



## Research article

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# **A new species of the genus *Liljeborgia* Spence Bate, 1862 (Crustacea: Amphipoda: Liljeborgiidae) associated with the burrows of the spoon worm *Urechis unicinctus* in the Sea of Japan**

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**Abstract.** A new symbiotic species of liljeborgiid amphipods, *Liljeborgia associata* sp. nov., is described from the burrows of the spoon worm *Urechis unicinctus* (Drasche, 1880) (Annelida: Polychaeta: Echiura: Urechidae) in the southern part of Peter the Great Bay and Posjeta Bay in the Sea of Japan. The new species is mostly similar and probably related to *Liljeborgia geminata* Barnard, 1969, known from the Californian coasts of the USA, and *Liljeborgia serratoides* Tzvetkova, 1967, described from Posjeta Bay in the Sea of Japan, but can be clearly distinguished from all congeners by morphological features of mouthparts, appendages and telson. The new species is only the fourth in the family Liljeborgiidae to be described from the Russian coast of the northwestern Pacific and the first in association with spoon worms (Echiura).

**Keywords.** Diversity, association, northwestern Pacific, boreal, barcoding.

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## Introduction

Representatives of the family Liljeborgiidae (Crustacea: Amphipoda) are relatively rare in the higher latitudes of the North Pacific. Only three genera, *Liljeborgia* Spence Bate, 1862, *Idunella* G.O. Sars, 1894 (= *Listriella* J.L. Barnard, 1959) and *Sextonia* Chevreux, 1920, are known from the area. About 10+ species of the family are known from the NE Pacific (eastern coasts of the Bering Sea; Pacific coasts of North America), including four described species of the genus *Liljeborgia*: *Liljeborgia cota* Barnard, 1962 known from the Gulf of Alaska to the northern Baja California, Mexico, at a depth of 366–2000 m; *L. geminata* Barnard, 1969 (*L. kinahani* Bate, 1862 is considered as a junior synonym), described from Goleta to the northern Baja California, at a depth of 3–70 m; *L. marcinabrio* Barnard, 1979, known from Bahia de Los Angeles, Gulf of California, at a depth of 46 m; and *L. pallida* Bate, 1857, having a wide area of distribution from NE Atlantic to the Central Californian coasts, at a depth of 40–611 m (Barnard 1959; Barnard & Karaman 1991; d’Udekem d’Acoz 2010; Cadien 2015). Moreover, two undescribed species are also presented in the NE Pacific: one free-living, deep-water species and one commensal

with *Pagurus hemphilli* (J.E.Benedict, 1892) (Crustacea: Decapoda: Paguridae), living along the shores of Central California (Cadien 2015).

The diversity of the family along the coasts of the NW Pacific (western coasts of the Bering Sea; Pacific coasts of Russia and Japan; Sea of Okhotsk; northern parts of the Sea of Japan and the Yellow Sea) is less studied and is obviously underestimated. Only three species of the family have been reported from Russian waters: *Liljeborgia serratoides* Tzvetkova, 1967 was described from the shallow waters of Posjeta Bay in the Sea of Japan (Tzvetkova 1967), *Sextonia caecus* Labay, 2017 was recently described from the Sea of Okhotsk, at a depth of 109–309 m (Labay 2017) and Gurjanova (1951) reported Northern Atlantic and Arctic *Lilljeborgia* cf. *fissicornis* (M. Sars, 1858) from the Bering Sea, representing the only known record of the species from the NW Pacific (see d’Udekem d’Acoz & Vader 2009). The geographically closest species of the family Liljeborgiidae to the aforementioned species are known from the Seto Inland Sea in southern Japan, where *Liljeborgia japonica* Nagata, 1965 and *L. serrata* Nagata, 1965, *Idunella chilensis* Chilton, 1921 and *I. curvidactyla* Nagata, 1965 were described by Nagata (1965), and the Yellow Sea, where *L. hwanghaensis* Kim & Kim, 1990 and *L. sinica* Ren, 1992 are also known (Kim & Kim 1990; Ren 1992, 2007; Ishimaru 1994).

A variety of symbiotic communities associated with soft-bottom deep-burrowing invertebrates were investigated in the Peter the Great Bay and Posjeta Bay in the Sea of Japan. The sampling of infaunal animals and symbiotic assemblage was carried out using a bait suction pump (yabby pump), which allowed us to study the diversity of burrowing crustaceans and their associates in Russian waters (e.g., Marin 2010, 2015, 2016, 2018a, 2018b; Marin *et al.* 2011, 2013; Marin & Kornienko 2014) with the description of several new crustacean (Marin 2013, 2017) and even a new phoronid species (Temereva & Chichvarkhin 2017). Numerous specimens of *Liljeborgia* sp. were also collected from the burrows of the spoon worm *Urechis unicinctus* (Drasche, 1880) (Annelida: Polychaeta: Echiura: Urechidae). Previously, this species was identified as *L. serratoides/geminata* in accordance with its similar morphology and distinctive coloration, however, the presence of several specific morphological features has allowed its separation into a new species described herewithin.

