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ГЛАСНИК ОДЈЕЉЕЊА ПРИРОДНИХ НАУКА, 23, 2019.

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*Dmitry Sidorov**

**GROUNDWATER DEPENDENT FAUNA OF COASTAL
RIVERS AND SPRINGS OF THE SOUTH KURIL ISLANDS:
FIRST EVIDENCE ON SUBTERRANEAN AMPHIPODS
(CRUSTACEA: AMPHIPODA)**

Abstract

A new species of groundwater amphipod, *P. (G.) koropokkuru* from interstitial waters of Kunashir and Shikotan islands is described and illustrated, and its taxonomic affinity with congeners briefly discussed. To define phylogenetic position of mentioned species DNA barcode data are obtained.

Keywords: Amphipoda, Pontogeneiidae, *Paramoera*, new species, islas Kuriles

INTRODUCTION

Interstitial amphipod species of the genus *Paramoera* Miers, 1875 (Malacostraca: Pontogeneiidae) are common inhabitants of the various biotopes near sea shore environments along the North Pacific beaches [8, 3, 6]. Kunashir Island and Lesser Kuril chain — these are several islands of the northwest Pacific that are practically unaffected by anthropogenic pressure. These island territories are characterized by pristine nature and undoubtedly of the great ethnocultural and recreational importance. Even more the islands are known for their interesting geological history which is reflected in the stratigraphic chronicle of the region [12]. There are many freshwater streams and springs and considerable thick sedimentary rocks creates fine conditions for karst relief forms (caves, grottoes and

* Dmitry Sidorov, Federal scientific center of the East Asia Terrestrial Biodiversity FEB RAS, Vladivostok 690022, Russian Federation. E-mail: biospeorossica@gmail.com

sinkholes). So, for example, turf-covered karst in the valleys of brooks in the western part of Kunashir and Shikotan may be an interesting microbiotope for future study (see Figure 1A, B).

Bibliographic data on amphipods of the South Kuril Islands is scattered. There were no special studies on the fauna of amphipods of the islands and data on species diversity from this territory is often random. The first data can be found in Ueno [11]. In the native literature indications for some species can be found in works of Tzvetkova [10], Bulycheva [1] and others. However, these publication provides information mostly on terrestrial Talitridae and brackish water Anisogammaridae. Data on freshwater amphipods of the South Kuril Islands are missing.

The goal of this work is preliminary taxonomic inventory of the amphipod's fauna of the Kunashir, Shikotan, Zelionyi and Yuri islands, to paying special attention to the groundwater dependent fauna of the coastal environments. One of the objectives of present work was the study of the taxonomical status of the subgenus *Ganigamoera* Sidorov, 2010 with aim of DNA sequencing tool.

The *Ganigamoera* was assigned for stygomorphic species from Primorye and its phylogenetic position amongst similar species with male transformed pleopod II remains unclear. Development of this problem could help in solving of a number of paleogeographical issues because this group has the modern marine relatives, its history seems may reflect the relatively recent eustatic sea level changes in the region.

MATERIAL AND METHODS

During the period from 04 to 19 August 2013 by the author in the framework of the 49th South Kuril Marine Expedition the hydrobiological works on Kunashir Island and a number of a small islands of the Lesser Kuril Chain was carried out. Collections of algae, fish and invertebrates were sampled (mostly including water mites, aquatic insects and ostracods). Amphipod collections were sampled in the following points (numbers in parentheses correspond to stations designation on map, Fig. 1): — KUNASHIR ISLAND. **Pervukhina** (04. VIII), Pervukhina Bay located on western side of the island (k01–03); **Valentina** (05. VIII), coast near the Valentina Lake on northwestern part of the island (k09); **Dlinnoye** (06. VIII), coast 2–3 km from the Dlinnoye Lake on northern part of the island (k11–12). —SHIKOTAN ISLAND. **Krabovaya** (07. VIII), the Krabovaya Bay on northwestern part of the island (k13); **Gorobetc** (08–09. VIII), environs of the Gorobetc Bay in central part of the island (k14–17); **Voloshina** (10. VIII), Voloshina Cape in the extreme southeast of the island (k18–20); **Otradnaya** (11, 16. VIII), environs of the Otradnaya Bay in northern part of the island (k21–25); **Ayvazovskiy** (14. VIII), coast near the Ayvazovskiy (Cerkovnaya) Bay in

southeast part of the island (k35–36); **Dimitrova** (15. VIII), Dimitrova Bay on northeast sea-side of the island (k38). — ZELIONYI ISLAND. **Glushnevskiy** (13. VIII), Srednee Lake not far from the Glushnevskiy Cape in south part of the island (k34). — YURI ISLAND. **Shirokaya** (12. VIII), environs of the Shirokaya Bay (k28–31).

