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A new species of *Neocervinia* (Copepoda: Harpacticoida: Cerviniidae) from the hyperbenthos of the Hatsushima cold-seep site in Sagami Bay, Japan

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Abstract

A new species of harpacticoid copepod, *Neocervinia itoi* (Cerviniidae), is described on the basis of females and copepodids collected from the Hatsushima cold-seep site in Sagami Bay, Japan. It is morphologically very close to its deep-sea congeners *N. tenuicauda* (Brotskaya, 1963) and *N. unisetosa* (Montagna, 1981). The new species differs primarily in the segmentation of the antennule and the endopod of both mandible and maxilliped, and in form and shape of the setae on leg 5. The presence of sensillar structures on the proximal part of the antennule is discussed. A key to the species of *Neocervinia* is presented.

Introduction

Since their initial discovery in 1977, hydrothermal vents have been explored at a quickening pace and diverse vent communties have now been documented from virtually all investigated areas of tectonic activity in the deep Pacific and Atlantic (Tunnicliffe, 1991). Cold seeps are similar aphotic environments under high hydrostatic pressure, however, differ from hydrothermal vents where the seeping medium is warmed by the close proximity of molten rock. At cold seep subduction zones, the oceanic crust lies well below the sediment as it is subducted below overlying continental crust along tectonically active continental margins. Consequently, seepage from accreted sediment wedges is more diffuse, lower in temperature and typically rich in dissolved methane.

Dense benthic communities dominated by colonies of the giant vesicomyid clam *Calyptogena soyoae* Okutani associated with a lithodid stone crab *Paralomis* were observed at the Hatsushima cold seep site discovered in the mid 1980s by the Japanese submersible *Shinkai 2000* at about 0.8–1.1 km depth in Sagami Bay off Tokyo. Subsequent investigation of the site by the Japan Marine Science and Technology Center (JAMSTEC) during a series of dives with the deep-sea submersible *Shinkai 2000* has resulted in the first data on the benthopelagic (hyperbenthic) zooplankton (Toda et al., 1994). By using specially designed opening-closing nets (Kikuchi et al., 1990) it was found that copepods represented the most dominant taxon accounting for over 90% of the total fauna (Toda et al., 1994).

In contrast to hydrothermal vents which harbour a diverse endemic copepod fauna (Humes (1987) and subsequent papers) very few copepods have been described from cold seeps. Humes (1988) discovered the siphonostomatoid *Bythocheres prominulus* Humes at a depth of 3260 m near the base of the West Florida Escarpment, a gigantic limestone cliff arising 2000 m above the seafloor. From the same site in the eastern Gulf of Mexico Humes (1989) also described the erebonasterid *Amphicrossus spinulosus* (Humes) (Poecilostomatoida). Finally, Toda et al. (1992) described the primitive clausidiid *Hyphalion sagamiense* Toda, Miura & Nemoto (Poecilostomatoida) from the mantle cavity of *Calyptogena soyoae* collected at the Hatsushima site. The hosts of *A. spinulosus* and *B. prominulus* are unknown.

Harpacticoid copepods were numerically dominant in the hyperbenthos at the Hatsushima cold-seep site in Sagami Bay (Toda et al., 1994). Among the more common species a new cerviniid was found, belonging to the recently erected genus *Neocervinia* Huys, Møbjerg & Kristensen (Huys et al., 1997). A description of the new cerviniid is provided below and its relationships within the genus discussed.

Materials and methods

Copepods were collected during dive 452 of the deep-sea submersible *Shinkai 2000* at the Hatsushima cold seep site (35°00′00″ N, 139°13′00″ E) at a depth of 1160 m in Sagami Bay, Japan, on 23 October 1989. Specimens were fixed in 10% formalin and subsequently preserved in 70% ethanol. Body length measurements were made in lactic acid. The holotype female and three paratypes were dissected in lactic acid and mounted on slides in polyvinyl lactophenol mounting medium. All drawings have been prepared using a camera lucida on a Zeiss Axioskop or an Olympus BH-2 differential contrast interference microscope. The terminology for body and appendages follows Huys & Boxshall (1991). Abbreviations used in the text are: P1–P6, first to sixth thoracopod; exp(enp)-1(2,3) to denote the proximal (middle, distal) segment of the exopod (endopod); ae, aesthetasc.

Taxonomic account

Family Cerviniidae Sars, 1903
Subfamily Cerviniinae Brotskaya, 1963
Genus *Neocervinia* Huys, Møbjerg & Kristensen, 1997
Neocervinia itoi, new species
Material examined. – Holotype ♀ (dissected on 1 slide; USNM no. 266459) and 4 paratypes (4 ♀♀ in alcohol; USNM no. 266460) are deposited in the National Museum of Natural History, Smithsonian Institution, Washington, D.C., U.S.A. Additional paratypes are dissected on slides: 1 ♀ deposited in The Natural History Museum, London (NHM reg. no. 1997.792) and 4 ♀♀ retained in the personal collection of the senior author. Eleven copepodites were also retained in the personal collection of the senior author.

Description

Dorsal habitus and P1–P4 are illustrated from the holotype. All other drawings are based on dissected paratypes.