## Material and methods

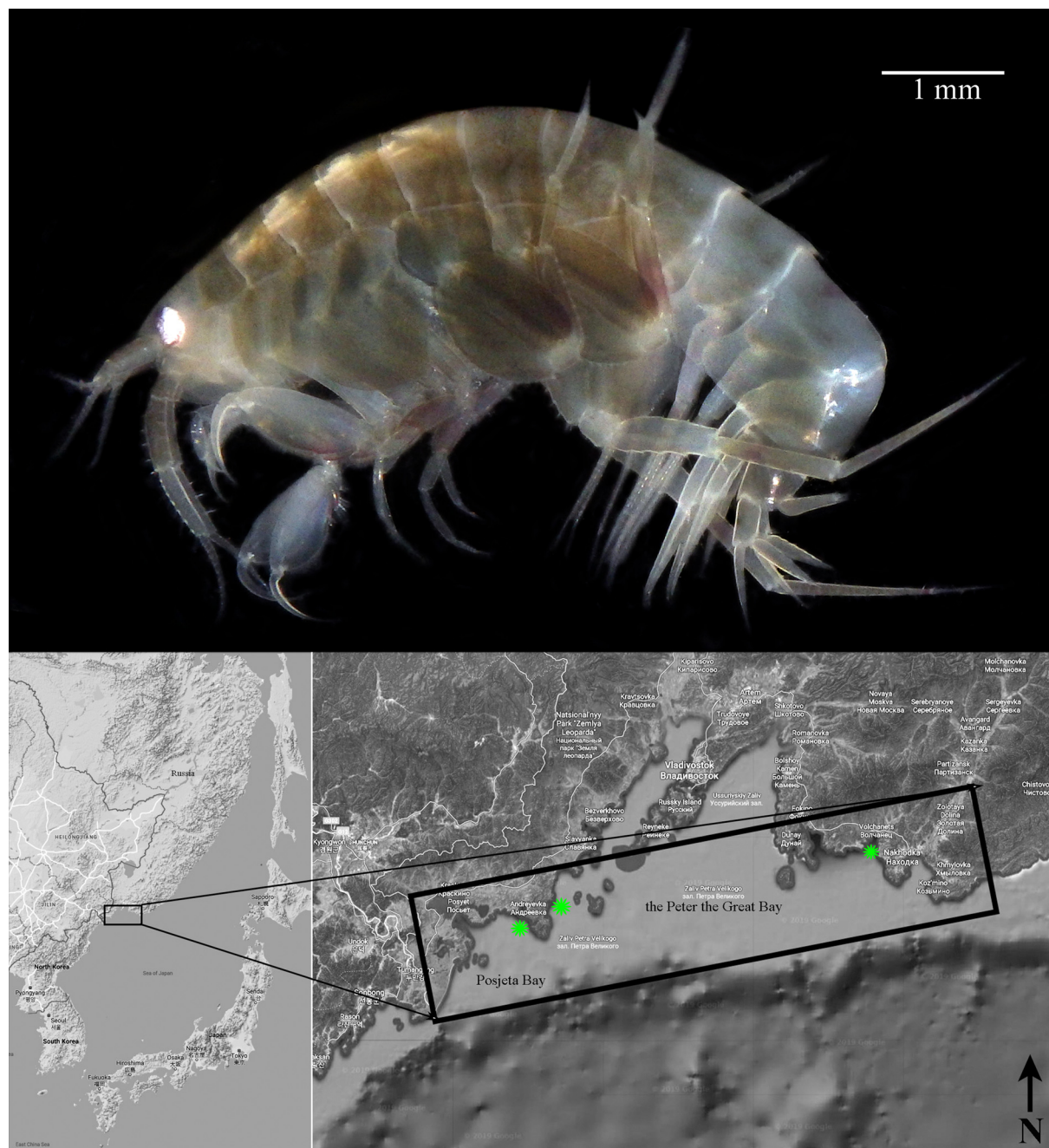
### Specimen sampling and imaging

Sampling was carried out in the estuary of Volchanka River in Vostok Bay near the scientific station “Vostok” (42°51’14.48” N, 132°46’47.24” E) and in Astafieva Bay (42°36’45.6” N, 131°12’24.8” E), both within Peter the Great Bay, as well as Troitza Bay (42°38’60.0” N, 131°07’27.8” E) located within Posjeta Bay in Sea of Japan (see Fig. 1). These bays are well known by large populations of the common spoon worm *U. unicinctus* and other burrowing animals. The hosts and symbiotic community were collected using a bait suction pump (yabby pump), which is actively used in the sampling of deep burrowing animals (e.g., Eleftheriou & McIntyre 2005). Unfortunately, in our study it was not possible to measure the length or volume of the host’s burrows. The collected specimens were photographed alive *in situ* using a Canon G16 digital camera and then fixed with 90% ethanol solution. The drawings were made using camera lucida attached to an Olympus SZX10 stereo microscope.

### Molecular study

To study the molecular genetic diversity, a fragment of the mitochondrial gene coding for cytochrome c oxidase subunit I (COI mtDNA) gene marker was amplified, sequenced and compared. Two female specimens (LEMMI) from Vostok Bay were used for the molecular-genetic examination. Total genomic DNA was extracted from abdominal and pereopod muscle tissue using the innuPREP DNA Micro Kit (AnalytikJena, Germany) following the manufacturer’s protocol. The COI gene marker was amplified with the help of primers «m13polylc»

(TGTAACGACGGCCAGTGAYTATWTTCAACAAATCATAAAGATATTGG) and «m13polyhc» (CAGGAAACAGCTATGACTAMACTTCWGGGTGACCAAARAATCA) (Carr *et al.* 2011); 16S with the help of +16SA ('CGCCTGTTTATCAAAAACAT') and -16SH ('CCGGTCTGAACTCAGATCACG'); 28S with the help of +C1 ('ACCCGCTGAATTTAAGCAT') and -D2 ('TCCGTGTTTCAAGACGG'). PCR products were performed on amplificator T100 (Bio-Rad, USA) under the following conditions: initial denaturation at 96°C for 1.5 min followed by 42 cycles of 95°C for 2 min, 49°C for 35 seconds, and 72°C for 1.5 min, followed by chain extension at 72°C for 7 min. The volume of 10 µL of reaction



**Fig. 1.** Habitus of live female specimen of *Liljeborgia associata* sp. nov. from Vostok Bay of the Sea of Japan with geographical indication of the collection sites in the Peter the Great Bay and Posjeta Bay of the Sea of Japan.

mixture contained 1 µL of total DNA, 2 µL of 5 × PCR mix (Dialat, Russia) and 1 µL of each primer. The amplification products were separated by using gel electrophoresis of nucleic acids on a 1.5% agarose gel in 1 × TBE, and then stained and visualized with 0.003% EtBr using imaging UV software. DNA nucleotide sequences were determined using Genetic Analyzer ABI 3500 (Applied Biosystems, USA) and BigDye 3.1 (Applied Biosystems, USA) with direct and reverse primers. Unfortunately, the number of sequences of the representatives of the family Liljeborgiidae deposited in genetic databases is still very small. As the phylogenetic relations of studied species are out of our interest, the obtained DNA (COI mtDNA) data are presented in the paper without any analysis.

### Type material and morphological study

The type material is deposited in the collection of Zoological Museum of Moscow State University, Moscow, Russia (ZMMU) and the Laboratory of Ecology and Evolution of Marine Invertebrates (LEMMI), A.N. Severtzov Institute of Ecology and Evolution, Russian Academy of Sciences, Moscow, Russia.

The body length (bl, mm), defined as the dorsal length from distal margin of head to the posterior margin of telson without the length of uropod III and antennae, is used as a standard measurement.

The terminology and the general model of the description are used in the study after d’Udekem d’Acoz & Vader (2009) and d’Udekem d’Acoz (2010).

### Abbreviations

|              |   |                     |
|--------------|---|---------------------|
| <i>A1</i>    | = | antenna 1           |
| <i>A2</i>    | = | antenna 2           |
| <i>bl</i>    | = | body length         |
| <i>Ep1–3</i> | = | epimeral plates 1–3 |
| <i>Gn1</i>   | = | gnathopod 1         |
| <i>Gn2</i>   | = | gnathopod 2         |
| <i>Md</i>    | = | mandible            |
| <i>Mx1</i>   | = | maxilla 1           |
| <i>Mx2</i>   | = | maxilla 2           |
| <i>Mxp</i>   | = | maxilliped          |
| <i>P1–P7</i> | = | pereiopods 1–7      |
| <i>U1–U3</i> | = | uropods 1–3         |

### Results

Phylum Arthropoda von Siebold, 1848  
Class Malacostraca Latreille, 1802  
Order Decapoda Latreille, 1802  
Family Liljeborgiidae Stebbing, 1899  
Genus *Liljeborgia* Spence Bate, 1862

*Liljeborgia associata* sp. nov.