In taxonomic sampling and morphological essay a common methods and technique were applied. The nomenclature for setal patterns on article 3 of the mandibular palp follows the standard described by Karaman [2]. Genomic DNA extraction, amplification, and sequencing procedures were performed on the basis of the Lower Plants Lab. at the EATB-Center FEB RAS [7]. All sequences have been submitted 31 Dec. 2018 [<http://www.ncbi.nlm.nih.gov>] to GenBank (accession numbers MK358130 — MK358136).

The description is based on the type series, with the material examined is deposited in the Muséum national d'Histoire naturelle, Paris, France (prefix MNHN; the holotype is here) and in the research collection of D. Sidorov (Vladivostok, Russia). An additional supplemental online materials can be accessed through RUSAB project of BOLD System and via Figshare DOI 10.6084/m9.figshare.7992185.

RESULTS OF DNA BARCODING

Appointing the subgenus *Ganigamoera* for subterranean amphipods with transformed pleopods II from Primorye we was failed to determine properly the taxonomic relations and was doubting if both Japanese and North American species belong to this group. To bring some clarity on this issue the partial mitochondrial “barcoding” (CO1) gene sequences for five lineages of *Paramoera* sampled from the several interstitial and epigeal environments were compared. The molecular analysis comprised 14 sequences of *Paramoera* and *Awacaris* Ueno, 1971 was selected as out-group taxa [5] of a total 658 bp in length available in GenBank (NCBI LG146870–LG146874, MG319669, MG317951), together with 3 individuals (NCBI MK358130–MK358132) representing a new species *P. (G.) koropokkuru* from two different insular localities, 2 individuals (NCBI MK358133, MK358134) of *P. (G.) myslenkovi* Sidorov, 2010 from Primorye, and individuals with uncertain taxonomic status (NCBI MK358135) from Kunashir identified as *P. cf. koysama* Kuribayashi et Kyono and (NCBI MK358136) as *A. cf. moneronensis* Labay from Shikotan.

Morphologically similar specimens of *P. (G.) koropokkuru*, sp. nov. from Kunashir (RUSAB003) and Shikotan (RUSAB004–05) were presented by two haplotypes with genetic divergence between them 9.4%, while divergence between Shikotan lineage (RUSAB004–05) and *Paramoera* sp. isolates G689, G690 accessions NCBI LC146873, LC146874 was lower: 6.7%, and between

morphologically related *P. (G.) koropokkuru*, sp. nov. and *P. erimoensis* Kuribayashi et Kyono, 1995 accessions NCBI LC146870, LC146871 divergence: 15.2–15.5% (Table 1). Monophyly of the *P. (G.) myslenkovi* and *P. (G.) koropokkuru*, sp. nov. within the *Ganigamoera* is strongly supported on BioNJ tree (Fig. 1). However, since phylogenetic relationships was assessed lacking *P. (G.) tiunovi* Sidorov, 2010, and lineage of *P. erimoensis* remained unresolved in our analysis insomuch the solution of the *Ganigamoera*-problem seems to difficult still.

TABLE 1. Estimates of evolutionary divergence (uncorrected *p*-distances) over sequence pairs between partial mitochondrial CO1 gene among species and haplotypes of the Far Eastern pontogeneiids, with standard error (SE) calculation.

Species	1	2	3	4	5	6	7	8	9	10	11
1. <i>P. bousfieldi</i>	—	0.017	0.014	0.016	0.015	0.016	0.015	0.015	0.013	0.014	0.013
2. <i>Paramoera</i> sp. G690	0.184	—	0.013	0.013	0.014	0.010	0.014	0.013	0.013	0.014	0.014
3. <i>P. erimoensis</i>	0.148	0.157	—	0.014	0.012	0.013	0.014	0.013	0.012	0.013	0.012
4. <i>P. (G.) koropokkuru</i> RUSAB003 Kunashir	0.184	0.107	0.155	—	0.015	0.011	0.014	0.012	0.013	0.013	0.014
5. <i>P. cf. koysama</i> RUSAB001 Kunashir	0.151	0.159	0.115	0.169	—	0.013	0.013	0.014	0.011	0.012	0.011
6. <i>P. (G.) koropokkuru</i> RUSAB004–05 Shikotan	0.186	0.067	0.152	0.094	0.155	—	0.013	0.013	0.012	0.012	0.013
7. <i>A. cf. moneronensis</i> RUSAB002 Shikotan	0.164	0.171	0.143	0.159	0.128	0.159	—	0.013	0.011	0.011	0.012
8. <i>P. (G.) myslenkovi</i> RUSAB006–07 Primorye	0.160	0.144	0.129	0.141	0.124	0.135	0.129	—	0.012	0.013	0.013
9. <i>A. rhyaca</i>	0.170	0.162	0.142	0.170	0.135	0.161	0.136	0.151	—	0.008	0.009
10. <i>A. japonica</i>	0.164	0.175	0.154	0.182	0.137	0.170	0.141	0.153	0.115	—	0.010
11. <i>A. yezoensis</i>	0.158	0.173	0.144	0.174	0.127	0.167	0.137	0.147	0.123	0.123	—

