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Ostracods (Crustacea) from Sarobetsu Marsh, Northern Hokkaido, Japan: Taxonomy and Phenology with Description of *Pseudocandona tenuirostris* sp. nov.

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We sampled ostracods in Sarobetsu Marsh, Rishiri Rebus Sarobetsu National Park, Hokkaido, Japan, from April 2005 to November 2007 and identified four species: *Cryptocandona* sp., *Pseudocandona tenuirostris* sp. nov., *Physocypria nipponica* Okubo, 1990, and *Metacypris digitiformis* Smith and Hiruta, 2004. *Cryptocandona* sp. is similar to *C. reducta* (Alm, 1914) and *C. brehmi* (Klie, 1934), but positive identification is postponed due to the absence of male specimens. In having three setae in the setal group of the second segment of the mandibular palp, *Pseudocandona tenuirostris* sp. nov. belongs to the *Ps. rostrata* species group, and it is characterized by a beak-like medial lobe on the distal end of each hemipenis. From literature based information, Japanese populations of *Physocypria nipponica* and *Ph. kraepelini* from Europe have morphological differences at their hemipenis. We regard *Ph. nipponica* as a distinct species, even though this taxon has been suggested to be a synonym of *Ph. kraepelini* Müller, 1993. We discuss the seasonal prevalence of the four species detected.

Key Words: Crustacea, Ostracoda, new species, seasonal changes, Hokkaido, Japan.

Introduction

Freshwater ostracods are small crustaceans, usually less than 1 mm in body length, with a bivalved carapace made of magnesium calcite (Karanovic 2012); different groups of ostracods are thought to have invaded non-marine habitats independently from marine ancestors (Martens *et al.* 1998). So far, approximately 1900 living species in 200 genera of freshwater ostracods are known worldwide (Martens *et al.* 2008), of which 90 have been reported from Japan (*e.g.*, Okubo 1990; Hiruta and Smith 2001; Smith and Hiruta 2004; Smith and Kamiya 2006; Smith and Janz 2008). Rather than in lotic environments including mountain rivers, they are more frequently found in lentic ecosystems such as freshwater wetlands (Meisch 2000).

Previous studies have detected a number of ostracod species in freshwater wetlands in Japan; for example, 16 non-marine species have been reported from Kushiro Marsh, Japan's largest freshwater wetland (Hiruta and Smith 2001; Smith and Hiruta 2004). Sarobetsu Marsh is the second largest; located in Hokkaido, northern Japan, it covers 67 km² and accounts for 15% of the total area of Japan's freshwater wetlands. The ostracod fauna in Sarobetsu Marsh, however, has not been previously investigated.

In this paper, we report the results of our three-year faunal survey of freshwater ostracods in Sarobetsu Marsh. We also show the seasonal prevalence of each species found within the research period; like in many other aquatic organisms, abundance and diversity of ostracods are known to vary with the seasons (*e.g.*, Hull 1997).

Material and Methods

We sampled at five sites (Figs 1, 2) once a month, from April to November, for three years from 2005 to 2007. The five sampling sites covered various environments in and around Sarobetsu Marsh (Table 1). Samples were quantitative, each consisting of 300 cm³ of material retained after straining both water and substrates through a 0.1 mm mesh sieve at each sampling site. We preserved samples in 70% ethanol and extracted ostracods from them under a stereoscopic microscope.

We dissected appendages, mounted them in Hoyer's solution on glass slides, and drew them with the aid of a camera lucida. We used a tragacanth gum solution to paste some carapaces onto microfossil slides. We treated carapaces (including the soft parts) with hexamethyldisilazane (HMDS) (Nation 1983) and examined them by SEM at 15–20 kV accelerating voltage. The material used in this study has been deposited in the Hokkaido University Museum, Sapporo (ZIHU). All specimens listed in "Material examined" sections were collected and prepared by S. F. Hiruta.

The chaetotaxic notation follows that of Broodbakker and Danielopol (1982), as revised for the antenna by Martens (1987) and for the thoracopods by Meisch (1996). Hemipenis terminology follows that of Danielopol (1969).

We use the same abbreviations for appendages as Meisch (2000): γ_{23} , aesthetasc of antennule; G_{1-3} , apical claws on penultimate segment of antenna; G_M , G_m , apical claws on terminal segment of antenna; Y, γ_1 , γ_2 , γ_3 , aesthetascs on antenna; h_{1-3} , terminal setae 1–3 on walking leg and cleaning

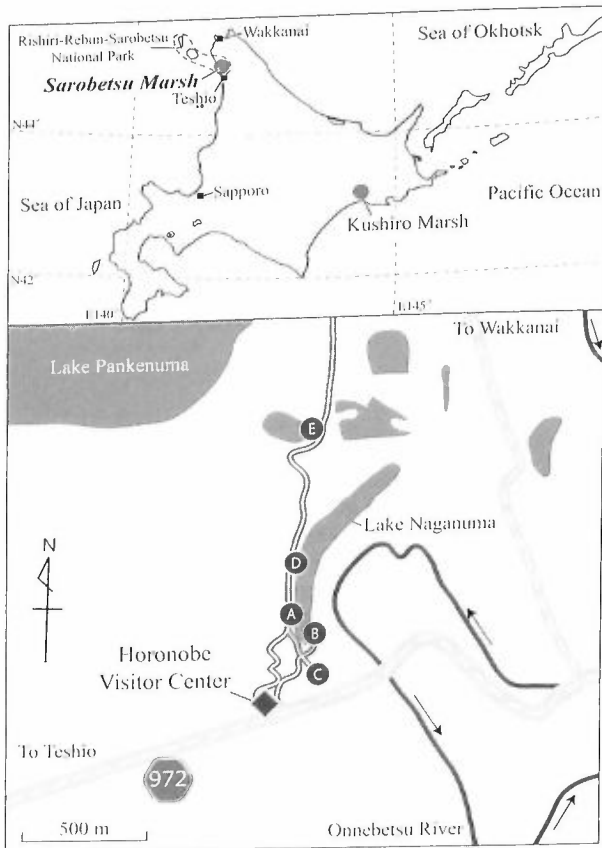


Fig. 1. Map showing the location of Sarobetsu and Kushiro Marshes (above); diagram of study area (below), with sampling sites A to E indicated by circles; black arrows indicate direction of river current.

leg; a, outer lobe of hemipenis; b, inner lobe of hemipenis; e, brusa complex; h, medial lobe of hemipenis; M, M process of hemipenis; CP, copulatory process; E, endopod; Pr, protopod; Exo, exopodite; DL, dorsal lobe; DR, dorsal ridge; MR, medial ridge; UR, upper claspig ramus; ZY, zygum.

Family **Candonidae** Kaufmann, 1900
Subfamily **Candoninae** Kaufmann, 1900
***Cryptocandona* sp.**
(Figs 3–6)

Material examined. ZIHU 3912, 1 female, site C (Table 1, Fig. 1), 5 November 2005, dissected, soft parts mounted on 16 slides; ZIHU 3913, 1 female, same site, 8 October 2005, soft parts mounted on 14 slides; ZIHU 3914, 3915, 2 females, same site, 6 November 2007, mounted on stubs for SEM observation.

Description of female. Carapace (Figs 3A, B, 4A, B) 0.97–0.99 mm long, 0.44–0.47 mm high ($n=4$); elongate in lateral view, highest just behind mid-length, broadly round-

Table 1. Locations and habitats of sampling sites.

Site	Latitude/longitude	Environment
A	45°00'47.2"N 141°43'53.4"E	Shallow (up to 50 cm) and vegetated
B	45°00'44.0"N 141°43'56.7"E	About 1 m in depth; vegetated
C	45°00'39.5"N 141°43'56.3"E	Hypogeous habitat
D	45°00'50.3"N 141°43'53.3"E	Open-water habitat
E	45°01'26.0"N 141°43'59.6"E	Very shallow (up to 20 cm) and heavily vegetated with reeds and sedges

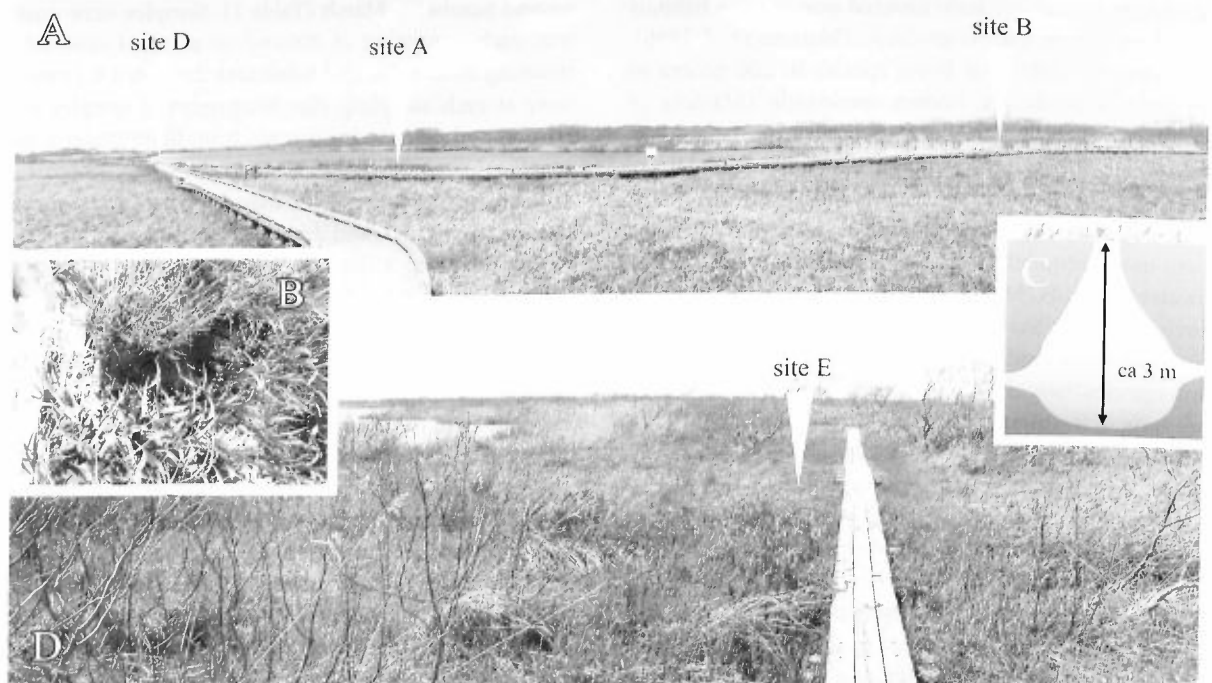


Fig. 2. Photographs of sampling sites in Sarobetsu Marsh. A, Southern part of Lake Naganuma, taken from a wooden path on 28 May 2005; B, small entrance hole to site C, a sink hole called a *yachi-manako*; C, diagram of the structure of a *yachi-manako*; D, site E, located in the northern part of Lake Naganuma.

