Zootaxa 4472 (1): 153–164
http://www.mapress.com/j/zt/

Article

ISSN 1175-5326 (print edition)
ZOOTAXA
ISSN 1175-5334 (online edition)

https://doi.org/10.11646/zootaxa.4472.1.8
http://zoobank.org/urn:lsid:zoobank.org:pub:8F2729F3-3C54-425E-B164-332DF2F8A66B

A new species of *Nemoura* Latreille (Plecoptera: Nemouridae) from Amur River Basin (South of the Russian Far East)

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Abstract

Nemoura sirotskii **sp. n**. (Plecoptera, Nemouridae) is described as a new stonefly species from the tributary streams of Zeya Reservoir (Amur River Basin) in the south of the Russian Far East. Detailed descriptions and illustrations are provided for the larvae and adult specimens. The diagnostic characters distinguishing it from sympatric species *N. arctica* are discussed.

Key words: Plecoptera, *Nemoura*, epiproct, larvae, Zeya River, Amur River, Far East, Russia

Introduction

Nemoura Latreille, 1796 is the most species-rich genus in the subfamily Nemourinae with more than 200 species recognized from the Holarctic and Oriental regions (DeWalt *et al.* 2018). Approximately half of the known *Nemoura* species occur in the Eastern Palaearctic. From the south of the Russian Far East (RFE), 21 species of *Nemoura* have been reported (Teslenko 2015, 2016). This number includes three species which were added to the stonefly fauna of Russia through the study of the Korean Plecoptera (Zwick 2010, Zwick & Baumann 2011). Most of the *Nemoura* species of the RFE have an East-Asian pattern of distribution; they also occur in neighbouring countries of China, Korea, and Japan. Two species, *N. arctica* Esben-Petersen 1910 and *N. sahlbergi* Morton, 1896, have a Holarctic distribution.

The Nearctic species of *Nemoura* was recently reviewed by Grubbs *et al*. (2018). They conclude that only four species occur in North America, including the Holarctic species *N. sahlbergi* and *N. arctica*. Grubbs *et al*. (2018) proposed the Nearctic *N. trispinosa* Claassen, 1923 as a subjective junior synonym of *N. arctica.*

As a contribution to the knowledge of the East Asian and Arctic *Nemoura* fauna, our paper describes and illustrates the adults and larvae of a new species, *N. sirotskii* **sp. n.**, which is morphologically similar to *N. arctica*. *Nemoura sirotskii* **sp. n.** was collected from foothill tributary streams of Zeya Reservoir (Amur River Basin) in the RFE.

Material and methods

Adults were collected with a sweep net. Larvae were sampled either by hand net or by picking specimens directly from the substrate with entomological forceps. All specimens were preserved in 75% ethanol. Abdomens of adults or larvae were removed and soaked in 10% NaOH overnight and rinsed with distilled water. Specimens were examined with the aid of a compound microscope in transmitted light. Association of larvae with adults was secured by extruding the epiproct from pre-emergent male larvae. The morphological terminology follows Baumann (1975) and Zwick (2004). Illustrations were produced using digital cameras (Nikon Coolpix 995 and Toup View 3.7) and Merlin scanning electron microscope in the Instrumental Centre of Biotechnology and Gene

Accepted by B. Kondratieff: 30 Jul. 2018; published: 7 Sept. 2018

Engineering of the Federal Scientific Center of the East Asia Terrestrial Biodiversity, Far Eastern Branch, Russian Academy of Sciences, Vladivostok, Russia (FSC EATB FEB RAS). The holotype and all paratypes are deposited in the collection of the FSC EATB FEB RAS.