TAXONOMIC PART

Order Amphipoda Latreille, 1816
Family Pontogeneiidae Stebbing, 1906
Genus *Paramoera* Miers, 1875

***Paramoera (Ganigamoera) koropokkuru*, sp. nov.**
 (Figures 2–4)

Diagnosis. Slender, prolonged, small-sized body of typical ‘far-eastern interstitial pontogeneiid’ amphipod (sexual dimorphism pronounced, i. e. males slightly smaller than females; both antennae with plate-like calceoli, pereopod VII and pleopod II sexually dimorphic with spiny carpus (article 5) and with gonopodous transformed outer ramus, respectively. Body segments smooth with sparse fine setae, light pinkish in color. Eyes reduced. Head inferior antennal sinus moderate, sub-rounded. Antenna II about as long as antenna I, gland cone sub-acute with 1 short apical seta, its tip reaching end of peduncular article 3. Maxilla I with symmetric narrow palps; outer lobe with only 10 pectinate spines. Epimera II and III with ventral margin armed. Telson slightly longer than uropod III peduncle. Small coxal gill VII present in both sexes. Sternal gills/humps are lacking. Body length 5.25–4.25 mm (female), 3.8–4.0 mm (males).

Material examined. **Holotype** MNHN-IU-2014–20753. RUSSIA: male, 3.8 mm, springs (seeps) on the beach, coast near the Valentina Lake (Sta. k09), freshwater, coarse-grained sand, small pebbles (not flooded), N44°16.831; E145°56.871, NW Kunashir Island, 05. VIII. 2013, leg. D. Sidorov. **Paratypes** 1–5/3sd-IBSS 3 females 5.25 mm, 4.5 mm, 4.25 mm (oostegites undeveloped), 1 male 4.0 mm, 10 juveniles, same data as for holotype; mouth of a small river near sea shore, Dimitrova Bay (Sta. k38), coarse-grained sand, stones, fast moving stream, N43°47.697; E146°49.665, NE Shikotan Island, 15. VIII. 2013, leg. D. Sidorov.

GenBank accession numbers: MK358130 — MK358132 for isolates RUSAB003–RUSAB005.

Etymology. The specific epithet ‘*koropokkuru*’ means a race of small ‘pit-dweller people’ in mythology of Ainu of the south Kuril Islands.

Description of holotype. GENERAL BODY MORPHOLOGY (Figs 2, 3L, 4A–F). Slender, weakly setose with minute setules *Paramoera*. Eyes reduced (consist of 8 detached ommatidia), pigmented. Body light pinkish, semi-transparent in color when alive with length up to 5.25 mm.

Head with inter-antennal (lateral) lobe evenly rounded, inferior antennal sinus not incised. Pereon and pleon unarmed, without dorsal spines. Coxal plates I–IV deeper than broad, with rounded distal margins, plate IV posterior margin

with prominent excavation, V and VI bilobate, plate VII smallest, semilunar. Coxal gills II–VII small, often less than corresponding coxal plates, VII the smallest. Median sternal processes (or sternal humps on ventral surface of pereonites) absent.

Epimeron I disto-posteriorly unproduced and rounded, ventral margin unarmed. Epimeron II disto-posteriorly minutely produced, ventral margin bearing 3 spines. Epimeron III distinctly produced, only with 2 spines on ventral margin.

Urosomites smooth, without cuticular medio-dorsal elevations and spines; ventral surface of urosomite I bearing 2 lateral spines and cluster of long thin setae at each side. Telson narrow, somewhat longer than uropod III peduncle, split to 40%, lobes sub-apically with 3 short setae, laterally single group of 2 setae.

ANTENNAE (Figs 2A, 3A, B). Antenna I 40% of body length, peduncle articles in relation 1.0: 0.65: 0.4, main flagellum of 18 articles; accessory flagellum reduced of 1-segmented minute scale-like article; peduncle articles with a few short and fine setae, such also the setation of flagellar articles, flagellar articles bearing calceoli and oblong aesthetascs. Antenna II slightly shorter of antenna I, peduncle articles 4 + 5 and flagellum in relation 0.2: 0.2: 1.0; articles 4 + 5 with 5 groups each of 2–4 small setae along entire lengths; flagellum modestly equipped with short setae, bearing calceoli.

MOUTH PARTS (Figs 3C–H). Mandibular palp with articles 2 and 3 subequal in length, article 3 bearing 1 group of 4 A-seta. Maxilla I sub-symmetric, palps narrow, apically with 5 spines; outer lobe with 10 subequal, pectinate spines. Maxilla II and maxilliped without peculiarities.