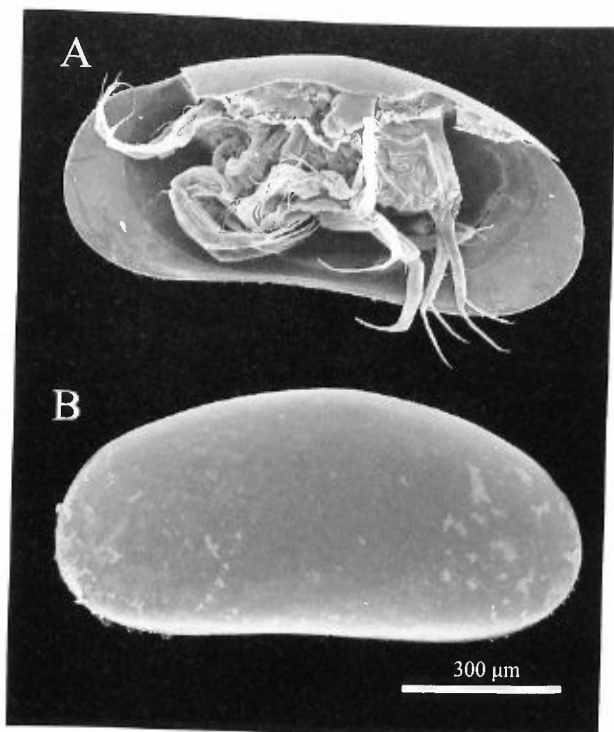


Fig. 3. *Cryptocandona* sp., SEM images. A, Female, ZIHU 3914, lateral view with left valve removed; B, female, ZIHU 3915, lateral view of left valve.

ed at anterior and posterior ends, slightly concave ventrally; conspicuously narrow in dorsal view, pointed at anterior and posterior ends. Valves very thin, transparent, with pearly luster; short, fine setae present on anterior, posterior, and ventral margins; other parts of surface with a few fine setae.

Antennule (Fig. 4C) seven-segmented. First two podomeres fused, with two dorsal setae and two long apico-ventral setae. Third podomere quadrate, with apico-dorsal seta. Fourth podomere quadrate, with apico-ventral seta. Fifth and sixth podomeres both quadrate; each with two long apico-dorsal setae and one apico-ventral seta. Seventh podomere elongate, with two long, one medium-long, and one short apical setae. Eighth podomere elongate and slender, with one long and two shorter apical setae and aesthetasc y_3 .

Antenna (Fig. 4D) four-segmented. First podomere with two antero-proximal setae and one long apico-ventral seta. Second podomere with one apico-ventral seta, aesthetasc Y, and exopodite (Exo); exopodite consisting of one long and two very short setae. Third podomere with long G_1 and G_3 , shorter G_2 , two apical setae, two mid-dorsal setae, four mid-ventral setae, apico-ventral aesthetasc y_2 , and postero-vental aesthetasc y_1 . Fourth podomere with G_M , shorter G_{III} , one apical seta, and aesthetasc y_3 .

Mandible (Fig. 5A, B) consisting of coxal plate and four-segmented palp. Coxal plate with seven strong teeth and several setae of various lengths at distal end, and one antero-lateral plumose seta (plumage not shown in Fig. 5A, B). First podomere with vibratory plate (Exo) externally, one long and one short inner-distal plumose setae, one long antero-distal seta, and alpha seta. Second podomere with

group of three setae, one medium-long and one beta inner-distal setae, and two outer apical setae. Third podomere with three outer middle setae, one outer apical seta, one mid-apical gamma seta, two medium-long setae, and two short inner apical setae. Fourth podomere with one distally plumed claw, one terminal claw, and one medium-long and one shorter apical setae.

Maxillula (Fig. 5C) with elongate vibratory plate (Fig. 5D), three masticatory processes, and two-segmented palp.

Maxilliped (Fig. 5E) with non-segmented palp, vibratory plate, one antero-proximal seta, one antero-distal seta, and one postero-distal seta. Palp with three apical setae. Vibratory plate (Exo) with two filaments. Masticatory process slender, with numerous setae.

Walking leg (Fig. 6A) five-segmented. Terminal claw (h_2) slender and long.

Cleaning leg (Fig. 6B) four-segmented. First podomere with three setae. Second podomere with one apical seta. Third podomere with one apical and one middle setae. Fourth podomere with one long (h_3), one medium-long (h_2), and one shorter (h_1) setae.

Uropodal ramus (Fig. 6C) with anterior seta and posterior seta. Two terminal claws with tiny denticles.

Remarks. Positive identification of our female specimens is difficult, as examination of male copulatory organs is necessary. Considering 12 species recognized in *Cryptocandona* (cf. Meisch 2000), nevertheless, our material is morphologically similar to *C. reducta* (Alm, 1914) (Alm 1914; Meisch 2000) and *C. brehmi* (Klie, 1934) (Namiotko and Danielopol 2002) in the overall proportions of the soft parts and carapace shape. Our specimens, however, exhibit a mix of character states from these two species (Table 2): the carapace length in our specimens is 0.97–0.99 mm (0.90–1.02 mm in *C. reducta*, 0.82 mm in *C. brehmi*); the number of setae on left and right antennule is symmetrical (symmetrical in *C. reducta*, asymmetrical in *C. brehmi*); G_M and G_3 on the antenna are shorter (longer in *C. reducta*, shorter in *C. brehmi*) than 0.8 times E_1 ; and the genital lobe protrudes slightly (protrudes slightly in *C. reducta*, flat in *C. brehmi*). Furthermore, in our specimens, the posterior seta on the uropod is relatively short; it is 12–14% the length of the anterior margin of the caudal ramus, while it is 16–25% and 15–16%, respectively, in *C. reducta* and *C. brehmi*. *Cryptocandona reducta* is widely distributed in Europe, with some populations reproducing sexually (Klie 1925, 1940; Pax 1942; Hartmann *et al.* 1999) and others solely parthenogenetically (e.g., Meisch 2000); *C. brehmi* is only known from the type series, consisting of two juveniles and two females, from a pool in a cave at “Hirowagara”, Sakuho-machi, Nagano Prefecture, Japan (Klie 1934).

Pseudocandona tenuirostris sp. nov.

(Figs 7–11)

Type material. Holotype: ZIHU 3916, male, site C (Table 1), Sarobetsu Marsh, Rishiri Reibun Sarobetsu National Park, Hokkaido, Japan (Fig. 1), 28 May 2005, soft parts mounted on 20 slides, carapace mounted on a micro-

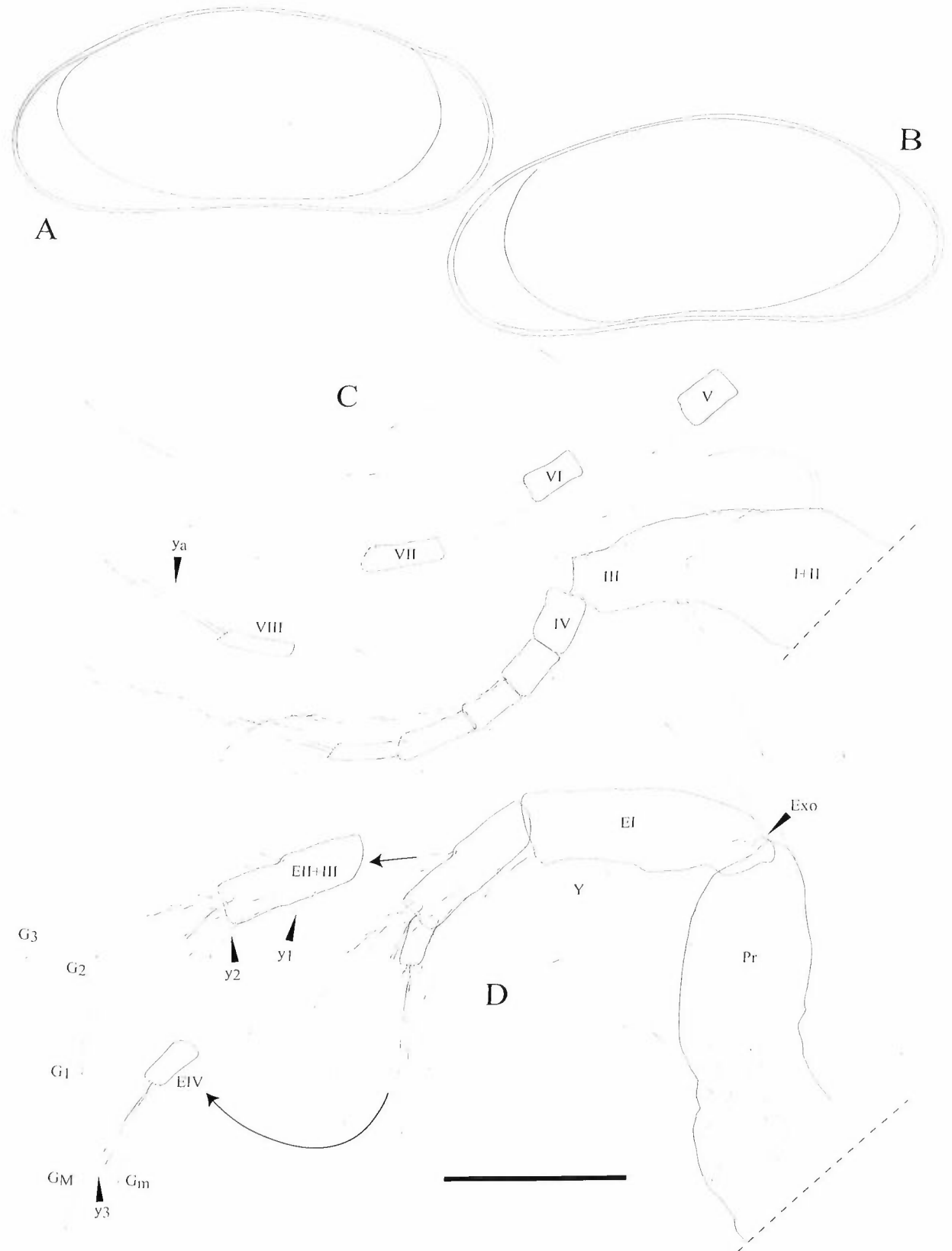


Fig. 4. *Cryptocandona* sp., female, ZIHU 3912. A, Lateral view of right valve; B, lateral view of left valve; C, antennule; insets, details of endopodites of fifth to eighth podomeres; D, antenna (inset, details of two terminal endopodal podomeres). Scale bar: A, B, 300 μ m; C, D, 100 μ m.