Female. Total body length of holotype 1.35 mm (Figure 1A) measured from tip of rostum to posterior margin of caudal rami. Maximum width 0.35 mm measured at posterior margin of P2-bearing somite. Prosome wider than urosome. Rostrum fused to cephalic shield; large and wide with pointed tip; shape triangular; with 2 pairs of sensillae dorsally and mid-dorsal pore near apex. P1-bearing somite clearly separated from cephalosome. Pleurotergites of somites bearing P2–P4 with dorsolateral rows of minute spinules anteriorly. Surface ornamentation consisting of densely arranged minute spinules present dorsally and laterally on genital double-somite and remaining urosomites, and ventrally on posterior part of anal somite (Figure 2D). Genital double-somite slightly longer than following three abdominal somites combined; suture marking original segmentation dorsally and laterally (Figure 2A–C). Posterior margin of genital double-somite, 2nd and 3rd abdominal somites with continuous spinular row. Anal somite with large spinules at posterolateral corners; anal operculum semicircular, with minute spinules.

Caudal rami divergent (Figure 2D), closely set proximally; cylindrical; longer than last 2 abdominal somites combined, about 7 times as long as average width; with 2 longitudinal rows of coarse spinules on dorsal face; densely covered with minute spinules on entire dorsal and ventral surface; with 3 tiny sensillae dorsally (near proximal end, 1/2 of ramus length, and near distal end, respectively); with 7 setae: setae I and II long, inserting at 1/3 and 2/3 of ramus length measured from proximal margin, respectively; setae III–VI arranged around distal margin, setae IV and V longest and pinnate; seta VII tri-articulate at base, located near inner subdistal corner.

Genital field (Figure 3C) located in anterior half of genital double-somite; gonopores paired slits, each covered by operculum derived from vestigial P6; P6 produced into elongate, narrow process with 1 rudimentary and 2 long setae. Copulatory pore large, leading via short chitinized copulatory duct to paired seminal receptacles; located slightly posterior to gonopores. Integument around genital field distinctly wrinkled. Ventral surface anterior to gonopores forming large protrusion (Figure 2A).

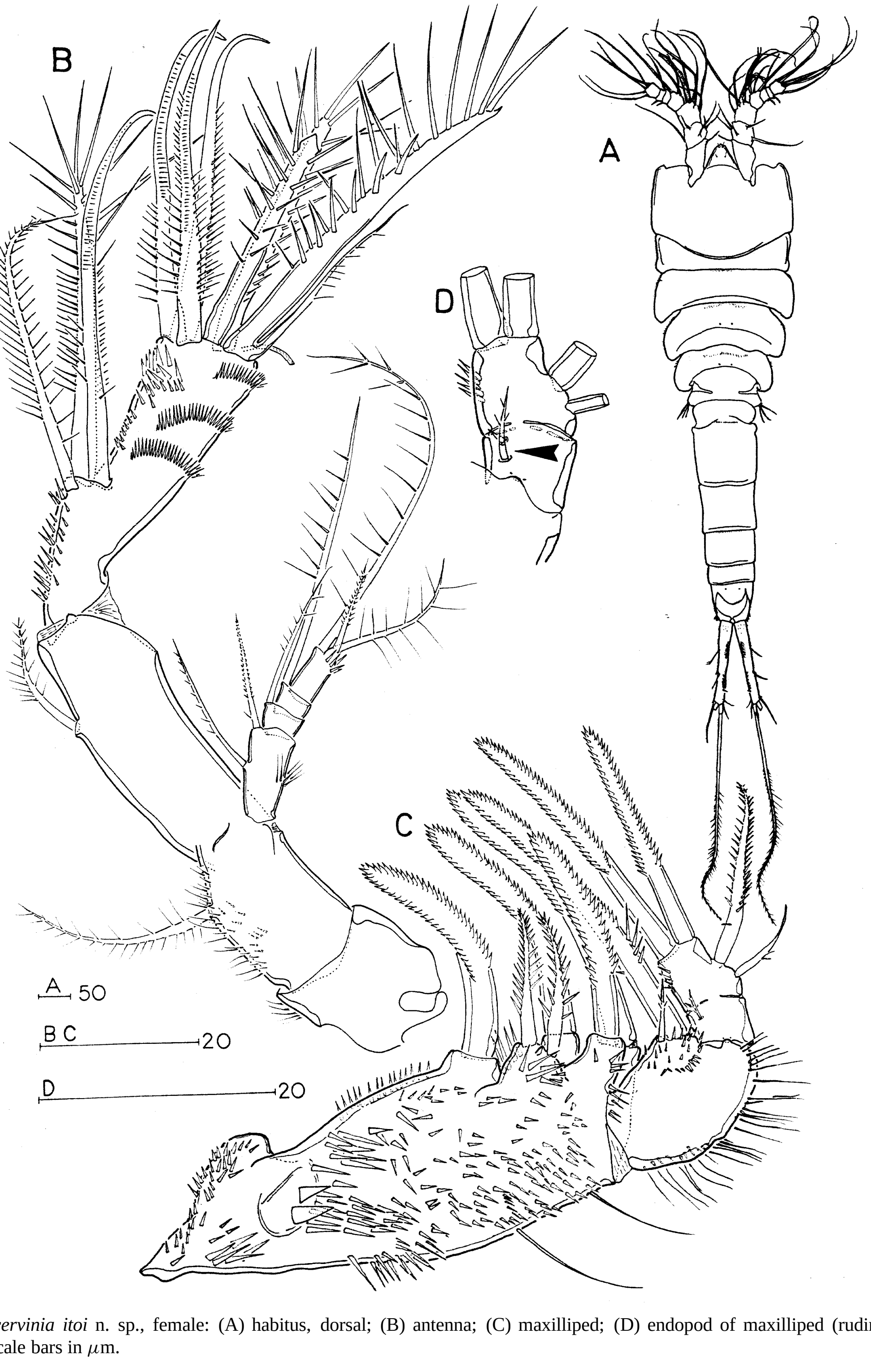

Figure 1. Neocervinia itoi n. sp., female: (A) habitus, dorsal; (B) antenna; (C) maxilliped; (D) endopod of maxilliped (rudimentary seta arrowed). All scale bars in μm.