GNATHOPODS I AND II (Figs 2A, D, E). Gnathopod I carpus (article 5) weak, about 0.4x as long as propodus (article 6), bearing 2 groups of rastellate setae; propodus sub-rectangular, palm slightly convex and armed with group of 8 distally-notched spines near palmar (or defining) angle; posterior margin longer than palm with 4 sets of setae; dactylus with 5 minute setae along inner margin, with 1 seta on outer face, nail indistinct. Gnathopod II similar to gnathopod I, but a little larger; carpus with 3 groups of rastellate setae; propodus armed with group of 10 distally-notched spines; dactylus similar to that of gnathopod I.

PEREOPODS (Figs 2A, 4A–F). Pereopods III and IV sub-similar; bases narrow, with sets of long setae on both margins. Lengths of pereopods V: VI: VII is 0.8: 0.95: 1.0. Pereopod VI length 0.4× body length. Bases V–VII broad, posterior margin with postero-distal lobes; marginal serrations with short setae posteriorly and with 4 single spines anteriorly; dactylus length about 0.3× of propodus length, inner margin with 1 thin seta at hinge. Pereopod VII basis (article 2) with 1 long seta on inner face; merus (article 4) stout, with group of 4 strong spines on posterior margin; carpus (article 5) slightly curved, anteroproximal surface densely lined with aggregation of stiff spines.

PLEOPODS AND UROPODS (Figs 2A, 3I-K, 4G-J). Pleopods ordinary, biramous, with two-hooked retinacles. Pleopod II gonopodous, transformed to passively prehensile an accessory sex construct consisting of terminal¹ *koczarga*-like structure accompanied by 3 spine-setae (2 long plumose and 1 short) on tip of outer ramus. Uropod I long, peduncle with 11 short spines, rami shorter than peduncle, with long apical and sub-apical spines. Uropod II outer ramus 0.8x as long as inner ramus, approximately 70% length of peduncle, both rami armed with 3–4 setae on outer margins. Uropod III biramous, about 70% as long as uropod I, peduncle twice as long as wide, with 3 spines distally, inner ramus (endopodite) 40% longer than peduncle, outer ramus (exopodite) a little shorter than inner; both rami lanceolate, bearing row of spines on both margins and 1 or 2 simple setae each sub-apically.

Variation. Not observed.

Sexual dimorphism. Females 5.25–4.25 mm long, both antennae without calceoli, oostegites (brood plates) weakly developed, small and present on pereopods II–V (non-setose in the material examined); pereopod VII and pleopod II lacking modified structures; in all other characters similar to male.

Distribution and ecology. *P. (G.) koropokkuru*, sp. nov. is known from interstitial groundwater habitats near sea shore in the Kunashir Is. (type loc.) and Shikotan Is. (see Fig. 1C, D).

Remarks. *P. (G.) koropokkuru*, sp. nov. most closely resembles *P. erimoensis* Kuribayashi et Kyono, 1995, but can be distinguished from the latter by (character states of *P. erimoensis* in parentheses): eyes of 7–8 ommatidia (4 ommatidia); maxilla I inner plate with 6 plumose setae (5 setae); maxilla I outer plate with 10 pectinate spines (11 spines); maxilla I palp narrow (broad); maxilla II inner plate with 4 plumose setae in oblique row (3 setae); maxilliped inner plate with 4 teeth distally (with 2 teeth); defining angle of gnathopods I and II armed with 8 and 10 spines (both with 13 spines); posterodistal corners of epimeral plates I–III sub-acute (corners of plates indistinct, rounded); epimeral plates II and III with spines on ventral margins (without spines); coxal gill VII present (absent); telson cleft 40% (55%).

***Paramoera cf. koysama* Kuribayashi et Kyono, 1995**

syn.: *Paramoera koysama* Kuribayashi et Kyono, 1995: p. 760, figs 1–5 (orig. descrip.).

Distribution. Previously recorded in Hokkaido, Japan [3].

¹ *Koczarga* (polish) — a tool from the fire-resistant material for movement of burning coals in a fire chamber.

New record. RUSSIA: three immature specimens 1–5/4sd-IBSS, 3.5 mm, 2×3.0 mm, springs (seeps) on the beach, coast near the Valentina Lake (Sta. k09), freshwater, coarse-grained sand, small pebbles (not flooded), N44°16.831; E145°56.871, NW Kunashir Island, 05. VIII. 2013, leg. D. Sidorov.

Accompanying fauna: *P. (G.) koropokkuru*, sp. nov. (Amphipoda: Pontogeneiidae) described above.

GenBank accession number: MK358135 for isolate RUSAB001.

