium 1. (g) Anterior view of parapodium 10. (h & i) Anterior and posterior views of parapodium 30. (j & k) Anterior and posterior views of parapodium 72. Scale bar = 1 mm.

Proboscis with pair of dark brown jaws, each with 8–9 teeth. Black paragnaths, usually with sharply pointed tip, present on both maxillary and oral rings; paragnath numbers in lectotype (range for all 10 mature specimens in parentheses) as follows. Group I: 2 (1–4); II: 6 on right and 11 on left side in two arched rows, total 17 (10–22); III: 28 (23–30) in transverse band; IV: 10 on right and 11 on left in two arched rows, total 21 (10–21); V: none; VI: 5 on right and 8 on left, in small clusters on gentle mound, total 13 (10–18); VII–VIII: 22 (19–26) in single transverse row. Paragnaths in group II conspicuously larger than the others, with hook-shaped tip. Two gentle mounds and 5 more proximal (on everted proboscis) conical mounds, resembling sometimes papillae, present as transverse rows parallel to paragnath row in group VII–VIII (Fig. 4c).

Parapodia of first 2 setigers uniramous, all following parapodia biramous (Figs 3d–j, 4e–k). Uniramous parapodia with reduced notopodia consisting of dorsal cirrus and superior ligule, with ordinary neuropodia.

Notopodia consisting of dorsal cirrus and three ligules in biramous parapodia, i.e. large superior ligule and upper and lower acicular ligules; all notopodial ligules subtriangular with tapering tip. Upper acicular ligule subequal to lower one in anterior setigers (Figs 3f, 4g); gradually diminishing in size in middle and posterior setigers (Figs 3g–j, 4h–k). Superior ligule thick in anterior setigers, thinner in middle and posterior setigers, most expanded in middle setigers. Dorsal cirri slender, tapering, gradually diminishing in size in posterior setigers.

Neuropodia consisting of ventral cirrus and three ligules throughout, i.e. prechaetal acicular ligule, postchaetal ligule and inferior ligule. Prechaetal acicular ligule and postchaetal ligule conical, of similar length, completely separate in anterior setigers (up to about first 10 setigers), fused at basal parts with their tapering tips remaining in middle and posterior setigers (Figs 3h,j, 4i,k). Inferior ligule conical. Ventral cirrus slender with tapering tip. Inferior ligule and ventral cirrus largest in anterior setigers, gradually diminishing in size in middle and posterior setigers.

Notochaetae all homogomph spinigers (Figs 5a, 6a, 7a,b); terminal sections (blades) very slender with finely serrated cutting edge. Numbers of notochaetae of atokes 10–40 in anterior and middle setigers, and less than 10 in posterior setigers in most specimens.

Neurochaetae in supra-acicular fascicle consisting of homogomph spinigers (around 10 in most cases, Fig. 5b) and heterogomph falcigers (up to 10 in most cases, Figs 5b, 6b, 7c) in anterior setigers. Neuropodial homogomph spinigers similar to notopodial ones in shape and size. Heterogomph falciger relatively thick, with serrated blade present in anterior and middle setigers (anterior 50–60 setigers in adults), and 1–3 thick simple chaetae with tapering tip (Figs 6c, 7d) present instead of heterogomph falcigers in posterior setigers (Fig. 5b). Incomplete simple chaetae found in several setigers (around setiger 40) just prior to posterior setigers with complete simple chaetae in juvenile, indicating formation of simple chaeta by deformation of tip of stalk of heterogomph falciger after detachment of its blade (Fig. 8).

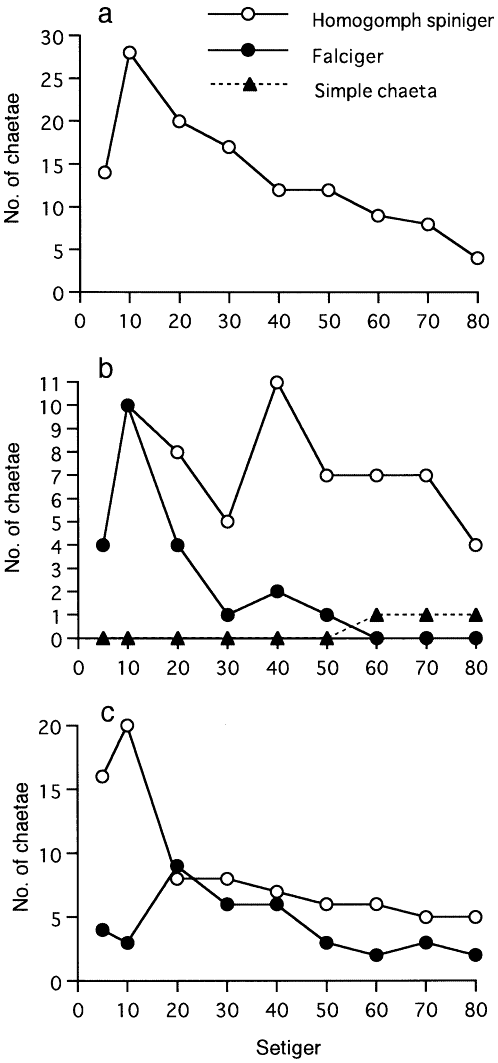


Figure 5. Number of chaetae throughout body of an immature specimen with total 94 setigers (body width 3.5 mm, body length 75 mm) of *Hediste japonica* (nontype, ZIHU-2101): (a) notopodial fascicles, (b) neuropodial supra-acicular fascicles and (c) neuropodial infra-acicular fascicles.

Neurochaetae in infra-acicular fascicle consisting of homogomph spinigers (5–20 in most cases, Figs 5c, 6d) at upper position, homogomph falcigers (up to 20 in most cases, Figs 6e, 7e) on middle portion, and heterogomph falcigers (up to 10 in most cases, Fig. 6f) at lower position; homogomph and heterogomph fal-

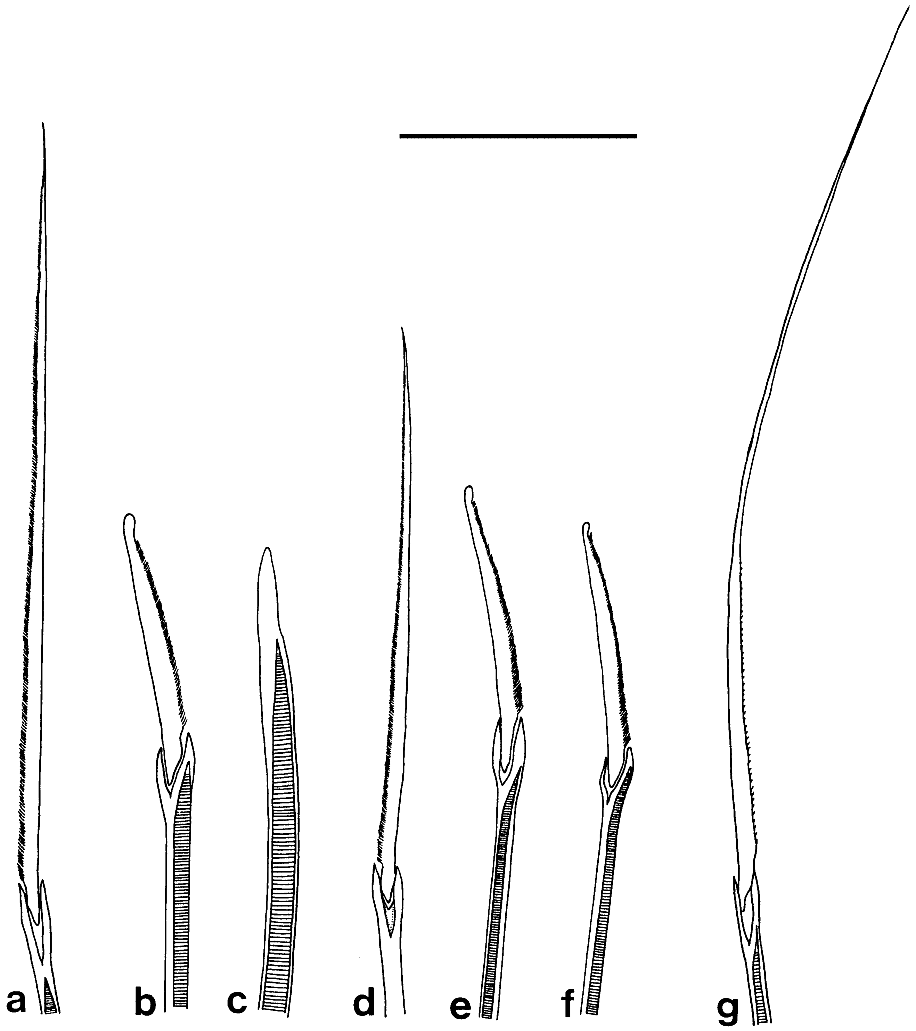


Figure 6. Chaetae of immature specimens (a–f, NSMT-Pol-107441) and a mature male (g, ZIHU-2103) of *Hediste japonica* (nontype with body width of 2.8–3.0 mm). (a) Homogomph spiniger from notopodial fascicle in parapodium 5. (b) Heterogomph falciger from neuropodial supra-acicular fascicle in parapodium 30. (c) Simple chaeta from neuropodial supra-acicular fascicle in parapodium 78. (d) Homogomph spiniger from neuropodial infra-acicular fascicle in parapodium 78. (e) Homogomph falciger from neuropodial infra-acicular fascicle in parapodium 30. (f) Heterogomph falciger from neuropodial infra-acicular fascicle in parapodium 5. (g) Epitokous homogomph spiniger from neuropodial infra-acicular fascicle in parapodium 50. Scale bar = 0.1 mm.

cigers relatively thin with a slender finely serrated blade, often indistinguishable between homogomph and heterogomph.

Acicula black except colourless proximal part; single acicula present in each ramus (occasionally 2 or 3 aciculae present in each ramus).

Pygidium with anus on dorsal side, with pair of cylindrical slender anal cirri (Figs 3c, 4d).

Epitokous metamorphosis at sexually mature stage: body colour changed to green in both males and females. Eyes slightly enlarged (Fig. 9a,b). Parapodial ligules (especially upper and lower acicular ligules of notopodia) enlarged slightly in both males and females (Fig. 9c,d); dorsal and ventral cirri also markedly elongated in males. Homogomph spinigers increasing in number (up to about 100 in notopodial fascicles, and up to about 50 each in neuropodial supra-acicular and infra-acicular fascicles) in some specimens (Figs 6g, 7a,b).

Allozyme pattern of LDH

Two bands of anodal migration, the wider more mobile and the narrower less mobile, were observed in all specimens examined (31 from Isahaya Bay, 88 from Daijugarami, 42 from Omutagawa River) (Fig. 10). No

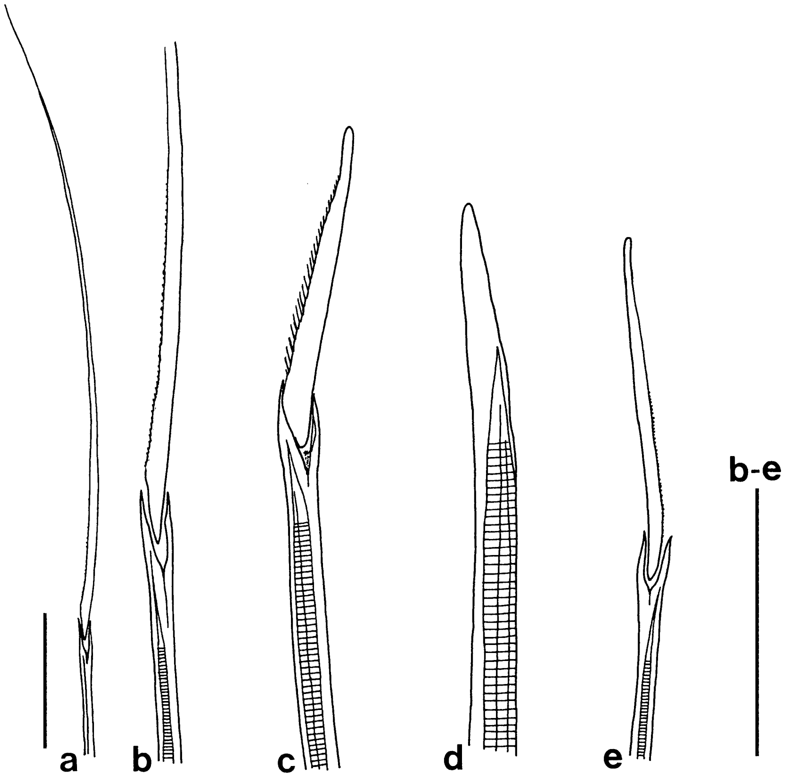


Figure 7. Chaetae of a mature female of *Hediste japonica* (Lectotype, UMUTZ-PLC-IZ-55A). (a) Homogomph spiniger from neuropodial supra-acicular fascicle in parapodium 30. (b) Enlarged part around base of blade of homogomph spiniger in a. (c) Heterogomph falciger from neuropodial supra-acicular fascicle in parapodium 30. (d) Simple chaeta from neuropodial supra-acicular fascicle in parapodium 71. (e) Homogomph falciger from neuropodial infra-acicular fascicle in parapodium 30. Scale bar = 0.1 mm.

variation of band pattern was observed, indicating that the loci for the allozymes of LDH were monomorphic.

Allometry

Body length (BL mm) was well correlated with body width (BW mm) according to the regression formula (Fig. 11a): BL = 29.4 BW − 17.9 ($r^2 = 0.95$, $P < 0.0001$, $n = 51$). Setiger number (SN) was correlated with BW, according to the regression formula with a high gradient in smaller specimens, i.e. juveniles (less than 2 mm in BW) (Fig. 11b): SN = 53.3 BW − 16.3 ($r^2 = 0.64$, $P < 0.0001$, $n = 22$). However, in the larger specimens, i.e. adults (2 mm or more in BW), setiger number was rather constant at around 90–100, according to the regression formula with a low gradient: SN = 4.5 BW + 78.4 ($r^2 = 0.17$, $P = 0.03$, $n = 27$). Maximum body sizes were 125 mm in BL, 4.9 mm in BW, and 108 in SN.

Eye width (EW mm) was correlated with BW according to the regression formula in immature specimens: EW = 0.03 BW + 0.04 ($r^2 = 0.66$, $P < 0.0001$, $n = 46$) (Fig. 12). EW (0.16–0.19 mm) of mature adults ($n = 8$) was significantly larger than EW (0.10–0.16) of immature specimens ($n = 7$) with corresponding BW of 2.1–2.9 mm (Mann–Whitney *U*-test, $P = 0.002$).

Paragnath number (PN) in each group increased rapidly with correlation to increasing body width in juveniles, with a high correlation coefficient and a high gradient of its formula, e.g. PN = 8.3 BW + 0.5 ($r^2 = 0.53$, $P < 0.0001$, $n = 22$) for group II, PN = 26.2 BW − 14.4 ($r^2 = 0.58$, $P < 0.0001$, $n = 21$) for group III, PN = 74.3 BW − 30.3 ($r^2 = 0.58$, $P < 0.0001$, $n = 21$) for all groups, but rather stable in adults, with a low correlation coefficient and a low gradient of its regression formula, e.g. PN = 1.0 BW + 11.7 ($r^2 = 0.06$, $P = 0.01$, $n = 102$) for group II, PN = 4.0 BW + 19.2 ($r^2 = 0.19$, $P < 0.0001$, $n = 101$) for group III, PN = 11.6 BW + 66.4 ($r^2 = 0.26$, $P < 0.0001$, $n = 99$) for all groups (Fig. 13).

Variation of paragnath number

Paragnath numbers in mature and immature adults collected from 8 localities are summarized in Table 2.

Karyotype

Diploid chromosome number was 28 (our unpublished data).

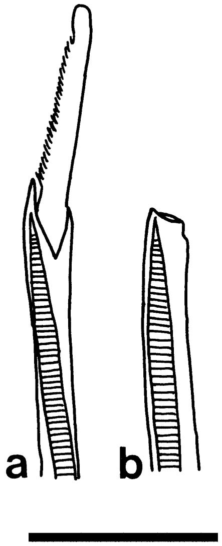


Figure 8. Chaetae of neuropodial supra-acicular fascicle of a juvenile of *Hediste japonica* (nontype with body width of 1.2 mm, ZIHU-2104). (a) Heterogomph falciger in parapodium 28. (b) Incomplete simple chaeta in parapodium 40, probably originated from a stalk of a heterogomph falciger after detach of its blade. (c) Typical simple chaeta in parapodium 50. Scale bar = 0.05 mm.

Reproduction

Reproductive swarming occurred at midnight just after high-tide during a few days throughout the spring tides in December at Kojima Bay, type locality (Izuka, 1908). In the present study, swarming was observed just after high-tide at around 20:30 on 29 December 2001 at Nanaura in the Ariake Sea. In both cases, mature males and females swam near the surface above the tidal flats which are the habitat of immature worms, and shed sperm and eggs there.

Mature or nearly mature adults were collected from the sediment samples in the Omutagawa River on 4 January and 21 December 1999, and only juveniles were collected in March in Daijugarami, indicating that sexual reproduction occurs during late December to January in the Ariake Sea, and that the life span is 1 year. Large specimens collected from Inchon, Korea on 19 January 1999 contained immature oocytes (up to 130 µm in diameter) or clusters of spermatocytes, indicating that sexual reproduction commences in late January or later in Korea.

Gamete morphology and development

The mature eggs (full-grown oocytes) were spherical and 180–210 µm in diameter in the fixed specimens (Fig. 14a). The mean diameter (± SD) of eggs contained in the body cavity of each swarming female was as follows: 187.1 ± 6.3 µm (n = 25) for the lectotype collected from Kojima Bay, 184.2 ± 4.4 µm (n = 14), 188.2 ± 4.8 µm (n = 20) and 188.7 ± 7.7 µm (n = 11) for three nontype specimens collected from the Ariake Sea. The oocytes contained 40–60 oil drops surrounding a germinal vesicle. The spermatozoa had a cone-shaped acrosome at the tip of the head (Fig. 14b). The sperm head was about 2.5 µm wide, about 3.5 µm long, and slightly angular at the base of acrosome.

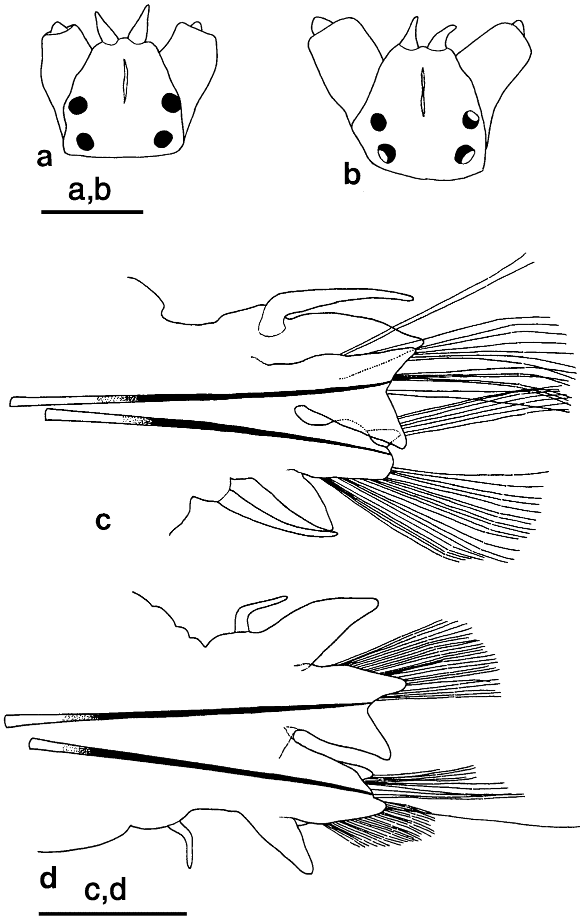


Figure 9. Mature adults of *Hediste japonica* (nontype, ZIHU-2102, 2103). (a & b) Dorsal views of prostomium of male and female. (c) Anterior views of parapodium 26 (left side) of a male. (d) Anterior view of parapodium 28 (left side) of a female. Scale bar = 1 mm.

After embryonic development (*c.* 2 days after fertilization), trochophore larvae swam actively, showing positive phototaxis; about 11 days after fertilization, most larvae with 4 or more setigers sank down to the estuary floor and became benthic (Izuka, 1908).

Habitat

All specimens were collected from muddy tidal flats in estuaries with a wide tidal range (around 3 m or more in the mean tidal range in spring tides). Where the

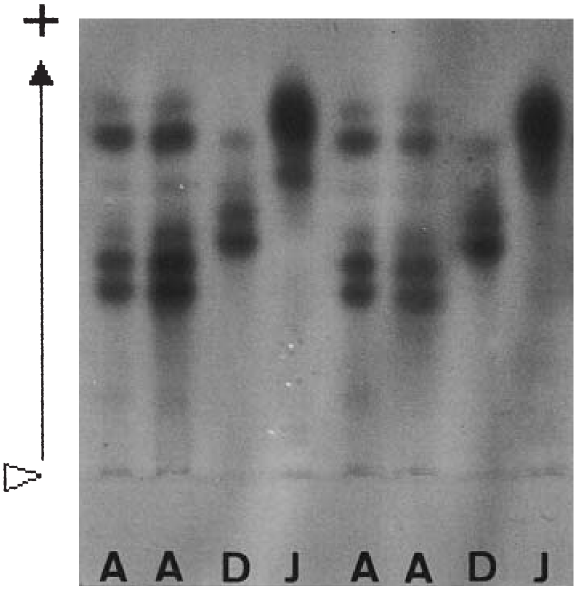


Figure 10. Typical electrophoretic patterns of lactate dehydrogenase in three Asian species of *Hediste*. A, ***H. atoka* sp. nov**. collected from the Omoigawa River; D, ***H. diadroma* sp. nov**. collected from the Omoigawa River; J, *H. japonica* collected from Daijugarami. White arrow indicates origin.

Omutagawa River flows into the Ariake Sea, this species occurred sympatrically with *Hediste diadroma* sp. nov.

Geographical distribution

The coasts of the Yellow Sea in Korea, the Seto Inland Sea and Ariake Sea in Japan (Fig. 15).

Remarks

Hediste japonica is easily distinguishable from all other *Hediste* species due to the presence of homogomph falcigers and the absence of heterogomph spinigers in the neuropodia, the presence of a digitate lobe at the tip of the postchaetal ligules throughout, and the fewer and larger paragnaths in group II.