Results and discussion

***Nemoura sirotskii* Teslenko, sp. n.**
(Figs. 1−22)

Material examined. South of the Russian Far East, Zeya R. Basin, Upper Amur River Basin: Holotype male: Amurskaya Oblast, Izhak River, Urkan River Basin, Zeya Reservoir Basin, N 53°58.409 E 128°57.391, 22.06.2004 (Fig. 24), coll. V. Teslenko (FSC EATB FEB RAS). Paratypes: 2 males, 1 female, Shirokovskaya R., Zeya Reservoir Basin, Zeya Nature Reserve, N 53°53.054 E 127°14.360, 23.06.2004 (Fig. 25), coll. V. Teslenko; 1 male (mounted), 1 female, 15 larvae, the same data and location as holotype, coll. V. Teslenko; 2 females (mounted), the same data and location as paratypes, coll. V. Teslenko; 1 male (mounted), Sukhodol stream, under the road bridge, Gulik R. Basin, Zeya R. Basin, N 53°58.179 E 127°04.491, 20.06.2004, coll. V. Teslenko; 1 male (mounted), the Jewish Autonomous Region, Khingan River, Amur R. Basin, 2 km from Obluchye sett. towards Khingansk sett., N 49°03.027 E 131°03.256, 26.06.2004, coll. V. Teslenko; 2 males, Khabarovsk Region, Okhota R., 7 km from the mouth, 25.06.1999, coll. T. Tiunova.

Description. Adult habitus: Body length of males 7.5–7.6 mm (n=6), females 8.4 mm (n=5). Macropterous, wings subhyaline, pale, veins brownish; forewing length of males 8.2 mm, females 8.4 mm. General body colour brown with pale markings dorsally (Fig. 1). Antenna and scape pale. Head brown, interocellar area and occiput medially with an indistinct light brown patch, occiput behind compound eyes dark brown. Ocelli pale. Pronotum pale with indistinct brownish X-shaped pattern, rugosities indistinct, lateral margins pale (Fig. 1). Legs pale. Mesonotum and metanotum dark brown with pale patches laterally, prescutum and scutellum pale.

Male. Subgenital plate of sternum 9 moderate in size, typical of the genus, with arcuate sides, narrowed distally to a pointed tongue-shaped tip. Vesicle broad apically, width equal to ½ width of subgenital plate. Tip of the subgenital plate lies between narrow finger-like inner paraproct lobes (Fig. 2). Outer paraproct lobe broad at the base with notch on the posterior margin, and two rounded posterolateral projections: the inner projection strongly sclerotized (Fig. 2). Cerci almost straight, plump, slightly prolonged, extended inward with a roundly swollen membranous apex covered with hairs; sclerotized laterally and terminating in a pair of appressed spines, outer spine absent (Figs. 2, 3); the vestigial terminal segment modified into an 'eye-spot' mark (Figs. 2, 3). Terga 1−2 and 9 mostly pale, terga 3–8 brownish with pale band widened to posterior margin of each segment (Fig. 1). Tergum 10 brown, medially with a pair of round shallow concavities below the epiproct; a few short setae and hairs scattered anteriorly and on an elevation encircling the concavities. Epiproct in dorsal view oval slightly widening in anterior half, becoming narrow basolaterally (Figs. 4, 5). Paired lateral arms of the dorsal sclerite directed obliquely down towards the middle, nearly touching, and sclerotized; the paired dorsal folds deep and covered with comb-like scales (Figs. 4, 19). Tip of the epiproct bilaterally symmetrical; each apical sclerite elongated, flat-lying, base extends beyond dorsal folds; directed oblique downward, slightly widened at the apex; the edge of distal apex rounded, rough, bears fine comb-like scales and 3–4 stout lateral spines (Figs. 4, 19). Both apical sclerites closely adhering to the ring sclerites and positioned ca. parallel to ring sclerites (Figs. 4, 5, 19). In ventral view, the ventral sclerite with straight margins, bears spines arranged in two longitudinal rows, about 16 spines in each row (Fig. 5).

Female. Sternum 7 extended medially forming a well-developed, broadly rounded and swollen pregenital plate, which is medium sclerotized, overlapping sternum 8 completely and anterior margin of sternum 9 partly (Fig. 6). In cleared slide-mounted genitalia, the subgenital plate and vaginal lobes on sternum 8 difficult to distinguish, hidden under the wall of the pregenital plate (Fig. 7). Vaginal pouch large, bell-shaped; anteriorly with paired triangular cross-elongated collar-like shields which surrounded by a pair of weakly sclerotized finger-shaped vaginal sclerites with beak-shaped apexes that are directed outward. The base of the finger-shaped each vaginal sclerite attached to rounded roughened pockets (Fig. 7). Sternum 9 well sclerotized, swollen anteromedially, the swelling flanked by pair of narrow dark patches (Fig. 6).