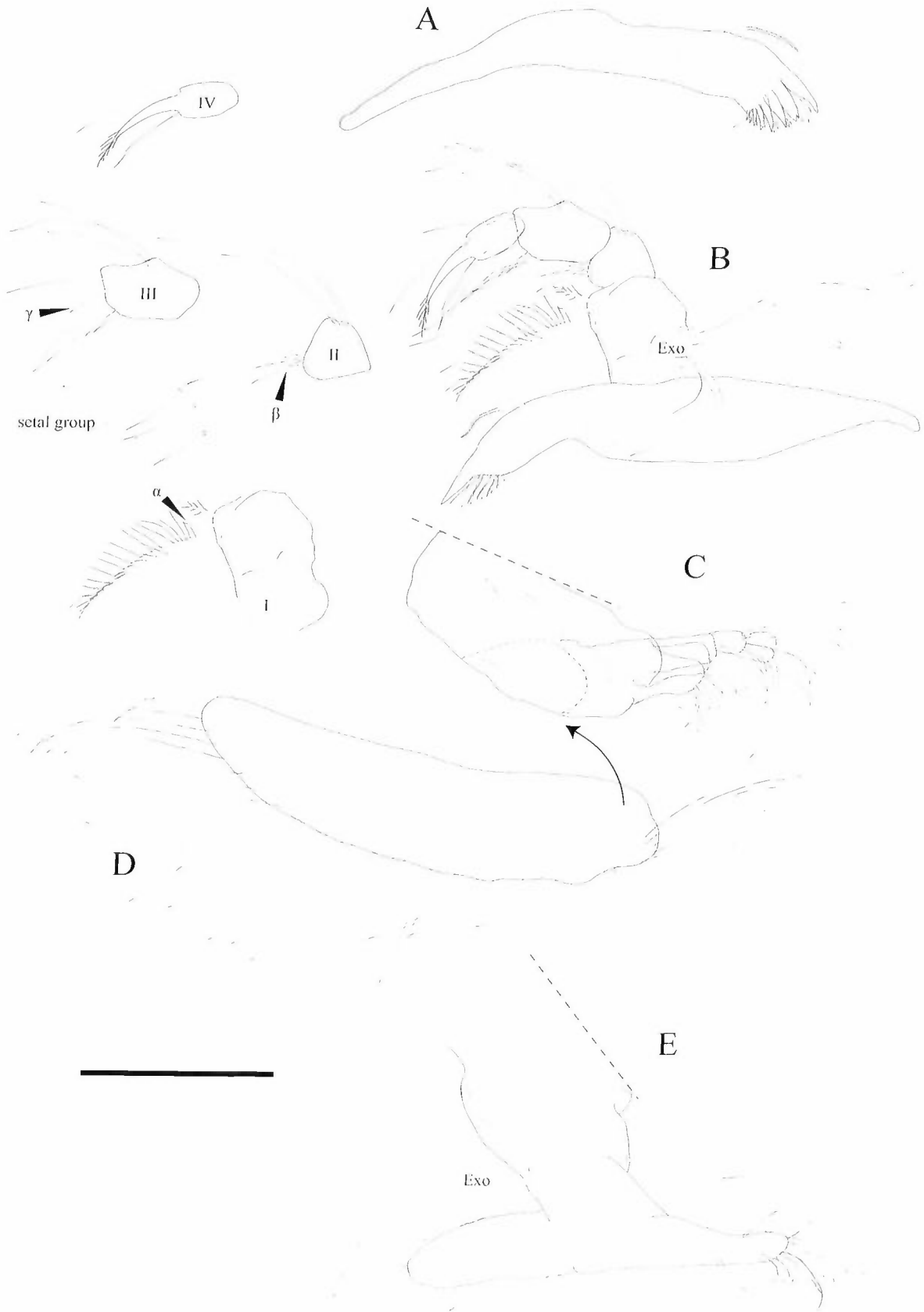


Fig. 5. *Cryptocandona* sp., female, ZIHU 3912. A, Coxal plate of mandible; B, mandible, with enlargements of four palp segments shown to the left; C, maxillula; D, vibratory plate of maxillula; E, maxilliped. Scale bar: 100 μ m.

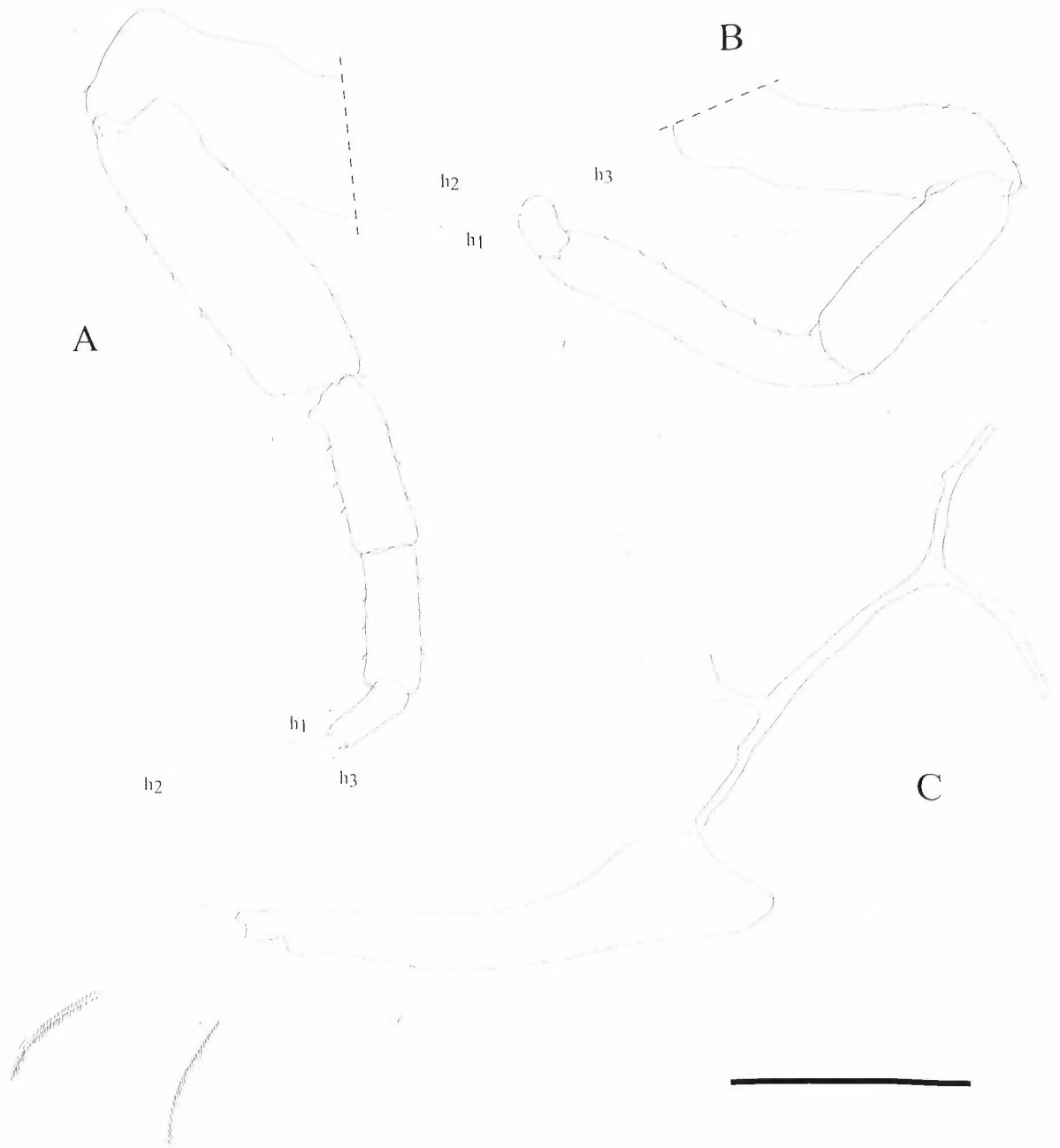


Fig. 6. *Cryptocandona* sp., female, ZIHU 3912. A, Walking leg; B, cleaning leg; C, uropod, with uropodal rami. Scale bar: 100 μ m.

Table 2. Character matrix of three *Cryptocandona* species.