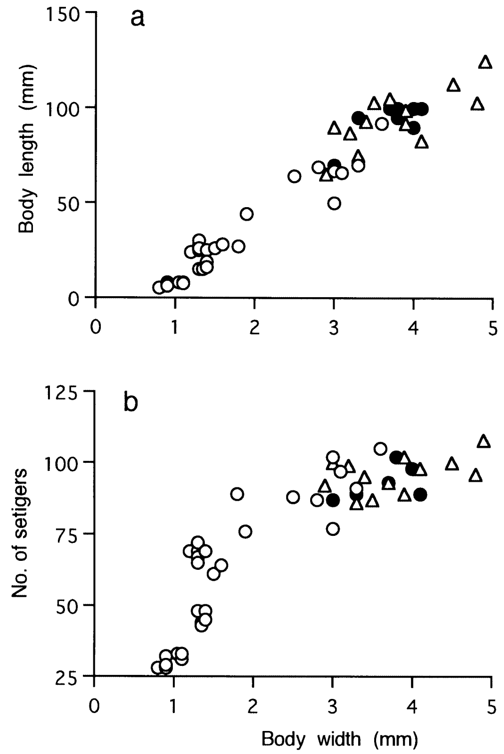


Figure 11. Relationships between body width and body length (a) and between body width and setiger number (b) in *Hediste japonica*. (●) Nine mature specimens collected from Kojima Bay (type series) and Ariake Sea; (○) 29 immature specimens collected from the Ariake Sea; (△) 13 immature specimens collected from Yellow Sea in Korea.

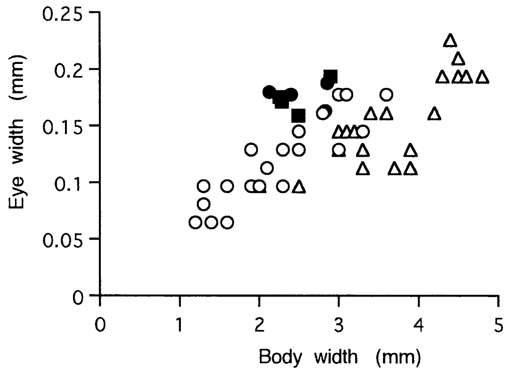


Figure 12. Relationship between body width and eye width in mature (closed symbols) and immature specimens (open symbols) of *Hediste japonica*. Four mature male (■), 4 female (●) and 22 immature specimens (○) were collected from the Ariake Sea; 23 immature specimens (△) were collected from the Yellow Sea in Korea.

Both the original and subsequent descriptions of this species (Izuka, 1908, 1912) are incomplete, presenting little information on chaetal morphology and paragnath number. As for falcigers in the neuropodia, only a typical heterogomph form was described, while the homogomph form was not. These morphological descriptions were probably based on specimens of different species, e.g. *H. diadroma* sp. nov. or *H. atoka* sp. nov. (see below), which were both present in Izuka's collections from the Sumida River and elsewhere (Table 1). It is also strange that the diameter of the mature eggs was described as 140–160 µm in the original description of *H. japonica* (Izuka, 1908). These values are characteristic of *H. diadroma* sp. nov. (see below), and different from those of the eggs remaining in the lectotype (180–200 µm).

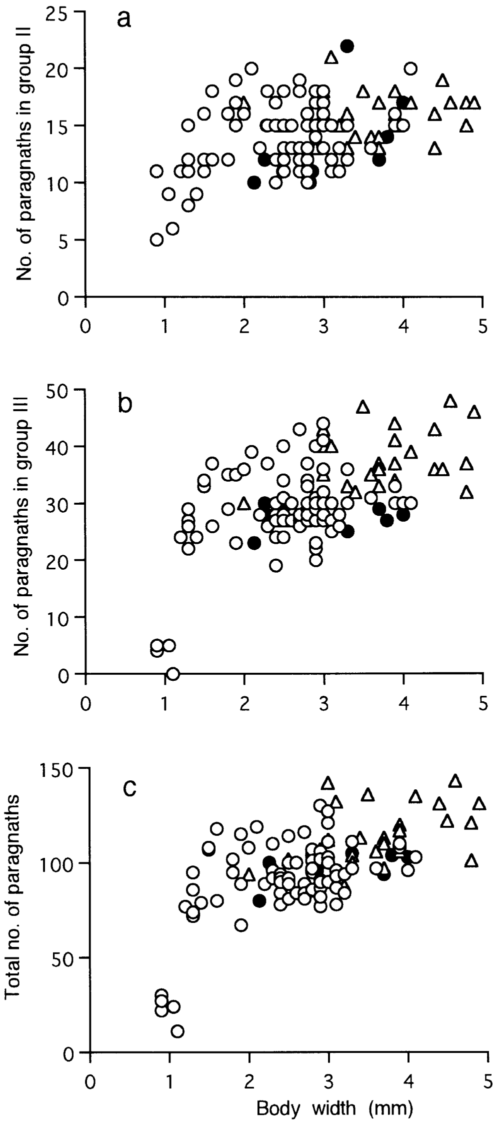


Figure 13. Relationships between body width and paragnath number in (a) group II (total number on right and left sides), (b) group III and (c) all groups in *Hediste japonica*. (●) Mature specimens collected from Kojima Bay (type series) and the Ariake Sea. (○) 86 immature specimens collected from the Ariake Sea. (△) 28 immature specimens collected from the Yellow Sea (Korea).

Our previous studies (Sato & Tsuchiya, 1991; Sato, 1999, 2000, 2001) showed the existence of three cryptic species (the small-egg, large-egg and Ariake forms) in Asian *Hediste*, and judged the first to be *H. japonica sensu stricto* based on its original description (Izuka, 1908), without examination of the type material. However, having examined the type, this judgement is here revised; the Ariake form is identical to *H. japonica sensu stricto*.

Neanthes japonica sensu Smith (1958) collected from two localities of Japan seems to contain two or more species. Smith (1958) detected the presence of homogomph falcigers and an increase in the number of homogomph spinigers in the epitokes of *H. japonica sensu stricto*, which were probably collected from Kojima Bay (type locality). However, the presence of homogomph falcigers was incorrectly regarded as an epitoke-specific characteristic, probably because Smith (1958) examined atokous specimens of different species (*H. diadroma* sp. nov. or *H. atoka* sp. nov.) for comparison with the epitokous ones of *H. japonica*.

Two populations of *Nereis japonica sensu* Khlebovich *et al*. (1982), which were collected from the Yellow Sea, may belong to *H. japonica sensu stricto* because of the small number of paragnaths found in group II (7–8 on the left side), and group IV (8–9 on the left side).

***HEDISTE DIADROMA* SP. NOV.** (FIGS 10, 16–27, 38–43)

Nereis japonica: Kagawa, 1955: 11–16, fig. 5; Okada, 1960: 63–71, pls 1–4.

Neanthes japonica: Sun *et al*., 1980: 100–110, figs 1–3; Sato & Osanai, 1986: 263–270, figs 1–7; Qiu & Wu, 1993: 360–367, figs 1, 2.

Small-egg type of *Neanthes japonica*: Sato & Tsuchiya, 1987: 29–42, figs 1–5; Sato & Tsuchiya, 1991: 371–382, figs 1, 2 and 4.

Small-egg form of *Neanthes japonica*: Sato & Ikeda, 1992: 299–307, figs 4, 7.

Small-egg form of *Hediste japonica*: Sato & Masuda, 1997: 163–170; Sato, 1999: 129–143, figs 1–13.

Hediste japonica: Sato, 2001: 66–86, Rouse & Pleijel, 2001: pl. 5c.

Type material

Holotype (NSMT-Pol-H 456): complete mature swarming female (BL: 52 mm, BW: 2.8 mm, 94 setigers), mouth of Omoigawa River, Aira-cho, Kagoshima Prefecture, 25 February 1986, coll. M. Sato. Paratypes: mature swarming 1 male (ZIHU-2093) and 1 female (ZIHU-2092) (BL: 37–50 mm, BW: 1.8–2.2 mm, 80–85 setigers), data as for holotype; mature swarming 5 males and 6 females (BL: 42–65 mm, BW: 2.3–3.3 mm, 84–98 setigers), Omoigawa River, Aira-cho, Kagoshima Prefecture, 19 February 1988, coll. M. Sato (CMNH-ZW-1891, MNHN-Poly-1368, 1369, NSMT-Pol-P457, OMNH-Iv-4228, SMF-12067, 12068, USNM-1008438, 1008439, ZMUC-Pol-1504, 1507); mature swarming 1 female (BL: 101 mm, BW: 4.6 mm, 88 setigers), Yahatagawa River, Itsukaichi-cho, Hiroshima Prefecture, 9 January 1982, coll. M. Sato (ZIHU-2094); mature swarming 2 males (BL: 37 mm, BW: 2.4–2.5 mm, 84 setigers), Kyobashigawa, Otagawa River, Hiroshima-shi, Hiroshima Prefecture, 9 January 1981, coll. M. Sato (ZIHU-2095); mature swarming 5 females (BL: 60–85 mm, BW: 3.3–4.7 mm, 78–105 setigers), Nanakitagawa River, Gamo, Sendai-

Table 2. Variation of paragnath numbers in mature (two localities) and immature specimens (six localities) of *Hediste japonica*

Locality	*n*	Body width (mm)	Mean paragnath number ± SD (range)						
			I	II[1]	III	IV[1]	VI[1]	VII–VIII	Total
MATURE									
Seto Inland Sea									
Kojima Bay (1906)	4	3.3–4.0	3.0 ± 0.8 (2–4)	16.3 ± 4.3 (12–22)	27.3 ± 1.7 (25–29)	19.0 ± 2.2 (16–21)	14.0 ± 3.4 (10–18)	22.0 ± 2.2 (19–24)	101.5 ± 5.1 (94–105)
Ariake Sea									
Nanaura (2001)	6	2.1–2.9	1.7 ± 0.8 (1–3)	11.5 ± 1.9 (10–15)	27.8 ± 2.6 (23–30)	13.7 ± 2.6 (10–16)	13.0 ± 1.7 (10–14)	23.2 ± 2.5 (20–26)	90.2 ± 8.1 (80–100)
IMMATURE									
Ariake Sea									
Isahaya Bay (1996)	31	2.0–3.0	2.2 ± 1.6 (0–7)	15.6 ± 3.0 (11–22)	26.3 ± 8.1 (4–39)	19.8 ± 2.9 (15–26)	13.9 ± 3.3 (7–20)	20.8 ± 4.7 (8–28)	98.6 ± 17.3 (53–122)
Rokkakugawa (1995)	6	2.0–3.0	2.5 ± 0.5 (2–3)	17.7 ± 1.5 (16–20)	38.7 ± 3.1 (36–44)	19.0 ± 2.1 (17–22)	13.2 ± 2.1 (10–15)	22.8 ± 2.1 (20–25)	113.8 ± 5.2 (108–121)
Daijugaram (1995)	88	2.7–3.6	1.7 ± 0.9 (1–5)	13.5 ± 2.8 (6–21)	26.1 ± 5.1 (16–38)	16.9 ± 3.3 (10–26)	12.9 ± 2.8 (5–21)	20.8 ± 4.4 (10–39)	92.0 ± 11.3 (58–121)
Omutagawa (1999)	37	2.2–4.1	1.8 ± 1.0 (1–4)	13.6 ± 2.2 (10–20)	27.6 ± 2.9 (20–34)	18.7 ± 2.3 (12–23)	12.1 ± 1.8 (9–15)	18.4 ± 3.1 (11–26)	92.3 ± 8.4 (78–110)
Korea									
Inchon (1999)	11	3.0–4.9	4.2 ± 1.8 (1–7)	16.7 ± 1.6 (13–19)	40.4 ± 5.3 (32–48)	23.5 ± 3.6 (17–28)	17.5 ± 3.5 (12–23)	25.5 ± 3.6 (19–30)	127.8 ± 12.6 (101–143)
Kunsan (2000)	17	2.0–3.9	2.2 ± 1.1 (1–5)	15.5 ± 2.1 (13–21)	34.5 ± 4.8 (27–44)	21.2 ± 3.4 (16–27)	14.6 ± 2.5 (9–19)	19.2 ± 3.4 (11–25)	107.2 ± 10.4 (88–132)

[1] Total number on right and left sides of proboscis.

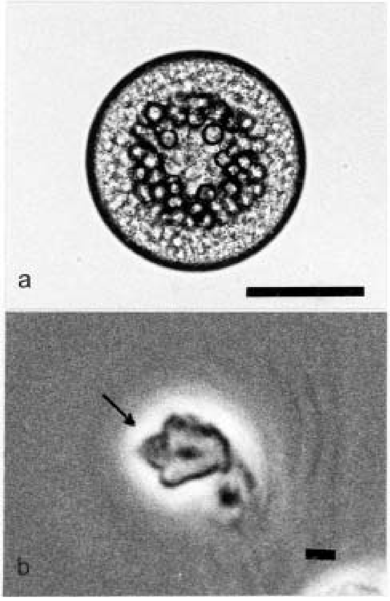


Figure 14. Gametes of *Hediste japonica* collected from the Ariake Sea. (a) Micrograph of a full-grown oocyte. Scale bar = 100 μm. (b) Phase-contrast micrograph of a sperm head with a conical acrosome (arrow). Scale bar = 1 μm.

shi, Miyagi Prefecture, 6 April 1981, coll. M. Sato (ZIHU-2097); mature swarming 2 males (BL: 50–65 mm, BW: 2.5–3.0 mm, 98 setigers), Usujiri, Minamikayabe-cho, Hokkaido, 17 June 1983, coll. K. Yokouchi (ZIHU-2096).

Other material examined

Epitokes collected during reproductive swarming: Usujiri, Minamikayabe-cho, Hokkaido, 28 June 1983, coll. K. Yokouchi, 1 specimen. Nanakitagawa River, Gamo, Sendai-shi, Miyagi Prefecture, 6 April 1981, coll. M. Sato, 15 specimens; 26 April 1994, coll. M. Ikeda, 52 specimens. Tonegawa River, Hagiwara, Chiba Prefecture, 23 March 1966, coll. unknown, 14 specimens. Fujimae, Shinkawa River, Nagoya-shi, Aichi Prefecture, 28 February 2002, coll. S. Sano *et al.* 12 specimens. Kyobashigawa, Otagawa River, Hiroshima-shi, Hiroshima Prefecture, 7–9 January 1981, coll. M. Sato, 13 specimens. Yahatagawa River, Itsukaichi-cho, Hiroshima Prefecture, 7 January 1981, coll. O. Kawamoto & M. Sato, 26 specimens; 9 January 1982, coll. M. Sato, 5 specimens. Nakagawa River, Fukuoka-shi, Fukuoka Prefecture, 3, 4 March 1980, coll. M. Sato, 40 specimens. Amorigawa River,

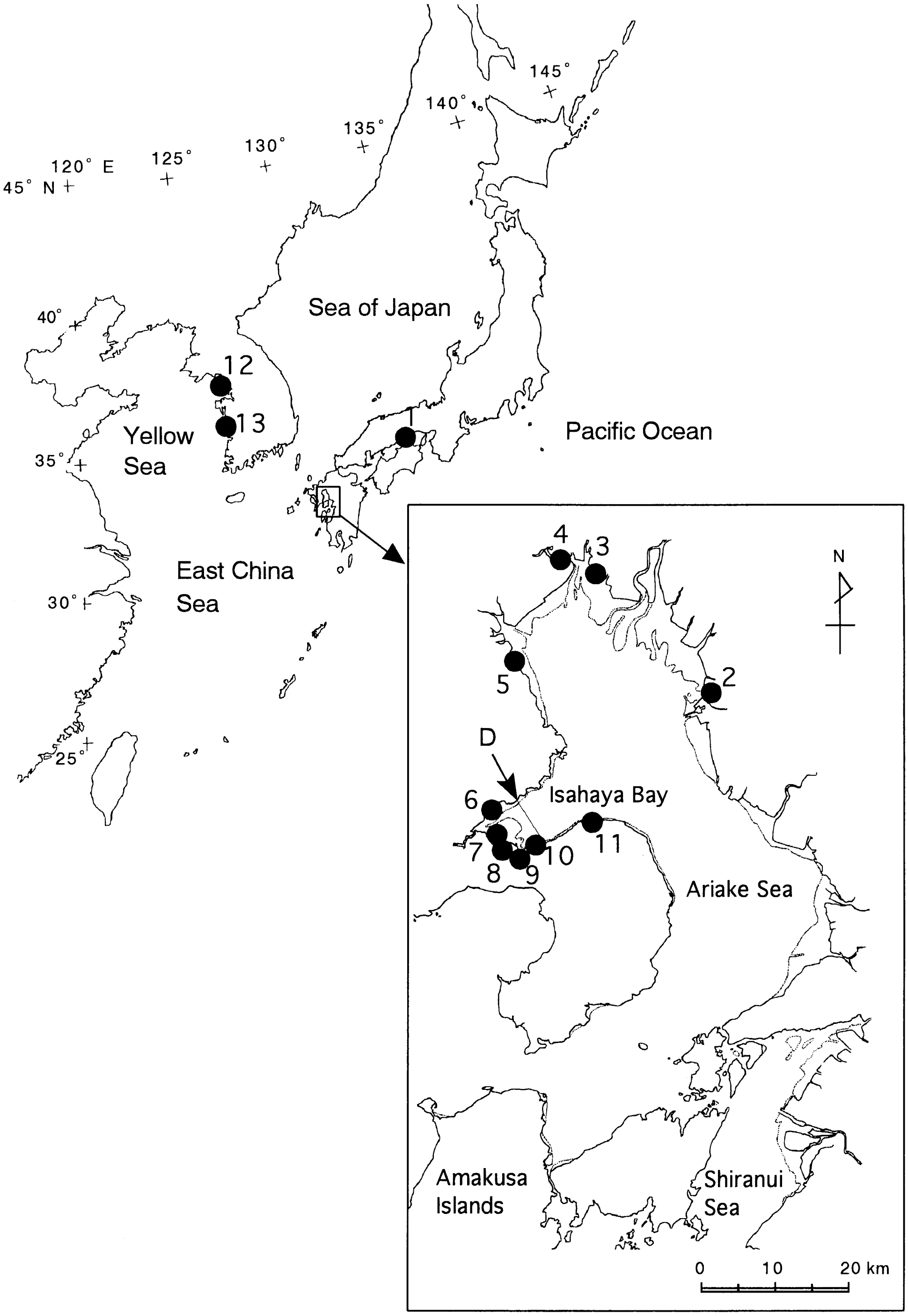


Figure 15. Distribution of *Hediste japonica*. 1. Kojima Bay (type locality). 2. Omutagawa River. 3. Daijugarami. 4. Rokkakugawa River. 5. Nanaura. 6. Honmyogawa River. 7. Onojima. 8. Nitandagawa River. 9. Ariakegawa River. 10. Yamadagawa River. 11. Kojirogawa River. 12. Yongyudo Island, Inchon. 13. Kumu River, Kunsan. D. Reclamation dyke, 7 km. All *Hediste* populations within the inner part of Isahaya Bay (6–10) were lost upon completion of dyke construction in April 1997. The habitat in the inner part of Kojima Bay (1, type locality) was also lost in 1959 when the land was reclaimed and a freshwater pond appeared due to the construction of a dyke similar to that of Isahaya Bay.

Hayato-cho, Kagoshima Prefecture, 21 February 1989, coll. M. Ikeda, 14 specimens. Byugawa River, Aira-cho, Kagoshima Prefecture, 21 February 1989, coll. M. Ikeda, 12 specimens; 13 March 1994, coll. M. Sato, 40 specimens. Omoigawa River, Aira-cho, Kagoshima Prefecture, 25 February 1986, coll. M. Sato, 77 specimens; 2 March 1987, coll. M. Sato, 1 specimen; 4, 18 March, 17 April 1988, coll. M. Sato & M. Ikeda, 23 specimens; 21, 22 February, 8, 9 March 1989, coll. M. Ikeda, 36 specimens; 1 March 1991, coll. M. Sato, 11 specimens. Kotsukigawa River, Kagoshima-shi, 22 March 1985, coll. K. Kusamura, 1 specimen; 25, 26 February 1986, coll. M. Sato *et al.* 6 specimens. Nagatagawa River, Taniyama, Kagoshima-shi, 20 February 1989, coll. M. Sato & M. Ikeda, 44 specimens; 18, 20 February, 6 March 1992, coll. M. Sato, 30 specimens. Kaminokawa River, Higashi-ichiki-cho, Kagoshima Prefecture, 22 February 1989, coll. M. Sato, 2 specimens; 12 February, 5 March 1992, coll. M. Sato, 10 specimens. Isakugawa River, Iriki-cho, Kagoshima Prefecture, 22 February 1989, coll. M. Ikeda, 7 specimens. Manosegawa River, Kaseda-shi, Kagoshima Prefecture, 22 February 1989, coll. M. Ikeda, 20 specimens; 14 March 1994, coll. M. Sato, 3 specimens.

Epitokes obtained after rearing in laboratory: Kominatogawa River, Asadokoro, Aomori Prefecture, fixed 10 February 1984 after rearing for 2 months, coll. M. Sato, 1 specimen. Natorigawa River, Yuriage, Miyagi Prefecture, fixed 15 April 1980 after rearing for 5 months, coll. M. Sato, 9 specimens.

Immature specimens (atokes): Kusamigawa River, Kitakyushu-shi, Fukuoka Prefecture, 30 March 1998, coll. M. Sato, 13 specimens. Wajirogawa River, Fukuoka-shi, Fukuoka Prefecture, 31 March 1998, coll. M. Sato, 1 specimen. Domengawa River, Omuta-shi, Fukuoka Prefecture, 24 November 1998, 4 January 1999, coll. K. Ichimiya, 50 specimens. Omutagawa River, Omuta-shi, Fukuoka Prefecture, 24 November 1998, 4 January 1999, coll. K. Ichimiya, 11 specimens. A small creek at Kobe, Mizuho-cho, Nagasaki Prefecture, 28 July 1998, coll. M. Sato, 13 specimens. Kikuchigawa River, Tamana-shi, Kumamoto Prefecture, 14 September 1996, coll. A. Nakashima, 53 specimens. Maekawa River, Yatsushiro-shi, Kumamoto Prefecture, 25 August 1998, coll. K. Ichimiya, 22 specimens. Omoigawa River, Aira-cho, Kagoshima Prefecture, 24 November 1995, 2 June, 28 September, 25 October, 25 November 1996, coll. A. Nakashima, 16 specimens; laboratory-bred worms fixed 4–48 months after fertilization at 23 April 1988, 9, 17 April 1989, 28 March 1990, coll. M. Sato, 14 specimens. Mangrove forest (northern limit in Pacific) at Atagogawa River, Kiire-cho, Kagoshima Prefecture, 2 July 1996, coll. A. Nakashima, 7 specimens.