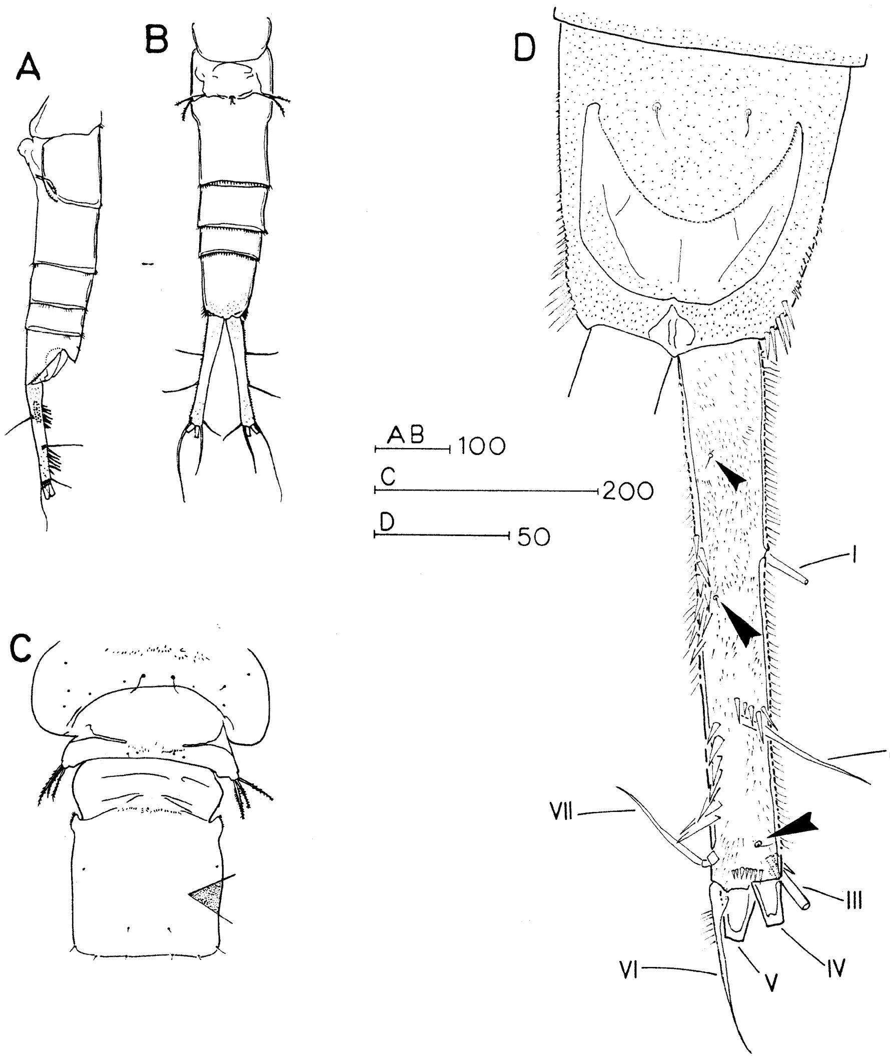

Figure 2. Neocervinia itoi n. sp., female: (A) abdomen, lateral; (B) abdomen, ventral; (C) P5-bearing somite and genital double-somite, dorsal; (D) anal somite and right caudal ramus, dorsal (sensillae arrowed). All scale bars in μm.

Antennules (Figure 3A) short, 6-segmented; with small outer sclerite and membranous area around basis of segment 1. Segment 1 with fine setules along anterior margin. Segment 2 longest; derived by fusion of segments 2 and 3 expressed in other Cerviniinae; original segmentation marked by incomplete, transverse, chitinous rib near anterior margin; with spinules along posterior margin and on ventral surface. Armature formula: 1-[1], 2-[16 + (1 + ae)], 3-[3], 4-[2], 5-[2], 6-[8]. Tiny sensilla present near posterior margin of segments 1 and 2 (arrowed in Figure 3A).

Antenna (Figure 1B). Coxa armed with minute setules. Basis and proximal endopod segment completely fused forming elongate allobasis; small surface suture present on outer surface; about 4 times as long as median width; abexopodal margin with 2 very widely separated pinnate setae and setules in basal part. Exopod located at 1/3 of allobasal length; 4-segmented; armature formula [2, 1, 1, 1 lateral + 2 apical]; exp-1 longest, as long as exp-2-4 combined, with fine spinules along outer margin; exp-2 and -3 short and subequal in length; exp-4 as long as exp-2 and -3 combined, with group of minute spinules at outer subapical corner. Free endopod shorter than allobasis, widening distally; with 3 transverse surface frills in distal half and 3 spinular patches around