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Figs 1–5

### Diagnosis

*A1* with stout articles 2 (about as long as wide) and 3 (about 1.5 times as long as wide), article 2 with dorsal projection produced into a rounded lobe; palp of *Md* with long and slender articles, article 3

almost equal to articles 1 and 2, about 5 times as long as wide; palp of *Mx1* with broad shovel-shaped article 1; stout articles 3 and 2 of *A2*, article 3 about as long as wide and article 2 about 3 times as long as wide; *P1* and *P2* with short and wide merus, about 3 times as long as wide; *PP5–7* with slender propodal segments, about 6, 7 and 10 times as long as wide, respectively; posterodorsal area of *Pleonites 1* and 2 produced into 3 small teeth of which the median one is the longest; *Urosomites 1* and 2 with well-marked dorsal crest; *Telson* with long distal teeth, accompanied by 2 interdental long and slender spines.

### Etymology

The species is named after its symbiotic lifestyle.

### Type material

#### Holotype

PACIFIC OCEAN • ♀ (bl 6.5 mm); Sea of Japan, Primorye, Peter the Great Bay, Vostok Bay, in front of the scientific station “Vostok”; 42°54′35.8″ N, 132°44′08.7″ E; depth 1–1.5 m; 30–31 Jul. 2017; I. Marin leg.; shore in front of the laboratory, sandy-gravel bottom overgrown with sea grass; yabby-pump; from burrows of spoon worm *U. uncinatus*; ZMMU Mb-1153.

#### Paratypes

PACIFIC OCEAN • 1 ♂; same data as for holotype; ZMMU Mb-1154 • 1 ♀; same data as for holotype; GenBank: MN704855, MN704856; ZMMU Mb-1155.

### Additional material

PACIFIC OCEAN • 8 ♀♀, 1 ♂; same data as for holotype; LEMMI • 5 ♀♀; Sea of Japan, Primorye, Posjeta Bay, Troitz Bay; 42°38′60.0″ N, 131°07′27.8″ E; depth 1–1.5 m; Jul. 2019; I. Marin leg.; muddy sand; inside burrow of spoon worm *U. uncinatus*; yabby-pump; LEMMI • 1 ♀; Peter the Great Bay, Astafieva Bay; 42°36′52.2″ N, 131°12′01.1″ E; depth 1–1.5 m; Jul. 2019; I. Marin leg.; sand bottom; inside burrow of spoon worm *U. uncinatus*; yabby-pump; LEMMI.

### Description

**HEAD** (Fig. 2c). Rostrum turned downward, distally acute; eye large (Fig. 1), with well-developed ommatidia, eye brightly white in alive specimens (Fig. 1).

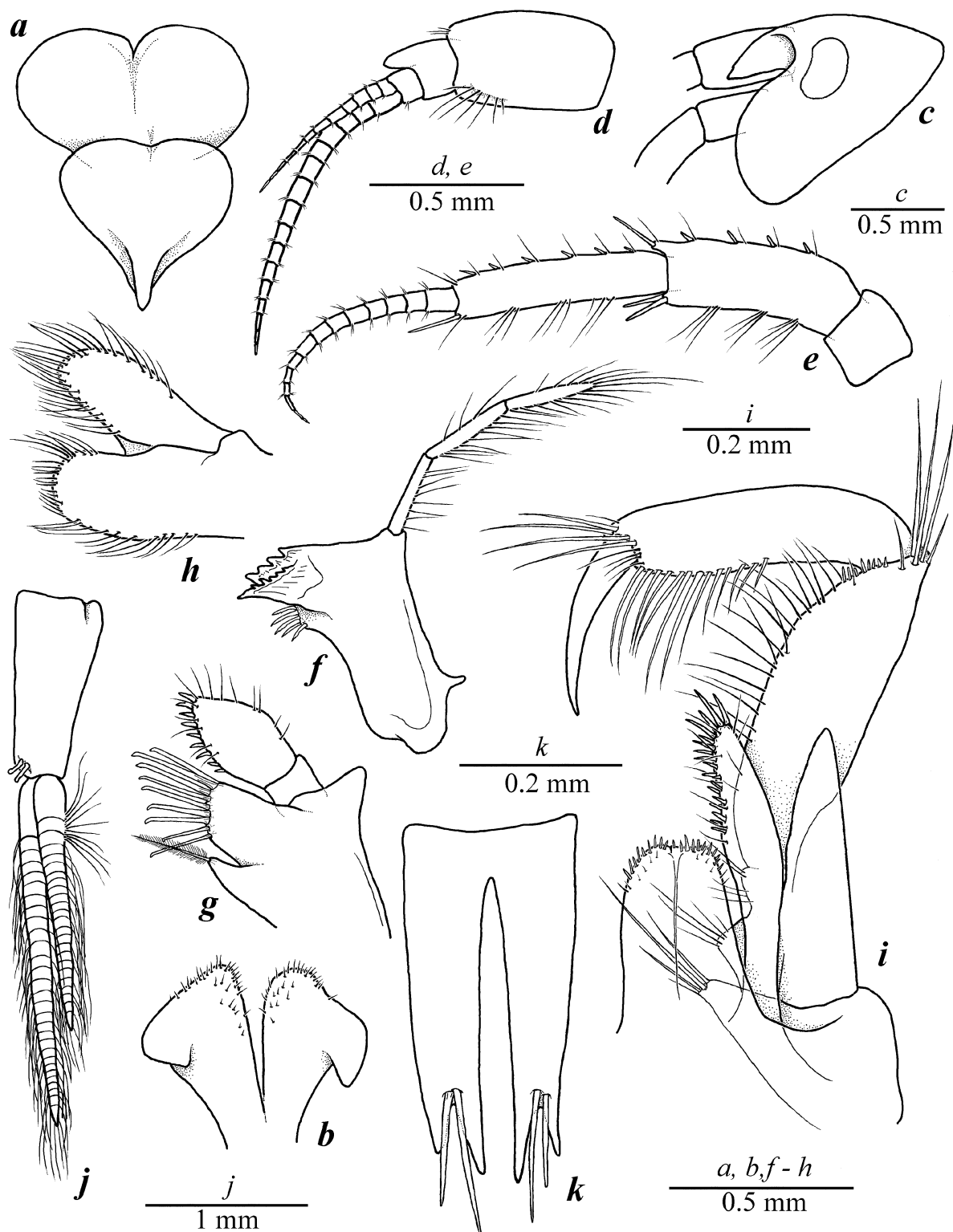
**ANTENNA 1** (Fig. 2d). Article 3 stout, about 1.5 times as long as wide; article 2 about as long as wide, with dorsal projection produced into a rounded lobe; major flagellum with 15–17 articles; accessory flagellum with 10–11 articles.

**ANTENNA 2** (Fig. 2e). Article 3 about as long as wide, unarmed; article 2 about 3 times as long as wide, with 4 small dorsal spines, with 1 long distodorsal and 2 long simple distoventral spines, with simple setae ventrally; article 1 about 4–4.5 times as long as wide, with small dorsal spines, with 1 distoventral long spine, unarmed ventrally; flagellum with 13–15 articles.

**LABRUM** (Fig. 2a). Upper lip with labrum broader than long and smaller than epistome, apical margin sinuous.

**EPISTOME** (Fig. 2b). With rounded lobes, protruding in lateral view, armed with small setae dorsally.

**MANDIBLE** (Fig. 2f). *Lacinia mobilis* large, anterior margin armed with 5 small strong teeth; palp consists of 3 long slender articles, similar in size, covered with long simple setae, article 1 almost equal to article 2, about 5–6 times as long as wide; article 2 about 5–6 times as long as wide; article 3 almost equal to articles 1 and 2, about 5 times as long as wide.