	<i>C. reducta</i>	<i>C. brehmi</i>	<i>Cryptocandona</i> sp.
Carapace length	0.90–1.02 mm	0.82 mm	0.97–0.99 mm
Setae on both side of antennule	symmetrical	asymmetrical	symmetrical
Relative length of G_{34} and G_4 on the antenna	longer	shorter	shorter
Genital lobe	slightly protruding	flat	slightly protruding
Relative length of posterior seta on uropod	longer (16–25%)	longer (15–16%)	shorter (12–14%)

fossil slide. Allotype: ZIHU 3917, female, same site, Sarobetsu Marsh, 10 May 2005, soft parts mounted on 15 slides, carapaces mounted on a microfossil slide. Paratypes: ZIHU

3918, 3919, two males, same data as for allotype, mounted on slides; ZIHU 3920, 3921, two females, same data as for allotype, mounted on slides; ZIHU 3922, 3923, two males,

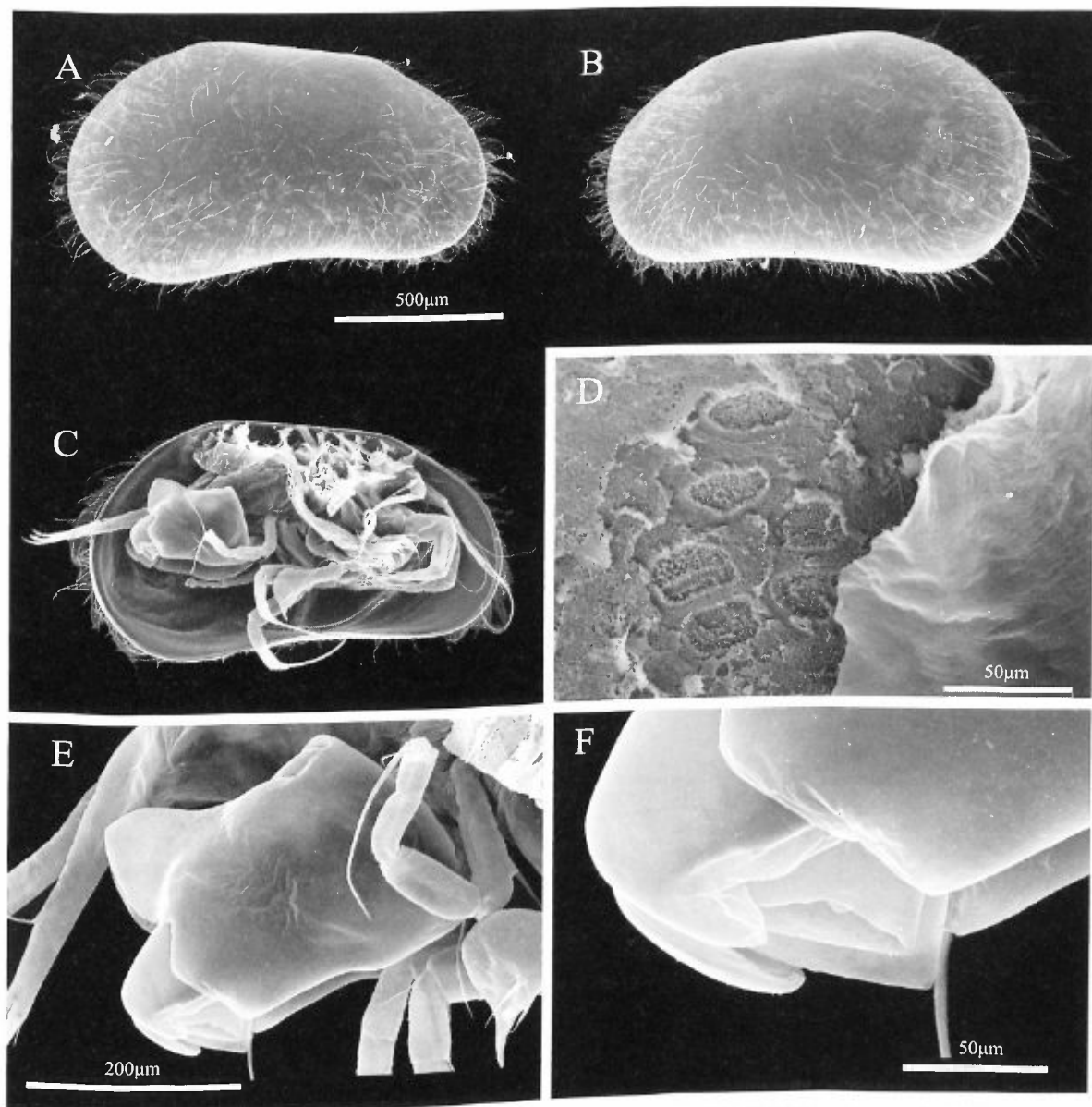


Fig. 7. *Pseudocandona tenuirostris* sp. nov., SEM images. A, B, E, F, Paratype, male, ZIHU 3922; C, D, paratype, male, ZIHU 3923. A, Lateral view of right valve; B, lateral view of left valve; C, lateral view of male with right valve removed; D, internal view of right valve with muscle scar; E, lateral view of hemipenis; F, enlarged image of end of hemipenis.

same data as for allotype, mounted on stubs for SEM observation.

Description of male. Carapace (Figs 7A, B, D, 8A–C) 1.24–1.27 mm long, 0.71–0.74 mm high ($n=4$; 1.25 mm long, 0.71 mm high in holotype); moderately compressed in dorsal view, posterior end round, anterior end slightly beak shaped (Fig. 8C); left valve overlapping right valve both anteriorly and posteriorly. Dorsal margin straight in lateral view, sloping towards anterior end; greatest height exceeds half the length. Surface of valves with long, stiff, perpendicularly attached setae and covered with numerous tiny pits observable by SEM (Fig. 7A, B).

Antennule (Fig. 8D) seven-segmented. First two podomeres fused, with two long dorsal setae and two long apico-ventral setae. Third podomere quadrate, with apico-dorsal

seta. Fourth podomere quadrate, with apico-dorsal seta. Fifth and sixth podomeres both quadrate, each with two long apico-dorsal setae. Seventh podomere with two long and one shorter apico-dorsal setae. Eighth podomere elongate and slender, with two long setae, one shorter seta, and aesthetasc γ .

Antenna (Fig. 8E) five-segmented. First podomere with one long, apico-ventral seta, one medium-long, antero-proximal, plumose seta, one long, antero-proximal seta, and one long latero-proximal seta. Second podomere with two apico-ventral setae, aesthetasc Y, and exopodite (Exo); exopodite consisting of one long and two short setae. Third podomere with two mid-apical male bristles (t2 and t3), one short apico-ventral and one apico-dorsal setae, and short mid-ventral seta. Fourth podomere with long G_1 and G_2 ,

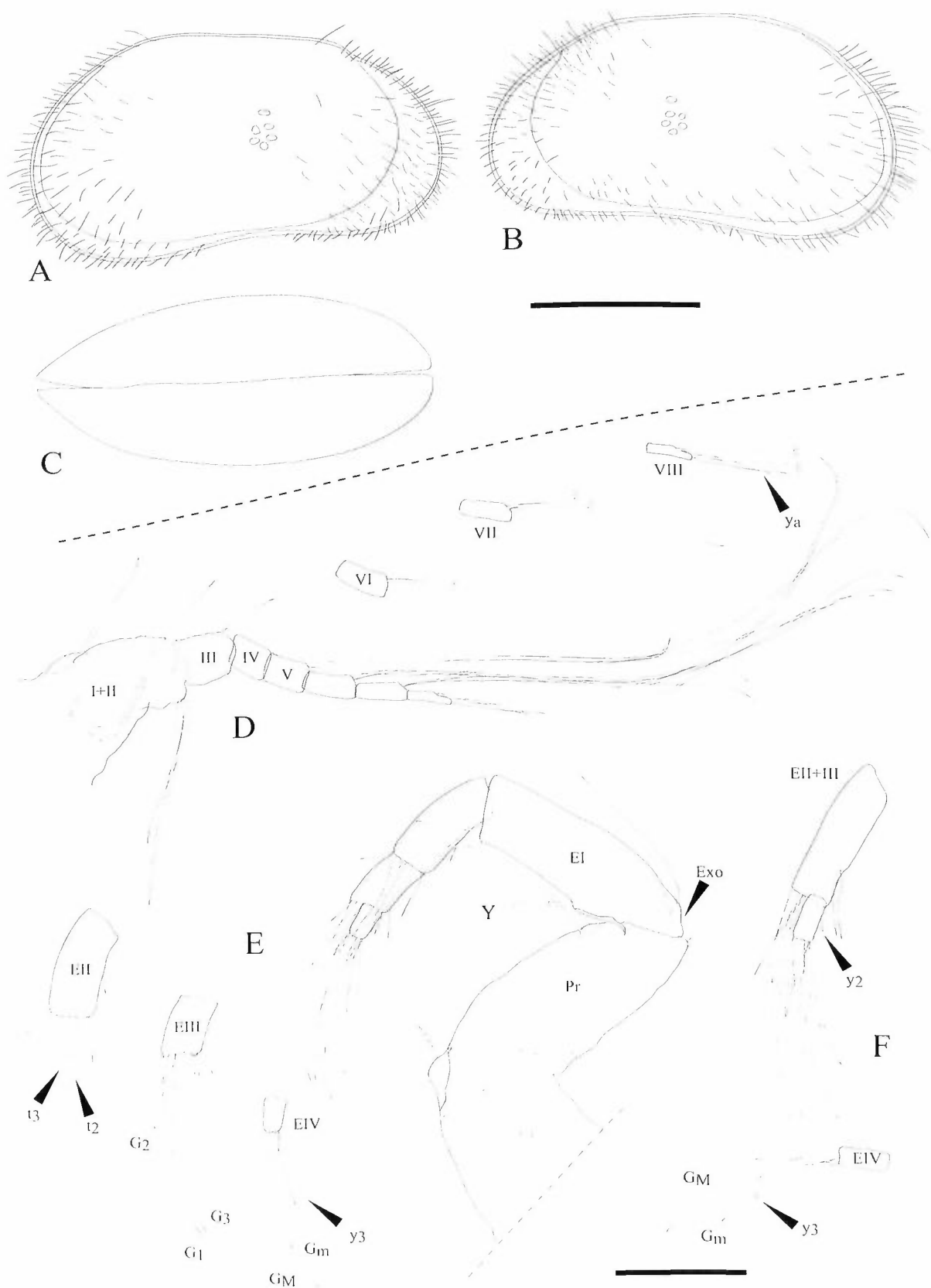


Fig. 8. *Pseudocandona tenuirostris* sp. nov. A–E, Holotype, ZIHU 3916; F, allotype, female, ZIHU 3917. A, Lateral view of right valve; B, lateral view of left valve; C, dorsal view of valves, head to the left; D, antennule (inset, details of endopodites of sixth to eighth podomeres); E, antenna (inset, details of endopodites of second to fourth podomeres); F, two terminal endopodal podomeres of antenna (t2 and t3 form male bristles). Scale bars: A–C, 500 μ m; D–F, 100 μ m.