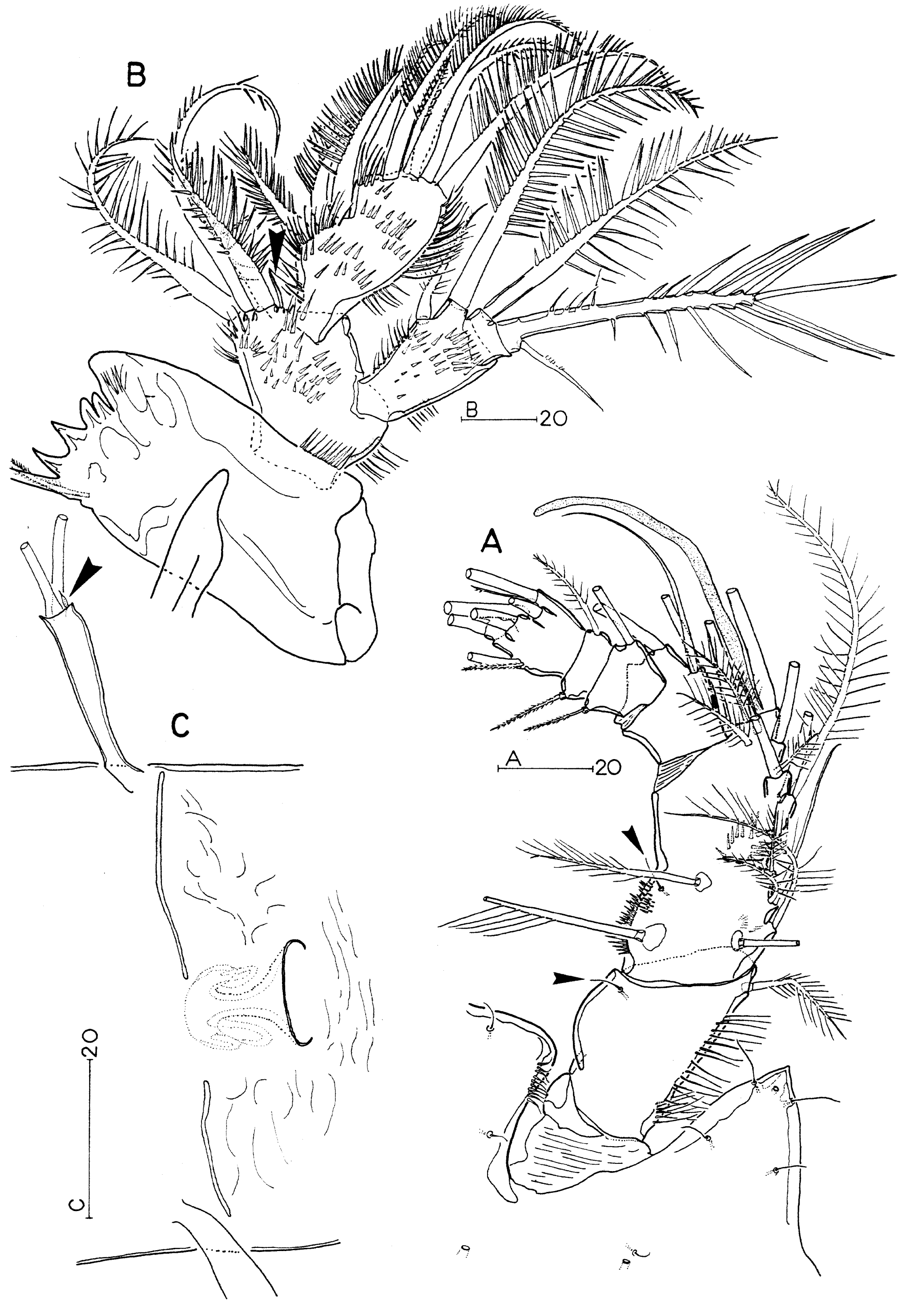

Figure 3. Neocervinia itoi n. sp., female: (A) antennule and rostrum, dorsal (sensillae arrowed); (B) mandible (basal vestigial seta arrowed); (C) genital field (P6 rudimentary seta arrowed). All scale bars in μm.