**Fig. 2.** *Liljeborgia associata* sp. nov., female (LEMMI) from Vostok Bay of the Sea of Japan. **a.** Upper lip. **b.** Lower lip. **c.** Head. **d.** Antenna 1. **e.** Antenna 2. **f.** Mandible. **g.** Maxilla 1. **h.** Maxilla 2. **i.** Maxilliped. **j.** Pleopod 1. **k.** Telson.



MAXILLA 1 (Fig. 2g). Outer plate with 8 large slender spines, ventrally denticulated; inner plate with a single long plumose seta; palp consists of 4 articles, article 1 shovel-shaped, with broad median part, about 1.5–2 times as long as wide, with 8–9 sharp robust spines along anterior margin, and simple small setae along dorsal margin.

MAXILLA 2 (Fig. 2h). Inner and outer plates distally rounded, robust, covered with numerous long simple setae along anterior and lateral margins.

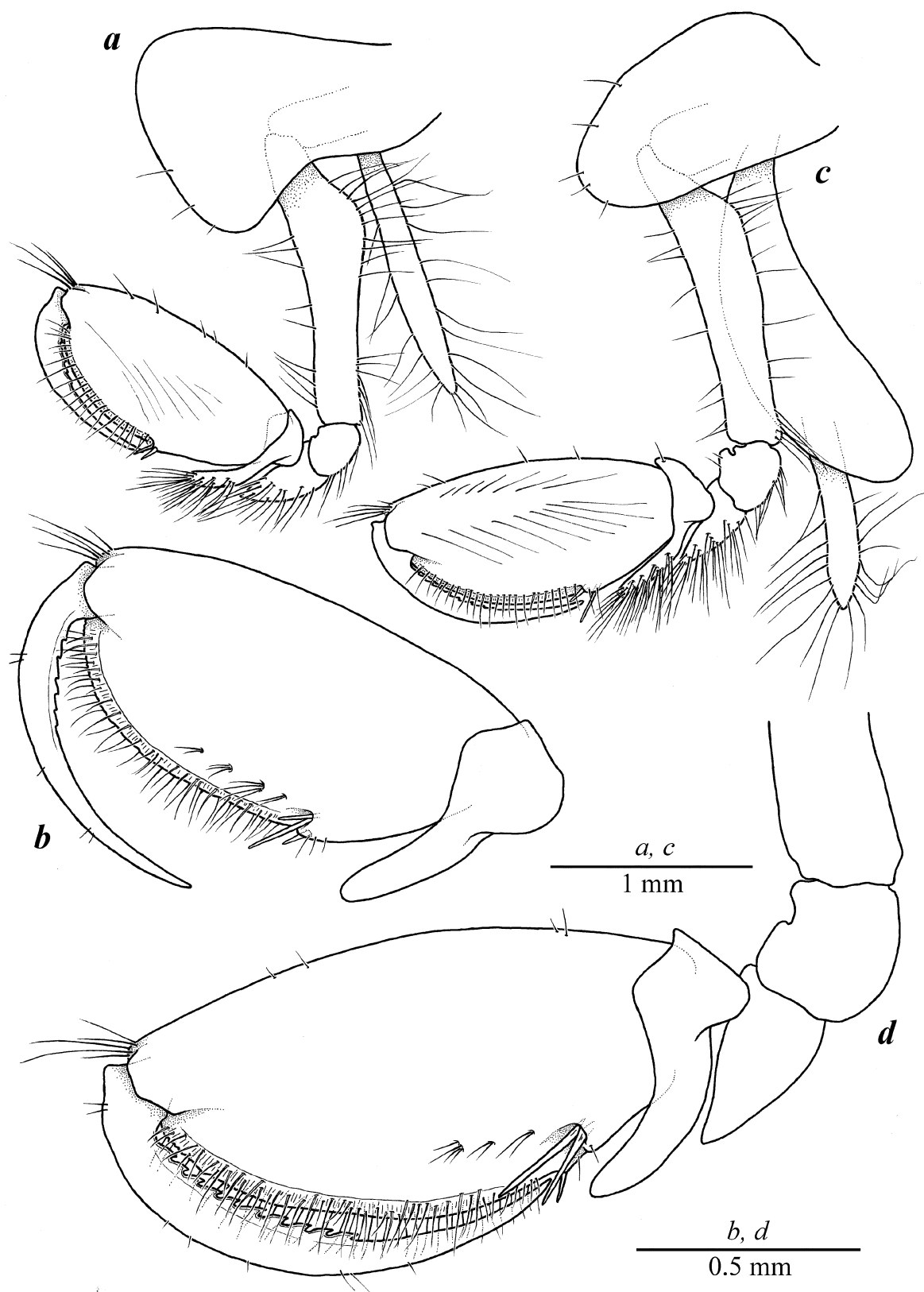
MAXILLIPED (Fig. 2i). Palp consists of 4 articles: article 1 of palp unarmed, article 2 with a cluster of long setae distodorsally, small robust setae distally and long simple setae along the inner margin, outer margin unarmed; article 3 with a cluster of long simple setae along anterior border and distal part of inner margin, outer margin unarmed, article 4 (dactylus) slender, curved, about 0.8 times as long as article 3, unarmed; outer plate with 13–15 robust spines along medial border (distal spines are narrow and rather long); inner plate covered with small setae along anterior margin.

GNATHOPOD 1 (Fig. 3a). Coxa trapezoidal, with anterior medial setae only, posterior border weakly concave; basis slender, about 6–7 times as long as wide, with ventral projection in proximal part, with long simple setae; ischium about as long as wide, with long simple setae along distoventral border; merus about 1.5 times as long as wide, sharpening distoventrally, with several groups of setae along ventral margin; carpus slender, with blunt distoventral process, reaching  $\frac{1}{3}$  of palm length, not reaching propodal group of strong spines, armed with several groups of setae; propodus about twice as long as wide both in male and females (Fig. 3b), with convex ventral margin, armed with small simple setae along almost all its length, with a depression and 3 long simple setae in proximal part level with end of dactylus, dorsal margin straight and unarmed; dactylus with 5–7 triangular teeth.

GNATHOPOD 2 (Fig. 3c). Very similar to *Gn1* in shape and slightly larger in size; coxa triangulo-elliptic; basis slender, about 6 times as long as wide, with long simple setae; ischium about as long as wide, with long simple setae along distoventral border; merus about 1.5 times as long as wide, sharpening distoventrally, with several groups of setae along ventral margin; carpus slender, with blunt distoventral process, reaching  $\frac{1}{4}$  of palm length, not quite reaching propodal group of strong spines, armed with several groups of setae; propodus about 2 times as long as wide in both male (Fig. 5a) and females (Fig. 3d), dorsal margin straight and unarmed, with convex ventral margin, armed with small simple setae along almost entire length, with a depression and 3 long simple setae in proximal part level with end of dactylus; dactylus with 10–11 triangular teeth.