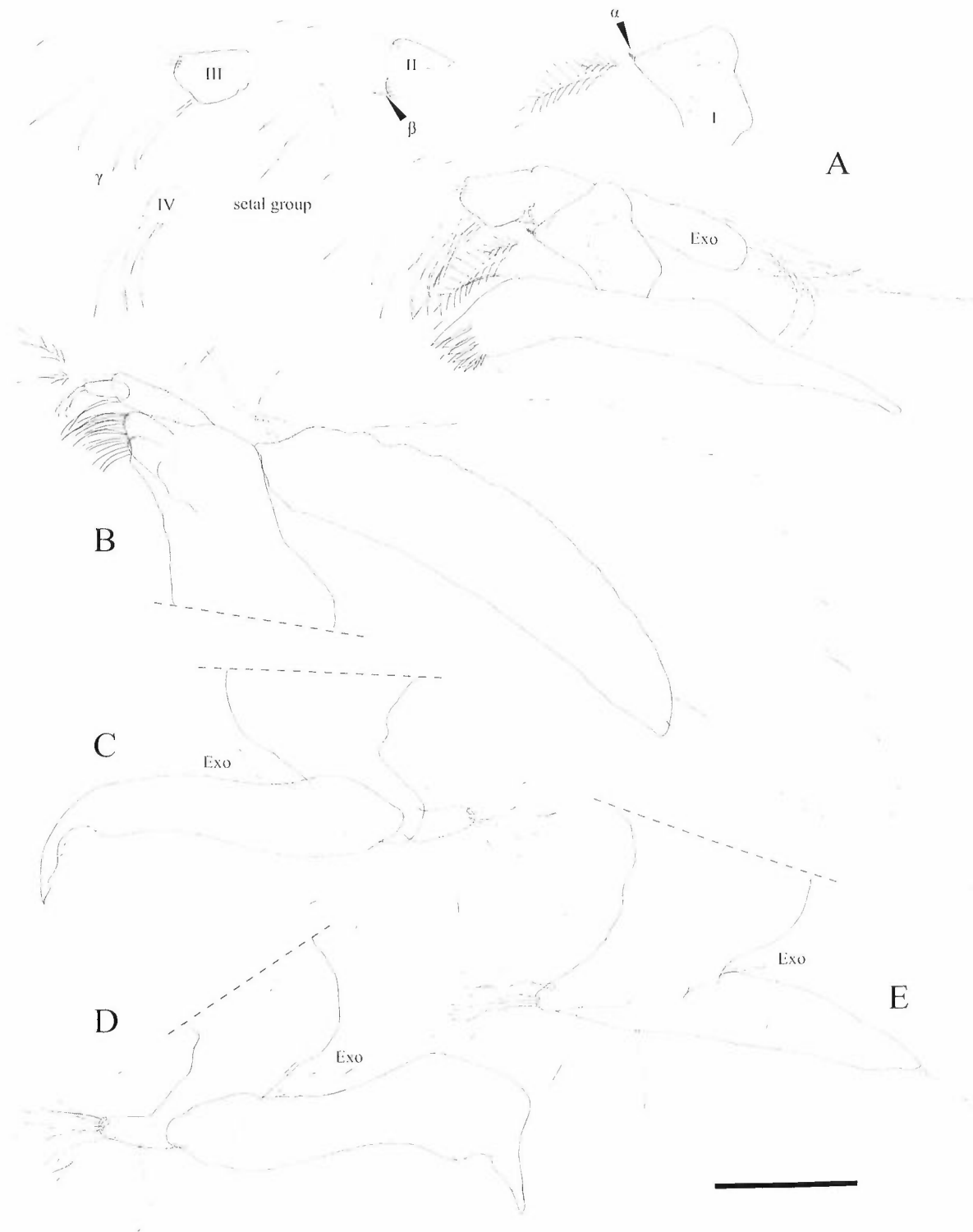


Fig. 9. *Pseudocandona tenuirostris* sp. nov. A–D, Holotype, ZIHU 3916; E, allotype, female, ZIHU 3917. A, Mandible (inset, enlargements of the four palp segments); B, maxillula; C, D, left and right male maxilliped, respectively; E, female maxilliped. Scale bar: 100 μ m.

short G_2 , and three apico-ventral setae. Fifth podomere with G_M and shorter G_m , one apical seta, and aesthetasc y_3 .

Mandible (Fig. 9A) consisting of coxal plate and four-segmented palp. Coxal plate with antero-lateral plumose seta and seven stout teeth, latter interspersed with several setae of various lengths. First podomere of palp with exopodal plate (Exo) and one long and one short inner-distal plumose

setae, one long antero-distal seta, and alpha plumose seta. Second podomere of palp with beta seta and group of three setae. Third podomere of palp with three outer apical setae, one mid-apical gamma seta, and two medium-long and one short inner apical setae. Fourth podomere of palp with one distally plumed claw, two apical setae, and one shorter apical seta.

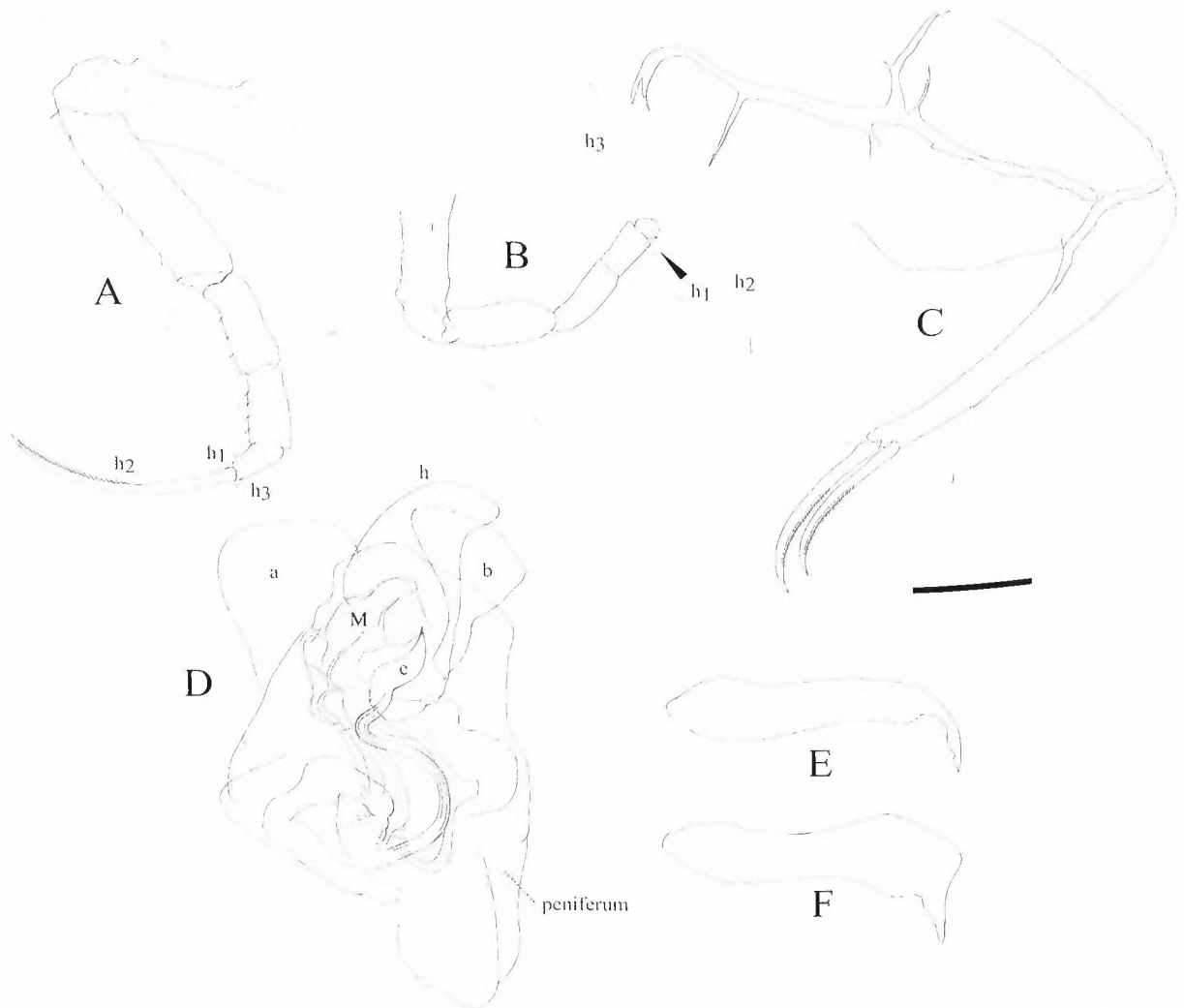


Fig. 10. *Pseudocandona tenuirostris* sp. nov., holotype, ZIHU 3916. A, Walking leg; B, cleaning leg; C, uropod, with uropodal rami; D, left hemipenis; E, left clasper organ; F, right clasper organ. Scale bar: 100 μ m.

Maxillula (Fig. 9B) with elongate vibratory plate, three masticatory processes, and two-segmented palp. First podomere with two long, apical plumose setae.

Maxilliped (Fig. 9C, D) with palp, vibratory plate (Exo), one antero-proximal seta, one antero-apical seta, and one postero-apical seta. Vibratory plate with two filaments. Palp transformed into asymmetrical clasper organs; left clasper organ (Fig. 10E) slender, curved, with two short distal setae; right clasper organ (Fig. 10F) helmet-shaped, with two short distal setae. Masticatory process with numerous setae.

Walking leg (Fig. 10A) five-segmented. Terminal claw (h_2) long, with denticles covering about half the length.

Cleaning leg (Fig. 10B) five-segmented. Penultimate segment subdivided. First podomere with three setae. Fourth podomere with one apical seta. Fifth podomere with two long (h_2 , h_3) and one shorter (h_1) setae.

Uropodal ramus (Fig. 10C) with anterior seta and well-developed posterior seta. Two terminal claws with tiny denticles.

Hemipenis (Fig. 10D) distally with three lobes. Medial lobe (h) longest among three, distally beak-shaped (Fig.

7F). Outer lobe (a) narrow, shortest, usually angling away somewhat obliquely (Figs 7E, 10D). Inner lobe (b) overlapping to medial lobe. Peniferum, inner lobe, and medial lobe, altogether tapering distally. M process (M) well developed, proximally branched for three parts. The brusa complex (c) pointed and forked at proximally.