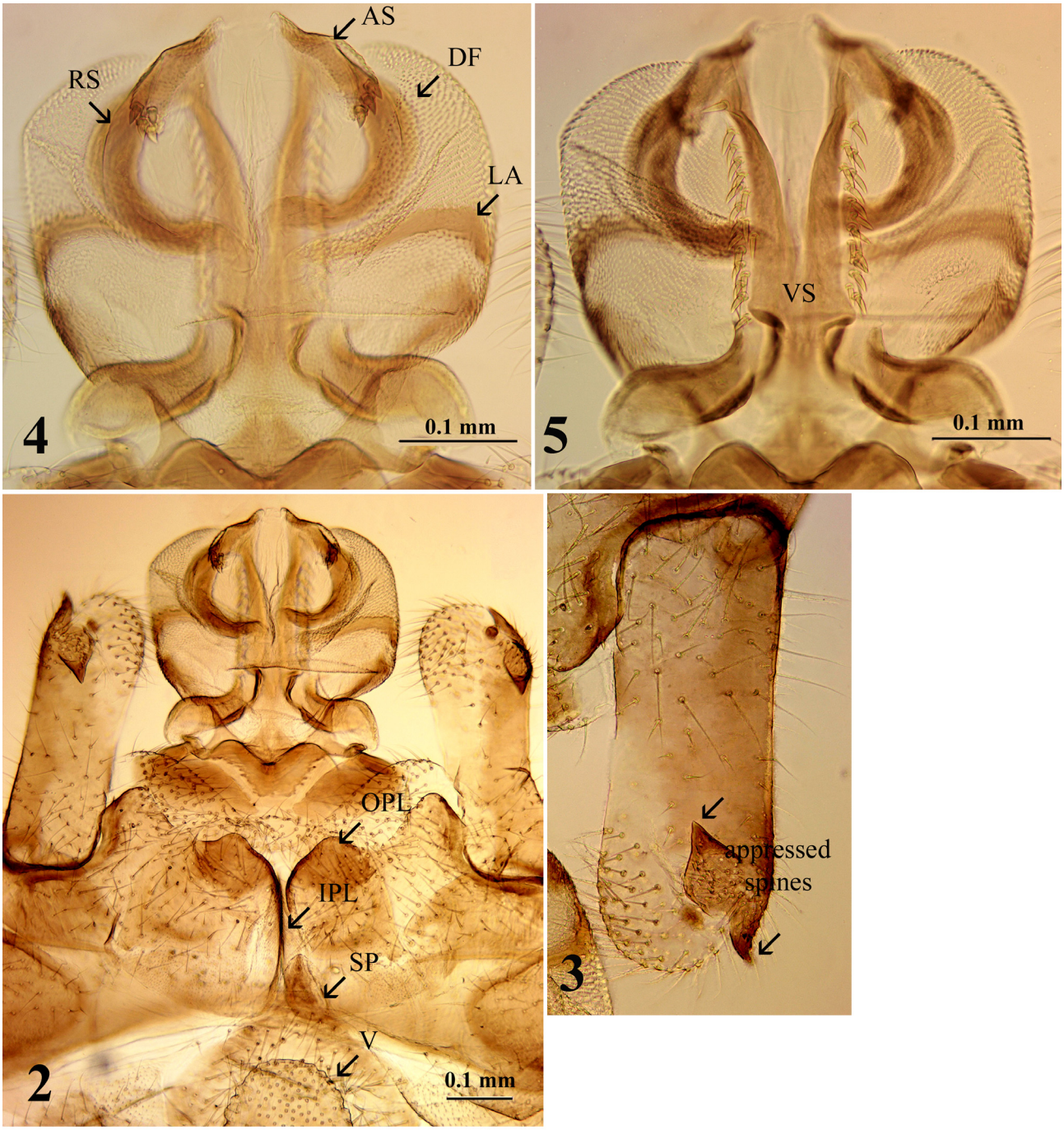
Mature larvae. Body length of males 6.5–8.5 mm (n=4), females 8.4–9.5 mm (n=5). General color brown (Fig. 9). Head with brown tentorial callosities, epicranial suture pale, a small light brown patch below epicranial

suture to the inside of each compound eye (Figs. 8, 9), occiput dark brown. Antenna, legs and cerci pale, scape light brown (Fig. 9). Pronotum approximately 1.6x wider than long, pattern indistinct, narrow dark brown band close to anterior pronotal margin mesally merging with X-shaped figure (Figs. 8, 9). Mesonotum and metanotum brown with diffuse pale patches. Fore femur 4.1x longer than wide (Fig. 13), hind femur 4.4x longer than wide. Abdomen relatively slender, integument light, matte in appearance, terga brown, a mesal row of darkish patches forming a longitudinal band on segments 1−7 (Figs. 8, 9). Cerci with 33 segments, length not exceeding 87% of body length (Fig. 8).