Remarks. Three juvenile specimens measuring ca. 3.0–3.5 mm in body length and without visible oostegites (brood plates) or any other sexual features were found in the material examined. Eyes large, reniform, black in color, but with a cincture of colorless ommatidia around (crystalline ommatidia). Specimens showed clear ontogenetic variation in the morphology of uropods: rami of uropods I and II bearing 1 extremely long saber-shaped spine apically; uropod III lacking plumose setae; remained appendages as gnathopods and pereopods being poorly armed.

Genus *Awacaris* Ueno, 1971

Awacaris cf. *moneronensis* (Labay, 1997)

syn.: *Sternomoera moneronensis* Labay, 1997: p. 754, figs 1–3 (orig. descrip.).
— Tomikawa, Sidorov, 2014, p. 36, figs 2–5.

Distribution. Previously recorded in Moneron, Russia [4].

New record. RUSSIA: ten specimens 1–5/5sd-IBSS, 10 females 3×8.0 mm, 4×6.0mm, 3×4.0 mm (oostegites developed, setose), stream near sea shore, Voloshina Cape (Sta. k20), stones, fine-grained sand, N43°42.613; E146°38.908, SE of Shikotan Island, 10. VIII. 2013, leg. D. Sidorov.

Accompanying fauna: *Eogammarus* sp. (Amphipoda: Anisogammaridae).

GenBank accession number: MK358136 for isolate RUSAB002.

Remarks. Although, *A. moneronensis* is a poorly known but the following features of Shikotan specimens are well correspond with description of this species [4, 9]: head inferior antennal sinus sharply incised; eyes very large, sub-rectangular (or reniform); character arrangement of sternal gills on pereonites II–VII as follows 1–4–4–4–5–3; carpus (article 5) of gnathopod II is slightly longer than corresponding propodus (article 6); oostegites II–V well developed, large, setose with short setae. As male morphologic features of *A. moneronensis* remains unclear a further field re-survey required.

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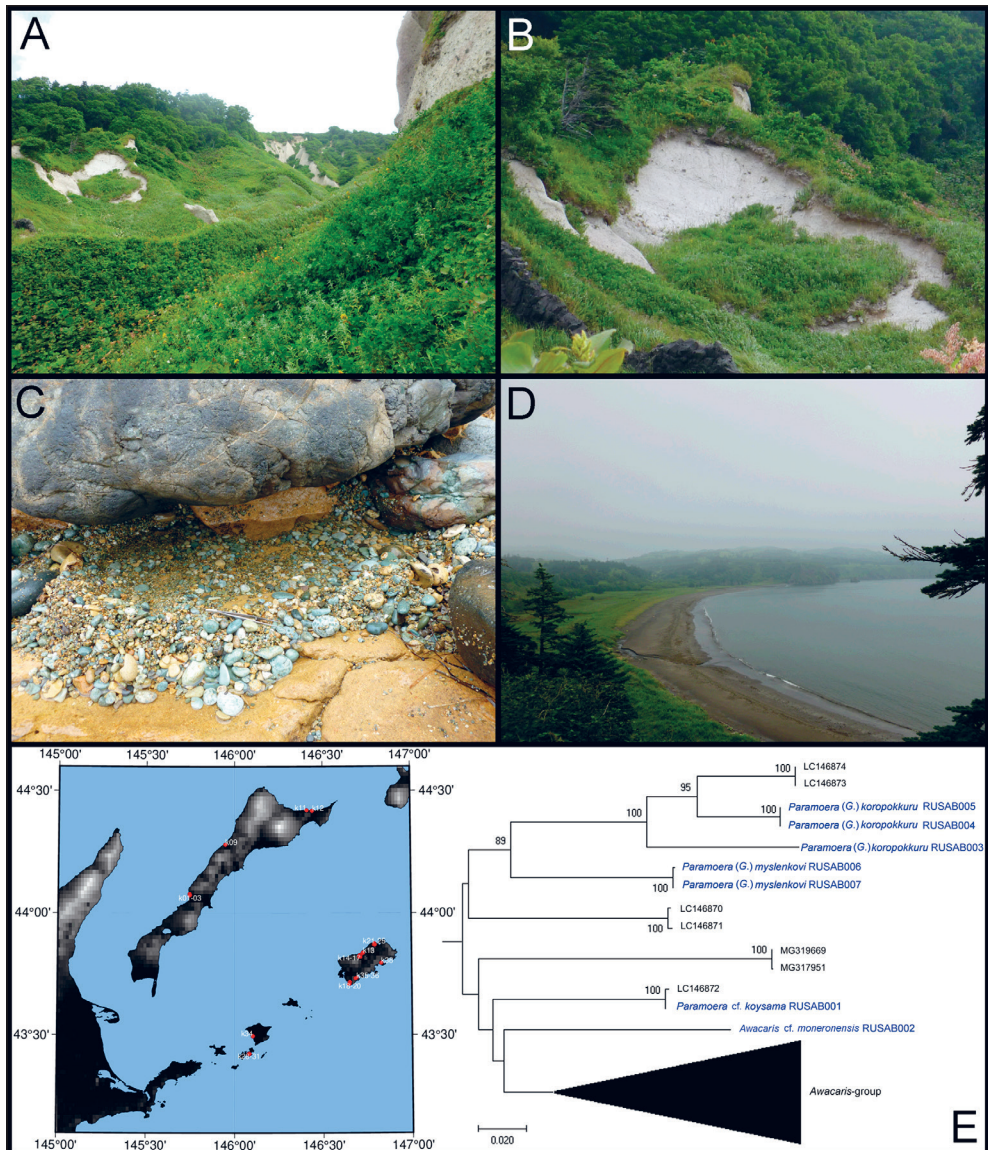