Zenker's organ (Fig. 11) with 5+2 internal rings of spines.

Description of female. Carapace 1.15–1.20 mm long, 0.63–0.64 mm high ($n=3$; 1.17 mm long, 0.63 mm high in allotype). Carapace slightly shorter and posteriorly lower and narrower than that of male.

Antenna (Fig. 8F) four-segmented. First and second podomeres similar to those of male. Third podomere with long G_1 and G_3 , shorter G_2 , one long and one short apico-dorsal setae, one apico-ventral aesthetasc y_2 , and two mid-ventral setae. Fourth podomere with G_M , shorter G_m , one apical seta, and aesthetasc y_3 .

Palp of Maxilliped (Fig. 9E) simple, non-segmented, with three apical setae.

The other parts of female similar to male.

Remarks. In having three setae in the setal group on

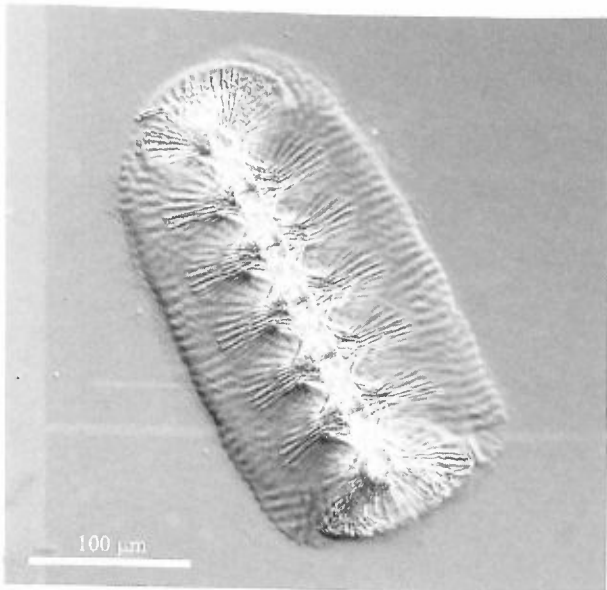


Fig. 11. *Pseudocandona tenuirostris* sp. nov., holotype, ZIHU 3916. Nomarsky optical image of Zenker's organ.

the second segment of the mandibular palp (Fig. 9A), *Pseudocandona tenuirostris* sp. nov. belongs to the *Ps. rostrata* (Brady and Norman, 1889) species group (Meisch 1996). The soft parts of *Ps. tenuirostris* (Fig. 7C) are similar in the morphology to those of *Ps. rostrata*, but the shape of the carapace is similar to that of *Ps. sarsi* (Hartwig, 1899) (Hartwig 1899; Meisch 2000). There are clear differences in the clasping organs and the hemipenis between *Ps. tenuirostris* and the latter two species. In *Ps. tenuirostris*, both the left and right male clasping organs are relatively long and slender (Fig. 10E, F); the fingers of both organs are shorter than in *Ps. rostrata* and more slender than in *Ps. sarsi*. In all three species, the hemipenis comprises three distal lobes, but in *Ps. tenuirostris* the medial lobe (h) is longest, very narrow, and beak-shaped; the outer lobe (a) is shortest. In contrast, in *Ps. rostrata*, the medial lobe (h) is stout and longest, whereas the inner lobe (b) is shortest; in *Ps. sarsi*, the outer lobe (a) is well developed and longer than the stout medial lobe (h).

Etymology. The new specific name is an adjective, derived from the combination of the Latin *tenuis* meaning "thin" or "fine" and *rostris* meaning "of beak", referring to the fine, beak-like medial lobe on the distal end of each hemipenis.

Subfamily **Cyclocypridinae** Kaufmann, 1900

Physocypria nipponica Okubo, 1990

(Figs 12–16)

Physocypria nipponica Okubo, 1990: 3, figs 1E–H, 2B, E, F;
Smith and Janz 2008: 2916, figs 26H–J, 32A–C.

Material examined. ZIHU 3909, 1 male, site E (Table 1, Fig. 1), 6 August 2005, dissected, soft parts mounted on 19 slides; ZIHU 3910, 1 female, same site, 6 August 2005, dissected, soft parts mounted on 18 slides; ZIHU 3911, 1 male,

same site, 19 June 2007; mounted on stubs for SEM observation.

Description of male. Carapace (Figs 12A, B, 13A, B) 0.57 mm long, 0.37 mm high (ZIHU 3909), 0.56 mm long, 0.39 mm high (ZIHU 3911). Dorsal margin of carapace more or less strongly arched. In dorsal view, left valve overlaps right valve at both ends. In frontal view, carapace distinctly asymmetrical; right valve overlaps left valve with conspicuous dorsal hump. Antero- and postero-ventral margins of right valve each with row of pustules; pustules situated on list formed by outer lamella of valve, rather than on selvage (Fig. 12C, D).

Antennule (Fig. 13C) seven-segmented. First two podomeres fused, with two long apico-ventral setae and one mid-dorsal seta. Third podomere with one apico-dorsal seta and externo-ventral Rome organ. Fourth podomere quadrate, with one apico-ventral seta and one apico-dorsal seta. Fifth podomere quadrate, with two apico-ventral setae and one very long and one long apico-dorsal setae. Sixth podomere quadrate, with two apico-ventral setae and two very long apico-dorsal setae. Seventh podomere quadrate, with two very long apico-ventral setae and two very long and one medium-long apico-dorsal setae. Eighth podomere elongate, with one claw, two long and one short apical setae, and aesthetasc y_4 .

Antenna (Fig. 13D) five-segmented. Penultimate segment subdivided. First podomere with two proximal setae (not depicted in Fig. 13D) and one long apico-ventral seta. Second podomere with one apico-ventral plumose seta and mid-ventral aesthetasc Y. Exopodite (Exo) situated on proximal part of second podomere; consisting one long and two short setae. Third podomere with one long and one shorter apico-ventral setae, two apico-dorsal setae, and two apical male bristles (t2 and t3); natatory setae (5+1) very long, extending beyond tips of terminal claws by ca 70% of their total length. Fourth podomere with long G_1 and G_3 , shorter G_2 , long apical seta, and mid-ventral seta. Fifth podomere with slender G_{M1} , shorter G_M , mid-ventral setae, and aesthetasc y_3 .

Mandible (Fig. 14A) consisting of coxal plate and 4-segmented palp. Coxal plate with seven teeth and antero-lateral seta. First podomere of palp with external vibratory plate (Exo), one inner seta and two plumose setae, and simple alpha seta. Second podomere of palp with two outer apical setae. Setal group on second podomere with three long plumose setae, one inner seta, and one plumose beta seta. Third podomere of palp with three outer apical setae, three mid-apical setae, and smooth externo-distal gamma seta. Fourth podomere of palp apically with two setae, one short seta, and one plume-tipped claw.

Maxillula (Fig. 14B) with elongate vibratory plate, three masticatory processes, and two-segmented palp.

Maxilliped (Fig. 14D, E) with asymmetrical clasping organ, vibratory plate (Exo), one long-plumed and two short antero-proximal setae, one plumose antero-apical seta, and one short postero-apical seta. Finger of right clasping organ (Fig. 15D) strongly developed, apically with one long process and one short, tooth-shaped process. Left finger (Fig.

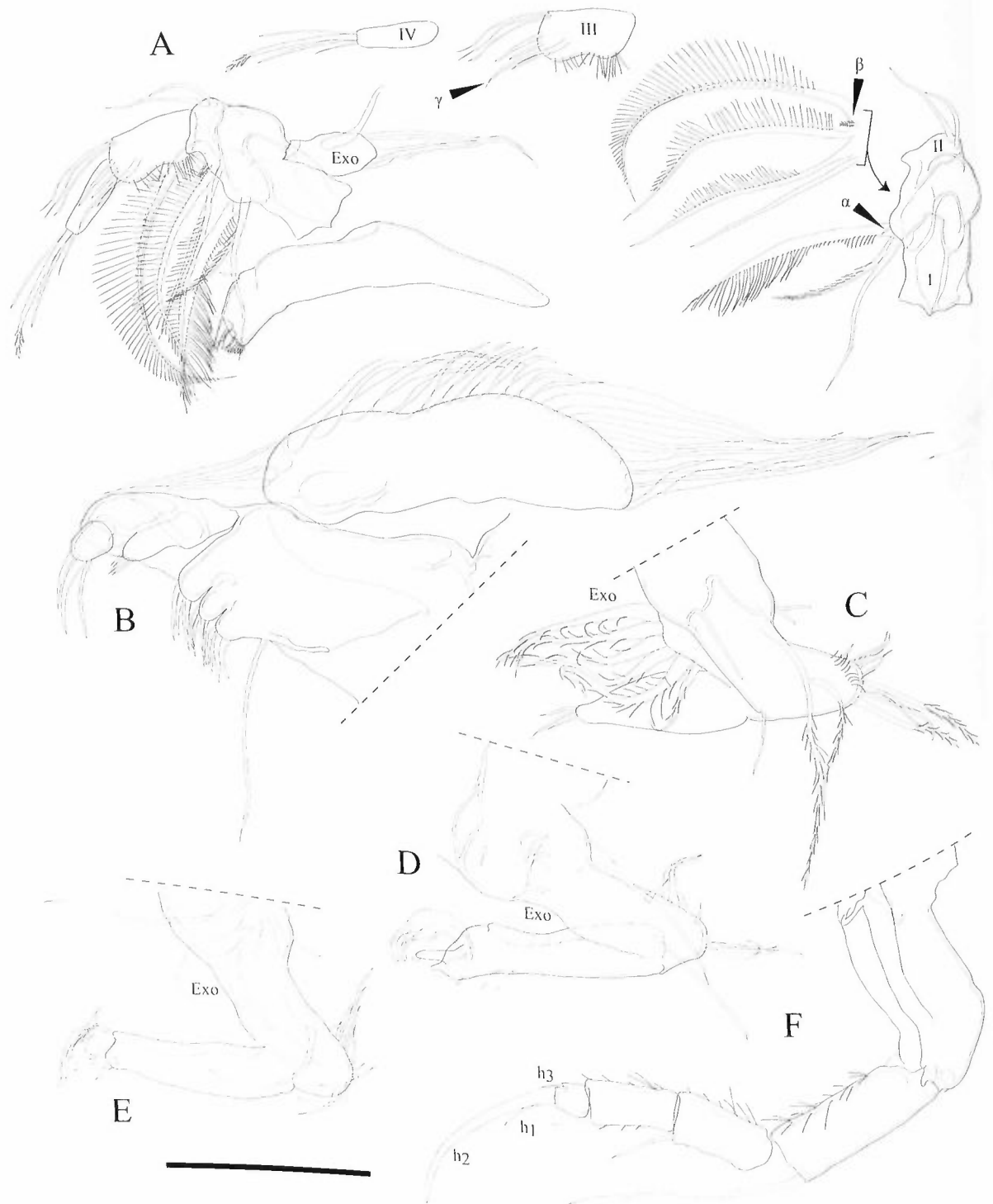


Fig. 14. *Physocypris nipponica* Okubo, 1990. A, B, D–F, Male, ZIHU 3909; C, female, ZIHU 3910. A, Mandible, with enlargements of the four palp segments; B, maxillula, with vibratory plate; C, maxilliped; D, E, left and right maxilliped, respectively; F, walking leg. Scale bar: 100 μ m.