PEREIOPOD 3 (Fig. 4a). Coxa elliptic and narrow; basis slender, about 8–9 times as long as wide, with straight margins, covered with long simple setae; ischium about as long as wide, with long simple setae along distoventral border; merus broad, about 3 times as long as wide, slightly longer than carpus and equal to propodus, dorsal margin produced forward, with long simple setae along dorsal and ventral margins; carpus about 3 times as long as wide, with straight margins, unarmed dorsally and with simple long setae along ventral margin; propodus about 4–4.5 times as long as wide, slightly curved, unarmed dorsally and ventrally, with several long simple setae at distodorsal angle; dactylus of normal length, slender, weakly curved, slightly shorter than propodus and equal to carpus in length.

PEREIOPOD 4 (Fig. 4b). Coxa wide, with parallel anterior and posterior border, ventral border with 3 well-marked serrations; basis slender, about 7–7.5 times as long as wide, with straight margins, covered with long simple setae; ischium about as long as wide, with long simple setae along distoventral border; merus broad, about 3 times as long as wide, longer than carpus and slightly shorter than propodus, dorsal margin produced forward, with long simple setae along dorsal and ventral margins; carpus about 2.5–3 times as long as wide, with straight margins, unarmed dorsally and with long simple setae along ventral margin; propodus about 5–5.5 times as long as wide, slightly curved, unarmed dorsally, with several



**Fig. 3.** *Liljeborgia associata* sp. nov., female (LEMMI) from Vostok Bay of the Sea of Japan. a. Gnathopod 1. b. Chela of Gn1. c. Gnathopod 2. d. Chela of Gn2.



long simple setae at distodorsal angle, with 5 tiny spines along ventral margin; dactylus of normal length, slender, weakly curved, slightly shorter than propodus and slightly longer than carpus.

PEREIOPOD 5 (Fig. 4c). Coxa subquadrate, with medially concave ventral margin, unarmed; basis broad, with anterior and posterior border convex; anterior border with 12–13 small spines accompanied by small setae, posterior border with well-marked serration; ischium about as long as wide, with large simple spine on anterodistal corner; merus about 4 times as long as wide, with 4 short anterior spines, 1 large distodorsal spine paired with a spinule, and 1 simple distoventral spine; carpus about 4 times as long as wide, slightly shorter than merus, unarmed posteriorly, with 3 small anterior spines, and with a distal pair of spines anteriorly and posteriorly; propodus (Fig. 4d) about 6 times as long as proximal width, slightly tapering distally, with 10 anterior spines and series of long medial setae; dactylus distinctly curved and of normal stoutness, with tip entire, about  $\frac{1}{2}$  of the length of propodus.

PEREIOPOD 6 (Fig. 4e). Coxa subquadrate, with medially concave ventral margin, unarmed; basis broad, with anterior and posterior border convex; anterior border with 10 small spines accompanied with small setae, posterior border with well-marked serration; ischium about as long as wide, with large simple spine on anterodistal corner; merus about 4 times as long as wide, with 5 short anterior spines, a pair of large distodorsal spines and 1 simple distoventral spine paired with a spinule; carpus about 4.5–5 times as long as wide, slightly shorter than merus, unarmed posteriorly, with 4 small anterior spines, and a distal pair of spines anteriorly and posteriorly; propodus (Fig. 4f) about 7 times as long as proximal width, tapering distally, with 4 long anterior spines and series of long medial setae; dactylus distinctly curved, sharp, about  $\frac{1}{3}$  of the length of propodus.

PEREIOPOD 7 (Fig. 4g). Coxa almost rectangular; basis greatly broad, almost round, with convex anterior border and strongly convex posterior border; anterior border with 12 small conical spines, posterior border serrated, distal border with strong long spine; ischium about as long as wide, unarmed; merus broad, about 2.5–3 times as long as wide, with 4 anterior small spines and pair of anterodistal simple spines, 3 posterior spines and a pair of long posterodistal long spines; carpus about 5.5 times as long as wide, slightly longer than merus, with single or paired anterior spines and long simple setae posteriorly; propodus about 10 times as long as wide, tapering distally, with 4 small anterior spines and long setae posteriorly; dactylus straight, very long and slender, entire, about  $\frac{1}{3}$  of the length of propodus.

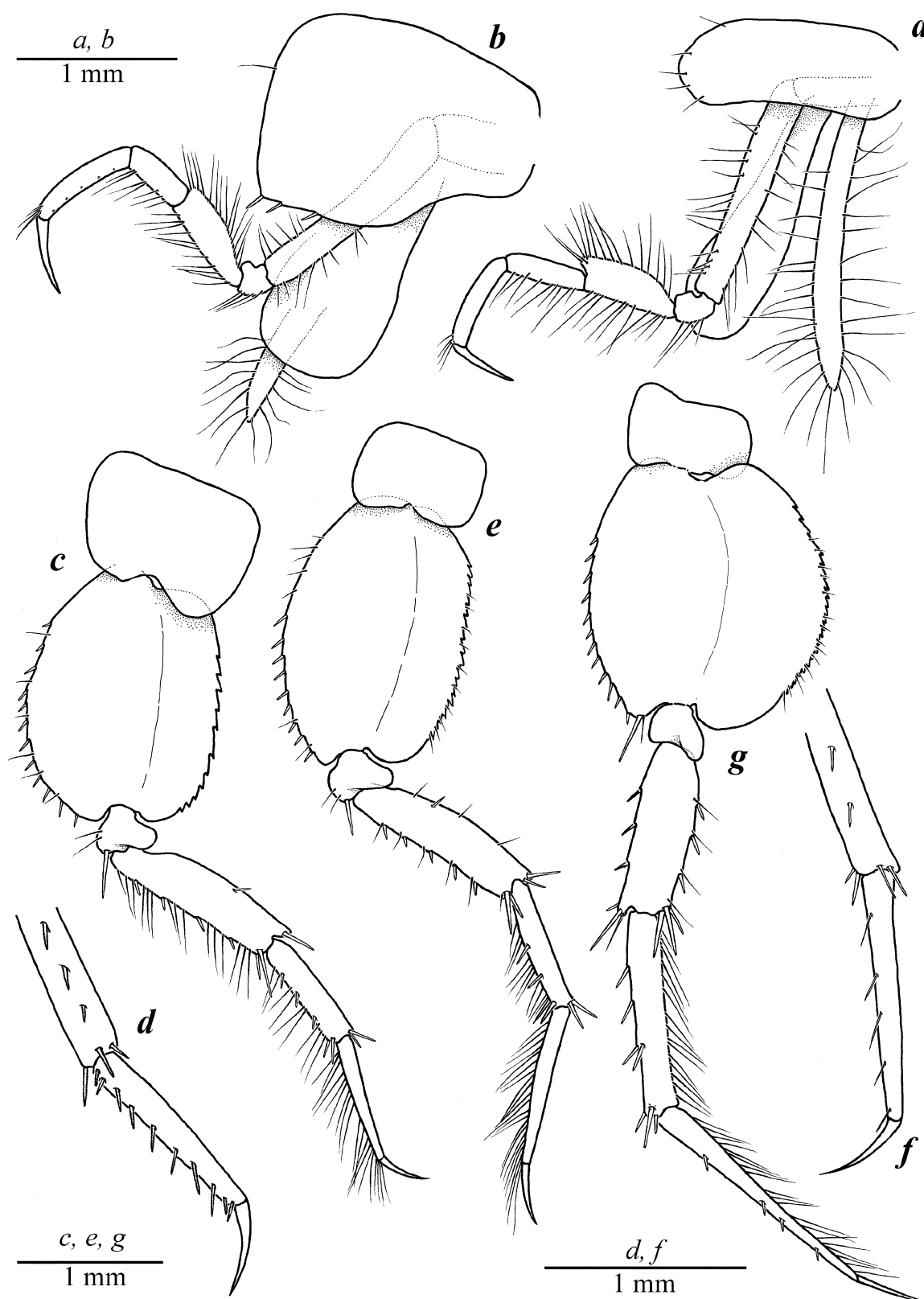
PLEONITE 1 (Fig. 5b). Posterodorsal area produced into 3 small teeth of which median is longest (Fig. 5e); *Ep1* with normally developed posteroventral tooth, with posterior border weakly convex, covered with numerous simple setae.