FIGURE 1. (A) sediment thickness in Pervukhina Bay, Kunashir (Malokurilskaya formation, Maastricht); (B) turf-covered sinkhole (dolines), *ibid.*; (C) microbiotope of *Paramoera koropokkuru*, sp. nov., Kunashir and (D) creek habitat in Dimitrova Bay, Shikotan; (E) left panel: map-scheme of the south Kuril Islands with localization of the collecting points and sequenced specimens; right panel: BioNJ cladogram reconstructed employing a HKY + Γ substitution model with bootstrap probabilities (values less than 50% not shown) based on the mitochondrial cytochrome c oxidase 1 (CO1) sequences for representatives of species within the Asian-Pacific pontogeniids. New sequences for this study are indicated in color. Scale bar indicates the number of substitutions per site.

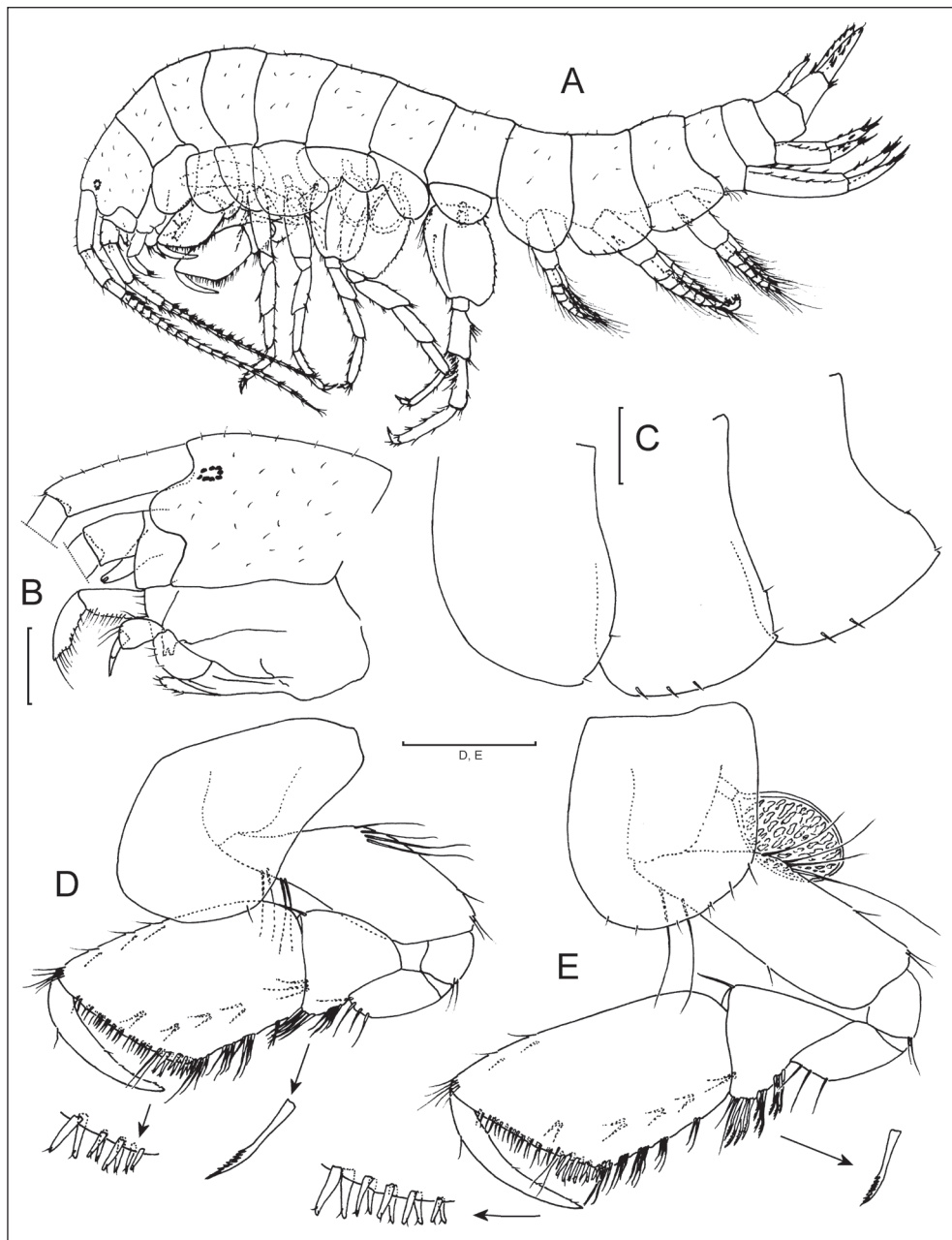


FIGURE 2. *Paramoera koropokkuru*, sp. nov., holotype, male, 3.8 mm: (A) habitus; (B) head; (C) epimeral plates I–III; (D) gnathopod I; (E) gnathopod II. Scales bars 0.2 mm.

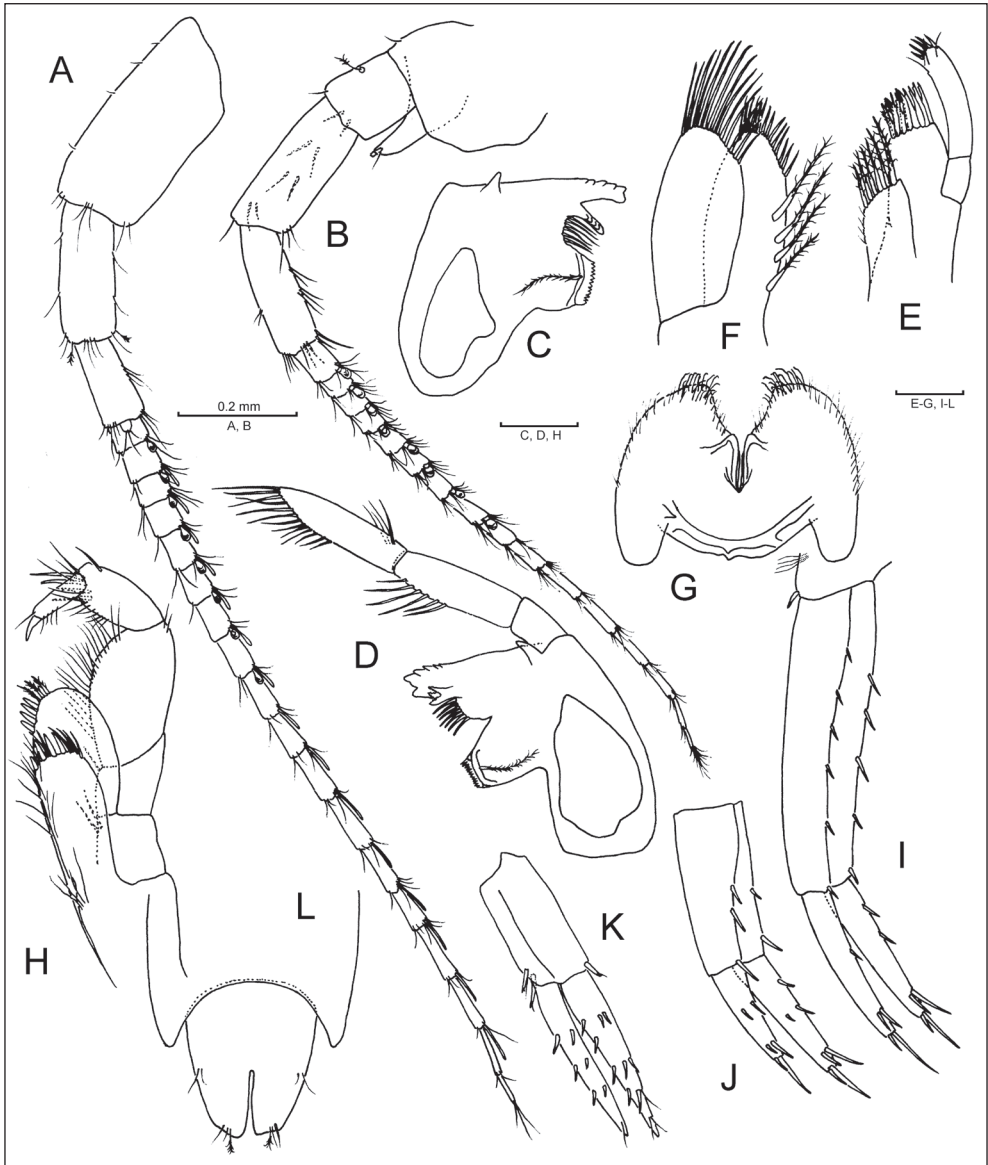


FIGURE 3. *Paramoera koropokkuru*, sp. nov., holotype, male, 3.8 mm: (A) antenna I; (B) antenna II; (C) mandible, left; (D) mandible, right; (E) maxilla I; (F) maxilla II; (G) paragnaths (=lower lip); (H) maxilliped; (I) uropod I; (J) uropod II; (K) uropod III; (L) telson. Scales bars 0.1 mm, if not indicated otherwise.