Diagnosis

Moderate number (10–20 in most cases) of paragnaths on both right and left sides of proboscis in group II. Homogomph falciger absent, heterogomph spinigers present in neuropodia. Neuropodial postchaetal ligule tapering to digitate lobe only in anterior setigers (up to setigers 15–25). At epitokous stage, delicate epitoke-specific sesquigomph spinigers added in all notopodial and neuropodial fascicles. Full-grown oocytes 130–170 µm in diameter. Reproductive swarming in winter or early spring.

Description

Body stout anteriorly, posteriorly gradually tapering toward pygidium (Fig. 16a,d). Dorsum convex, venter relatively flat with longitudinal midventral groove. Colour in preserved specimens whitish cream with dark brown pigmentation on anterior dorsal surface; colour in life at sexually immature stage reddish brown.

Prostomium pyriform, broader than long, with pair of tapered antennae situated at anterior end. Pair of palps with massive palpophores about twice as long as antennae and short round palpostyles. Two pairs of round or oval eyes almost equal in size, arranged trapezoidally (the space between the anterior pair about 1.3 times as wide as that between the posterior pair). Longitudinal mid-dorsal groove present on anterior dorsum of prostomium. Partial dark pigmentation present on dorsal anterior surface of prostomium and palpophore (Fig. 16a).

Peristomium nearly as long as following setigers, with 4 pairs of tentacular cirri of unequal length; posterior dorsal tentacular cirri longest, reaching back to setigers 6–12; anterior dorsal tentacular cirri next longest, reaching back to setigers 3–4.

Proboscis with pair of dark brown jaws, each with 6–9 teeth (Fig. 16b,c). Black paragnaths, usually with pointed tip, present on both maxillary and oral rings; paragnath numbers in holotype (range for all 188 mature specimens in parentheses) as follows. Group I: 2 (1–8); II: 16 on right and 15 on left sides in two or three arched rows each, total 31 (18–52); III: 37 (22–61) in transverse band; IV: 24 on both sides in three arched rows each, total 48 (25–67); V: none; VI: 6 on both sides in small clusters, total 12 (3–17); VII–VIII: 26 (11–41) in single transverse row.

Parapodia of first 2 setigers uniramous, all following parapodia biramous (Fig. 16e–l). Uniramous parapodia with reduced notopodia consisting of dorsal cirrus and superior ligule, and with ordinary neuropodia.

Notopodia consisting of dorsal cirrus and three ligules in biramous parapodia, i.e. large superior ligule and upper and lower acicular ligules; all notopodial ligules subtriangular with tapering tip. Upper acicular ligule subequal to lower one in anterior

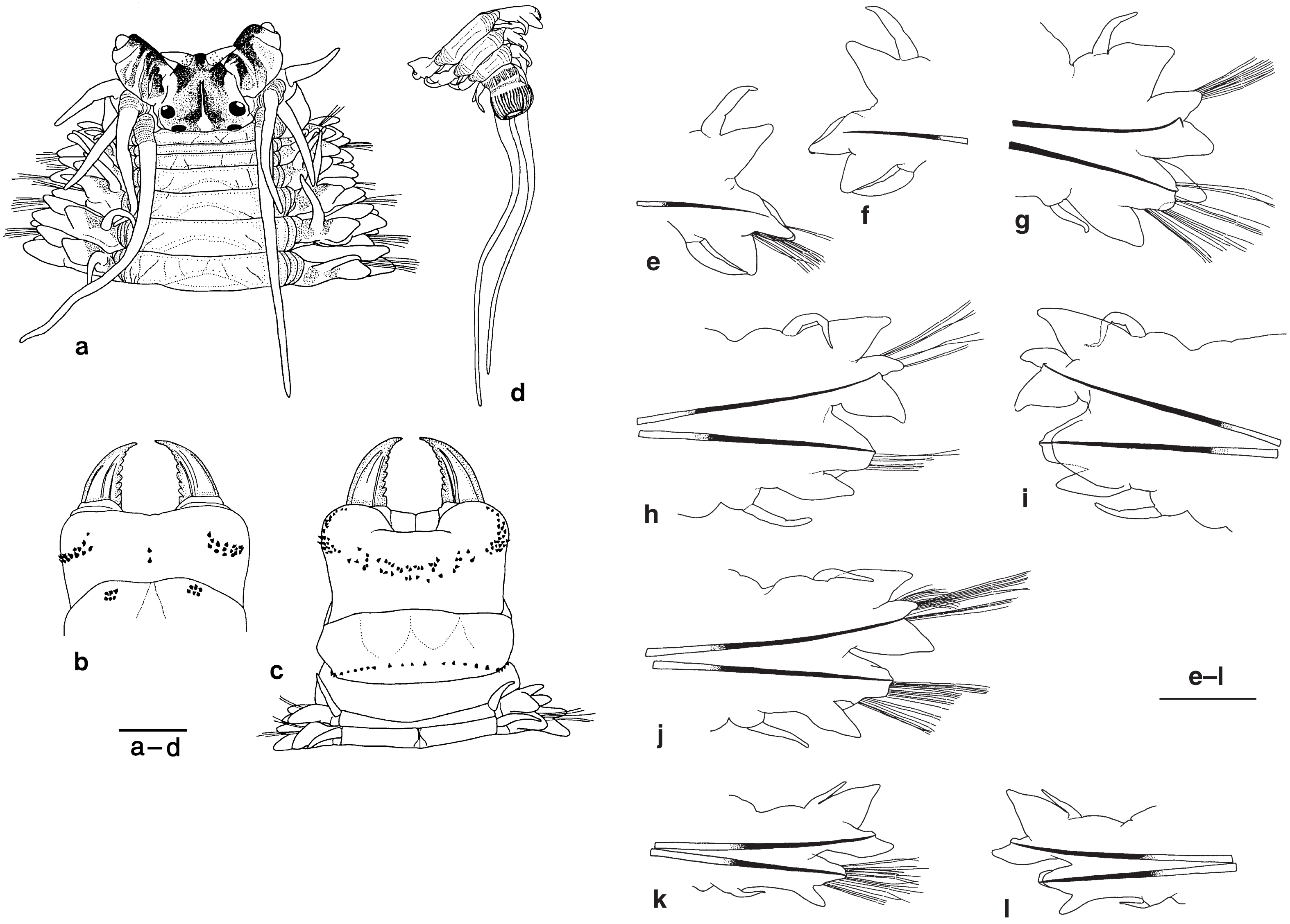


Figure 16. Epitokous female of ***Hediste diadroma* sp. nov**. (holotype, NSMT-Pol-H456). (a) Dorsal view of anterior end; everted proboscis not shown here. (b & c) Dorsal and ventral views of the everted proboscis. (d) Dorsal view of Posterior end. (e–l) Parapodia of the left side (chaetae not shown in f, i, l). (e & f) Anterior and posterior views of parapodium 2. (g) Anterior view of parapodium 10. (h & i) Anterior and posterior views of parapodium 32. (j) Anterior view of parapodium 41. (k & l) Anterior and posterior views of parapodium 72. Scale bar = 1 mm.

setigers (Fig. 16g), gradually diminishing in size in middle setigers and reduced to a small protrusion in posterior setigers (Fig. 16h–l). Superior ligule thick in anterior setigers, thinner in middle and posterior setigers (from around setiger 15), most expanded in middle setigers. Dorsal cirri slender, tapering, gradually diminishing in size in posterior setigers.

Neuropodia consisting of ventral cirrus and three ligules throughout, i.e. prechaetal acicular ligule, postchaetal ligule and inferior ligule. Prechaetal acicular ligule and postchaetal ligule conical with tapering tip, of similar lengths, completely separate in anterior setigers (up to about first 10 setigers, Fig. 16e–g), fused at following setigers; tapering tip of postchaetal ligule diminishing to digitate lobe, present up to around setiger 20, and absent in following ones (Fig. 16i,l); prechaetal acicular ligule with blunt tip. Inferior ligule conical. Ventral cirrus slender with tapering tip. Inferior ligule and ventral cirrus gradually diminishing in size in posterior setigers.

Notochaetae all homogomph spinigers, neurochaetae homogomph and heterogomph spinigers, heterogomph falcigers and simple chaetae, except addition of epitoke-specific chaetae to all notopodial and neuropodial fascicles in mature adults (see below) (Table 3).

Notopodial homogomph spiniger with slender serrated blade (Fig. 17a,b), 10–50 in number in most setigers, and less than 10 in anteriormost and posteriormost setigers (Fig. 18a).

Neurochaetae of supra-acicular fascicle consisting of homogomph spinigers (up to 20, Fig. 18b) and heterogomph falcigers (up to 10, Fig. 19b) in anterior and middle setigers (anterior 40–50 setigers in adults). Neuropodial homogomph spinigers similar to notopodial ones in shape and size. Heterogomph falcigers with serrated blade; 1–3 thick simple chaetae with tapering tip present instead of heterogomph falcigers in posterior setigers (Figs 17f, 19a).

Neurochaetae of infra-acicular fascicle consisting of homogomph spinigers (up to 15, Fig. 18c) at upper position, heterogomph spinigers (up to 5, Figs 17c,d, 18d) in middle portion, and heterogomph falcigers (up to 40, Figs 17e, 19c) at lower position.

Aciculae black except colourless proximal part; single acicula present in each ramus (occasionally 2 aciculae present in each ramus).

Pygidium with anus on dorsal side, with pair of cylindrical slender anal cirri (Fig. 16d).

Epitokous metamorphosis at sexually mature stage: body wall becoming thin and transparent; ripe eggs

Table 3. Numbers of chaetae in parapodia of setigers 10–11 and 30–31 of mature females (F, $n = 5$) and males (M, $n = 5$) in ***Hediste diadroma* sp. nov.** collected from Omoigawa River and ***H. atoka* sp. nov.** collected from Niidagawa River. *Abbreviations:* Ho-S, homogomph spiniger; He-S, heterogomph spiniger; Fal, heterogomph falciger; Ep, epitoke-specific sesquigomph spiniger

	Notopodial fascicle		Neuropodial supra-acicular fascicle			Neuropodial infra-acicular fascicle			
	Ho-S	Ep	Ho-S	Fal	Ep	Ho-S	He-S	Fal	Ep
S10–11									
H. diadroma									
F	18.4 ± 9.4	0	6.8 ± 4.3	1.8 ± 1.1	0	8.0 ± 5.9	0.8 ± 1.1	7.0 ± 4.1	0
M	18.6 ± 11.5	0	5.2 ± 4.9	3.8 ± 3.6	0	7.6 ± 5.1	1.8 ± 0.8	7.6 ± 5.3	0
Diff.[1]	ns		ns	ns		ns	ns	ns	
H. atoka									
F	26.0 ± 15.8	0	14.8 ± 6.4	6.6 ± 2.3	0	13.6 ± 3.6	3.2 ± 1.1	21.4 ± 4.8	0
M	42.2 ± 7.0	0	20.6 ± 3.0	8.2 ± 1.9	0	23.6 ± 4.3	3.0 ± 2.5	17.8 ± 4.4	0
Diff.[1]	ns		ns	ns		**	ns	ns	
S30–31									
H. diadroma									
F	18.4 ± 5.8	18.6 ± 17.2	11.8 ± 3.1	1.4 ± 0.6	3.8 ± 5.9	1.4 ± 1.3	0.8 ± 1.3	14.2 ± 5.8	13.8 ± 5.0
M	17.2 ± 4.3	33.8 ± 17.3	9.2 ± 1.3	2.4 ± 1.3	5.0 ± 3.6	2.6 ± 1.5	0.2 ± 0.4	16.2 ± 2.5	30.0 ± 9.0
Diff.[1]	ns	ns	ns	ns	ns	ns	ns	ns	*
H. atoka									
F	15.8 ± 7.0	0	11.4 ± 3.9	3.4 ± 1.7	0	2.4 ± 1.8	1.4 ± 1.1	18.2 ± 6.3	0
M	20.4 ± 8.4	0	18.4 ± 3.5	6.2 ± 1.6	0	4.2 ± 3.5	0.8 ± 0.8	24.4 ± 5.4	0
Diff.[1]	ns		*	*		ns	ns	ns	

[1] Significance of difference between females and males was tested by Mann–Whitney *U*-test; ns, not significant ($P > 0.05$), *significant ($P < 0.05$), **significant ($P < 0.01$).

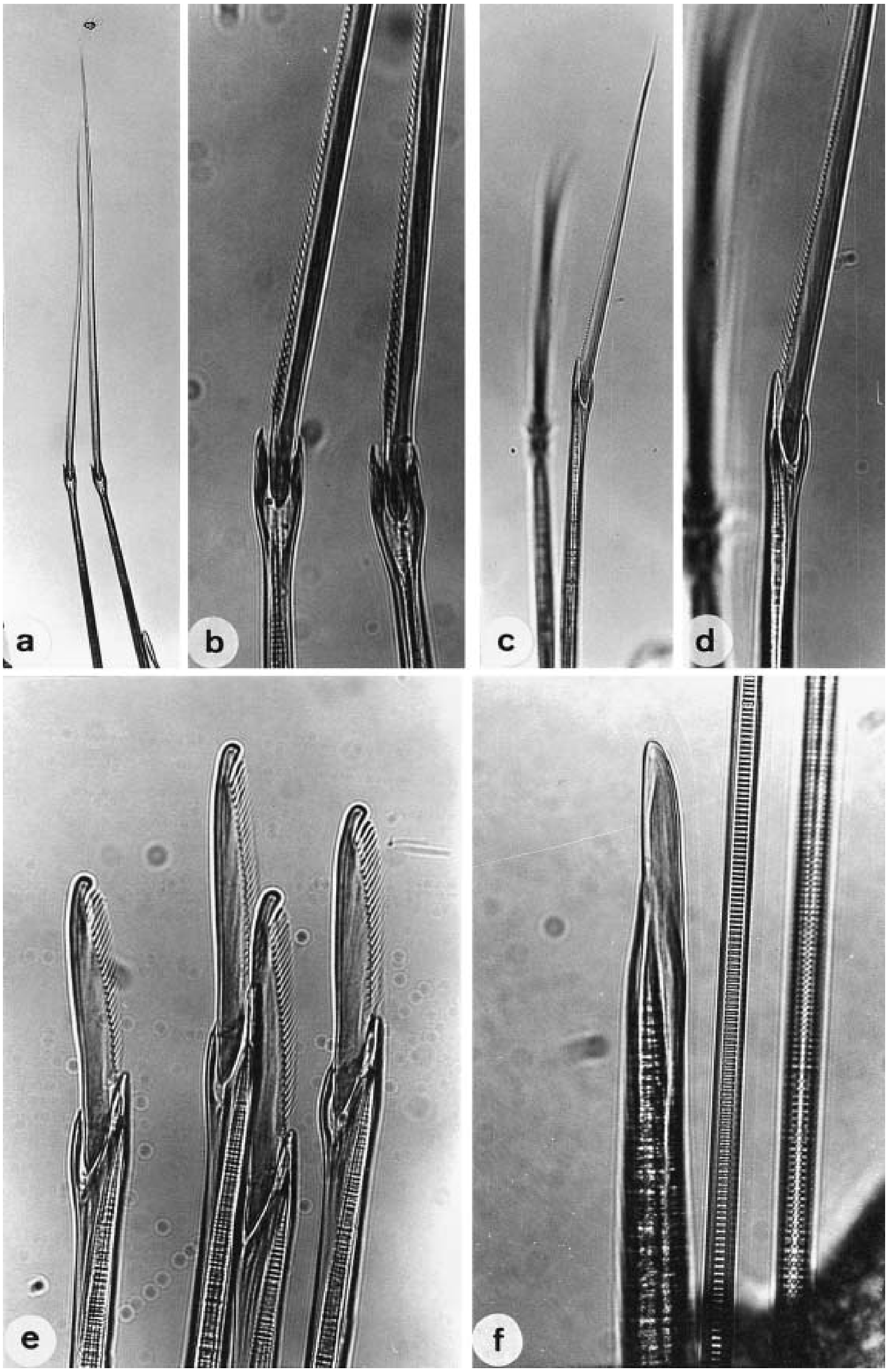


Figure 17. Chaetae of an epitokous female of ***Hediste diadroma* sp. nov**. (paratype, ZIHU-2092). (a) Homogomph spinigers from notopodial fascicle in parapodium 71. ×40. (b) Enlargement of base of blade in a. ×580. (c) Heterogomph spiniger from neuropodial infra-acicular fascicle in parapodium 3. ×290. (d) Enlargement of base of blade in c. ×580. (e) Heterogomph falcigers from neuropodial infra-acicular fascicle in parapodium 16. ×580. (f) Simple chaeta from neuropodial supra-acicular fascicle in parapodium 75. ×580.

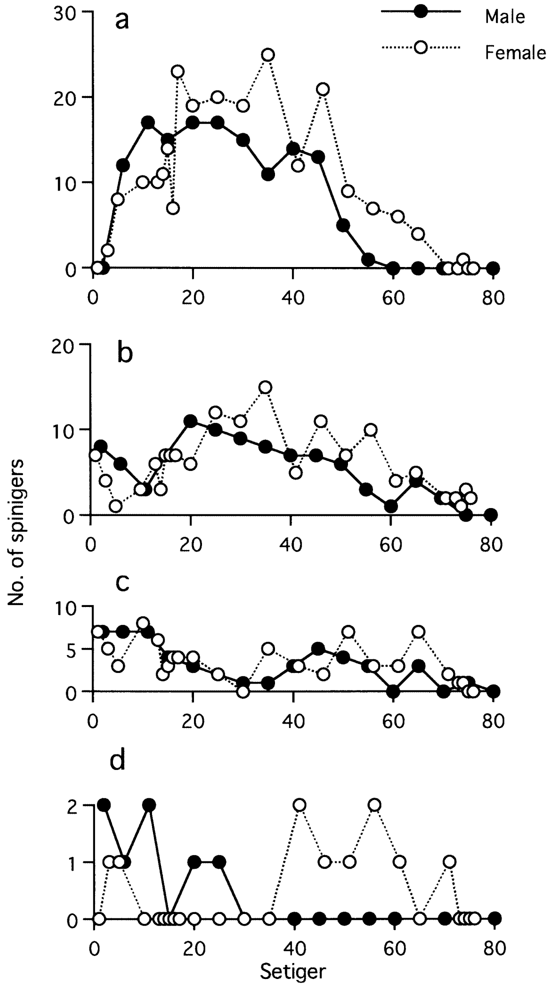


Figure 18. Number of spinigers in each parapodium throughout body of a mature male (●, BL 37 mm with 85 setigers, paratype, ZIHU-2093) and a mature female (○, BL 50 mm with 80 setigers, paratype, ZIHU-2092) of ***Hediste diadroma* sp. nov**. Homogomph spinigers in (a) notopodial fascicle, (b) neuropodial supra-acicular fascicle and (c) neuropodial infra-acicular fascicle. (d) Heterogomph spinigers in neuropodial infra-acicular fascicle.

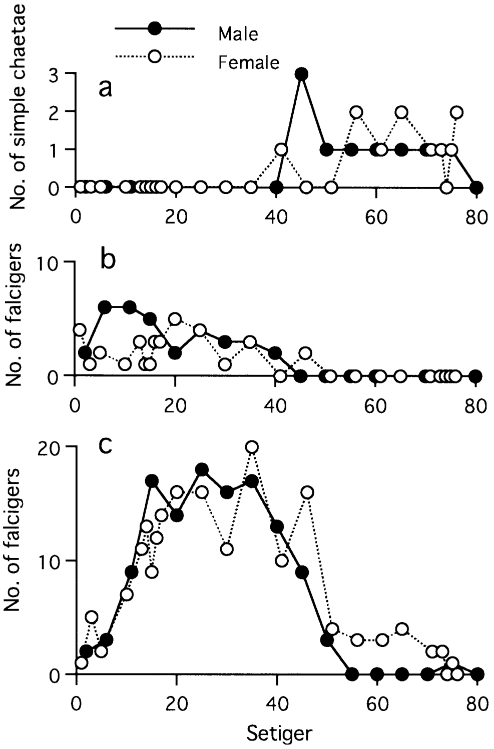


Figure 19. Number of simple chaetae and falcigers in each parapodium throughout body of a mature male (●) and a mature female (○) of ***Hediste diadroma* sp. nov.** (same specimens as those shown in Fig. 18). (a) Simple chaetae in neuropodial supra-acicular fascicle. (b) Heterogomph falcigers in neuropodial supra-acicular fascicle. (c) Heterogomph falcigers in neuropodial infra-acicular fascicle.

(fully mature oocytes) green colour in females, sperm whitish in males (Sato & Tsuchiya, 1987; Sato, 1999). Eyes slightly enlarged in both males and females. Parapodial ligules (especially upper and lower acicular ligules of notopodia) enlarged slightly in both males and females; dorsal and ventral cirri also elongated in males (Fig. 20). Delicate transparent epitoke-specific sesquigomph (intermediate between homogomph and heterogomph) spinigers with less serrated blade added posterior to all fascicles of atokous chaetae in notopodia (up to 60) and in neuropodia (up to 60) in middle setigers in both males and females (Figs 21, 22; Table 3). Number of epitoke-specific spinigers in neuropodial infra-acicular fascicle more abundant in males than in females.

Allozyme pattern of LDH

Total of 309 specimens from 12 localities was examined electrophoretically. Three conspicuous bands of anodal migration were observed in most specimens, with two bands of lower mobility closer together (Fig. 10). Two loci relating to the LDH pattern seemed to be almost monomorphic (the frequency of the dominant allele exceeded 0.9 for each locus) (Sato & Masuda, 1997).

Allometry

Body length (BL mm), wet weight before shedding gametes (WW g) and setiger number (SN) in mature adults correlated with body width (BW mm) according to the following regression formulae (Fig. 23): $BL = 24.9\ BW - 8.1$ ($r^2 = 0.65$, $P < 0.0001$, $n = 193$), $WW = 0.1\ BW^{2.1}$ ($r^2 = 0.61$, $P < 0.0001$, $n = 46$), $SN = 4.1\ BW + 75.0$ ($r^2 = 0.12$, $P = 0.0006$, $n = 96$).