PLEONITE 2 (Fig. 5b). Posterodorsal area produced into 3 small teeth of which median is longest (Fig. 5e); *Ep2* with normally developed posteroventral triangular tooth, with posterior border distinctly convex, covered with numerous simple setae.

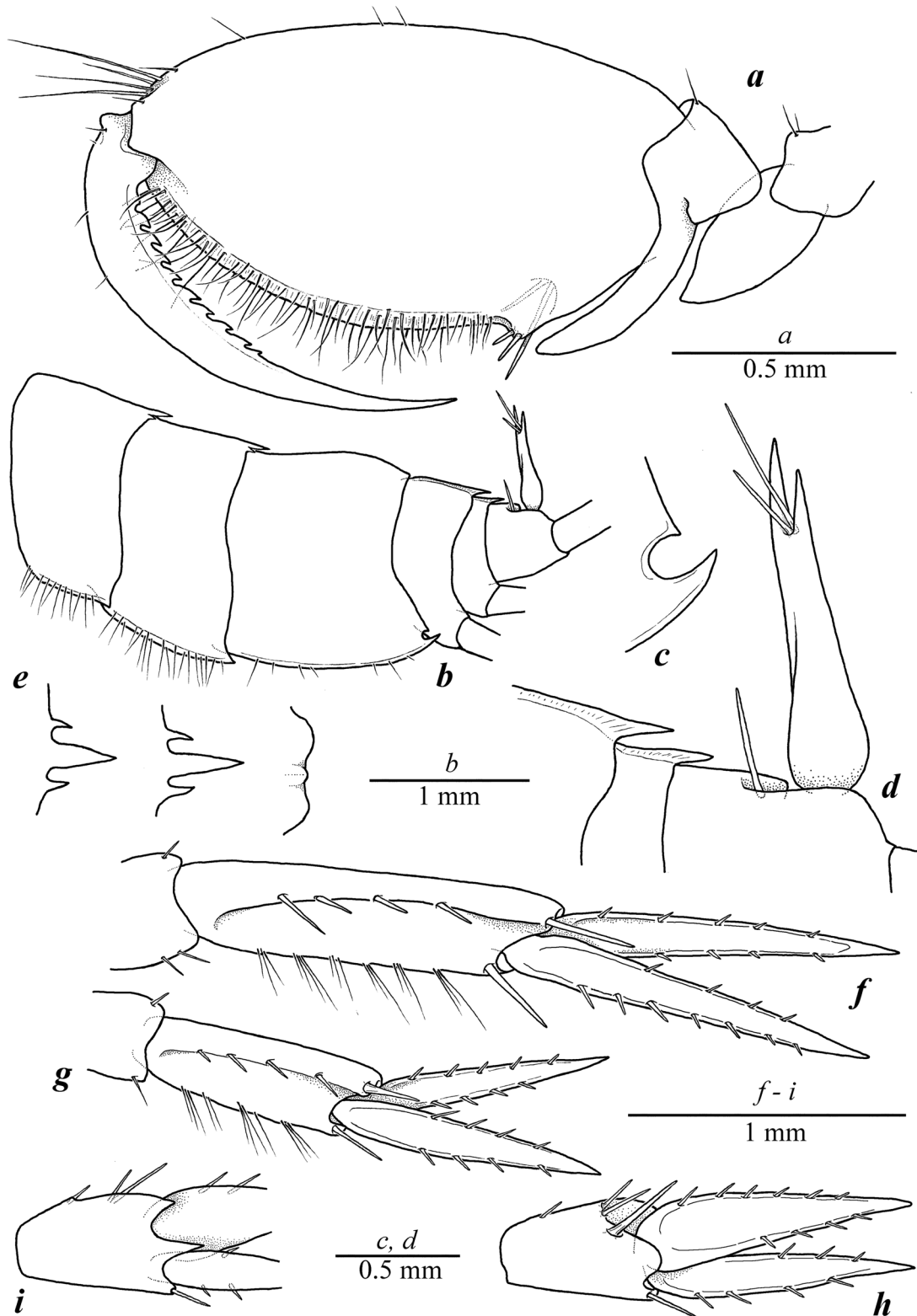
PLEONITE 3 (Fig. 5b). Posterodorsal area toothless (Fig. 5e); *Ep3* with small posteroventral tooth and distinct rounded notch (Fig. 5c), with posterior border convex.

UROSOMITE 1. With well-developed dorsal lamina and posterodorsal tooth (Fig. 5d); peduncle of *U1* with medial distal corner rounded, with 4 dorsolateral spines of which proximal is longest, with 1 dorsomedial spine and 1 ventromedial spine; outer ramus with 5 small outer spines and 4 small medial spines; inner ramus with 4 small outer spines and 7 small medial spines.

UROSOMITE 2. With well-developed dorsal lamina and posterodorsal tooth (Fig. 5d); peduncle of *U2* with 4 slender dorsolateral distal spines, with 1 dorsomedial spine and 1 ventromedial spine; outer ramus



**Fig. 4.** *Liljeborgia associata* sp. nov., female (LEMMI) from Vostok Bay of the Sea of Japan. **a.** Pereopod 3. **b.** Pereopod 4. **c.** Pereopod 5. **d.** Distal segments of *P5*. **e.** Pereopod 6. **f.** Distal segments of *P6*. **g.** Pereopod 7.



**Fig. 5.** *Liljeborgia associata* sp. nov., male (a) (LEMMI) and female (b-i) (LEMMI) from Vostok Bay of the Sea of Japan. **a.** Chela of *Gn2*. **b.** Pleonites 1-3 and urosomites 1-3. **c.** Ventroproximal margin of pleonite 3. **d.** Urosomites 1-3 and telson. **e.** Dorsoproximal margins of pleonites 1-3. **f.** Uropod 1. **g.** Uropod 2. **h.** Uropod 3. **i.** Peduncle of uropod 1, outer view.

with 5 small outer spines and 4 small medial spines; inner ramus with 4 small outer spines and 4 small medial spines.