synonymy of these species. The description of *Ph. kraepelini* which distributed around Palearctic region by Meisch (2000) indicates that the structure of the hemipenis is different between the two taxa. In *Ph. kraepelini*, the peniferum is quite angular and the outer edge is stout proximally. However, there is no information on variation in the outer edge of hemipenis in either of these two taxa. For the time being, we regard *Ph. nipponica* is a valid species.

Family **Limnocytheridae** Klie, 1938
Subfamily **Timiriaseviinae** Mandelstam, 1960
Metacypris digitiformis Smith and Hiruta, 2004

(Fig. 17)

Metacypris digitiformis Smith and Hiruta, 2004: 38, figs 2, 3, 4A, B, 5; Smith and Janz 2009: 230, fig. 5I, J.

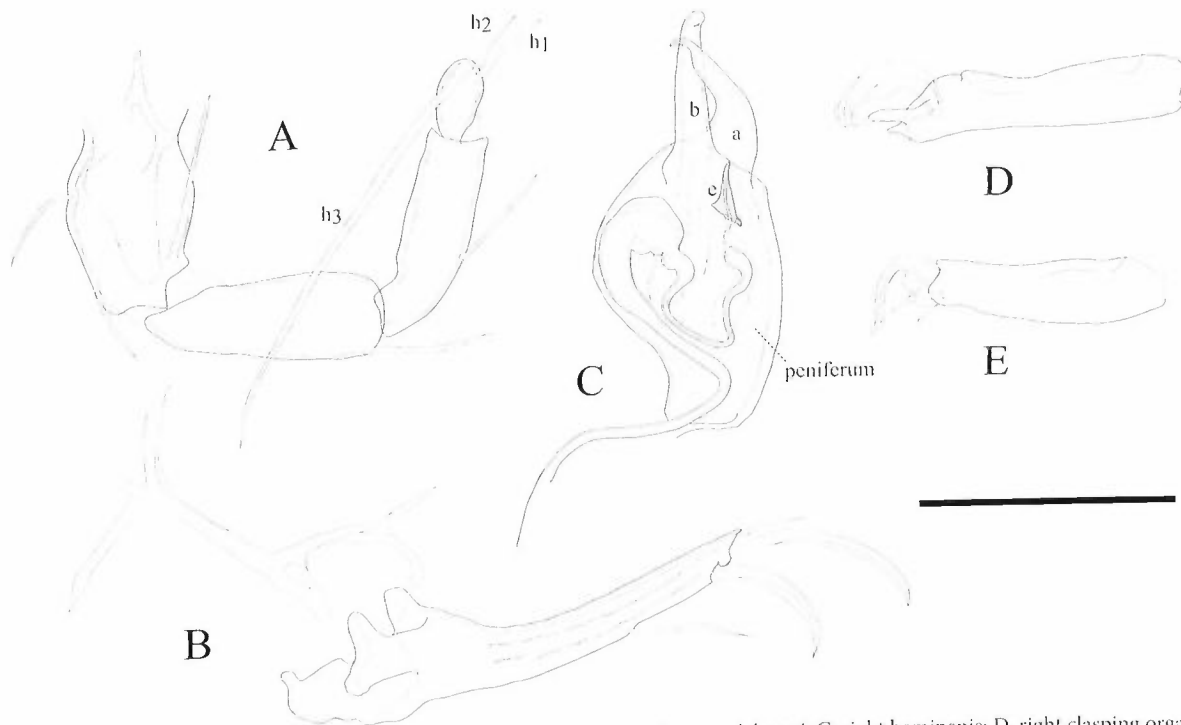


Fig. 15. *Physocypria nipponica* Okubo, 1990. A, Cleaning leg; B, uropod with uropodal rami; C, right hemipenis; D, right clasper; E, left clasper. Scale bar: 100 μ m.

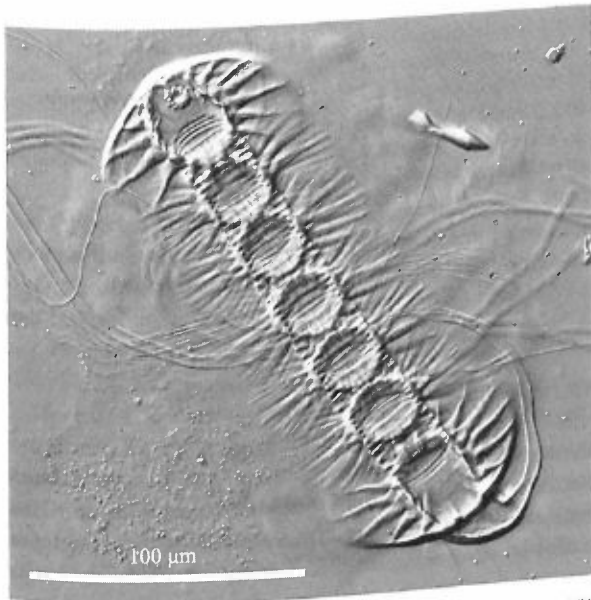


Fig. 16. *Physocypria nipponica* Okubo, 1990, male, ZIHU 3909. Nomarski optical image of Zenker's organ.

Material examined. ZIHU 3903, 1 male, site E (Table 1, Fig. 1), 6 August 2005, dissected, soft parts mounted on 17 slides; ZIHU 3904, 1 male, site B (Table 1, Fig. 1), 13 May 2006, dissected, soft parts mounted on 16 slides; ZIHU 2006, dissected, soft parts mounted on 16 slides; ZIHU 3905, 1 female, site E, 6 August 2005, dissected, soft parts mounted on 16 slides; ZIHU 3906 (1 male), ZIHU 3907 (1 female), ZIHU 3908 (1 female), site E, 17 July 2007, mounted on stubs for SEM observation.

Comparative material. Holotype, ZIHU 2063, male. Paratype, ZIHU 2064–2067, 4 females; ZIHU 2071–2072, 2

males.

Remarks. There are a few minor morphological differences between the holotype from Kushiro Marsh and our specimens from Sarobetsu Marsh. In the Kushiro Marsh specimens, the medial ridge of the hemipenis attaches to about the middle of the dorsal ridge or at the end of the zygom, and the entire outer margin of the hemipenis is gradually curved (Fig. 17B). In the Sarobetsu Marsh specimens, the medial ridge attaches to the end of the dorsal ridge, and the hemipenis has the outer margin gradually curved over the proximal two-fifths, but nearly straight distally (Fig. 17A). The shape of the upper clasper ramus (UR) also shows subtle variation. This backward-directed ramus is usually straight in the Sarobetsu population (Fig. 17A), but in a few specimens it is identical in form to that in the Kushiro population (Fig. 17B). The carapace and soft parts are identical between the two populations, and we attribute the above morphological differences to geographical variation in a single species.

Ecology

Figure 18A shows the combined seasonal occurrence of the four ostracod species treated in this study; the total number of specimens collected each month was far from constant across years. We observed *Cryptocandona* sp. at site C (Fig. 2B, C) from early autumn to winter (Fig. 18B), and at site E, where very small individuals up to the third instar stage appeared from May to July. Population density began to increase in late September, and adults appeared in October. We found no males. This hypogean species lacks natato-