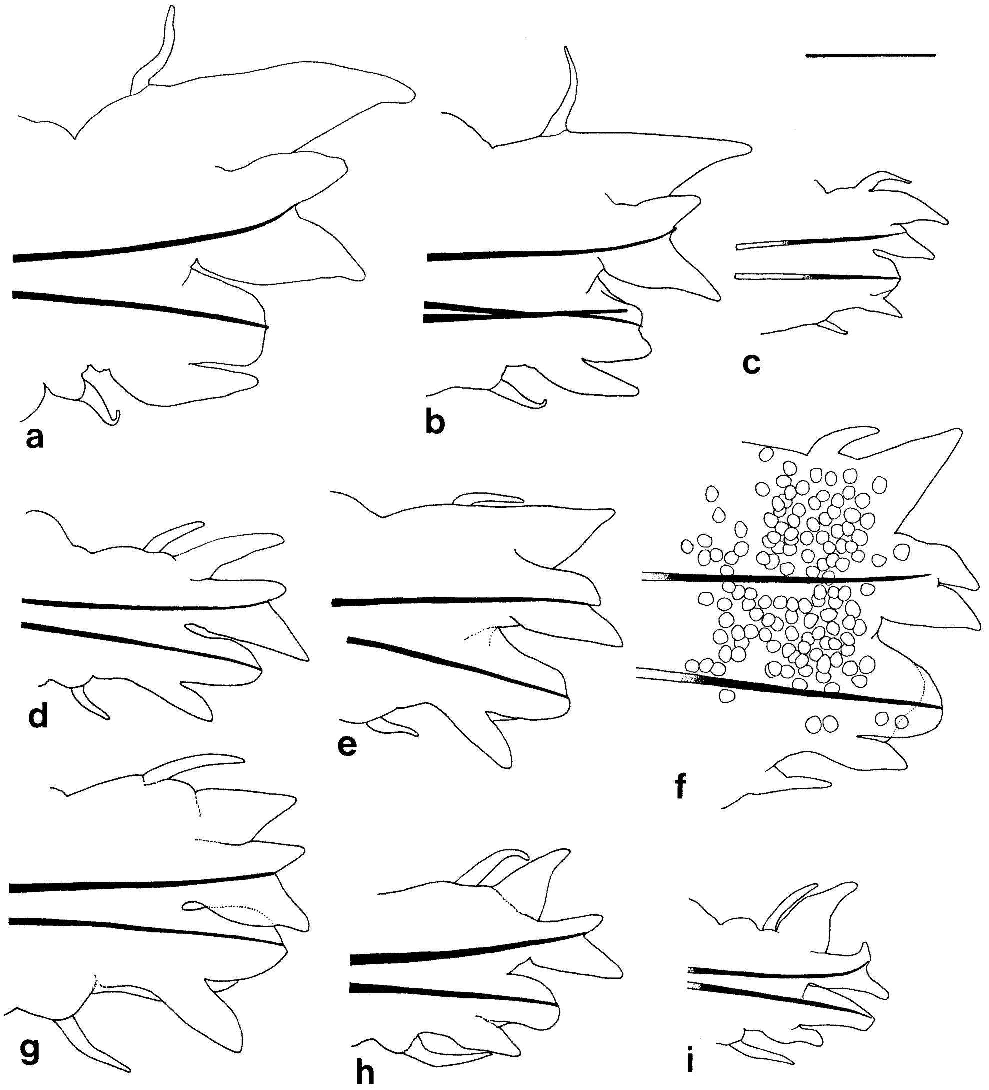


Figure 20. Anterior views of parapodium 28–32 of various sized immature atokous specimens (a–c), epitokous mature females (d–f) and males (g–i) of ***Hediste diadroma*** **sp. nov**. Chaetae not shown. (a) 2-year-old specimen after artificial fertilization, Omoigawa River, BW 5.1 mm. (b) 2-year-old specimen after artificial fertilization, Omoigawa River, BW 4.2 mm. (c) 1-year-old specimen after artificial fertilization, Omoigawa River, BW 2.0 mm. (d) Paratype (USNM-1008439), after spawning, BW 2.6 mm. (e) Paratype (OMNH-Iv-4228), after spawning, BW 2.7 mm. (f) Paratype (ZIHU-2094), before spawning, BW 4.6 mm. (g) Paratype (MNHN-Poly-1369), after spawning, BW 3.3 mm. (h) Paratype (SMF-12067), after spawning, BW 2.3 mm. (i) Paratype (ZIHU-2095), after spawning, BW 2.4 mm. Scale bar = 1 mm.

Maximum sizes were 150 mm in BL, 5.1 mm in BW, and 114 in SN for mature adults, and 160 mm in BL, 5.3 mm in BW and 118 in SN for immature specimens which were reared in laboratory for 2 years after artificial fertilization.

Eye width (EW mm) was correlated with BW according to the following regression formula in immature specimens: EW = 0.04 BW + 0.09 ($r^2 = 0.57, P = 0.0001$, $n = 18$) (Fig. 24). EW (0.19–0.32 mm) of mature adults ($n = 24$) was not significantly correlated with BW ($r = 0.26$, $P = 0.2$), but significantly larger than EW (0.11–0.26 mm) of immature specimens ($n = 13$) with corresponding BW of 2.3–4.9 mm (Mann–Whitney *U*-test, $P = 0.0006$).

Total paragnath number was not significantly correlated with body size in mature adults collected from a single locality, i.e. Omoigawa River ($r = 0.1$, $P = 0.3$, $n = 64$), and Nagatagawa River ($r = 0.2$, $P = 0.3$, $n = 35$) (Fig. 25).

Variation of paragnath number

Paragnath numbers in mature (10 localities) and immature specimens (5 localities) are summarized in Table 4.

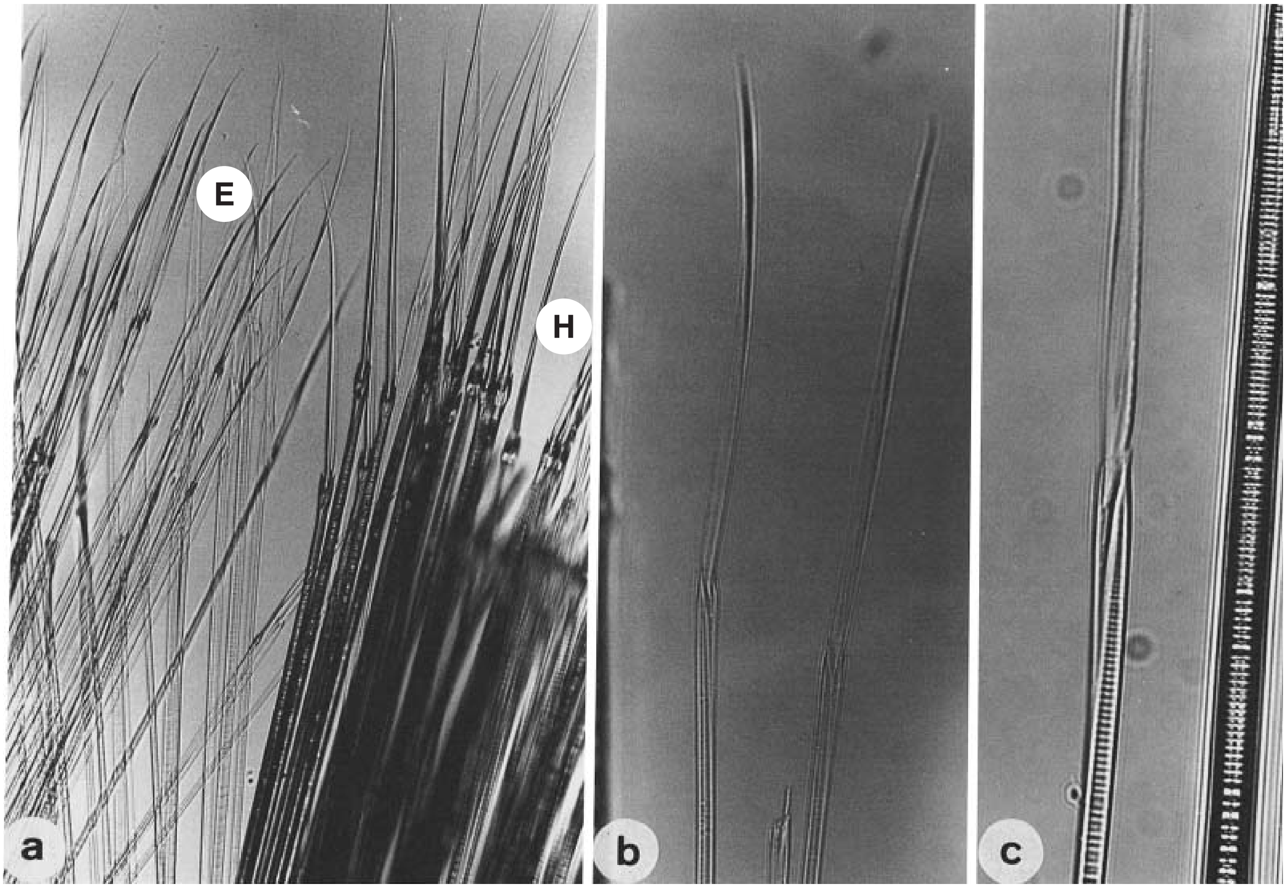


Figure 21. Epitoke-specific sesquigomph spinigers of mature adults of ***Hediste diadroma* sp. nov**. (a) Many transparent epitoke-specific sesquigomph spinigers (E) added behind the notopodial fascicle of ordinary homogomph spinigers (H) in parapodium 30 (paratype, ZIHU-2096). ×175. (b) Epitoke-specific sesquigomph spinigers in parapodium 20 of a female (paratype, SMF-12068). ×350. (c) Enlarged part around base of blade of an epitoke-specific sesquigomph spiniger (specimen same as b). ×700.

Karyotype and sex determination

Diploid chromosome number was 28, including a pair of heteromorphic sex chromosomes (Sato & Ikeda, 1992). The sex seemed to be determined by a simple system of male heterogamy, i.e. an XX–XY system, where the Y submetacentric chromosome was larger than the X. A sex ratio of almost 1 : 1 was observed (Sato, 1999). The autosomes consisted of 9 metacentric pairs including 3 distinctly larger pairs and 4 submetacentric pairs.

Reproduction

Reproductive swarming occurred during the cold season, winter or early spring, i.e. January to April, except for June in Hokkaido, northern Japan. Many epitokous males and females simultaneously swam up to the surface just after high-tide at night during the spring tides, and were transported downstream by the ebb tide (Kagawa, 1955; Sato & Tsuchiya, 1987; Sato, 1999). Eggs or sperm were shed into seawater around the river mouth, where fertilization occurred. After spawning, the adults died. The wet weight of the adult body after spawning reduced to 15–50% of that before spawning (average ± SD: 32.9 ± 0.09% in 9 males, 22.7 ± 0.07% in 16 females) (Fig. 26). Fecundity (entire number of eggs produced by a female) ranged from over 10 000 to 1 million eggs (Sato, 1999).

Development

The full-grown oocytes were 130–170 μm in diameter (Sato & Tsuchiya, 1991; Sato, 1999). The ooplasm was relatively transparent and contained 20–40 oil drops around a germinal vesicle. The spermatozoa had a cone-shaped acrosome at the tip of the head (Sato, 1999). The sperm head was about 3 μm wide, about 4 μm long, and angular at the base of acrosome. The gamete ultrastructure and early events of fertilization are described in Sato & Osanai (1986) and Sato (1999).

In early development, a larval planktonic phase was present during the trochophore, metatrochophore and

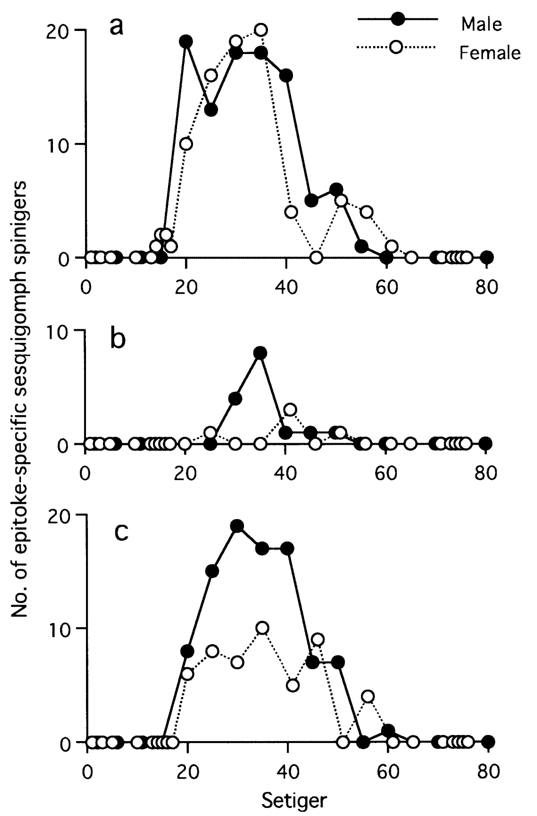


Figure 22. Number of epitoke-specific sesquigomph spinigers in (a) notopodial, (b) neuropodial supra-acicular and (c) neuropodial infra-acicular fascicles of each parapodium throughout body of a mature male (●) and a mature female (○) of ***Hediste diadroma* sp. nov**. (specimens same as Fig. 18).

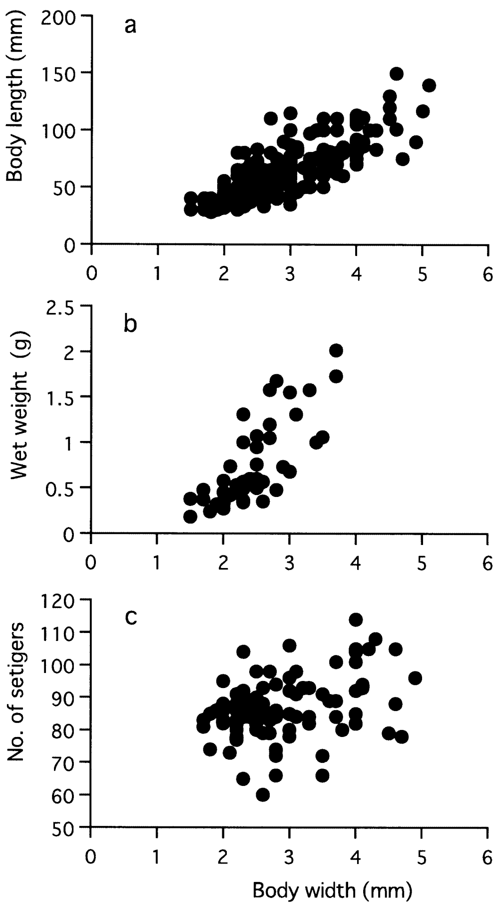


Figure 23. Relationships between body width and (a) body length, (b) wet body weight before shedding of gametes and (c) setiger number in mature adults of ***Hediste diadroma* sp. nov**. (a) 193 specimens collected from 12 localities, (b) 46 specimens collected from 3 localities, (c) 96 specimens collected from 11 localities.

nectochaeta stages (Kagawa, 1955; Sato & Tsuchiya, 1991). Salinity approaching full-strength seawater (more than 20‰) was essential for early development. The nectochaeta larvae of the 6-setiger stage, which seemed to gain a tolerance to lower salinity, settled into brackish waters of the adult habitat, moving upstream on rising tides about 1 month after fertilization (Kagawa, 1955).

Habitat and life history

Adults and juveniles showed euryhaline distributions in estuaries; they lived within burrows mainly in sandy or muddy tidal flats, commonly coexisting with *H. atoka* sp. nov. along the Japanese coast (excepting that of the Ariake Sea, Kyushu) and coexisting with *H. japonica* in the Omutagawa River where it flowed into the Ariake Sea. The life-cycle is migratory and diadromous, planktonic (embryos and larvae) in the sea and benthic (juveniles and adults) in brackish waters (Sato, 1999). This seemed to result in a frequent gene flow from river to river via the sea, and consequently in the lower level of genetic differentiation among geographically separated populations (Sato & Masuda, 1997).

The life-span of 1 year was suggested by a monthly population survey in China (Qiu & Wu, 1993).

Geographical distribution

The coasts of Japan and China (Fig. 27).

Etymology

The specific name refers to the diadromous life history of this species.

Remarks

Though *Hediste diadroma* sp. nov. is very similar to *H. atoka* sp. nov., they are distinguishable at the

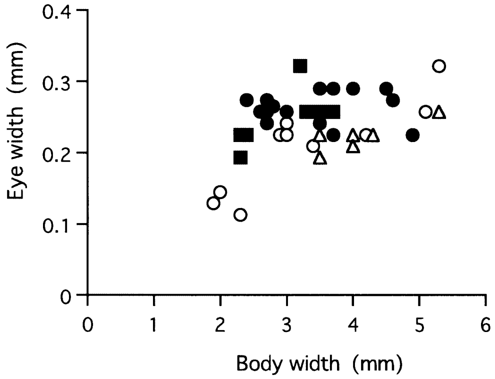


Figure 24. Relationship between body width and eye width in mature (closed symbols) and immature specimens (open symbols) of ***Hediste diadroma* sp. nov**. Mature specimens were collected from Nanakitagawa River, Otagawa River, and Omoigawa River (6 males,■, 18 females,●). Immature specimens were collected from Omutagawa River ($n = 6$, △), or laboratory-bred from artificial fertilization of gametes collected from Omoigawa River ($n = 12$, ○).

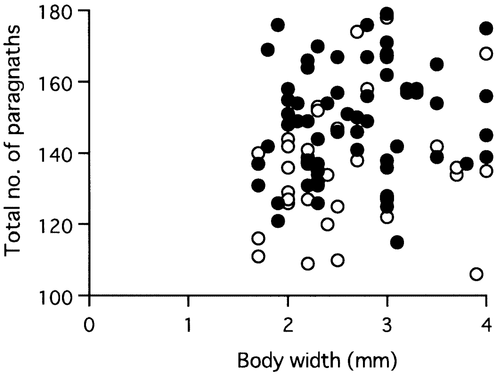


Figure 25. Relationship between body width and total paragnath number of all groups on proboscis of mature adults of ***Hediste diadroma* sp. nov**. (●) Specimens collected from Omoigawa River ($n = 64$). (○) Specimens collected from Nagatagawa River ($n = 35$).

mature stage due to the occurrence of epitokous metamorphosis, with the addition of epitoke-specific spinigers and a smaller egg size in *H. diadroma* sp. nov. They are morphologically indistinguishable at the immature stage. The difference in paragnath numbers in the maxillary ring of the proboscis is significant between the two species of sympatric populations, but cannot strictly distinguish them (see below). However, they are clearly distinguishable by complete allele substitution at several allozyme loci, which is detectable by electrophoretic analysis (Sato & Masuda, 1997). The electrophoretic pattern of LDH is available as a diagnostic character for identification of immature worms (Fig. 10).

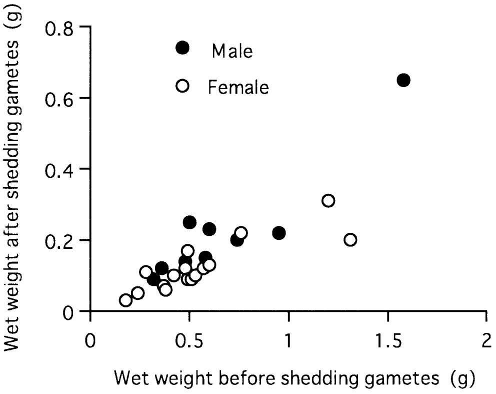


Figure 26. Relationship between wet body weights before and after shedding of gametes in mature males (●, $n = 9$) and females (○, $n = 16$) of ***Hediste diadroma* sp. nov**. Specimens were collected from Omoigawa River in 1986.

Nereis japonica sensu Kagawa (1955) and Okada (1960), and *Neanthes japonica sensu* Sun *et al*. (1980) and Qiu & Wu (1993), seem to be identical to *H. diadroma* sp. nov. because of the occurrence of reproductive swarming and a mature-egg diameter of less than 180 μm.

HEDISTE ATOKA SP. NOV. (FIGS 1, 10, 28–43)

Neanthes japonica: Kikuchi, 1998: 125–146, figs 1–12.
Large-egg type of *Neanthes japonica*: Sato & Tsuchiya, 1987: 29–42, figs 1, 4, 5; Sato & Tsuchiya, 1991: 371–382, figs 1, 3, 4.
Large-egg form of *Neanthes japonica*: Sato & Ikeda, 1992: 299–307, figs 5–7.
Large-egg form of *Hediste japonica*: Sato & Masuda, 1997: 163–170: Sato, 1999: 129–143, figs 1–5, 7–13.

Type material

Holotype (NSMT-Pol-H454): complete mature female (BL: 70 mm, BW: 3.2 mm, 94 setigers), Shinjogawa River, Aomori-shi, Aomori Prefecture, 3 June 1992, coll. M. Sato. Paratypes: mature 4 males and 8 females (BL: 56–100 mm, BW: 2.0–3.3 mm, 76–90 setigers), data as for holotype (CMNH-ZW-1889, 1890, MNHN-Poly-1367, NSMT-Pol-P455, OMNH-Iv-4225∽4227, SMF-12065, 12066, USNM-00, ZMUC-Pol-1503); mature 1 male (ZIHU-2099) and 1 female (ZIHU-2098) (BL: 50 mm, BW: 3.0–3.2 mm, 86 setigers), Niidagawa River, Hachinohe-shi, Aomori Prefecture, 6 June 1982, coll. M. Sato; mature female (BL: 63 mm, BW: 2.6 mm, 80 setigers), Kotsukigawa River, Kagoshima-shi, Kagoshima Prefecture, 11 October, 1989, coll. M. Sato (ZIHU-2100).