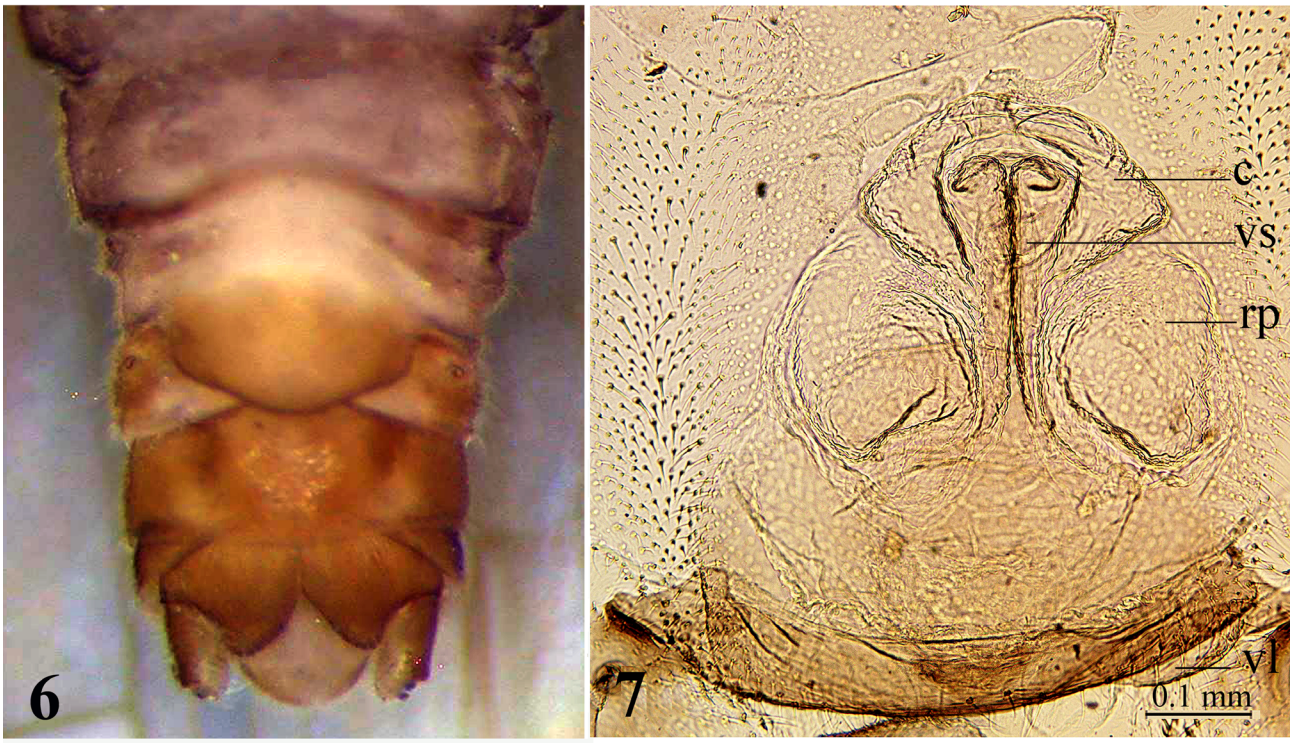
FIGURE 1. Male of *Nemoura sirotskii* **sp. n.**, habitus, dorsal view, not to scale. Photo by VT.