UROSOMITE 3. With sharp medial projection (Fig. 5i), with 1–2 lateral spines, 1 long posterolateral tooth and 3 posterodorsal spines; outer ramus with 3–4 small outer spines and 4 small medial spines; inner ramus with 6 small outer spines and 4 small medial spines (Fig. 5h).

TELSON (Fig. 2k). With cleft reaching 0.85–0.9 of telson length; distal teeth about  $\frac{1}{4}$  of the length of telson, medial teeth longer than outer teeth, 2 interdental spines long and slender, overreaching outer teeth.

COLORATION. General coloration of body and all appendages translucent white with large light and dark brown spots; some segments of appendages dark red or with dark red spots; eyes brightly white (Fig. 1).

BODY SIZE. Largest collected female = bl 6.5 mm; largest collected male mostly similar in size to largest female, with bl 6.5 mm.

### Taxonomic remarks

The new species is distinctly morphologically similar to *L. geminata* and *L. serratoides*. At the same time, the new species can be clearly distinguished by several morphological features not known from the latter species.

From *L. geminata* (see description, presented by Barnard (1969)), the new species can be distinguished by the following characters: smaller dorsal crest on urosomites 1 and 2 (Fig. 5d); *Mx1* with wide, shovel-shaped article 3 of palp (Fig. 2h); shorter and wider merus of *P1* and *P2* (see Fig. 4a–b); slender propodal segments of *PP5*–*7* (see Fig. 4c–g); absence of long proximal seta of peduncle of *U1* (see Fig. 5f); shape of medial teeth of telson (Fig. 2k), which are significantly longer and accompanied by 2 long interdental spines. At the same time, *L. geminata* seems to be more closely related to the new species by its very similar body coloration, the presence of a small dorsal crest on urosomites 1 and 2, short articles of peduncle of *A1*, relatively stout distal segments of *PP3*–*4* (especially merus), and other morphological features that clearly separate both species from their relative *L. serratoides*.

From *L. serratoides* (after Tzvetkova 1967), the new species is easily separated by the following characters: presence of a dorsal crest on urosomites 1 and 2; stout articles of peduncle of *A1* and *A2* (Fig. 2d–e); wide distal article of palp of *Mx1*; stout and wider distal segments, especially merus, of *PP1*–*2*; slender propodal segment of *PP5*–*7*; length and shape of medial teeth of telson, which are significantly longer and accompanied by 2 long interdental spines. In addition, relatively stout and wide segments of *PP3*–*4* are distinguishing features that separate the aforementioned species from tropical relatives, such as *L. japonica* and *L. serrata* (e.g., Nagata 1965; Azman & Othman 2013).

From *L. japonica* and *L. serrata*, known from the Seto Inland Sea in southern Japan (see Nagata 1965), the new species can be distinguished by longer rostrum, mandibular palp with longer and slender articles, especially distal one; shorter and stouter articles of peduncle *A1* and *A2*; stouter distal segments of *PP3*–*4*, especially stout and wide merus of *P3*; significantly shortened dactyli of *PP5*–*7*; slender rami of *U3* and longer median teeth of telson.

From *Liljeborgia hwanghaensis* Kim & Kim, 1990, known from the Korean coasts of the Yellow Sea (37°23' N, 126°35' E) (see Kim & Kim 1990), the new species can be clearly separated by the posterodorsal armature of pleonites 1 and 2 (3 teeth in the new species vs 5 teeth in *L. hwanghaensis*) and shorter median teeth of telson.

### Habitat and ecology

Specimens of *Liljeborgia associata* sp. nov. were collected inside the burrows of *Urechis unicinctus* (Drasche, 1880) (Polychaeta: Echiura: Urechidae), representing the first case of an association between liljeborgiid amphipods and spoon worms (Echiura). The spoon worm *U. unicinctus* lives in large U-shaped burrows constructed in muddy and sandy sediments in the inter- and subtidal zones in the Sea of Japan and the Yellow Sea (Abe *et al.* 2014; pers. obs.). About 20% of the studied spoon worms burrows were inhabited by a single individual of the new species, which may indicate aggressive territorial behavior.

### Distribution

The specimens of *Liljeborgia associata* sp. nov. were found in the southern part of the Peter the Great Bay and the northern part of Posjeta Bay in the Sea of Japan (Russian Federation) (see Fig. 1), where the present study was accomplished. The distribution of the new species is probably related with the distribution range of its host, *U. unicinctus* (see Abe *et al.* 2014).

### Key to the species of the family Liljeborgiidae from the boreal waters of the NW Pacific (with species of *Liljeborgia* from the NE Pacific)

1. *Gn1* significantly larger than *Gn2*; *U3* large, paddle-like; postero-dorsal margin of pleonites 2–3 armed with marked teeth; eyes absent. NW Pacific: Sakhalin, the Sea of Okhotsk, depth 109–309 m ..... *Sextonia caecus* Labay, 2017
- *Gn1* similar to *Gn2*; *U3* smaller than *U1* and *U2*; postero-dorsal margin of pleonites 2–3 unarmed; eyes present (except *L. cota*) ..... 2
2. Carpus of *G1* and *G2* with strongly produced slender ventral lobe extending along hind margin of propodus ..... 3 (*Liljeborgia* Spence Bate, 1862)
- Carpus of *G1* and *G2* lacking produced ventral lobe. Known from the NE Pacific only ..... *Idunella* G.O. Sars, 1894 (= *Listriella* J.L. Barnard, 1959)
3. Posterodorsal area of pleonites 1 and 2 produced into 5 teeth of which the median one is the longest. Known from the Korean coasts of the Yellow Sea .... *Liljeborgia hwanghaensis* Kim & Kim, 1990
- Posterodorsal area of pleonites 1 and 2 produced into 3 teeth of which the median one is the longest ..... 4
4. Telson cleft only  $\frac{1}{4}$  to  $\frac{1}{3}$ , lacking terminal spines on lobes of telson; basis of *PP5*–*7* more than twice as long as wide; blind. NE Pacific: from the Gulf of Alaska to the northern Baja California, depth 366–2000 m ..... *Liljeborgia cota* Barnard, 1962
- Telson cleft nearly to base, lobes with imbedded terminal spine; basis of *PP5*–*7* about 1–1.5 times as long as wide; with eyes ..... 5
5. Epimeral plate 1 concave above postero-ventral tooth. From NE Atlantic to the coasts of Central California, depth 40–611 m ..... *Liljeborgia pallida* Bate, 1857
- Epimeral plate 1 convex above postero-ventral tooth ..... 6
6. Cusps of lobes of the telson longer medially than laterally; eyes reniform. NE Pacific: from Bahia de Los Angeles, Gulf of California, depth 46 m ..... *Liljeborgia marcinabrio* Barnard, 1979
- Cusps of lobes of the telson subequal longer laterally than medially; eyes oval to subquadrate ..... 7
7. Urosomites 1–2 with well-marked dorsal crest, peduncle of *A1* with short and stout articles (article 3 about 1.5 times as long as wide; article 2 about as long as wide) ..... 8

- Urosomites 1–2 without dorsal crest, peduncle of *A1* with slender articles (article 3 about 2–2.5 times as long as wide; article 2 about 1.5 times as long as wide). NW Pacific: Posjeta Bay in the Sea of Japan, intertidal ..... *Liljeborgia serratooides* Tzvetkova, 1967
- 8. Medial teeth of telson about 0.15 length of telson, less than 1.5 times longer than outer teeth and accompanied by 1 long interdental spine. NE Pacific: from Goleta to the northern Baja California, depth 3–70 m ..... *Liljeborgia geminata* Barnard, 1969
- Medial teeth of telson about 0.25 length of telson, about 2 times longer than outer teeth and accompanied by 2 long interdental spines. NW Pacific: the Peter the Great Bay and the northern part of Posjeta Bay in the Sea of Japan, intertidal ..... *Liljeborgia associata* sp. nov.