Table 4. Variation of paragnath numbers in mature (10 localities) and immature specimens (5 localities) of ***Hediste diadroma*** **sp. nov.**

			Mean paragnath number ± SD (range)						
Locality	*n*	Body width (mm)	I	II[1]	III	IV[1]	VI[1]	VII–VIII	Total
MATURE									
Usujiri (1983)	1	2.5	2 –	23 –	49 –	41 –	11 –	22 –	148 –
Nanakitagawa (1981)	12	3.0–4.9	1.1 ± 0.3 (1–2)	26.5 ± 5.2 (18–39)	41.7 ± 8.0 (31–61)	38.2 ± 6.4 (25–47)	9.6 ± 1.8 (6–12)	26.0 ± 4.8 (19–35)	143.9 ± 15.2 (109–166)
Tonegawa (1966)	10	2.3–4.0	1.2 ± 0.6 (1–3)	22.9 ± 4.0 (18–32)	39.5 ± 9.6 (29–61)	35.0 ± 6.1 (29–50)	10.1 ± 3.6 (3–14)	25.9 ± 6.2 (18–38)	134.6 ± 26.0 (108–198)
Otagawa (1981)	5	1.7–3.3	2.6 ± 1.1 (1–4)	42.6 ± 8.5 (32–52)	38.0 ± 6.1 (30–45)	60.8 ± 6.6 (50–67)	11.4 ± 3.4 (6–15)	30.2 ± 6.3 (26–41)	185.6 ± 11.6 (169–199)
Yahatagawa (1981)	15	2.0–4.6	2.4 ± 1.1 (1–4)	41.1 ± 6.5 (28–50)	46.6 ± 4.8 (40–57)	58.6 ± 8.4 (44–76)	12.0 ± 2.4 (9–17)	26.6 ± 5.5 (20–40)	187.7 ± 14.2 (167–221)
Nakagawa (1980)	16–25	3.1–5.1	1.6 ± 0.8 (1–3)	28.6 ± 6.2 (21–41)	35.5 ± 7.9 (25–49)	40.3 ± 7.1 (30–55)	10.1 ± 3.0 (3–16)	23.1 ± 6.5 (15–38)	139.8 ± 23.9 (109–182)
Omoigawa (1986–89)	66–72	1.8–3.2	2.1 ± 1.3 (1–8)	28.7 ± 5.3 (20–44)	40.6 ± 6.2 (25–56)	44.2 ± 6.5 (29–62)	10.0 ± 2.0 (4–15)	23.5 ± 3.7 (16–31)	149.1 ± 15.2 (115–179)
Kotsukigawa (1986)	6	2.5–4.5	2.0 ± 0.6 (1–3)	28.3 ± 6.3 (19–38)	36.5 ± 6.5 (29–47)	39.2 ± 1.5 (37–41)	9.2 ± 1.2 (7–10)	22.0 ± 1.4 (20–24)	137.2 ± 14.4 (118–160)
Nagatagawa (1992)	29–30	1.7–3.9	1.4 ± 0.7 (1–3)	28.4 ± 4.4 (20–39)	34.0 ± 5.7 (22–47)	40.6 ± 6.1 (30–56)	9.1 ± 1.8 (5–13)	20.8 ± 4.8 (11–30)	134.2 ± 15.8 (106–174)
Kaminokawa (1992)	12	2.6–4.0	1.4 ± 0.7 (1–3)	26.2 ± 4.3 (18–33)	41.1 ± 5.6 (34–53)	41.9 ± 5.9 (36–54)	9.1 ± 2.0 (6–14)	23.2 ± 4.9 (16–30)	142.8 ± 18.2 (120–181)
IMMATURE									
Omutagawa (1998–99)	11	3.4–5.2	1.5 ± 0.8 (1–3)	20.0 ± 4.3 (11–27)	35.6 ± 7.8 (19–48)	33.8 ± 6.6 (21–44)	9.3 ± 2.1 (6–12)	21.5 ± 2.7 (16–25)	121.6 ± 19.7 (77–151)
Kikuchigawa (1996)	54	1.6–5.0	1.4 ± 0.6 (0–3)	24.7 ± 4.1 (17–35)	30.7 ± 5.4 (22–47)	36.7 ± 6.3 (15–54)	8.9 ± 2.0 (4–14)	21.0 ± 2.4 (15–26)	123.4 ± 14.7 (91–162)
Maekawa (1998)	9–22	1.5–2.6	1.8 ± 1.0 (1–4)	30.2 ± 5.1 (23–40)	38.7 ± 5.7 (32–51)	44.5 ± 6.4 (38–60)	11.4 ± 1.4 (8–14)	24.4 ± 3.7 (19–33)	154.4 ± 17.3 (129–190)
Omoigawa (1995–96)	13	2.5–5.5	1.6 ± 1.4 (1–6)	27.8 ± 5.4 (21–37)	38.3 ± 7.3 (27–53)	43.7 ± 6.5 (32–52)	9.7 ± 3.6 (3–19)	21.5 ± 2.2 (17–25)	142.6 ± 15.6 (113–166)
lab-bred[2]	9	1.8–3.4	1.3 ± 1.1 (0–4)	23.8 ± 3.6 (19–29)	25.7 ± 3.2 (20–30)	34.7 ± 3.7 (31–41)	9.8 ± 2.0 (6–12)	22.1 ± 4.3 (17–28)	117.3 ± 11.2 (98–130)
Atagogawa (1996)	7	1.6–2.8	3.0 ± 0.8 (2–4)	29.9 ± 5.0 (22–37)	35.9 ± 5.6 (25–41)	37.1 ± 4.5 (30–43)	9.4 ± 1.3 (8–12)	20.3 ± 2.0 (17–23)	135.6 ± 14.4 (105–146)

[1] Total number on right and left sides of proboscis.

[2] Reared for 4–12 months after artificial fertilization in laboratory.

Other material examined

Mature adults collected from field: Obuchinuma Lake, Rokkasho-mura, Aomori Prefecture, 31 May 1992, coll. M. Sato, 18 specimens. Niidagawa River, Hachinohe-shi, Aomori Prefecture, 14 June 1980, coll. M. Sato, 4 specimens; 6 June 1982, coll. M. Sato, 12 specimens. A small creek, Otsuchi-cho, Iwate Prefecture, 15 June 1988, coll. M. Sato, 6 specimens. Nata-Bridge, Yoshinogawa River, Tokushima Prefecture, 30 May 1999, coll. M. Sato, 2 specimens. Kotsukigawa River, Kagoshima-shi, Kagoshima Prefecture, 4 March 1985, coll. M. Sato, 1 specimen; 6 October 1986, coll. M. Sato, 15 specimens (postspawners); 5 November 1987, coll. T. Nagahama, 2 specimens; 10–28 September, 3–24 October 1988, coll. M. Sato, 32 specimens; 14, 15 September, 11 October 1989, coll. M. Sato, 15 specimens. Nagatagawa River, Taniyama, Kagoshima-shi, Kagoshima Prefecture, 25 September, 21 October 1991, coll. M. Sato, 28 specimens. Oosatogawa River, Ichiki-cho, Kagoshima Prefecture, 4 October 1986, coll. M. Sato, 1 specimen (postspawner); 26 October 1988, coll. M. Sato, 2 specimens (postspawners). Kaminokawa River, Higashi-ichiki-cho, Kagoshima Prefecture, 22 September 1991, coll. M. Sato, 12 specimens. Isakugawa River, Iriki-cho, Kagoshima Prefecture, 17 September 1989, coll. M. Ikeda, 1 specimen (postspawner).

Mature adults obtained after rearing in laboratory: Shinjogawa River, Aomori-shi, Aomori Prefecture, fixed 20 June, 4 July, 17 August 1981 after rearing for 1–3 months, coll. M. Sato, 21 specimens. Omoigawa River, Aira-cho, Kagoshima Prefecture, fixed 29 October 1987, 18 October 1991, 25 March 1992 after rearing for 1–6 months, coll. M. Sato, 3 specimens.

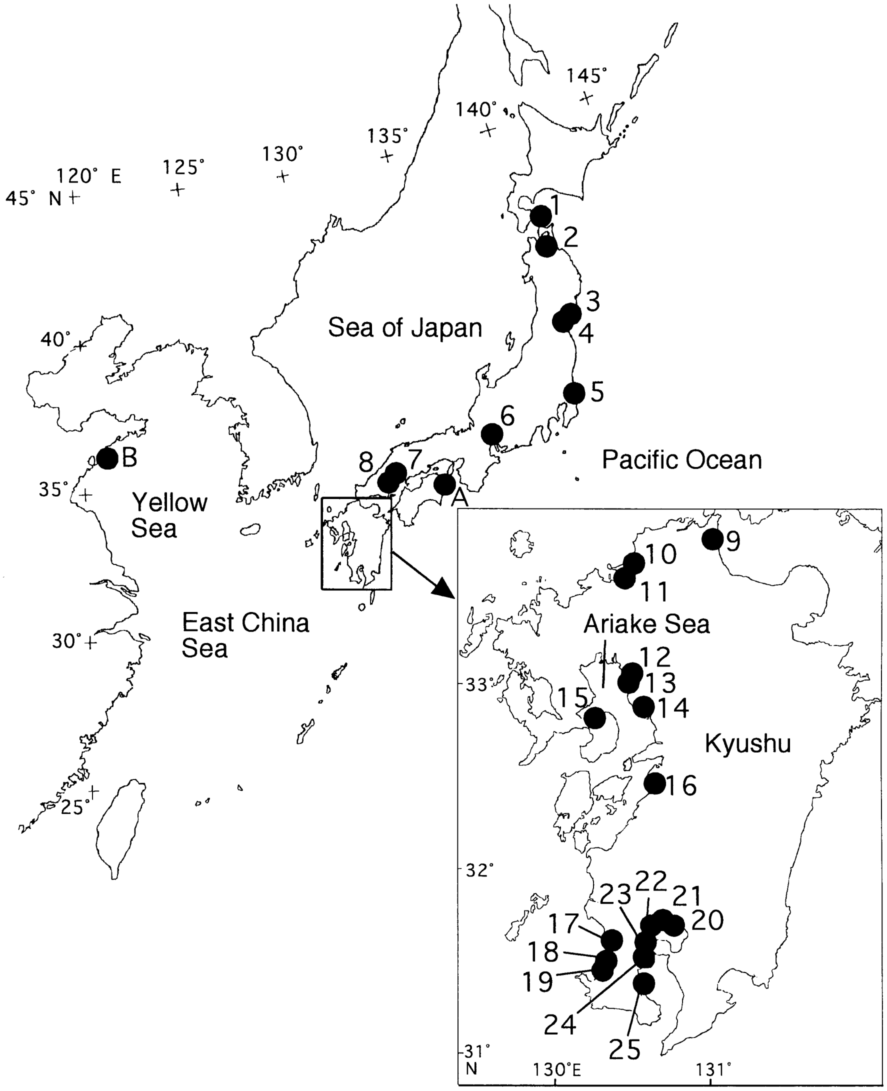


Figure 27. Distribution of ***Hediste diadroma* sp. nov**. 1. Usujiri. 2. Kominatogawa River. 3. Nanakitagawa River. 4. Natorigawa River. 5. Tonegawa River. 6. Shinkawa River. 7. Otagawa River. 8. Yahatagawa River. 9. Kusamigawa River. 10. Wajirogawa River. 11. Nakagawa River. 12. Domengawa River. 13. Omutagawa River. 14. Kikuchigawa River. 15. Kobe. 16. Maekawa River. 17. Kaminokawa River. 18. Isakugawa River. 19. Manosegawa River. 20. Amorigawa River. 21. Byugawa River. 22. Omoigawa River (type locality). 23. Kotsukigawa River. 24. Nagatagawa River. 25. Atagogawa River. A, Tokushima-shi (Kagawa, 1955; Okada, 1960). B, Qingdao (Sun *et al.*, 1980; Qiu & Wu, 1993).

Immature specimens: Nanakitagawa River, Sendai-shi, Miyagi Prefecture, 14 July 1994, coll. M. Ikeda, 51 specimens. Kokufugawa River, Sado Island, Niigata Prefecture, 19 October, 1990, coll. M. Sato, 18 specimens. Kumanogawa River, Kumano-shi, Mie Prefecture, 3 January 1997, coll. M. Ikeda, 5 specimens. Yahatagawa River, Itsukaichi-cho, Hiroshima Prefecture, 25 February 1998, coll. M. Sato, 15 specimens. Kusamigawa River, Kitakyushu-shi, Fukuoka Prefecture, 30 March 1998, coll. M. Sato, 27 specimens. Tsurikawa River, Genkai-cho, Fukuoka Prefecture, 31 March 1998, coll. M. Sato, 9 specimens. A small creek, Shiohama, Tsuyazaki-cho, Fukuoka Prefecture, 31 March 1998, coll. M. Sato, 12 specimens. A small creek, Tsuyazaki, Tsuyazaki-cho, Fukuoka Prefecture, 31 March 1998, coll. M. Sato, 16 specimens. Kazurugawa River, Koga-machi, Fukuoka Prefecture, 31 March 1998, coll. M. Sato, 16 specimens. Wajirogawa River, Fukuoka-shi, Fukuoka Prefecture, 31 March 1998, coll. M. Sato, 12 specimens. Kikuchigawa River, Tamana-shi, Kumamoto Prefecture, 14 September 1996, coll. A. Nakashima, 1 specimen. Maekawa River, Yatsushiro-shi, Kumamoto Prefecture, 25 August 1998, coll. K. Ichimiya, 4 specimens. Minamatagawa River, Minamata-shi, Kumamoto Prefecture, 11 September 1995, coll. A. Nakashima, 31 specimens. Manosegawa River, Kaseda-shi, Kagoshima Prefecture, 16 May 1995, coll. A. Nakashima, 61 specimens. Omoigawa River, Aira-cho, Kagoshima Prefecture, 24 November 1995, 19 March, 18 April, 16 May, 20 June, 17 July, 28 August, 28 September, 25 October, 25 November 1996, coll. A. Nakashima, 485 specimens. Nagatagawa River, Taniyama, Kagoshima-shi, Kagoshima Prefecture, 30 May 1995, coll. A. Nakashima, 38 specimens; laboratory-bred worms fixed 2–4 months after fertilization at 22 October 1991, coll. M. Ikeda, 29 specimens. Mangrove forest (northern limit in Pacific) at Atagogawa River, Kiire-cho, Kagoshima Prefecture, 2 July 1996, coll. A. Nakashima, 3 specimens. Honjogawa River, Kushima-shi, Miyazaki Prefecture, 13 May 1995, coll. M. Sato, 14 specimens. Oyodogawa River, Miyazaki-shi, Miyazaki Prefecture, 9 November 1997, coll. S. Yamada, 5 specimens. Koshuku-okawa River, Naze-shi, Amami-oshima Island, Kagoshima Prefecture, September 1989, coll. M. Sato, 9 specimens.

Diagnosis

Large number (more than 20 in most cases) of paragnaths on right and left sides of proboscis in group II. Homogomph falciger absent and heterogomph spinigers present in neuropodia. Neuropodial postchaetal ligule tapering to digitate lobe only in anterior setigers (up to setigers 15–25). Sexual maturity without any epitokous metamorphosis in both males and females. Full-grown oocytes 200–250 μm in diameter. Reproduction without swarming. Direct development without true pelagic stage.

Description

Body stout anteriorly, posteriorly gradually tapering toward pygidium (Fig. 28a,d). Dorsum convex, venter relatively flat with longitudinal midventral groove. Colour in preserved specimens whitish cream with dark brown pigmentation on anterior dorsal surface; colour in life at sexually immature stage reddish brown.

Prostomium pyriform, broader than long, with pair of tapered antennae situated at anterior end. Pair of palps with massive palpophore about twice as long as antennae and short round palpostyle. Two pairs of round or oval eyes almost equal in size, arranged trapezoidally (space between anterior pair about 1.4 times as wide as that between posterior pair). Longitudinal mid-dorsal groove present on anterior dorsum of prostomium. Partial dark pigmentation present on dorsal anterior surface of prostomium and palpophore (Fig. 28a).

Peristomium nearly as long as following setigers, with 4 pairs of tentacular cirri of unequal length; posterior dorsal tentacular cirri longest, reaching back to setiger 8 or 9; anterior dorsal tentacular cirri next longest, reaching back to setiger 3 or 4.

Proboscis with pair of dark brown jaws, each with 7–9 teeth (Fig. 28b,c). Black paragnaths, usually with pointed tip, present on both maxillary and oral rings; paragnath numbers in holotype (range for all 176 mature specimens in parentheses) as follows. Group I: 3 (0–8); II: 21 on both right and left sides, in two or three arched rows, total 42 (29–82); III: 36 (23–79) in transverse band; IV: 25 on right and 26 on left in three arched rows, total 51 (34–92), V: none; VI: 6 on right and 5 on left, in small clusters, total 11 (1–17); VII–VIII: 14 (9–35) in single transverse row.

Parapodia of first 2 setigers uniramous, all following parapodia biramous (Fig. 28e–k). Uniramous parapodia with reduced notopodia consisting of dorsal cirrus and superior ligule, and with ordinary neuropodia.

Notopodia consisting of dorsal cirrus and three ligules in biramous parapodia, i.e. large superior ligule and upper and lower acicular ligules; all notopodial ligules subtriangular with tapering tip. Upper acicular ligule subequal to lower one in anterior setigers (Fig. 28g), gradually diminishing in size in middle setigers and absent in posterior setigers (Fig. 28h–k). Superior ligule thick in anterior setigers, thinner in middle and posterior setigers (from around setiger 15), most expanded in middle setigers. Dorsal cirri slender, tapering, not reaching tip of superior ligule.

Neuropodia consisting of ventral cirrus and three ligules throughout, i.e. prechaetal acicular ligule, postchaetal ligule and inferior ligule. Prechaetal acicular ligule and postchaetal ligule conical with tapering tip, of similar lengths, completely separate in anterior setigers (up to about first 10 setigers, Fig. 28e–g), fused at following setigers; tapering tip of postchaetal ligule diminishing to digitate lobe, present up to around setiger 20, and absent in following setigers (Fig. 28i,k); prechaetal acicular ligule with blunt tip. Inferior ligule conical. Ventral cirrus slender with tapering tip. Inferior ligule and ventral cirrus gradually diminishing in size in posterior setigers.

Notochaetae all homogomph spinigers with slender serrated blade (Fig. 29a,b), 10–50 in number in most setigers, and less than 10 in anteriormost and posteriormost setigers (Fig. 30a; Table 3).

Neurochaetae of supra-acicular fascicle consisting of homogomph spinigers (up to 30, Fig. 30b) and heterogomph falcigers (up to 10, Fig. 31b) in anterior and middle setigers (anterior 40–50 setigers in adults). Neuropodial homogomph spinigers similar to notopodial ones in shape and size. Heterogomph falcigers with

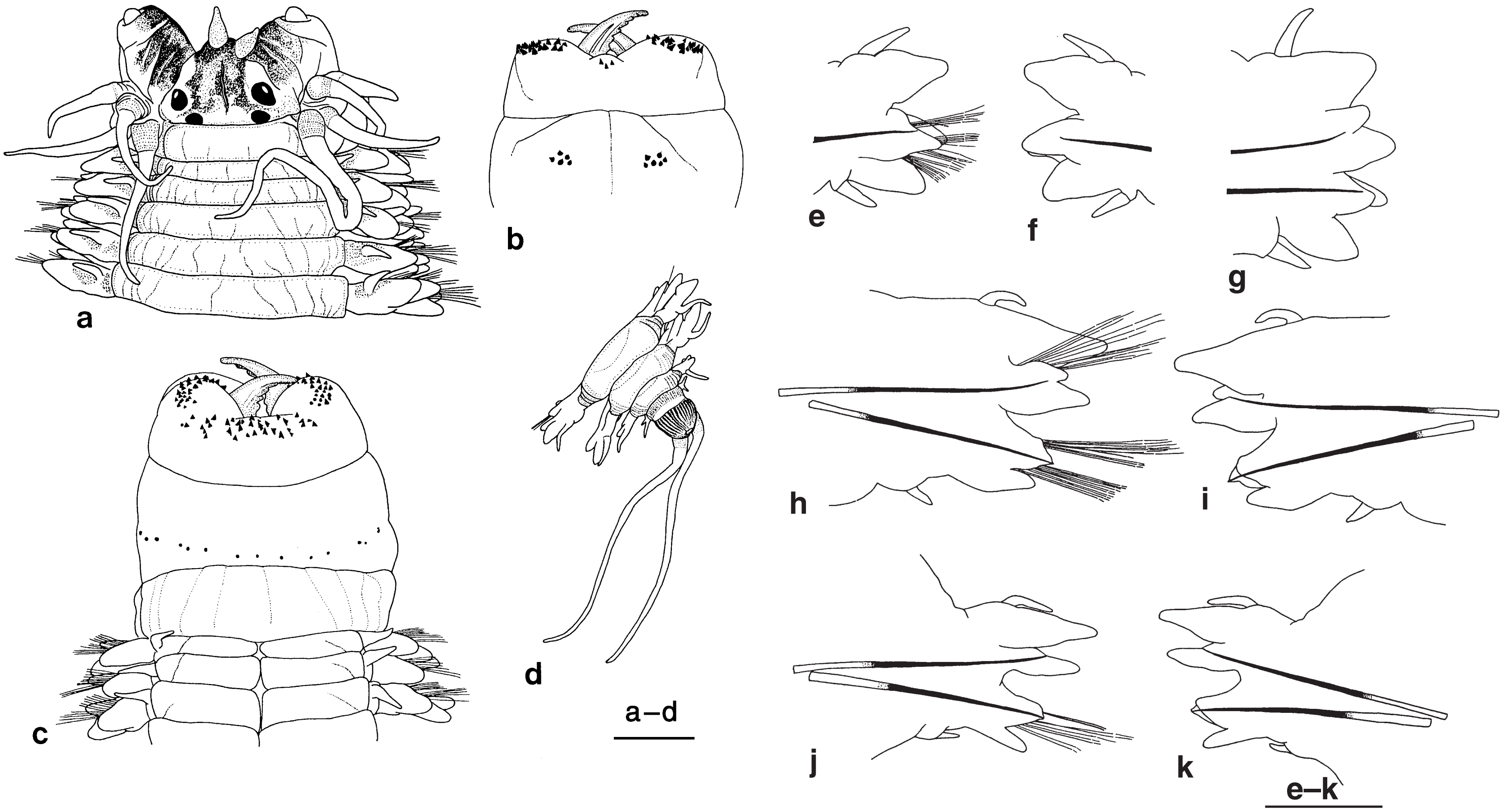


Figure 28. Mature female of ***Hediste atoka* sp. nov**. (holotype, NSMT-Pol-H454). (a) Dorsal view of anterior end; everted proboscis not shown here. (b & c) Dorsal and ventral views of everted proboscis. (d) Dorsal view of posterior end. (e–k) Parapodia of the left side (chaetae not shown in f, g, i, k). (e & f) Anterior and posterior views of parapodium 2. (g) Anterior view of parapodium 10. (h & i) Anterior and posterior views of parapodium 30. (j & k) Anterior and posterior views of parapodium 71. Scale bar = 1 mm.