Setation. Setal fringe around the pronotum relatively regular and dense, consisting of occasional long hairs, and bristles narrowed and pointed apically (Figs. 10, 11). Length of the longest pointed bristles at the anterior and posterior pronotal corners attains 5.5% of pronotal width. Pronotum disc bears short blunt setae and occasional tiny procumbent setae with acute hooked apices (Figs. 10, 11). Outer wing pad margin covered with acute bristles; long and tiny acute spine-like setae in the lines on wing pads (Fig. 12). Legs with uniform chaetotaxy: all femora covered mainly with short acute bristles, occasionally fine procumbent setae and few thin long hairs. Setal fringe on outer femur margin consisting of short and thin spine-like bristles in an irregular arrangement in the apical half on fore leg, short spine-like bristles occupy whole outer femur margin on hind leg; the longest spine-like bristles attain 36% of femur width on the fore leg (Fig. 13) and 32% of femur width on the hind leg. Setation of tibiae and

tarsi very similar to femur. Terga covered with short acute bristles and fine procumbent setae. Posterior tergal margins feature relatively sparse, mostly spine-like bristles and one pair of conspicuous relatively long spine-like bristles (Fig. 14). Length of the longest bristles on terga 5−6 does not exceed 36.5% of segment length (Fig. 14). Cercal chaetotaxy uniform; apical whorl comprises a set of acute bristles, long thin hairs absent; intercalary setae very sparse (Figs. 15−18). Length of acute bristles on the margins longer than in the middle of apical whorl and slightly exceeding of segment length on cercal segments 8−10 (Fig. 16); marginal stout acute bristles of medial cercal segments not exceeding 60% of the segment length on cercal segment 16, length of sparse intercalary bristle 23% of segment length (Fig. 17). Apical cercal segments bear apical whorls with a few thin spine-like bristles and tiny setae with rounded apices. Thin intercalary bristles increase in number and in size from the basal to the apical part of the cerci, length of intercalary bristles reaches about 25% of apical segment length (Fig. 18).



FIGURES 2–5. Male of *Nemoura sirotskii* **sp. n.** 2. Abdominal tip, ventral with epiproct in dorsal. 3. Cercus, ventral. 4. Epiproct, dorsal. 5. Epiproct, ventral. Photos by VT. AS—apical sclerite, DF—dorsal fold, IPL—inner paraproct lobe, OPL—outer paraproct lobe, RS—ring sclerite, SP—subgenital plate, V—vesicle, VS—ventral sclerite.



FIGURES 6–7. Female of *Nemoura sirotskii* **sp. n.** 6. Abdominal tip, ventral. 7. Vaginal complex, cleared, dorsal: c—collar, vs—vaginal sclerite, rp—rounded roughened pocket, vl—vaginal lobe.

Diagnosis. The apical sclerites of the epiproct of *N. sirotskii* resemble those of *N. arctica.* The apical sclerites of *N. sirotskii* (Figs. 2, 4, 19) are substantially larger relative to the epiproct than those of *N. arctica* (Figs. 20, 21). They adhere closely to the ring sclerites in parallel positions, and are located above them in dorsal view (Figs. 2, 4, 19); the apical sclerites of *N. arctica* are located rather in front of the ring sclerites in dorsal view (Figs. 20, 21). A salient difference is found in the shape of cerci: those of *N. sirotskii* bear only two apical spines (Figs. 2, 3); cerci of *N. arctica* bear three spines (Figs. 22, 23), with the unpaired outer spine being highly variable in size and shape (Lillehammer 1974, fig. 25; Zhiltzova 1972, figs. 1, 2; Zhiltzova 2003, figs. 427−428; Kondratieff & Baumann 2004, fig. 2; Grubbs *et al.* 2018, figs. 1−16).