Some other species of the genus *Liljeborgia* are known from the region and the Sea of Japan (e.g., Ishimaru 1994; Ren 2007). For example, Ishimaru (1994) notes *Liljeborgia aequabilis* Stebbing, 1888 from the tropical part of the Seto Inland Sea influenced by the Kuroshio Current. This species is actually recognized in Australia, New Zealand and the Sulu Sea as a common associate of hermit crabs (Stebbing 1888; after Hurley 1954; see also Vader 1995) and by most morphological features, especially long and slender segments of *PP3–4* and the form of telson (see above), belong to the tropical representatives of the genus, such as *L. japonica* and *L. serrata*, or possibly represent an undescribed species. These species are known from warm (tropical) waters and probably are not present in the boreal zone of the northern Pacific Ocean.

## Discussion

The symbiotic communities associated with deep-burrowing invertebrates are very diverse and still include many undescribed species. Burrows of spoon worms of the genus *Urechis* as well as almost all large echiuroid worms are inhabited by various symbiotic animals, such as shrimps, crabs, bivalves and even fishes (e.g., Fisher & MacGinitie 1928; Anker *et al.* 2005, 2015; Itani *et al.* 2005; Goto & Kato 2012; Marin 2014; Goto *et al.* 2017). The symbiotic fauna associated with *U. unicinctus* in the Peter the Great Bay has not yet been fully studied, whereas the discovered symbiotic assemblage includes unidentified acoela turbellarian worms (Platyhelminthes: Rhabditophora: Polycladida), crabs *Pinnixa rathbuni* Sakai, 1934 (Crustacea: Decapoda: Pinnotheridae) and *Sestrostoma balssi* (Shen, 1932) (Crustacea: Decapoda: Varunidae), copepod *Goidelia* cf. *japonica* Embleton, 1901 (Crustacea: Copepoda: Poecilostomatoida: Catiniidae) and goby *Gymnogobius heptacanthus* (Hilgendorf, 1879) (Pisces: Gobiidae) (pers. obs.). *Liljeborgia associata* sp. nov. is probably only associated with the burrows of the spoon worms, since no free-living specimens of the genus *Liljeborgia* or any other liljeborgiidae amphipods were sampled on the surface of muddy or sand-bottom substrates in the Peter the Great Bay and Posjeta Bay in the Sea of Japan during the present or previous studies (e.g., Gurjanova 1951; Tzvetkova 1967). Moreover, Tzvetkova (1967) described a quite common *L. serratooides* from mussel communities, overgrowing rocks and hard substrates in Posjeta Bay of the Sea of Japan, while *Liljeborgia* species were not recorded on muddy and soft-bottom substrates that dominate the bay. This fact indirectly proves that *Liljeborgia associata* sp. nov. rarely emerges from the host's burrows and does not inhabit the surrounding soft-bottom substrates as a free-living species.

At the same time, representatives of the family Liljeborgiidae have been observed in association with other marine invertebrates. Often *Liljeborgia* spp. are observed in association with hermit crabs (Taylor 1979; Vader 1995). *Liljeborgia aequabilis* Stebbing, 1888, associated with the hermit crab *Dardanus arrosor* (Herbst, 1796) (Crustacea: Decapoda: Diogenidae), has a specific colorful body coloration (e.g., Vader & Tandberg 2015: fig. 4; Taylor 1979: fig. 1), similar to other symbiotic species of the family (Barnard 1959; Mills 1962: fig. 2), while none of the free-living species are known to have bright pigmentation in life (e.g., d'Udekem d'Acoz & Vader 2009; d'Udekem d'Acoz 2010; Cadien 2015). *Liljeborgia psaltrica* Krapp-Schickel, 1975 is recorded among associates of the orange coral

*Astroides calycularis* (Pallas, 1766) (Cnidaria: Scleractinia: Dendrophylliidae) in the Mediterranean (Terrón-Siglera *et al.* 2016). *Idunella clymenellae* (Mills, 1962) was found and described inside the tube arrays of the polychaete worm *Clymenella torquata* (Leidy, 1855) (Polychaeta: Maldanidae) in the intertidal flats of Barnstable Harbor, Massachusetts (Mills 1962; Sanders *et al.* 1962). These amphipods inhabit about 20–25% of tubes with live polychaetes (Mills 1962) and show a positive response to polychaetes mucus (Batcheler & Mills 1965). Also, *Idunella albina* (J.L. Barnard, 1959) was commonly collected from dense beds of echiuroid worms off Californian coasts (Barnard 1959), however, the direct relationship of this species with spoon worms is not shown. An undescribed species of the genus *Idunella* is reported from the burrows of the mud shrimp *Upogebia affinis* (Say, 1818) (Crustacea: Decapoda: Upogebiidae) (Fox & Bynum 1975), while individuals of the genus *Idunella* (= *Listriella*) are common inhabitants of maldanid and terebellid polychaete tubes (Barnard 1969; Bousfield 1973) and are reported with holothurians (Fox & Bynum 1975; Vader 1978). It is probable that associations of these animals with marine invertebrates are much more common than is currently known.

As mentioned above, according to morphological data, the new species is closely related to *L. geminata*. Unfortunately, minimal genetic data on the family Liljeborgiidae are presented in GenBank (NCBI) database, and genetic data are not available for the relative congeners of the described species. At the same time, it can be assumed that *Liljeborgia associata* sp. nov. and *L. geminata* represent a pair of closely related vicariant taxa known as boreal amphipacific species. Such species had a wide distribution area in the northern part of the Pacific Ocean during the Pliocene–Middle Pleistocene period, characterized by a higher water temperature and sea level, which allowed the boreal fauna to move from east to west and even through the Arctic basin [as Great Trans-Arctic Biotic Interchange (GTAI)] (Einarsson *et al.* 1967; Durham & MacNeil 1967; Briggs 2003). During the temperature decreasing in the Sea of Okhotsk and the Bering Sea in the Late Pleistocene (about 2 Ma ago), the ranges were subsequently divided into the western and the eastern distribution ranges (Zenkevitch 1963; Briggs 2003; Maggs *et al.* 2008; Marin 2018a). Further genetic data will allow a more detailed description of the phylogenetic relationships within the representatives of the family from the northern part of the Pacific Ocean.

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