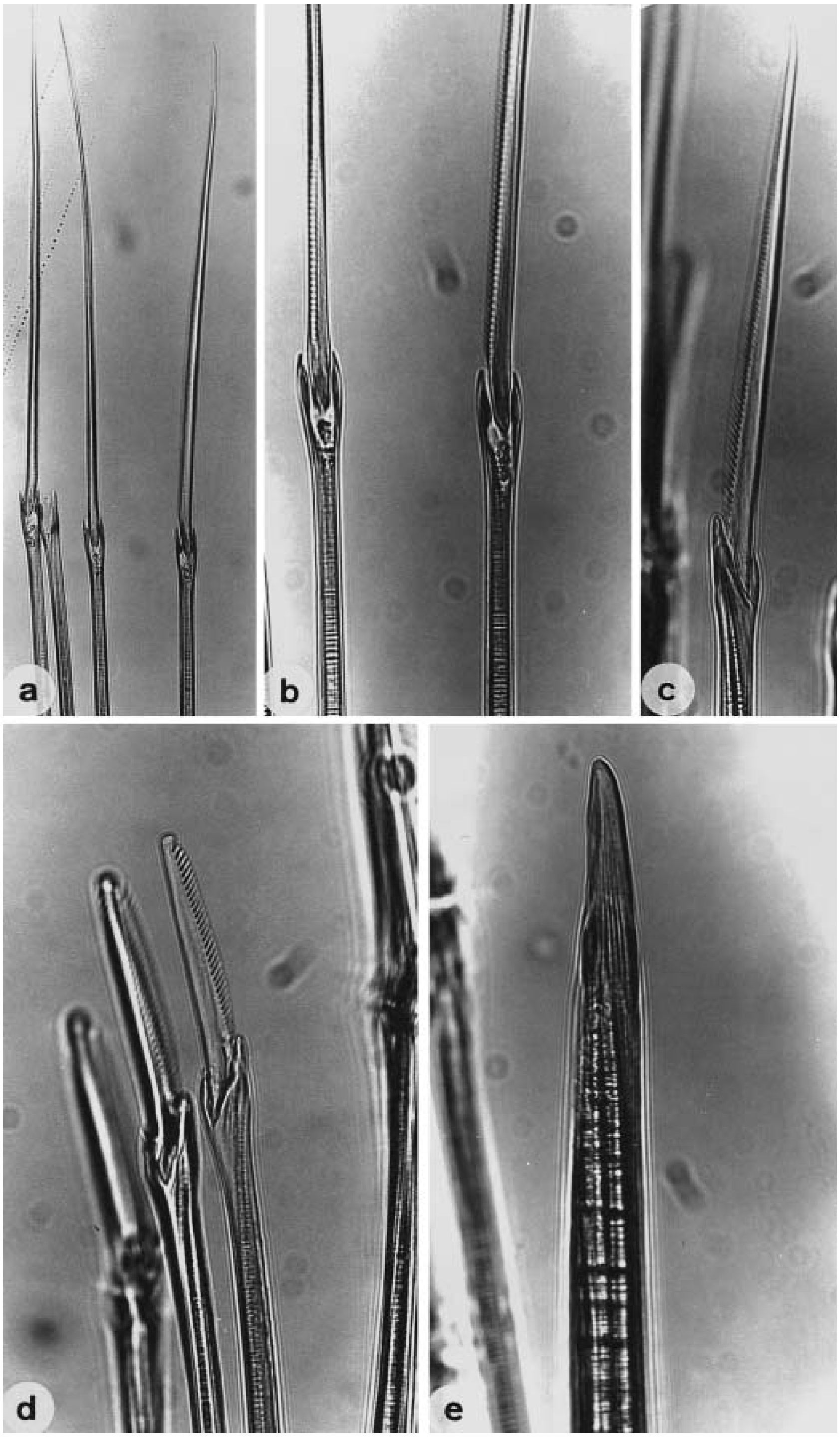


Figure 29. Chaetae of a mature female of ***Hediste atoka* sp. nov.** (paratype, ZIHU-2098). (a) Homogomph spinigers from notopodial fascicle in parapodium 3. ×290. (b) Enlargement of base of blade in a. ×580. (c) Heterogomph spiniger from neuropodial infra-acicular fascicle in parapodium 1. ×580. (d) Heterogomph falcigers from neuropodial infra-acicular fascicle in parapodium 3. ×580. (e) Simple chaeta from neuropodial supra-acicular fascicle in parapodium 64. ×580.

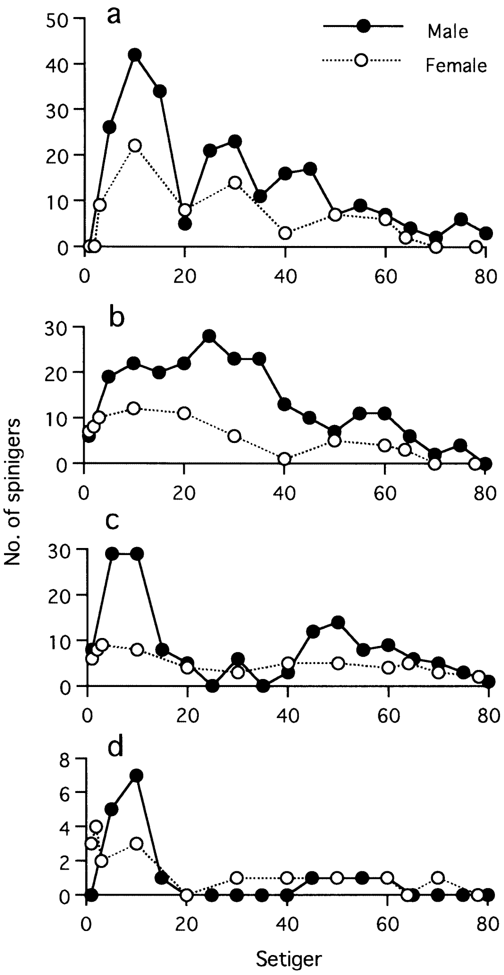


Figure 30. Number of spinigers in each parapodium throughout body of a mature male (●, incomplete specimen, BW 3.2 mm, with 83 setigers, paratype, ZIHU-2099) and a mature female (○, complete specimen, BW 3.0 mm, with 86 setigers, paratype, ZIHU-2098) of ***Hediste atoka*** **sp. nov**. homogomph spinigers in (a) notopodial fascicle, (b) neuropodial supra-acicular fascicle, and (c) neuropodial infra-acicular fascicle. (d) Heterogomph spinigers in neuropodial infra-acicular fascicle.

serrated blade; 1 or 2 thick simple chaetae with tapering tip present instead of heterogomph falcigers in posterior setigers (Figs 29e, 31a). Form of blade of original heterogomph falciger remaining at tip of simple chaeta in setiger 71 of holotype, showing fusion between blade and stalk of heterogomph falciger (Fig. 32).

Neurochaetae in infra-acicular fascicle consisting of homogomph spinigers (up to 30, Fig. 30c) at upper position, heterogomph spinigers (up to 10, Figs 29c, 30d) on middle portion, and heterogomph falcigers (up to 35, Figs 29d, 31c) at lower position.

Aciculae black except colourless proximal part; single acicula present in each ramus.

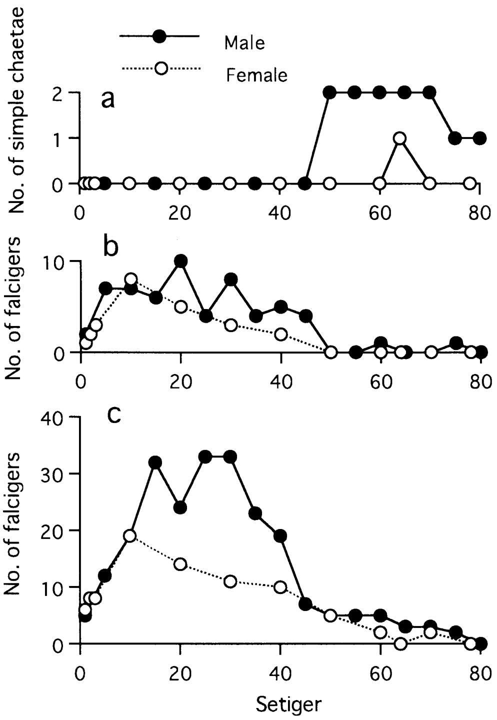


Figure 31. Number of simple chaetae and falcigers in each parapodium throughout body of a mature male (●) and a mature female (○) of ***Hediste atoka*** **sp. nov**. (same specimens as those shown in Fig. 30). (a) Simple chaetae in neuropodial supra-acicular fascicle. (b) Heterogomph falcigers in neuropodial supra-acicular fascicle. (c) Heterogomph falcigers in neuropodial infra-acicular fascicle.

Pygidium with anus on the dorsal side, with pair of cylindrical slender anal cirri (Fig. 28d).

Sexually mature stage: ripe eggs green colour in females, sperm whitish in males (Sato & Tsuchiya, 1987; Sato, 1999). No epitokous metamorphosis observed in chaetal arrangement, size in eyes, parapodial ligules and dorsal and ventral cirri in both males and females (Fig. 33). Numbers of homogomph spinigers and heterogomph falcigers in neuropodial fascicles more abundant in males than those in females (Table 3).

Allozyme pattern of LDH

Total of 937 specimens from 23 localities was examined electrophoretically. Three characteristic bands of anodal migration were observed in most specimens, with two bands of lower mobility closer together (Fig. 10). Two loci relating the LDH pattern seemed to be almost monomorphic (the frequency of the dominant allele exceeded 0.99 for each locus) (Sato & Masuda, 1997). These allozyme patterns are clearly different from those of *Hediste diadroma* sp. nov. and *H. japonica*.

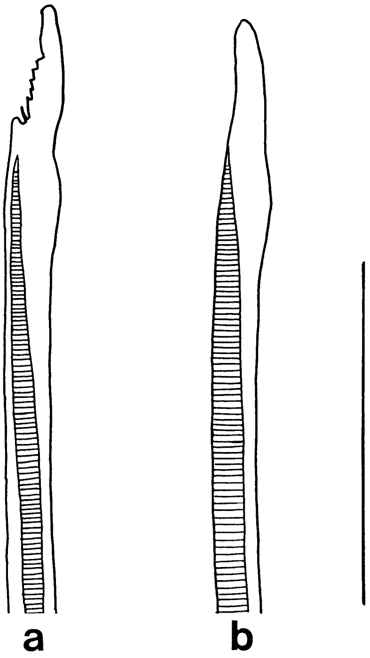


Figure 32. Two simple chaetae found in left neuropodial supra-acicular fascicle of setiger 71 of holotype of ***Hediste atoka* sp. nov.** (NSMT-Pol-4454). (a) Incomplete simple chaeta, originated from fusion of a blade and a stalk of a heterogomph falciger. (b) Typical simple chaeta. Scale bar = 0.05 mm.

Allometry

Body length (BL mm), wet weight (WW g) and setiger number (SN) were correlated with body width (BW mm) according to the following regression formulae (Fig. 34): $BL = 17.9\ BW^{1.1}$ ($r^2 = 0.72$, $P < 0.0001$, $n = 44$), $WW = 0.05\ BW^{2.0}$ ($r^2 = 0.49$, $P < 0.0001$, $n = 52$), $SN = 5.9\ BW + 66.1$ ($r^2 = 0.35$, $P = 0.0002$, $n = 34$). Maximum sizes were 100 mm in BL, 4.2 mm in BW, and 94 in SN for mature adults.

Eye width (EW mm) was correlated with BW according to the following regression formula: $EW = 0.06\ BW + 0.08$ ($r^2 = 0.43$, $P < 0.0001$, $n = 39$) for immature specimens, $EW = 0.06\ BW + 0.10$ ($r^2 = 0.45$, $P = 0.0004$, $n = 22$) for mature adults (Fig. 35). EW (0.18–0.35 mm) of mature adults ($n = 22$) was not significantly different from that (0.13–0.27 mm) of immature specimens ($n = 39$) with corresponding BW of 1.5–3.3 mm (Mann–Whitney *U*-test, $P = 0.31$).

Total paragnath number was not significantly correlated with body size in mature adults collected from a single locality, i.e. Kotsukigawa River ($r = 0.04$, $P = 0.7$, $n = 61$), Nagatagawa River ($r = 0.1$, $P = 0.6$, $n = 28$), and Shinjogawa River ($r = -0.3$, $P = 0.2$, $n = 20$) (Fig. 36).

Variation of paragnath number

Paragnath numbers in mature (9 localities) and immature specimens (8 localities) are summarized in Table 5.

Karyotype and sex determination

Diploid chromosome number was 28, including a pair of heteromorphic sex chromosomes (Sato & Ikeda, 1992). The sex seemed to be determined by a simple system of male heterogamy (XX–XY system), where the Y submetacentric or subtelocentric chromosome was larger than the X metacentric or submetacentric chromosome. A sex ratio of almost 1 : 1 was observed (Sato, 1999). The autosomes consisted of 10 metacentric pairs including 4 distinctly larger pairs and 3 submetacentric pairs.

Reproduction

Spawning occurred within the brackish waters of the adult habitat without reproductive swarming (Sato & Tsuchiya, 1987; Sato, 1999). We observed that females spawned within the burrows in glass aquaria in the laboratory. Males seemed to release sperm near the openings of the burrows of females. However, Kikuchi (1998) observed that mature males and females crawled out of the sediment and released gametes while they crept on the sediment surface in aquaria in the laboratory, and he also collected mature males and females which were swimming in water in a field.

In Kagoshima Prefecture, southern Japan, many mature adults were collected in late September–October, but a few were found throughout almost the whole year. Kikuchi (1998) also collected mature adults from February to October, showing two main reproductive periods in spring and autumn in Miyagi Prefecture, northern Japan. We collected many mature adults in June in Aomori Prefecture, northern Japan.

This species is semelparous, though spent adults are alive within burrows for a while after spawning. Fecundity was several thousand eggs (Sato, 1999).

Development

The full-grown oocytes were 200–250 μm in diameter (Sato & Tsuchiya, 1991; Sato, 1999). The ooplasm was relatively opaque and contained more than 100 oil drops around a germinal vesicle. The spermatozoa had a cone-shaped acrosome at the tip of the head (Sato, 1999). The sperm head was about 3 μm wide, about 4 μm long, and round at the base of acrosome. The gamete ultrastructure and early events of fertilization are described in Sato (1999).

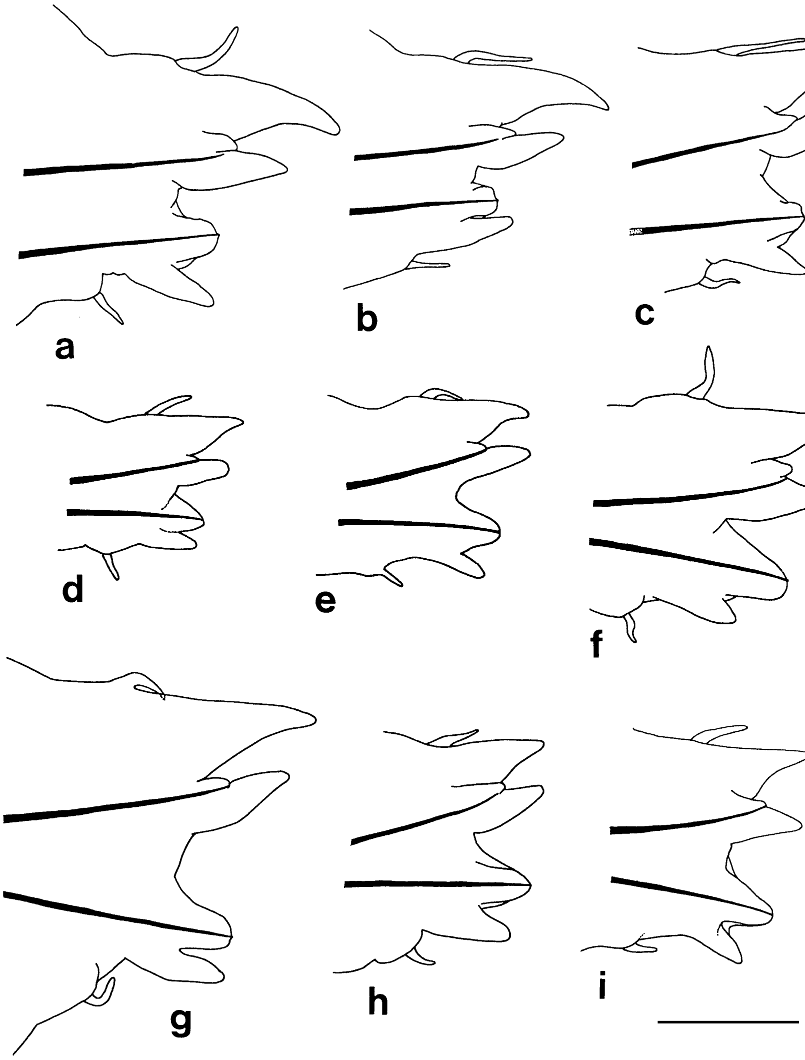


Figure 33. Anterior views of parapodia 30–32 of various sized immature specimens (a–c), mature females (d–f) and mature males (g–i) of ***Hediste atoka* sp. nov.** Chaetae not shown. All measurements body width. (a) 4-month-old specimen after artificial fertilization, Nagatagawa River, 3.1 mm. (b) 3-month-old specimen after artificial fertilization, Nagatagawa River, 2.5 mm. (c) 3-month-old specimen after artificial fertilization, Nagatagawa River, 2.6 mm. (d) Female, Kotsukigawa River, 2.5 mm. (e) Female, Kotsukigawa River, 2.5 mm. (f) Female (paratype, CMNH-ZW-1890), 3.0 mm. (g) Male (paratype, USNM-1008436), 3.3 mm. (h) Male (paratype, SMF-12066), 3.0 mm. (i) Male, Kotsukigawa River, 2.3 mm. Scale bar = 1 mm.

Embryonic development occurred within the jelly layer, which was thinner and more solid than that of *H. diadroma* sp. nov., showing a direct development without a true planktonic phase (Sato & Tsuchiya, 1991; Sato, 1999). The 3-setiger nectochaetae hatched out of the jelly layer and soon began a benthic life, though advanced juveniles with 4 or more setigers could float up towards the surface during flood tides (Sato, 1999). Kikuchi (1998) also showed that juveniles with 4 or 5 setigers could disperse most actively by their swimming behaviour. The early development could proceed under a wide range of salinity, with the optimal salinity around 15‰ (Sato & Tsuchiya, 1987).

Habitat and life history

Adults and juveniles showed euryhaline distributions in estuaries; they lived within burrows mainly in sandy and muddy tidal flats. *H. atoka* sp. nov. commonly coexisted with *H. diadroma* sp. nov. along the Japanese coast, except within the Ariake Sea.

The life-cycle appeared to reach its final stage within brackish waters in a river, resulting in limited gene flow and a consequently higher level of genetic differentiation among geographically separated populations (Sato & Masuda, 1997).

The life-span seemed to usually be about 6 months, yielding two generations per year (Kikuchi, 1998).

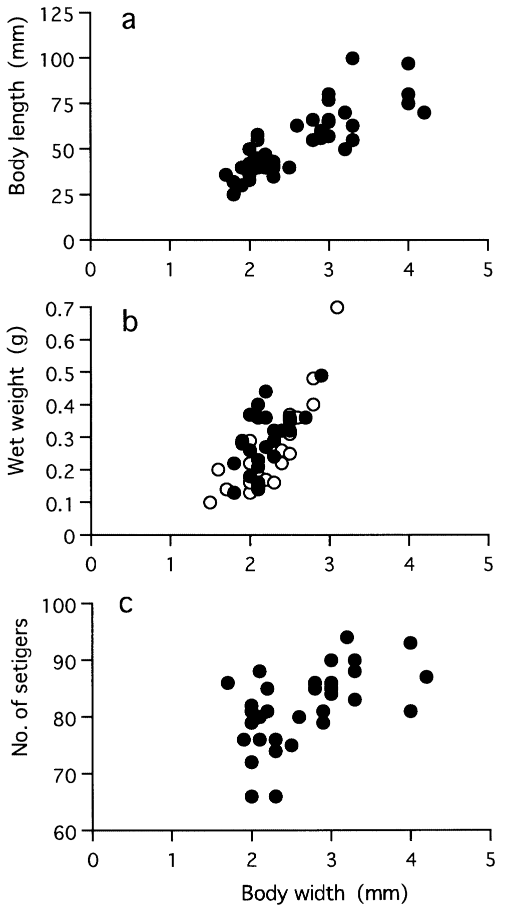


Figure 34. Relationships between body width and (a) body length, (b) wet body weight before shedding of gametes and (c) setiger number in mature adults (●) and immature specimens (○) of ***Hediste atoka* sp. nov.** (a) Mature adults (*n* = 44) collected from six localities. (b) Mature adults (*n* = 27) collected from three localities. Immature specimens (*n* = 25) were laboratory-bred from artificial fertilization of gametes collected from Nagatagawa River. (c) Mature adults (*n* = 34) collected from five localities.

Geographical distribution

The coasts of Japan (Fig. 37).

Etymology

The specific name refers to the atokous form throughout the life history of this species, without any epitokous metamorphosis at the sexually mature stage.

Remarks

While *H. atoka* sp. nov. is very similar to *H. diadroma* sp. nov., it is morphologically distinguishable at the sexually mature stage as unlike the latter, it does not undergo epitokous metamorphosis (see remarks in description of *H. diadroma* sp. nov.).

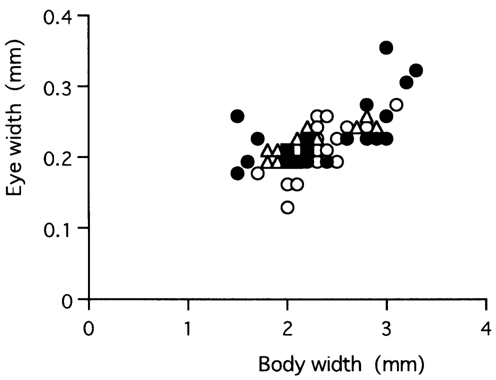


Figure 35. Relationship between body width and eye width in mature (closed symbols) and immature specimens (open symbols) of ***Hediste atoka* sp. nov.** Mature specimens (5 males, ■, 17 females, ○) were collected from Shinjogawa River (holotype and paratype) and Kotsukigawa River. Immature specimens were collected from Manosegawa River (*n* = 15, Δ), or laboratory-bred from artificial fertilization of gametes collected from Nagatagawa River (*n* = 24, ○).