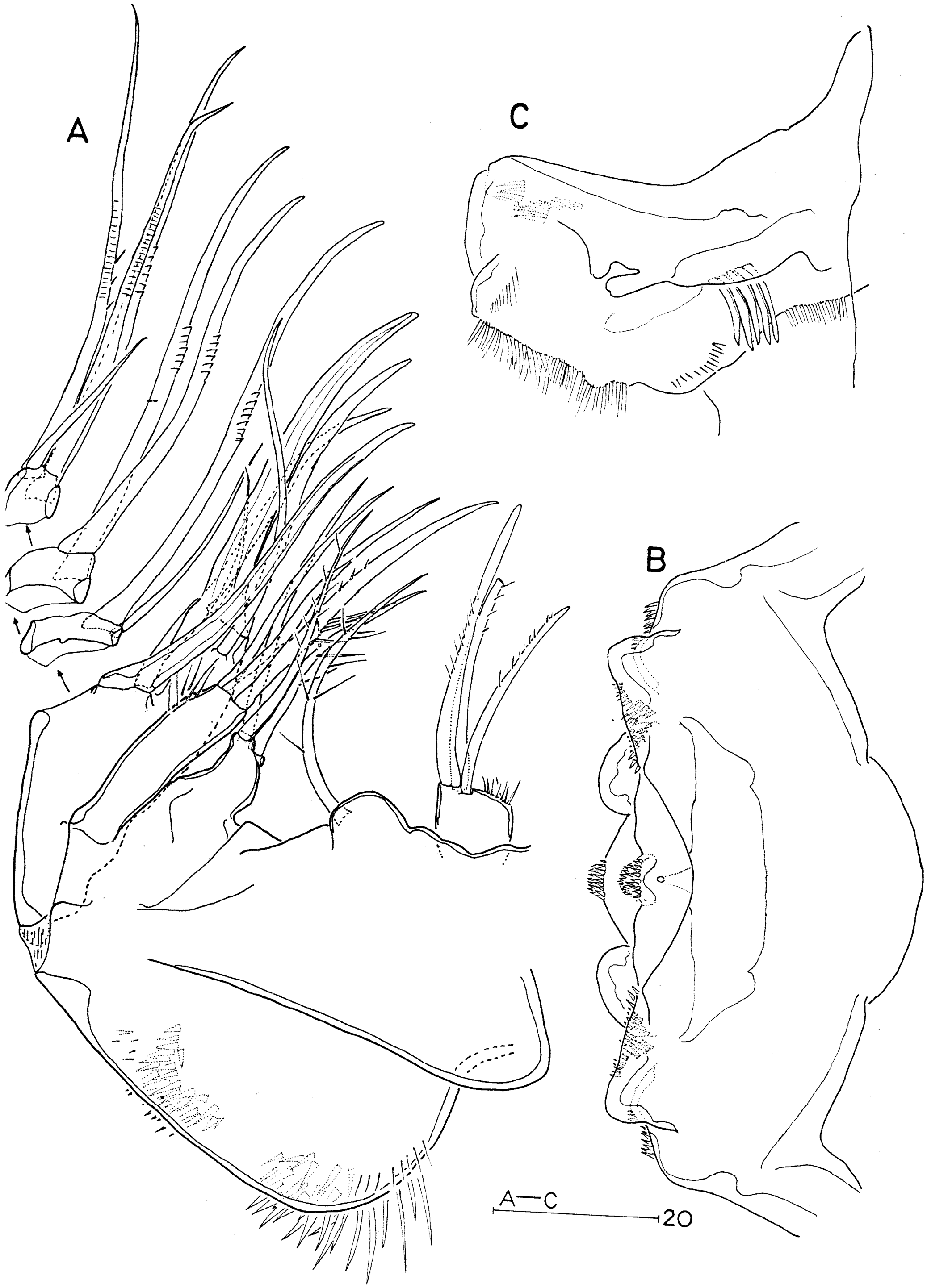

Figure 4. *Neocervinia itoi* n. sp., female: (A) maxilla (with disarticulated endopod); (B) labrum, anterior; (C) right paragnath, anterior. Scale bar in μm.