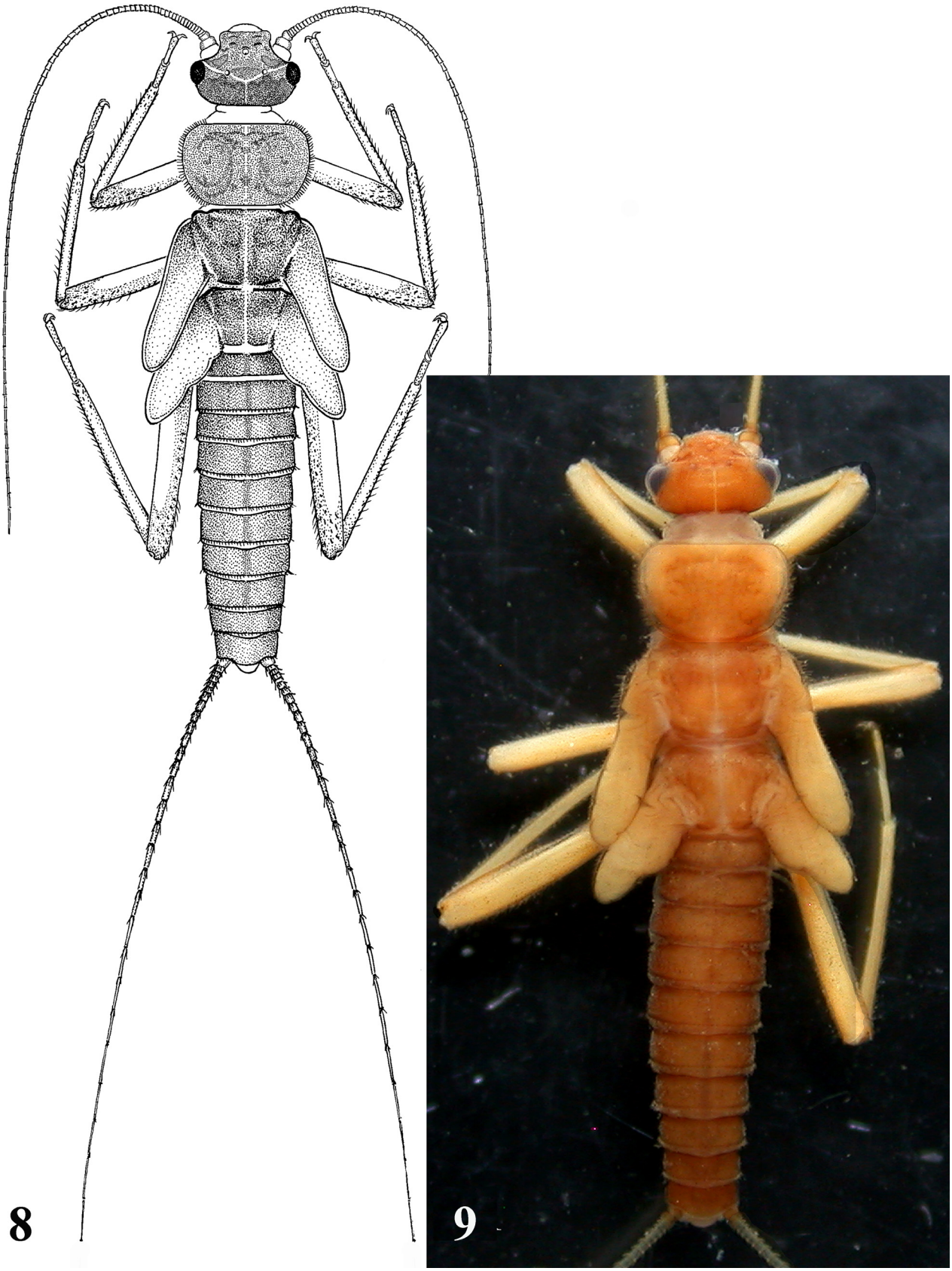
Distribution. *Nemoura sirotskii* was collected in the zone of continuous permafrost with rare thawed patches, in cold mountain streams or rivers where the water temperature varies from 8.0 to 10.0°C in June. The streams flow from the Dzhagdy and Tukuringa Ranges into Zeya Reservoir and are tributaries of the Zeya River (Amur River Basin) (Figs. 24, 25). Additional collecting sites are at Khingan River, left tributary of Amur River (Fig. 26) and Okhota River (the Sea of Okhotsk Basin).

Nemoura sirotskii occurs together with cryophilic *Mesocapnia* sp., *N. arctica, Megarcys pseudochracea* (Zhiltzova, 1977), and *Suwallia talalajensis* (Zhiltzova, 1976) in the streams of the Zeya Reservoir Basin and Okhota River.