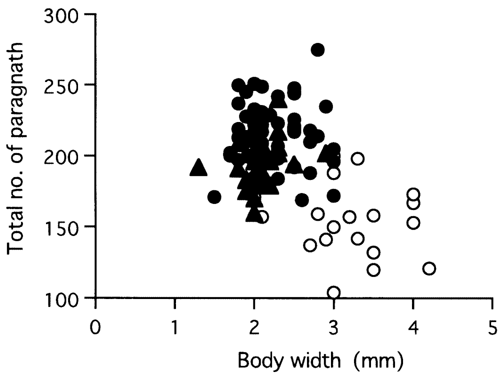


Figure 36. Relationship between body width and total paragnath number of all groups on proboscis of mature adults of ***Hediste atoka* sp. nov.** (●) Specimens collected from Kotsukigawa River (*n* = 61). (▲) Specimens collected from Nagatagawa River (*n* = 28). (○) Specimens collected from Shinjogawa River (*n* = 20).

Neanthes japonica sensu Kikuchi (1998) was judged to be identical to *H. atoka* sp. nov. owing to his description of the egg diameter of up to 260 μm, and his personal communication on the LDH electrophoretic patterns of his material.

COMPARISON OF CHARACTERISTICS OF THE THREE *HEDISTE* SPECIES

BODY SIZE

Mean body width (BW) and number of setigers in mature adults were compared in specimens taken

Table 5. Variation of paragnath numbers in mature (9 localities) and immature specimens (8 localities) of ***Hediste atoka* sp. nov.**

			Mean paragnath number ± SD (range)						
Locality	*n*	Body width (mm)	I	II[1]	III	IV[1]	VI[1]	VII–VIII	Total
MATURE									
Obuchinuma (1992)	17–18	2.1–3.4	2.7 ± 1.0 (2–5)	47.2 ± 6.0 (37–62)	37.7 ± 6.7 (26–51)	55.6 ± 6.3 (47–70)	9.9 ± 1.9 (8–13)	21.1 ± 3.9 (15–27)	174.6 ± 16.4 (148–201)
Shinjogawa (1981, 1992)	25–27	2.0–4.2	1.6 ± 0.8 (0–4)	40.5 ± 7.0 (29–53)	35.9 ± 8.0 (24–53)	49.6 ± 9.8 (34–68)	9.8 ± 2.1 (5–13)	14.8 ± 3.9 (9–24)	156.5 ± 27.3 (104–202)
Niidagawa (1980, 1982)	15–17	2.5–3.5	1.8 ± 0.9 (0–3)	39.4 ± 5.5 (30–46)	38.3 ± 8.8 (23–55)	54.8 ± 10.7 (39–75)	9.6 ± 2.4 (5–16)	14.2 ± 3.4 (9–22)	158.5 ± 25.9 (122–204)
Otsuchi (1988)	6	3.0–4.0	2.3 ± 1.5 (1–5)	57.2 ± 11.7 (45–77)	54.3 ± 5.4 (49–62)	69.8 ± 12.1 (59–92)	11.5 ± 2.7 (8–16)	16.7 ± 2.8 (14–21)	211.8 ± 24.3 (181–248)
Osatogawa (1986, 1988)	3	1.8–2.5	1.3 ± 0.6 (1–2)	51.0 ± 8.2 (42–58)	54.3 ± 3.5 (51–58)	68.0 ± 10.8 (59–80)	11.0 ± 2.6 (8–13)	21.3 ± 0.6 (21–22)	207.0 ± 14.2 (191–218)
Kaminokawa (1991)	11–12	1.7–2.8	2.3 ± 1.0 (1–4)	52.4 ± 3.0 (47–57)	52.8 ± 6.3 (44–64)	56.2 ± 7.0 (46–66)	11.1 ± 1.4 (9–13)	20.6 ± 3.6 (16–26)	195.6 ± 17.2 (173–226)
Omoigawa (1987, 1991)	1–3	1.6–2.8	4.0 (4–4)	59.0 (51–67)	68.0 (66–70)	68.0 –	13.0 (11–15)	21.0 –	245.0 –
Kotsukigawa (1986–89)	61–62	1.5–3.0	3.1 ± 1.5 (1–8)	57.2 ± 8.8 (40–82)	59.9 ± 8.3 (44–79)	60.7 ± 7.9 (45–79)	11.1 ± 2.4 (5–17)	20.5 ± 3.0 (14–26)	213.1 ± 22.7 (169–275)
Nagatagawa (1991)	28	1.3–2.9	2.6 ± 0.9 (1–5)	53.8 ± 6.4 (40–66)	49.6 ± 5.9 (41–62)	56.6 ± 7.1 (45–74)	10.5 ± 2.7 (1–15)	21.7 ± 4.3 (14–35)	194.8 ± 15.7 (160–240)
IMMATURE									
Kikuchigawa (1996)	1	3.2	3 –	59 –	58 –	62 –	12 –	12 –	206 –
Maekawa (1998)	1–4	2.0–2.3	3.5 ± 1.3 (2–5)	54.0 ± 2.6 (52–57)	61.7 ± 3.2 (58–64)	60 –	13.0 ± 2.0 (12–16)	28.3 ± 4.0 (24–33)	219 –
Minamatagawa (1995)	33	2.0–4.0	2.7 ± 0.8 (1–4)	54.8 ± 8.2 (33–70)	51.5 ± 6.6 (39–66)	60.6 ± 7.4 (45–72)	12.5 ± 2.9 (8–22)	19.6 ± 3.6 (14–30)	201.8 ± 16.0 (162–228)
Manosegawa (1995)	61	2.0–4.5	1.7 ± 0.6 (1–4)	50.3 ± 6.9 (35–66)	46.4 ± 7.6 (30–67)	50.8 ± 7.0 (32–66)	11.0 ± 2.0 (4–15)	19.8 ± 3.5 (13–32)	180.0 ± 19.2 (127–215)
Omoigawa (1995–96)	483	1.0–4.5	3.3 ± 1.2 (0–8)	63.7 ± 9.1 (24–95)	62.9 ± 9.3 (21–92)	62.2 ± 8.1 (7–87)	11.9 ± 2.5 (0–21)	23.0 ± 3.5 (11–35)	227.1 ± 24.5 (65–292)
Nagatagawa (1995)	38	2.5–4.5	2.8 ± 1.0 (1–6)	45.4 ± 12.4 (17–68)	46.7 ± 11.7 (18–68)	48.9 ± 14.6 (12–76)	8.8 ± 3.0 (2–13)	18.1 ± 7.2 (0–31)	170.7 ± 44.5 (60–250)
lab-bred[2]	28–29	1.5–3.1	1.3 ± 0.5 (0–2)	36.6 ± 11.8 (4–60)	29.4 ± 8.0 (4–39)	36.5 ± 12.8 (1–56)	6.0 ± 2.7 (0–11)	13.3 ± 3.7 (3–19)	121.1 ± 38.1 (15–177)
Atagogawa (1996)	3	1.3–2.2	5.3 ± 0.6 (5–6)	59.0 ± 6.6 (53–66)	54.3 ± 7.8 (48–63)	49.0 ± 8.2 (42–58)	10.7 ± 1.5 (9–12)	29.0 ± 2.0 (27–31)	207.3 ± 13.6 (199–223)
Honjogawa (1995)	15	1.9–3.4	2.9 ± 0.9 (2–5)	59.6 ± 9.0 (45–76)	62.1 ± 6.1 (52–70)	65.7 ± 8.2 (52–77)	11.1 ± 1.6 (8–13)	15.9 ± 3.6 (10–21)	217.4 ± 22.2 (180–260)

[1] Total number on both right and left sides of proboscis.

[2] Reared for 2–4 months after artificial fertilization in laboratory.

from 18 localities (Fig. 38). Though the BW varied among populations within a species, the mean BW of *H. diadroma* sp. nov. was significantly larger than that of *H. atoka* sp. nov. in the sympatric populations (Mann–Whitney *U*-test, $P < 0.001$). In both species, the difference in BW between males and females was not significant. The smaller body size in *H. atoka* sp. nov. seems to be related to the shorter life-span of this species (see description for each species).

No marked difference in the number of setigers was detected among the three species, though available data were limited owing to frequent detachment of the posterior tip of the specimens.

Paragnath number

The number of paragnaths on the proboscis of each group was compared in mature adults collected from 18 localities throughout Japan, and in immature specimens (comparable to the mature adults in body size) collected from 19 localities in Korea and in Kyushu, Japan (Tables 2, 4, 5; Figs 39, 40).

Though the mean paragnath numbers (PN) varied among populations within a species, marked differences among species were detected in groups on the maxillary ring for both mature and immature specimens: the total PN for both right and left sides of the proboscis in group II were large in the populations of

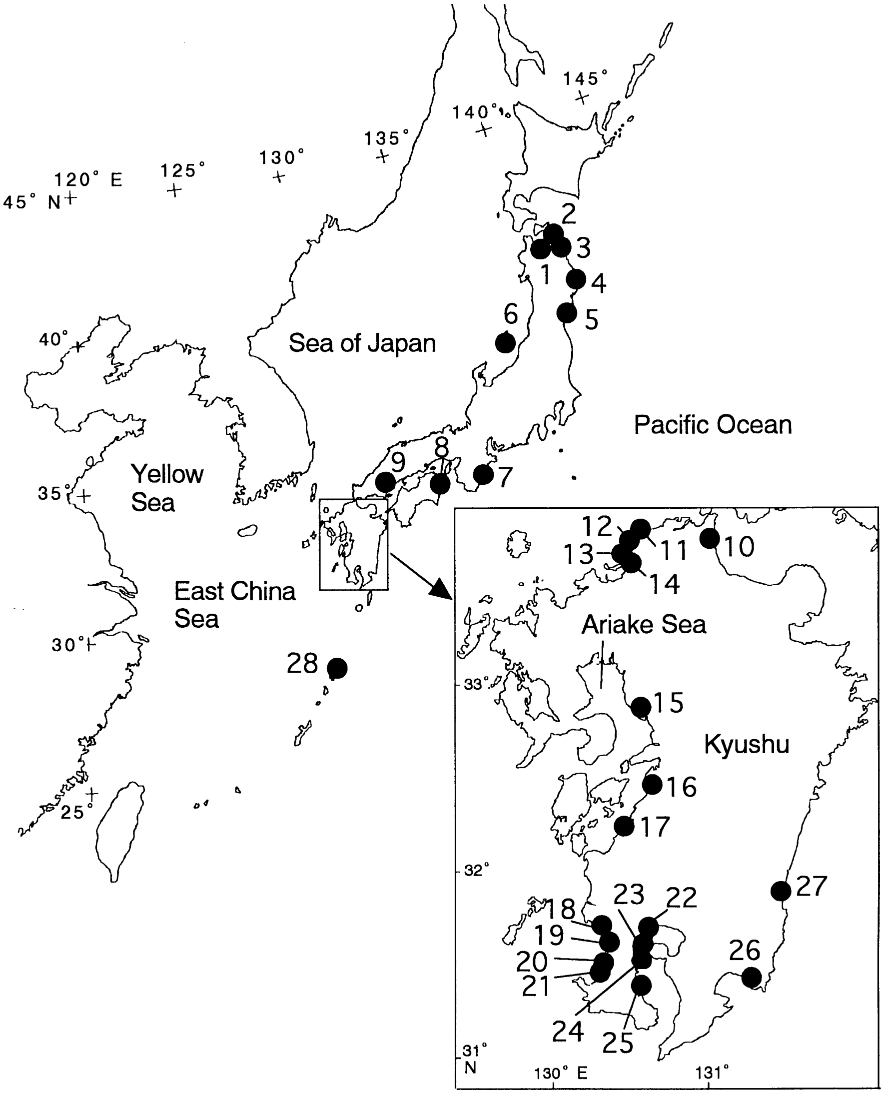


Figure 37. Distribution of ***Hediste atoka* sp. nov**. 1. Shinjogawa River (type locality). 2. Obuchinuma. 3. Niidagawa River. 4. Otsuchi. 5. Nanakitagawa River. 6. Kokufugawa River. 7. Kumanogawa River. 8. Yoshinogawa River. 9. Yahatagawa River. 10. Kusamigawa River. 11. Tsurikawa River. 12. Shiohama and Tsuyazaki. 13. Kazurugawa River. 14. Wajirogawa River. 15. Kikuchigawa River. 16. Maekawa River. 17. Minamatagawa River. 18. Osatogawa River. 19. Kaminokawa River. 20. Isakugawa River. 21. Manosegawa River. 22. Omoigawa River. 23. Kotsukigawa River. 24. Nagatagawa River. 25. Atagogawa River. 26. Honjogawa River. 27. Oyodogawa River. 28. Koshuku-okawa River.

H. atoka sp. nov. (more than 40 in most cases, except for two populations in Aomori Prefecture), intermediate in populations of *H. diadroma* sp. nov. (20–40 in most cases, except for two populations in Hiroshima Prefecture), and small in populations of *H. japonica* (less than 20 in most cases) (Fig. 41). The interspecific differences were significant in all combinations of sympatric populations with sufficiently large sample sizes for statistical testing, i.e. between *H. diadroma* sp. nov. and *H. atoka* sp. nov. (three localities with mature and four with immature specimens, Mann–Whitney *U*-test: $P < 0.05$), and between *H. diadroma* sp. nov. and *H. japonica* (a single locality with immature specimens, $P = 0.0001$) (Figs 39a, 40a). Total paragnath numbers on both right and left sides of the proboscis in group IV were also large in populations of *H. atoka* sp. nov. (more than 40 in most cases), intermediate in populations of *H. diadroma* sp. nov. (30–50 in most cases, except for two populations in Hiroshima Prefecture and one in Miyagi Prefecture), and small in populations of *H. japonica* (less than 30) (Figs 39b, 40b). The interspecific differences were significant in all combinations of sympatric populations with sufficiently large sample sizes, i.e. between *H. diadroma*

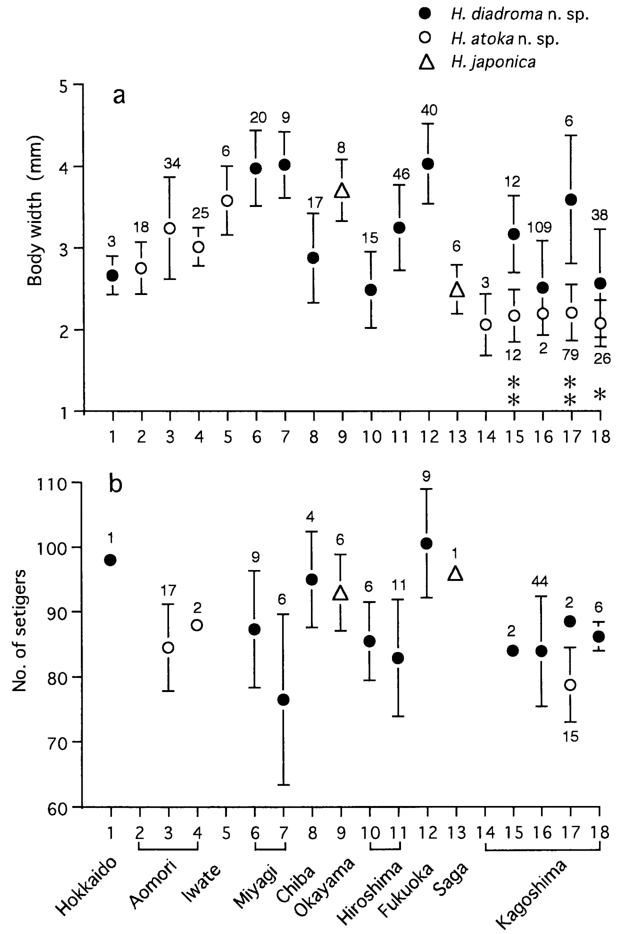


Figure 38. Body width (a) and number of setigers (b) of mature adults of 3 *Hediste* species collected from 18 localities in Japan. Average and SD bar are shown in each locality. The locality numbers are arranged from north to south. The number on each datum indicates sample size. Differences in body width between the sympatric two species in three localities (15, 17, 18) in Kagoshima Prefecture are significant (Mann–Whitney *U*-test, $^{*}P < 0.001$, $^{**}P < 0.0001$). 1. Usujiri. 2. Obuchinuma. 3. Shinjogawa River. 4. Niidagawa River. 5. Otsuchi. 6. Nanakitagawa River. 7. Natorigawa River. 8. Tonegawa River. 9. Kojima Bay. 10. Otagawa River. 11. Yahatagawa River. 12. Nakagawa River. 13. Nanaura. 14. Osatogawa River. 15. Kaminokawa River. 16. Omoigawa River. 17. Kotsukigawa River. 18. Nagatagawa River.

sp. nov. and *H. atoka* sp. nov. (three localities with mature and two with immature specimens, $P < 0.05$), and between *H. diadroma* sp. nov. and *H. japonica* (a single locality with immature specimens, $P < 0.0001$). Similar significant interspecific differences were also observed for group III, i.e. between *H. diadroma* sp. nov. and *H. atoka* sp. nov. (three localities each, with both mature and immature specimens, $P < 0.05$), and between *H. diadroma* sp. nov. and *H. japonica* (a sin-

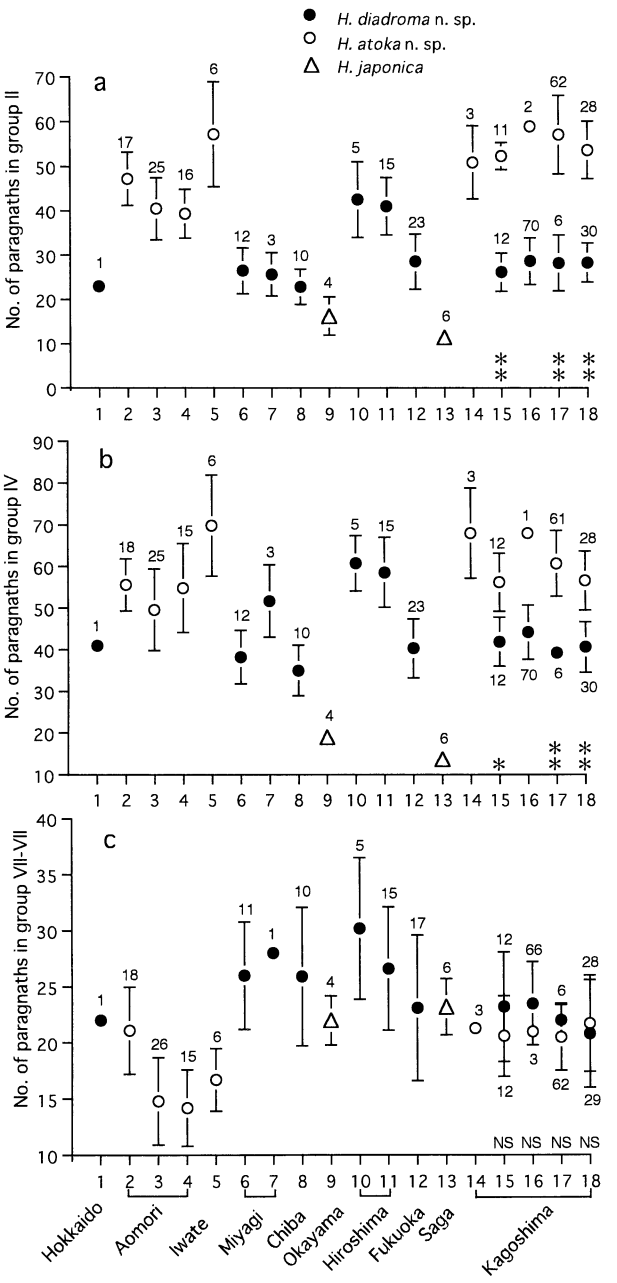
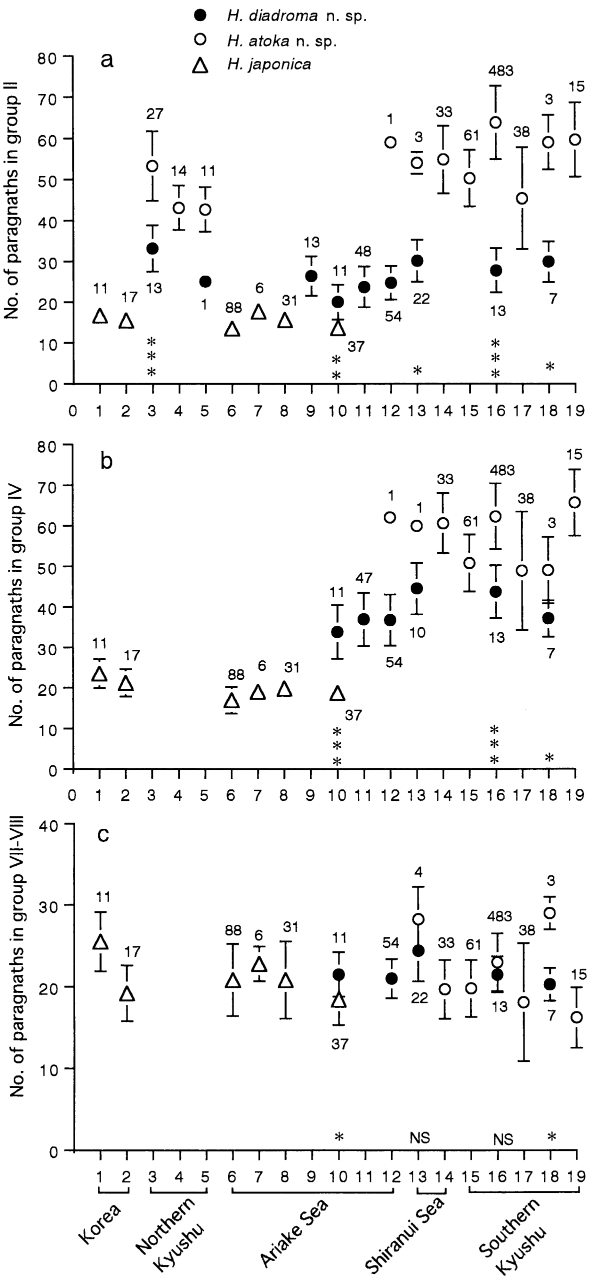


Figure 39. Number of paragnaths on groups II (a), IV (b) and VII–VIII (c) in proboscis of mature adults of 3 *Hediste* species collected from 18 localities in Japan. Total numbers on right and left sides are shown for groups II and IV. Average and SD bar are shown in each locality. Data from Tables 2, 4 and 5. The locality numbers are the same as in Fig. 38, and are arranged from north to south. The number on each datum indicates sample size. Significance of difference of mean numbers between the sympatric populations of ***H. diadroma* sp. nov**. and ***H. atoka* sp. nov**. in three or four localities in Kagoshima Prefecture was tested by the Mann–Whitney *U*-test; differences are not significant (NS, $P > 0.05$) or significant ($^{*}P < 0.001$, $^{**}P < 0.0001$).



gle locality with immature specimens, $P = 0.0002$). Paragnath numbers in group I were significantly larger in *H. atoka* sp. nov. than in *H. diadroma* sp. nov. in 4 out of 6 sympatric localities ($P < 0.05$), but not significantly different between the species in a sympatric locality ($P = 0.2$).