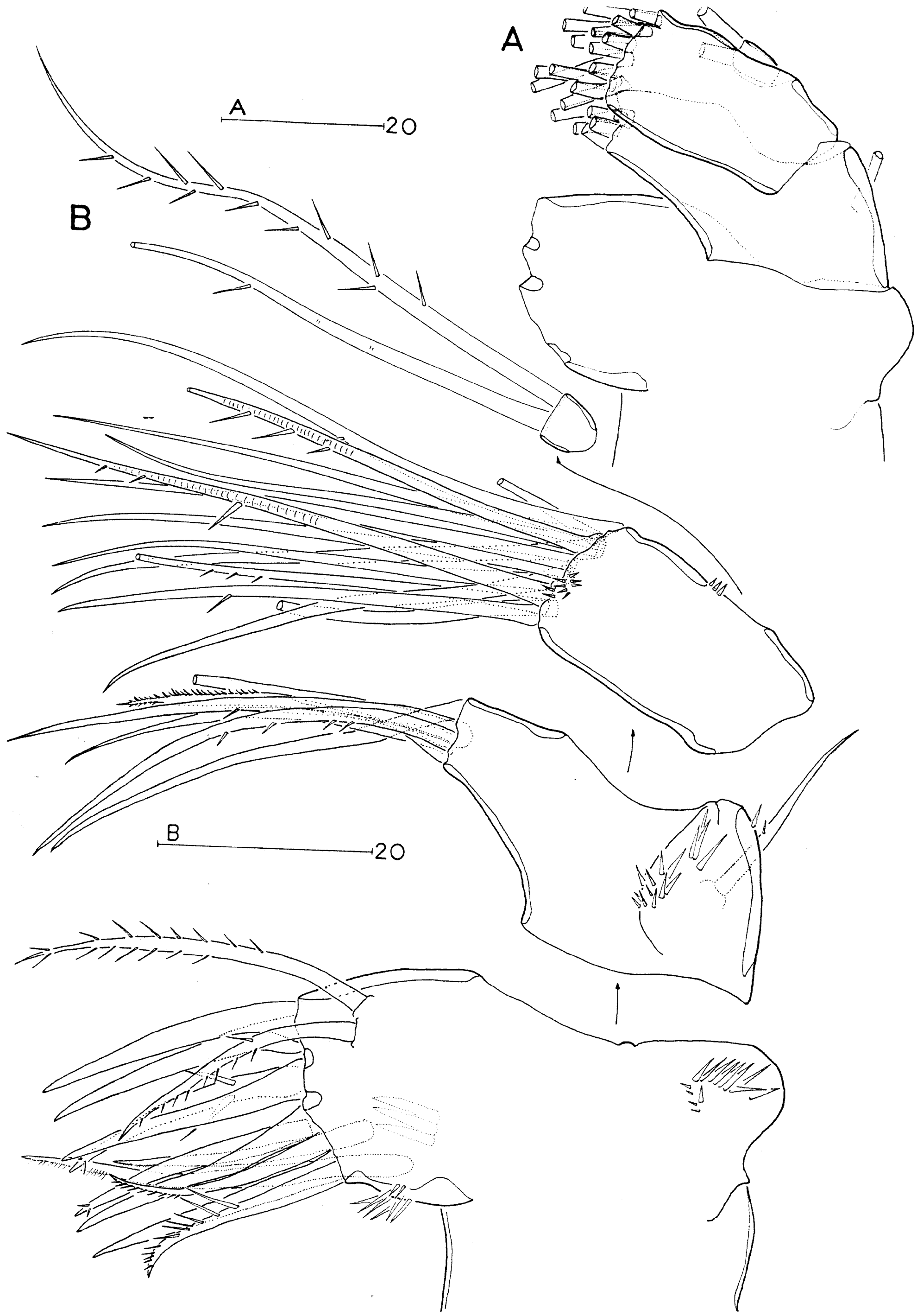

Figure 5. *Neocervinia itoi* n. sp., female: (A) Contours of maxillule, armature largely omitted; (B) maxillule, disarticulated. All scale bars in μm.

abexopodal margin; lateral armature consisting of 2 pinnate setae (1 weakly geniculate) and 1 spinulose spine; distal armature consisting of 3 pinnate setae (2 weakly geniculate) and 3 spinulose spines, the largest of which is basally fused to a slender, unipinnate seta and bearing distinctive tube pore.

Labrum (Figure 4B) well developed with three lobes; posterior lobe smooth; ventral margin ornamented with short and dense median spinules and short lateral spinules in middle lobe; anterior lobe with short dense median spinules.

Mandible (Figure 3B). Coxal gnathobase well developed; with 1 large tooth ventrally, 2 blunt plus 2 pointed teeth medially, and bipinnate seta covered by elongate tooth dorsally. Coxa with spinules around insertion site of palp. Palp biramous, comprising basis, 1-segmented endopod and 3-segmented exopod; all segments (except exp-2 and -3) with spinules on anterior face. Basis with 3 well developed bipinnate setae and distal vestigial seta (arrowed in Figure 3B). Exopod slightly shorter than endopod; exp-1 largest, with short proximal and long distal seta laterally; exp-2 with 1 lateral seta; exp-3 minute, drawn out into strong spinulose spine apically and with short seta at outer subapical corner. Endopod swollen, with 3 setae laterally and 6 setae apically; all setae pinnate; slender spinules present along outer and inner margins.

Paragnaths (Figure 4C) well developed chitinized lobes; long setules on inner lateral margin, anteriorly; flattened stout spinules on anterior face under chitinized lobe; small spinules on outer distal margin, posteriorly.

Maxillule (Figure 5A–B). Praecoxa with well developed arthrite bearing 11 spines around distal margin and 2 setae on anterior surface. Coxa with epipodite represented by 1 seta; endite short, with 6 apical setae. Basis and endopod completely fused forming 'maxillulary allobasis'; with total of 14 apical setae. Exopod minute, with 2 long bipinnate setae.

Maxilla (Figure 4A). Syncoxa with 4 endites (2 praecoxal, 2 coxal); outer margin with 2 spinular patches; enditic setal formula [3, 1, 3, 3]; distal praecoxal endite largely incorporated, distal coxal endite cylindrical. Allobasis drawn out into long curved claw; accessory armature consisting of 1 curved spine and 2 setae distally, and 2 curved spines proximally near articulation with endopod. Endopod 3-segmented; enp-1 with 1 geniculate spiniform seta and 1 slender seta; enp-2 with 2 geniculate spiniform setae; enp-3 with 1 geniculate spiniform seta, 1 short, naked and 2 long, striated setae.

Maxilliped (Figure 1C). Syncoxa elongate; with anterior pattern of various spinules; with 4 strong, bilaterally serrate spines and 2 bipinnate setae. Basis armed with 1 strong, bilaterally serrate spine and 1 small seta. Endopod indistinctly 2-segmented; original segmentation marked by incomplete surface sutures anteriorly and posteriorly (Figure 1D); proximal part (= enp-1) with rudimentary seta and additional spinule (possibly representing vestigial armature element); distal part (= enp-2) with 2 strong bilaterally serrate spines apically, and 2 setae laterally along outer edge (proximal one about half the length of distal bipinnate one).

Swimming legs P1–P4 (Figure 6A–D). Coxae strongly developed, with distinct pattern of spinules as illustrated in Figure 6A–D. Bases with outer bipinnate seta; inner margin with long setules (P2–P3) or strong spinules (P4); in P1 with inner seta accompanied by spinules. Rami 3-segmented with exopods longer than (P1, P3, P4) or about equal in length to endopod (P2). Outer margins of exopodal segments (particularly exp-1) with one or several spinule rows. Outer margins of endopodal segments and inner margin of P2–P4 enp-1 with long setules or spinules.

Leg 1 (Figure 6A) shortest. Intercoxal sclerite armed bilaterally with 5 prominent spinules around distal margin and smaller spinules on anterior surface. Basis without spiniform projection on distal edge between exopod and endopod; outer seta not extending to distal margin of exp-1 segment; inner seta similar to outer seta, extending beyond distal margin of enp-1. Distal setae of exp-3 not curly.