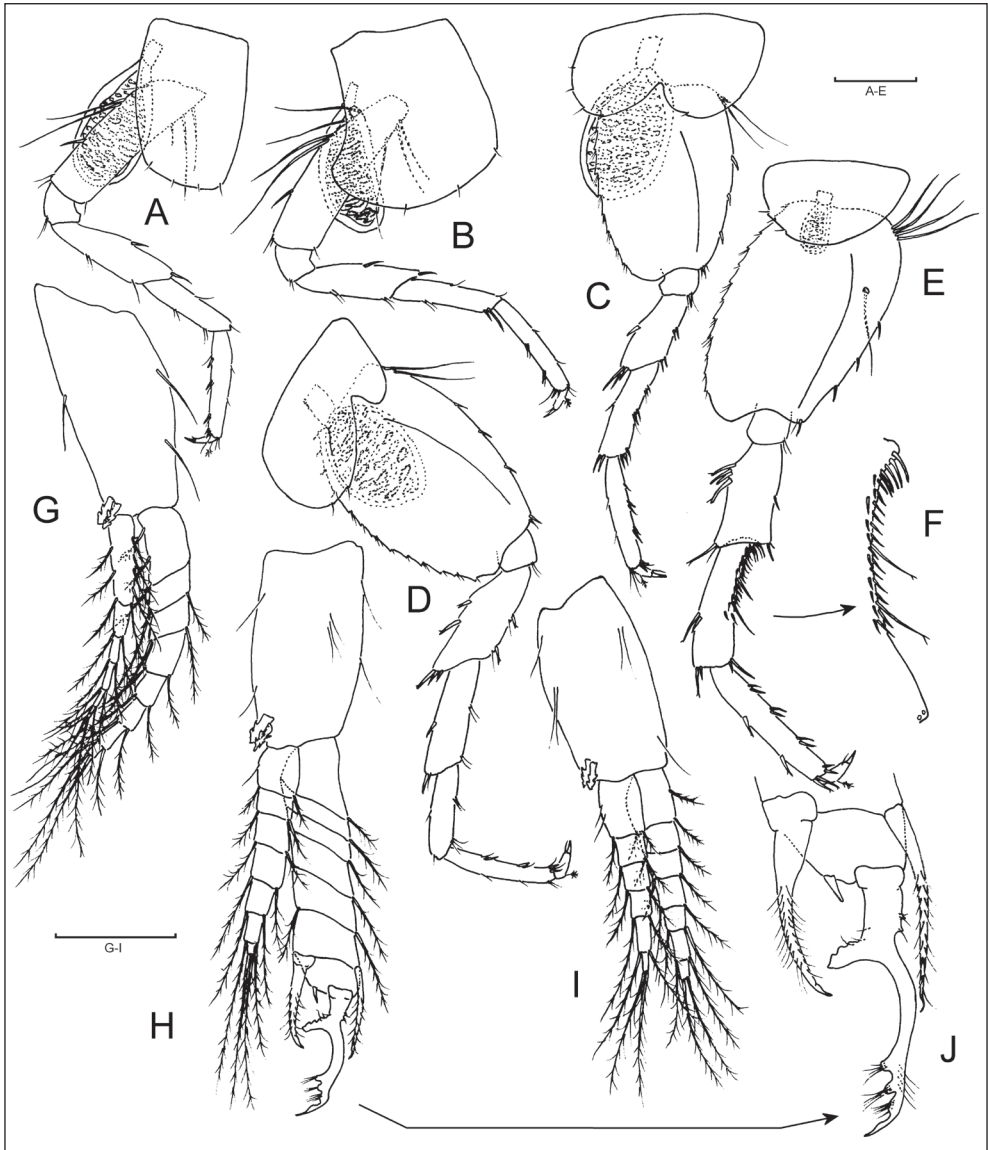


FIGURE 4. *Paramoera koropokkuru*, sp. nov., holotype, male, 3.8 mm: (A) pereopod III; (B) pereopod IV; (C) pereopod V; (D) pereopod VI; (E) pereopod VII; (F) spine-bearing anterior surface of carpus (article 5) of pereopod VII; (G) pleopod I; (H) pleopod II; (I) prehensile structure on tip of pleopod II outer ramus; (J) pleopod III. Scales bars 0.2 mm.