In contrast, numbers in groups VI and VII–VIII in the oral ring were not significantly different among the three species in most sympatric localities for both mature and immature specimens (Figs 39c, 40c). However, the total numbers for both right and left sides of the proboscis in group VI were significantly larger in *H. japonica* (average ± SD: 12.1 ± 1.8, $n = 37$) than in *H. diadroma* sp. nov. (9.3 ± 2.1, $n = 11$) in the Omutagawa River ($P = 0.0002$).

Figure 40. Number of paragnaths on groups II (a), IV (b) and VII–VIII (c) in proboscis of immature specimens of three *Hediste* species collected from 19 localities in Korea and Kyushu, Japan. Total numbers in right and left sides are shown for groups II and IV. Average and SD bar are shown in each locality, based on data shown in Tables 2, 4 and 5 and additional data for three localities (3–5). Body sizes of all specimens are comparable to those of mature adults (see Tables 2, 4 and 5). The locality numbers are arranged from north to south. The number on each datum indicates sample size. Significance of difference of mean numbers between the sympatric populations of ***H. diadroma* sp. nov.** and ***H. atoka* sp. nov.** in four localities (3, 13, 16, 18), and ***H. diadroma* sp. nov.** and *H. japonica* in a single locality (10), was tested by the Mann–Whitney *U*-test. Differences: not significant (NS, $P > 0.05$); significant ($^{*}P < 0.05$, $^{**}P < 0.001$, $^{***}P < 0.0001$). 1. Inchon. 2. Kunsan. 3. Kusamigawa River. 4. Tsuyazaki. 5. Wajirogawa River. 6. Daijugarami. 7. Rokkakugawa River. 8. Honmyogawa River. 9. Kobe. 10. Omutagawa River. 11. Domengawa River. 12. Kikuchigawa River. 13. Maekawa River. 14. Minamatagawa River. 15. Manosegawa River. 16. Omoigawa River. 17. Nagatagawa River. 18. Atagogawa River. 19. Honjogawa River.

Figure 41. Typical arrangements of paragnaths in right side of group II in 3 Asian *Hediste* species. (a) ***H. atoka* sp. nov.** collected from Omoigawa River. (b) ***H. diadroma* sp. nov.** collected from Kikuchigawa River. (c) *H. japonica* collected from Daijugarami. Scale bar = 1 mm.

A reduction of paragnath numbers was observed in the laboratory-bred specimens when compared with those taken from the field with a corresponding body size (Tables 4, 5; Fig. 42). In *H. atoka* sp. nov., numbers in all groups in the laboratory-bred specimens were significantly smaller than those of the field-derived mature and immature ones ($P < 0.001$). In *H. diadroma* sp. nov., paragnath numbers of the laboratory-bred specimens were significantly reduced only in groups of the maxillary ring ($P < 0.04$), and not significantly different in groups of the oral ring ($P > 0.3$), in comparison with the field-derived mature and immature specimens, except for no significant difference in

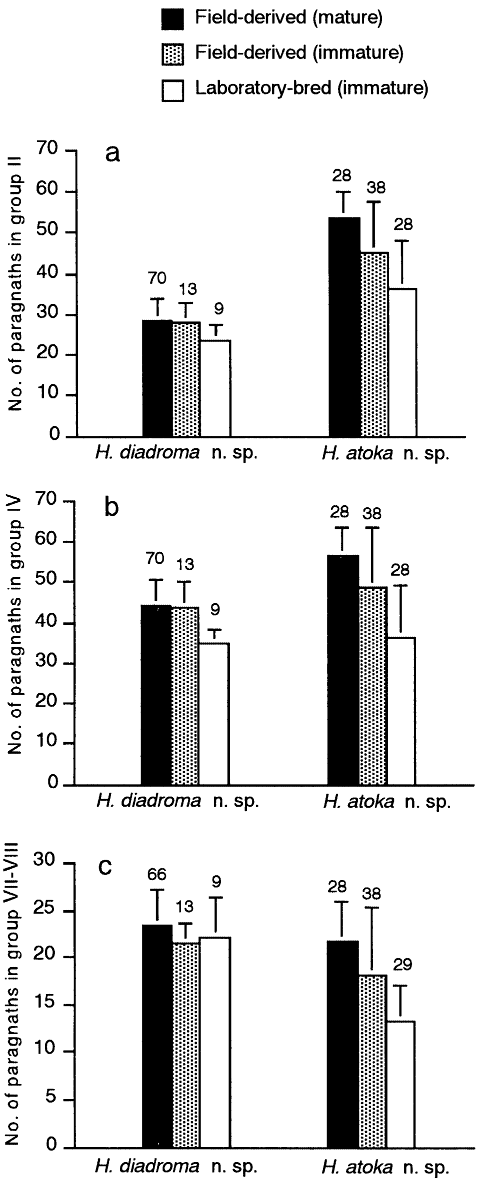


Figure 42. Comparisons of number of paragnaths on groups II (a), IV (b) and VII–VIII (c) in field-derived mature (■), immature (▦) and laboratory-bred immature (□) specimens of ***H. diadroma* sp. nov.** and ***H. atoka* sp. nov.** Total numbers in right and left sides are shown for groups II and IV. All specimens of *H. diadroma* sp. nov. were derived from the Omoigawa River, Kagoshima Prefecture. All specimens of *H. atoka* sp. nov. were derived from the Nagatagawa River, Kagoshima Prefecture. Average and SD bar are shown for each bar, based on data shown in Tables 4 and 5. The body sizes of all immature specimens are comparable to those of mature adults (see Tables 4 and 5). The number on each datum indicates sample size.

groups I and II of the maxillary ring in comparison with the field-derived immature specimens ($P > 0.07$).

Paragnath numbers in groups II and III in the laboratory-bred specimens were significantly larger in *H. atoka* sp. nov. than in *H. diadroma* sp. nov. ($P < 0.03$), though the numbers in group IV were not significantly different ($P > 0.1$).

DISCUSSION

ECOLOGICAL SIGNIFICANCE OF *HEDISTE* SPECIES DIVERSITY

The present study shows that the Asian *Hediste* involves at least three different species, and that these commonly coexist in the same place, forming a species complex. The coexistence of *H. diadroma* sp. nov. and *H. atoka* sp. nov. is widespread in Japan, while their ratio is variable among different sites (our unpublished data). *Hediste diadroma* sp. nov. also coexisted with *H. japonica* in a limited area of the Ariake Sea.

The species diversity in Asian *Hediste*, which is one of the most dominant genera in estuarine benthic communities, appears to be ecologically significant. *Hediste* species often attain high density and biomass in local populations, even in places where only few other benthic animals live, e.g. biomass in wet weight up to 620 g/m^2 in Mikawa Bay, Japan (Nagai & Nagai, 1981), and 390 g/m^2 in Qingdao, China (probably *H. diadroma* sp. nov. only, Qiu & Wu, 1993), and play an important role in the activity of nutrient cycling in an estuary ecosystem (Tsuchiya & Kurihara, 1979; Kurihara, 1983; Sayama & Kurihara, 1983; Kikuchi, 1986). The considerable biomass of the *Hediste* species is produced by the uptake of detritus or other organic matter, and is predated on by carnivores such as birds and bottom-dwelling fish.

Tidal flats in Asian estuaries supply important foraging places for many migratory birds such as *Charadrius* and *Calidris* from autumn to spring, because they are located along their flight paths (Hanawa & Takeishi, 2000). The *Hediste* species appear to constitute a major part of the food resource for these birds (unpublished data); the involvement of three different species with different life-cycles within the same or neighbouring estuaries further enhances this resource. For example, birds cannot prey on adults of *H. diadroma* sp. nov. just after their spawning in winter or early spring, but can prey on adults of *H. atoka* sp. nov. at that time.

INTERSPECIFIC DIFFERENTIATION IN PARAGNATH NUMBER

The paragnaths on the eversible proboscis seem to function for effective feeding and burrowing (Barnes &

KEY TO ASIAN SPECIES OF *HEDISTE*

1 Homogomph falcigers present, heterogomph spinigers absent in neuropodia. Neuropodial postsetal ligule tapering to digitate lobe throughout........ *H. japonica* (Izuka, 1908)
– Homogomph falcigers absent, heterogomph spinigers present in neuropodia. Neuropodial postsetal ligule tapering to digitate lobe only in anterior setigers........ 2
2 Epitoke-specific sesquigomph spinigers added to all notopodial and neuropodial fascicles in sexually full-mature adults. Mature eggs less than 170 µm in diameter........ ***H. diadroma* sp. nov**.
– Sexual maturity without addition of epitoke-specific chaetae and any other epitokous change. Mature eggs more than 200 µm in diameter........ ***H. atoka* sp. nov**.

Head, 1977; Hateley *et al*., 1992). Our results show that their numbers in the maxillary ring (at least in groups II, III, and IV) are significantly different among the three species in the sympatric populations, and that this difference (at least in groups II and III) was heritable in the laboratory-bred specimens of *H. diadroma* sp. nov. and *H. atoka* sp. nov. These findings suggest that there is a distinct genetic basis for determination of the paragnath numbers. However, the general reduction of paragnath numbers in the laboratory-bred specimens indicates that the actual number is influenced by environmental factors (e.g. food quality). The heritability of paragnath patterns was also demonstrated in *H. diversicolor*, which showed interpopulational differences (Hateley *et al*., 1992).

In *H. diversicolor*, the paragnaths of the maxillary ring are relatively long, strongly curved, and directed backwards and towards the lumen of the gut, while the paragnaths of the oral ring are shorter and show little curvature (Barnes & Head, 1977; Hateley *et al*., 1992). A similar tendency was observed in the three species in the present study. The paragnaths of the maxillary ring seem to grip relatively large food items effectively when the proboscis is retracted, while only the oral ring is everted during burrowing or browsing the sediment surface (Hateley *et al*., 1992). Marked differences in paragnath numbers in the maxillary ring between the three species imply a difference in diet or dominant mode of feeding. The paragnaths in the maxillary ring of *H. japonica* are fewer in number and larger in size in comparison with not only the two other Asian species, but also many populations of *H. diversicolor* in Europe and *H. limnicola* in North America (Smith, 1958; Barnes & Head, 1977; Khlebovich *et al*., 1982; Gillet, 1990; Hateley *et al*., 1992), suggesting some specialized feeding habits in *H. japonica*.

A number of feeding modes have been observed in *H. diversicolor*: predator and scavenger, a surface deposit-feeder (feeding on detritus or microorganisms deposited in the sediment surface), and a facultative filter-feeder (= suspension-feeder) (Harley, 1950; Rönn *et al*., 1988; Riisgård, 1991; Nielsen *et al*., 1995). When the phytoplankton concentration is sufficiently high, this species shifts from predatory/surface deposit-feeding to suspension-feeding. It is probable that the Asian *Hediste* species also have various feeding modes like *H. diversicolor*, though only surface deposit-feeding has been documented (Tsuchiya & Kurihara, 1979).

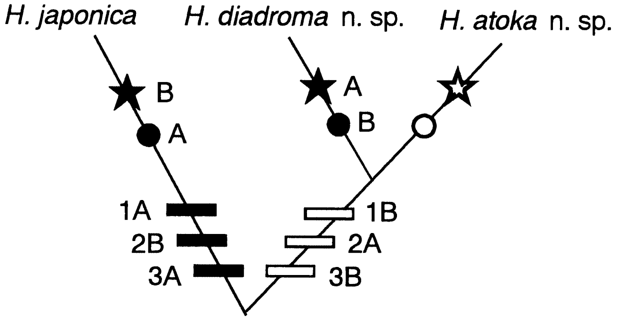


Figure 43. Hypothesis of the phylogenetic relationships in three Asian *Hediste* species. Stars indicate reproductive characteristics; (★) reproductive swarming of epitokous adults with (A) or without (B) addition of epitoke-specific sesquigomph spinigers; (☆) reproduction without swarming and any epitokous metamorphosis. Circular symbols indicate developmental characteristics; (●) pelagic development probably lecithotrophic from middle-sized eggs (180–210 µm in diameter) (A), or probably planktotrophic from small eggs (130–170 µm) (B); (○) direct development from large eggs (200–250 µm). (▬ & ▭) indicate the contrasting character combinations in immature specimens. 1. Neuropodial homogomph falcigers present (A) or absent (B). 2. Neuropodial heterogomph spinigers present (A) or absent (B). 3. Neuropodial postsetal ligule tapering to digitate lobe throughout (A) or only in anterior setigers (B).

EVOLUTION OF EPITOKY IN *HEDISTE*

H. japonica is morphologically distinguishable from the other two species even at the immature stage by three independent characteristics, while *H. diadroma* sp. nov. and *H. atoka* sp. nov. are indistinguishable,

Table 6. Comparison of epitokous metamorphosis among Asian nereidids

Species	Enlargement of eye	Modification of parapodial ligules and cirri	Modification of chaetae	References
Hediste japonica	Slight	Slightly enlarged	Increase in number of homogomph spinigers	Present study
***H. diadroma* sp. nov.**	Slight	Slightly enlarged	Addition of epitoke-specific sesquigomph spinigers in middle setigers	Present study
Neanthes virens	?	Slightly enlarged	Addition of epitoke-specific sesquigomph spinigers	Imajima, 1972
Namalycastis (3 species)	?	?	Presence of epitoke-specific sesquigomph or heterogomph spinigers	Glasby, 1999
Namanereis (2 species)	?	?	Presence of epitoke-specific slender capillaries	Glasby, 1999
Many species of 10 genera*	Marked in most cases	Markedly enlarged and deformed in middle or posterior setigers	Replaced by paddle-like chaetae in middle or posterior setigers	Imajima, 1972, 1996 Wu *et al.* 1985

**Tylorrhynchus, Nicon, Rullierinereis, Leonnates, Ceratonereis, Platynereis, Perinereis, Nectoneanthes, Nereis, Neanthes*.

suggesting that the latter two species are most closely related (Fig. 43). The similar electrophoretic patterns of LDH also suggest a closer relationship between *H. diadroma* sp. nov. and *H. atoka* sp. nov. They share a common dominant allele at one of two LDH loci (Sato & Masuda, 1997), while *H. japonica* seems to share no common allele with them (Fig. 10).

Epitokous metamorphosis (slight enlargement of eyes and parapodial ligules) occurs in mature adults of *H. diadroma* sp. nov. and *H. japonica* in combination with their reproductive behaviour of swarming. Elongation of the dorsal and ventral cirri occurs only in males of both species. The number of spinigers increased in the epitokous adults in different ways between the two species, i.e. an increase in the number of ordinary homogomph spinigers in *H. japonica*, and the addition of epitoke-specific sesquigomph spinigers in *H. diadroma* sp. nov. In either case, the epitokous metamorphosis was markedly different from the typical one found in many other nereidids (Table 6).

Epitokous metamorphosis in many other nereidids involves a common drastic morphological change to the 'heteronereid form' in both males and females or in males only, i.e. the marked enlargement of eyes, the division of the body into two or three parts, the enlargement and/or deformation of the parapodial ligules and cirri (most marked in the middle or posterior setigers), and all or most chaetae replaced by paddle-like chaetae (Imajima, 1972, 1996; Schroeder & Hermans, 1975; Wu *et al*., 1985). The change to the heteronereid form seems to be adaptive, serving to increase swimming ability during reproductive swarming.

However, epitokous metamorphosis in the two *Hediste* species is relatively inconspicuous, and characterized by the addition of slender spinigers instead of replacement by paddle-like chaetae. Metamorphosis with the addition of epitoke-specific spinigers similar to that of *H. diadroma* sp. nov. is known in *Neanthes virens* (Imajima, 1972) and three species of *Namalycastis* (Glasby, 1999) (Table 6). The addition of spinigers may be for defence against predators, and thus appear to add little to the worms' swimming ability. In fact, the *Hediste* epitokes swim relatively slowly during swarming (pers. observ.). The epitokous metamorphosis and reproductive swarming in *Hediste* may have evolved independently of those in many other nereidids, as suggested by Sato (1999).

Reproductive swarming towards the sea seems to be important for *H. diadroma* sp. nov., which migrates between brackish waters and the sea within its lifecycle, to perform successful fertilization and early development in salinity approaching that of full-strength seawater (at least 20‰) (Sato & Tsuchiya, 1987; Sato, 1999). However, there is no evidence showing the migration of adults during reproductive swarming in *H. japonica.* In Kojima Bay (Izuka, 1908) and the Ariake Sea (present study), the reproductive swarming of *H. japonica* was observed only within the habitat of immature worms. Therefore, the evolutionary significance of reproductive swarming in *H. japonica* is unknown.

The reproductive swarming of mature adults is necessarily followed by pelagic early development, because eggs and sperm are shed during the swarming. Judging from the description of Izuka (1908), the early development is probably lecithotrophic with a relatively short pelagic life (about 10 days) in *H. japonica* with an intermediate egg size

(180–210 µm in diameter). It is different from the probable planktotrophic development with a longer pelagic life (about 1 month) in *H. diadroma* sp. nov. with a smaller egg size (130–170 µm) (Kagawa, 1955; Sato & Tsuchiya, 1991), and from the direct development in *H. atoka* sp. nov. with a larger egg size (200–250 µm) (Sato & Tsuchiya, 1991).

The distribution of *H. japonica* is restricted to muddy tidal flats (Fig. 15), where tidal ranges are the greatest in Asia (mean tidal range of around 3 m or more in the spring tides) (Fig. 44). In these tidal flats, the fine muddy particles of sediments are severely disturbed by water movement produced by the strong tidal current during each flooding and ebbing (Sato & Takita, 2000). In these places, *H. atoka* sp. nov. has never been found, while *H. diadroma* sp. nov. coexists with *H. japonica* in some areas. In *H. atoka* sp. nov., spawning, fertilization and early development to 3-setiger juveniles occur within a burrow (Sato & Tsuchiya, 1991; Sato, 1999), and may be unsuccessful in such unstable muddy sediments because of the difficulty of maintaining an intact burrow during this process. The reproductive swarming followed by pelagic development in *H. japonica* may be adaptive to a unique habitat with extremely unstable sediments.

SPECIES ENDANGERED BY HUMAN IMPACT

The Ariake Sea (1700 km^2 in area) in Kyushu is unusual in that it is characterized by a considerable tidal range (a maximum of over 6 m in spring tides) and has the largest extent of tidal flats (about 200 km^2) in Japan. About 20 indigenous species which are endemic to it, or inhabit only the Chinese and Korean coasts, are known. These include fishes and invertebrates (Sato & Takita, 2000). The distribution of most of these species is limited to the innermost part of the Ariake Sea. The distributional range of *H. japonica* (Fig. 15) is similar to that of the other indigenous species. The restricted distribution may be due not only to their narrow habitat-preference related to the great tidal range, but also to the rapid reduction of available habitats resulting from recent human impact.

In Asia, many tidal flats have been heavily damaged by pollution and reclamation. In Japan, about half of

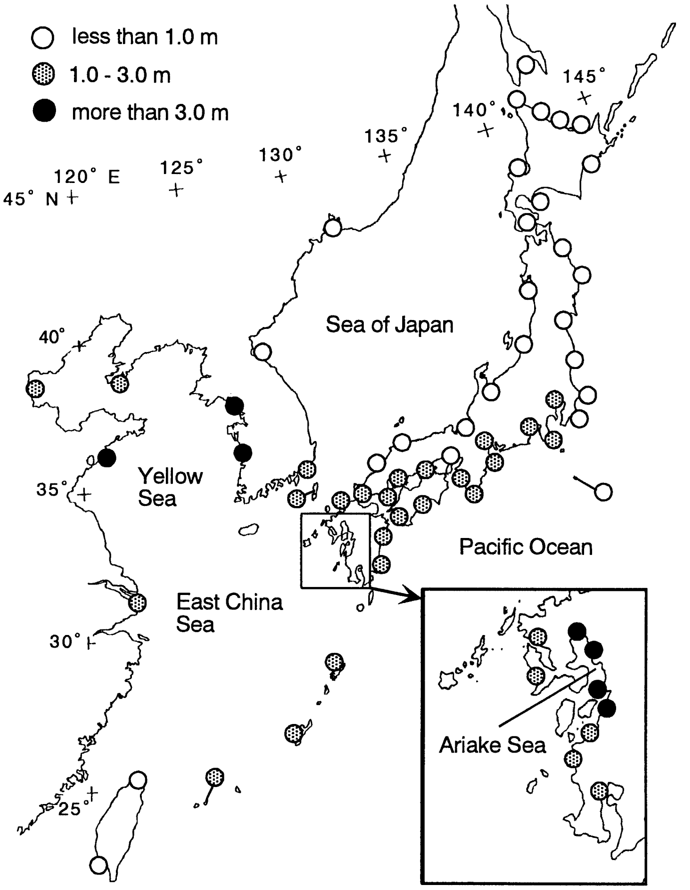


Figure 44. Mean tidal range (vertical distance between mean low and high tide marks) in spring tides in Japan and neighbouring countries. After Sato & Takita (2000).

the tidal flats have already disappeared, mostly during the past 100 years, because of artificial reclamation (Sato & Takita, 2000). The tidal flats in Kojima Bay (the type locality of *H. japonica*) in the Seto Inland Sea were lost in 1959 to reclamation. Within Japan, we have not succeeded in collecting *H. japonica* outside the inner part of the Ariake Sea. A recent reclamation project begun in 1997 has caused the loss of a muddy shallow area of about 36 km^2, including the most important habitat for *H. japonica*, in Isahaya Bay in the inner part of the sea (Fig. 15). The Korean tidal flats, where the tidal range is the greatest in Asia (a maximum of over 9 m in spring tides), are also inhabited by *H. japonica*; these have also been seriously damaged by reclamation on an even larger scale (Kellermann & Koh, 1999; Hong, 2000).

A reduction in the distributional range of muddy shallow-water fauna in Japan is well documented in some bivalves such as *Tagillarca granosa*, for which many fossil records are available. *Tagillarca granosa* is one of the most common species found in shell mounds around Tokyo (Morse, 1879), and living specimens were recorded in many bays in western Japan until 100 years ago (S. Sato, 2000). At present, however, its distribution is restricted probably only to the inner part of the Ariake Sea. It is possible that *H. japonica* may become as rare as *T. granosa* in Japan.

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