Leg 2 (Figure 6B). Intercoxal sclerite armed bilaterally with 3 stout spinules and several smaller ones arranged around the distal margin and both anterior and posterior surfaces. Basis with prominent triangular projection between exopod and endopod; inner margin produced into lobate process. Enp-1 enlarged; distal inner corner produced into curved spinous process; inner element modified into curved spine. Proximal seta of enp-2 markedly shorter than distal.

Leg 3 (Figure 6C). Intercoxal sclerite armed bilaterally on both anterior and posterior face with 1 stout spinule and several smaller ones. Basis with spinous projection between exopod and endopod. Enp-1 almost as long as enp-2 and -3 combined; inner seta spiniform.

Leg 4 (Figure 6D). Intercoxal sclerite armed bilaterally with 1 stout and 1 slender spinule on posterior face. Basis without spinous projection between exo-

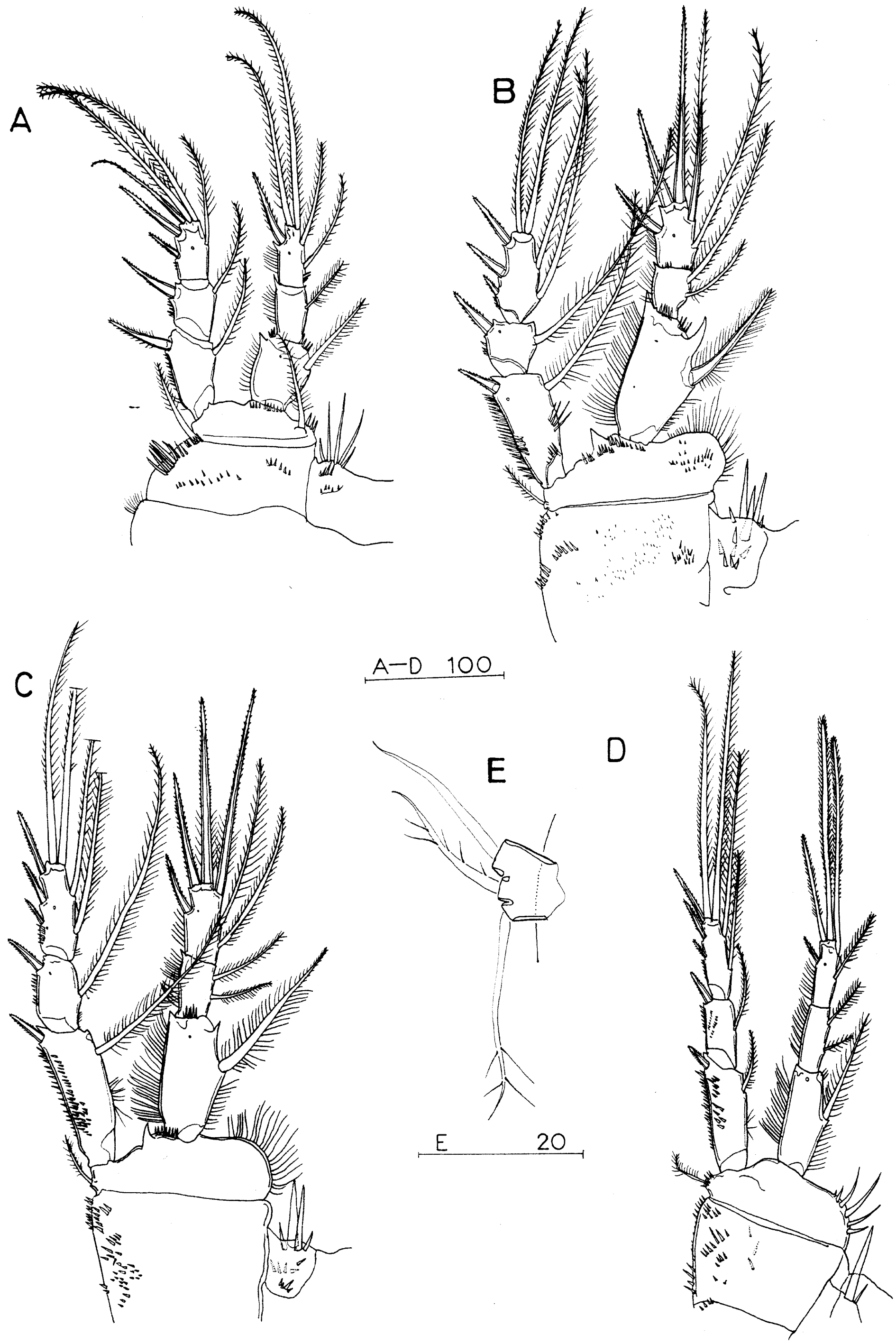

Figure 6. Neocervinia itoi n. sp., female: (A) P1, anterior; (B) P2, posterior; (C) P3, posterior; (D) P4, posterior; (E) P5. All scale bars in μm.

pod and endopod. Endopod slender. Spine and seta formula as follows:

	Coxa	Basis	Exopod			Endopod		
			1	2	3	1	2	3
P1	0-0	1-1	I-1;	I-1;	III-2-1	0-1;	0-1;	I-2-2
P2	0-0	1-0	I-1;	I-1;	III-2-2	0-I;	0-2;	I-2-2
P3	0-0	1-0	I-1;	I-1;	III-2-2	0-1;	0-2;	I-2-3
P4	0-0	1-0	I-1;	I-1;	III-2-2	0-1;	0-2;	I-2-2

Leg 5 (Figure 6E) laterally displaced; largely incorporated into somite; not defined at base; represented by small subrectangular lobe with 3 sparsely plumose or pinnate setae around distal margin; middle one shortest.