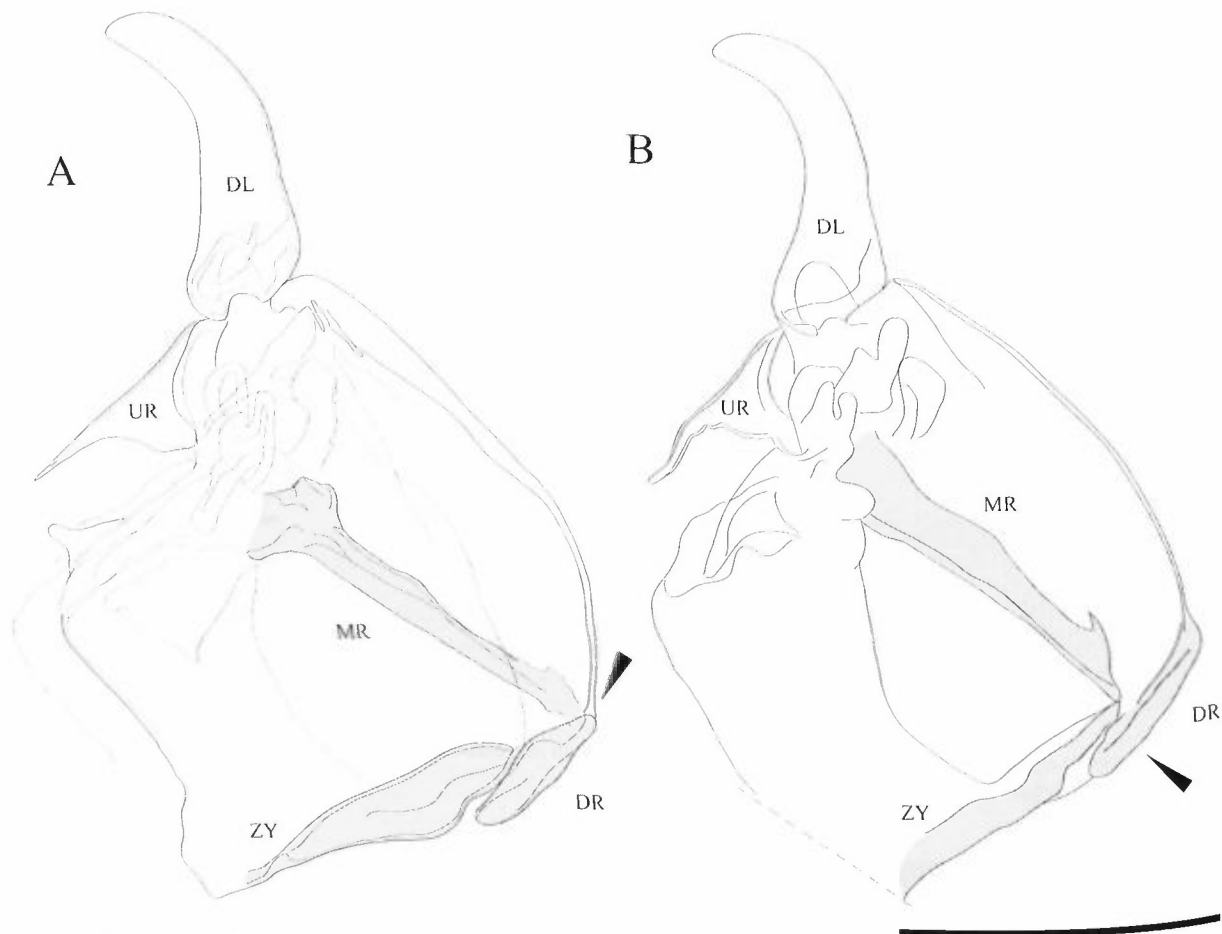


Fig. 17. Comparison of the hemipenis of *Metacypris digitiformis* Smith and Hiruta, 2004 between specimens from the the Sarobetsu and Kushiro populations. A, Right hemipenis typical of the Sarobetsu population, male, ZIHU 3903; B, hemipenis typical of the Kushiro population, redrawn from Smith and Hiruta (2004); arrowheads indicate point of attachment to MR. Scale bar: 100 μ m.

ry setae and cannot swim; it usually occurs in groundwater.

We collected individuals of *Pseudocandona tenuirostris* sp. nov. in all sampling periods and at every site except site D (Fig. 2A); adults and juveniles occurred simultaneously in all month with the maximum number of individuals in May (Fig. 18C). This temporary broad occurrence of both juveniles and adults suggests that this species survives at least over one winter. The Antenna of this species completely lack natatory setae, which are important for swimming, and we observed individuals crawling in muddy sediments.

We collected *Physocypria nipponica* only at site E and only in June and August; we obtained very few individuals, all of which were adults, including males. Site E was the shallowest and most heavily vegetated site (Fig. 2D). *Physocypria nipponica* has well-developed natatory setae and is a very good swimmer.

We collected *Metacypris digitiformis* at sites A, B (Fig. 2A), and E (Fig. 2D); it appeared in May after the snow had melted, rapidly decreased in number in August, and finally disappeared in September (Fig. 18D). Most of adult males appeared early in July, a little earlier than adult females. We observed adult females brooding eggs in the carapace in July and numerous young juveniles toward the end of this month. The spinneret seta on the exopodite of each antenna secretes a viscous liquid used for adherence to the substrate

and for locomotion. This species cannot swim and prefers a heavily vegetated environment.

Discussion

Based on current information, the freshwater ostracod fauna of Hokkaido appears to be similar in composition to that of Europe, rather than to that of Honshu, Japan. Other than a few minor groups, freshwater ostracods are represented by two major families, Candonidae and Cyprididae. Compared with ostracod faunas in other regions of Japan, that of Hokkaido is relatively rich in candonids, with only a few cypridids (Hiruta and Smith 2001); for instance, from southern part of Honshu, Okubo (1990) reported one species in Candonidae, versus 13 species in Cyprididae. Three of the four species we collected from Sarobetsu Marsh belonged to Candonidae, but none to Cyprididae. A similar tendency in the composition of freshwater ostracods has been reported with respect to the European fauna (Martens *et al.* 1998). The occurrence of *Metacypris* adds further faunal similarity between Europe and Hokkaido. The genus belongs to Limnocytheridae, which is the only family in the superfamily Cytheroidea that exclusively inhabits freshwater environments. Over 130 fossil species in the genus have

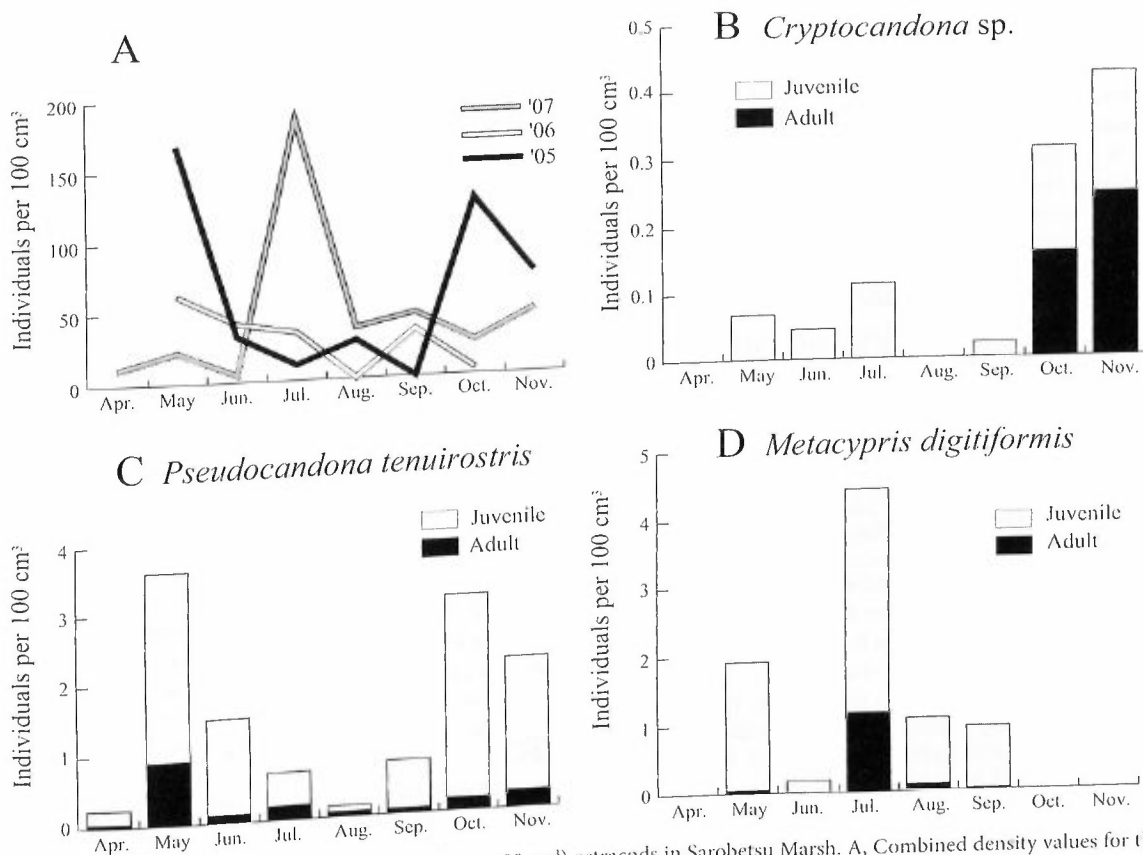


Fig. 18. Seasonal occurrence and density (individuals per 100 cm³) ostracods in Sarobetsu Marsh. A, Combined density values for the four species detected (*Cryptocandona* sp.; *Pseudocandona tenuirostris* sp. nov.; *Physocypria nipponica* Okubo, 1990; *Metacypris digitiformis* Smith and Hiruta, 2004), with values for different years shaded differently; B–D, densities of adults and juveniles (individuals per 100 cm³), by species; B, *Cryptocandona* sp.; C, *Ps. tenuirostris* sp. nov.; D, *M. digitiformis* (this species was collected at sites A, B, and E). The monthly densities are the combined values from all five sampling sites, 2005–2007.

been reported from Cretaceous to Recent (Kempf 1980), whereas their abundance and diversity had been decreased and only three extant species are known, each in Europe (*M. cordata* Brady and Robertson, 1870), Thailand (*M. srisumomae* (Savatenalinton, Borgonie, and Martens, 2008)), and Hokkaido (*M. digitiformis*) (Karanovic 2012).

The ostracod diversity detected in Sarobetsu Marsh was lower than that in the other large wetland in Hokkaido, Kushiro Marsh. Sarobetsu Marsh is about one fourth the area of Kushiro Marsh, and the difference in species richness may be due to the difference in size. Aridification in the core region of Sarobetsu Marsh, where recently there have been considerable human disturbances, may also be a factor in the lower richness. Soil amelioration for grazing has taken place around the marshland; as a result, the water level in the core region of the marsh has rapidly decreased and the vegetation has been changing (Fujimura *et al.* 2012). We observed that the dwarf bamboo, *Sasa veitchii* (Carrière) Rehd., is intruding into the core region, which indicates aridification. These changes have caused a reduction in shallow vegetative environments. We found only a few valves of ostracods at site D, an open-water habitat, which may indicate the importance of shallow vegetative environments for tiny aquatic animals, including ostracods.

The seasonal occurrence and abundance of ostracods in Sarobetsu Marsh may depend on the amount of precipita-

tion in the form of snow in winter. When deposited snow melts in the spring, the water level in the marsh is higher than at any other time of the year. After all the snow melts, the water level gradually decreases from spring to autumn (Saito *et al.* 2008). The ostracod fauna observed at Sarobetsu Marsh was not consistent from year to year. For example, we observed *M. digitiformis* and *Pseudocandona tenuirostris* at sites A and B in 2005, following a winter with high precipitation (Japan Meteorological Agency 2005). During the following two years, however, we found no ostracods at site A or B. Due to low precipitation in the winters of 2006 and 2007, the marshland water level was relatively low both years (Japan Meteorological Agency 2006, 2007). We assume that the decreased water level caused the environments at sites A and B to become more like that at site D.

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