Male. Unknown.

Etymology

The species is named in honour of the late Dr Tatsunori Itô in recognition of his excellent contributions to cerviniid taxonomy.

Discussion

Huys et al. (1997) recently analysed the phylogenetic relationships within *Cervinia* Norman and concluded that the genus is paraphyletic. They re-instated *Pseudocervinia* Brotskaya as a valid genus and proposed two new genera, *Neocervinia* and *Brotskayaia*, to accommodate various species that previously had been placed in *Cervinia*. The new cold seep cerviniid is placed in the genus *Neocervinia* Huys, Møbjerg & Kristensen, 1997 on account of the following combination of characters: rostrum well developed; setae of P1 exp-3 not curly; P2–P4 without distinct spinous process on the inner margin of the basis; P1–P4 endopods 3-segmented; P5 minute, laterally displaced and not defined at base. The spine and seta formulae of the swimming legs is also in agreement with the generic dignosis.

The genus *Neocervinia* currently encompasses two species, *N. tenuicauda* (Brotskaya, 1963) from 5680-5690 m in the Japan Trench and *N. unisetosa* (Montagna, 1981) described from the bathyal zone of the Beaufort Sea off Alaska. *N. itoi* can be readily distinguished from its congeners by the 6-segmented antennules in the female. The reduction in antennulary segmentation results from fusion (rather than failure to separate) of segments 2 and 3 expressed in the other species of the genus and the original segmentation plane is marked by an incomplete, transverse, chitinous rib near the anterior margin. As a result, the aesthetasc present on segment 3 in both *N. tenuicauda* and *N. unisetosa* is found on segment 2 in the cold seep species. Huys et al.'s (1997) original diagnosis of the genus *Neocervinia* should therefore be corrected accordingly: 'Antennule 6- or 7-segmented in female, with aesthetasc on segment 2, or 3, respectively'.

The presence of sensillae (marked in Figure 3A) on the 1st and 2nd antennulary segments of *N. itoi* is remarkable. Sensillar structures are typically found on the rostrum, the cephalic shield and around the posterior margin of the body somites (except on the penultimate somite) but have thus far not been reported on appendages in harpacticoids. It is unlikely that the sensillae represent vestigial setae since the first antennulary segment in harpacticoids (except for the highly condensed antennules of the polyarthran families) never bears more than 1 seta. Because of their small size they could have easily been overlooked in other cerviniids, including the other species of *Neocervinia*, however scrutinous re-examination of the closely related *Cervinia synarthra* Sars revealed that they are absent, suggesting that these sensillar structures might well be an autapomorphy for *Neocervinia*. The fact that they are absent in copepodid V females and therefore do not appear until the final moult indicates a possible role in mate location or recognition. Interestingly, similar sensillae have recently been described in the hyperbenthic calanoid *Stygocyclopia balearica* Jaume & Boxshall where they are found on the proximalmost segments of the antennules in both sexes (Jaume & Boxshall, 1995). In *N. itoi* additional tiny sensillae were also observed on the dorsal surface of the caudal rami (Figure 2D).

Montagna (1981) stressed the usefulness of mouthpart setation characters in the Cerviniidae since even in the earliest descriptions they appear to be accurately figured. Our observations on *N. itoi* revealed that despite the generally large size of the mouthparts in this family, simple setal counts should be used with caution since various elements can be reduced in size or even be rudimentary such as some of the setae found on the mandibular basis and the maxillipedal endopod. Hence, several differences listed below might be more apparent than real and would require confirmation. The most significant discrepancy is the number of lateral setae on the basis of the mandible, being 4

(1 vestigial) in *N. itoi*, 3 in *N. tenuicauda* but only 1 in *N. unisetosa* (cf. name). Huys et al. (1977) already remarked on the doubtful validity of this character in *N. unisetosa*. Further differences can be found in the endopodal segmentation of the mandible, being 4-segmented in both *N. unisetosa* and *N. tenuicauda*, but only 3-segmented in *N. itoi* due to fusion of the 1st and 2nd segments. Both *N. unisetosa* and *N. tenuicauda* have a 2-segmented maxillipedal endopod, yet fusion of these segments (marked by an incomplete transverse suture) has produced a 1-segmented endopod in *N. itoi.* Moreover, the seta found in the proximal half of the maxillipedal endopod in *N. itoi* is strongly reduced (Figure 1D) in comparison to that of its congeners. The apical armature elements on the P5 of *N. tenuicauda* and *N. unisetosa* are very similar to each other in aspects of length and form, i.e. the two apical setae being reduced in *N. tenuicauda* and *N. unisetosa*, but normal in *N. itoi*. The female P6 of *N. itoi* is elongate with 1 rudimentary element and 2 apical setae, but short and with 3 normal setae apically in *N. unisetosa* and *N. tenuicauda.*

On account of the general reduction in segmentation of the antennule and both mandibular and maxillipedal endopods, *N. itoi* can be considered as the most advanced species in the *Neocervinia* lineage.

Key to the Species of Neocervinia *(based on females)*

1. Antennule 7-segmented, and 2 apical setae of P5 reduced **2**
   Antennule 6-segmented, and all apical setae of P5 normal .... *N. itoi* n.sp.
2. Caudal rami as long as last 4 urosomites combined (L/W = 12) .... *N. tenuicauda*
   Caudal rami as long as last 2 urosomites combined (L/W = 8) .... *N. unisetosa*

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