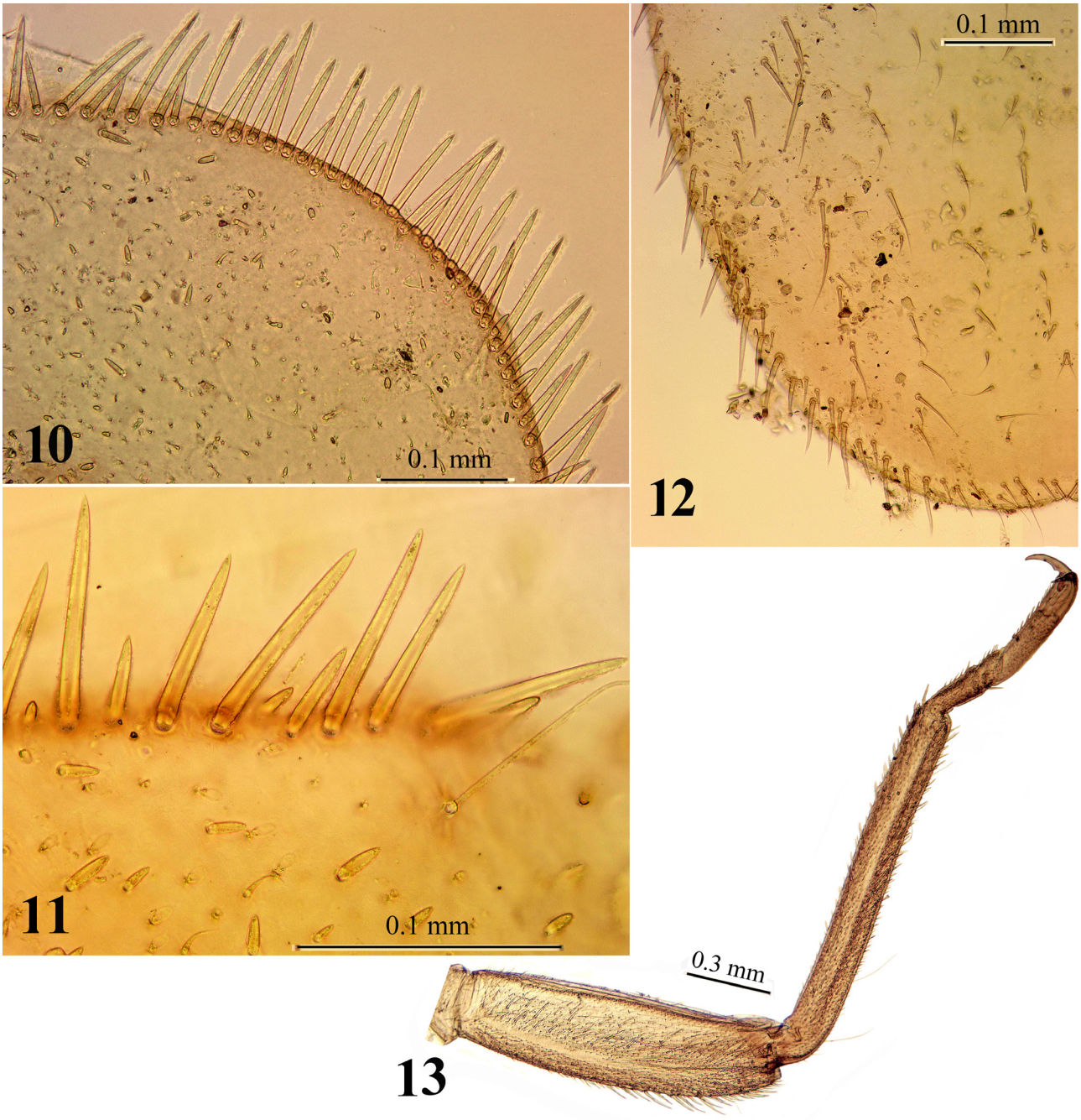
Etymology. The species is named in honor of the outstanding hydroecologist Sergei Yegorovich Sirotsky, the leader and organizer of the monitoring hydrobiological research in the Bureya and Zeya River Basins.

Discussion

The new species *N. sirotskii* is morphologically similar to *N. arctica.* The latter taxon may be a species complex, even though Grubbs *et al*. (2018) conclude that all specimens they studied from five Eurasian and twelve North American Arctic localities belong to a single variable morphological species. As a note of reference, the listing of *N. arctica* and other Arctic stoneflies species as occurring in Latvia is due to an error at the time of the compilation of the Fauna Europea checklist fauna-eu.org (pers. comm. Mārtiņš Kalniņš 10 February 2012).



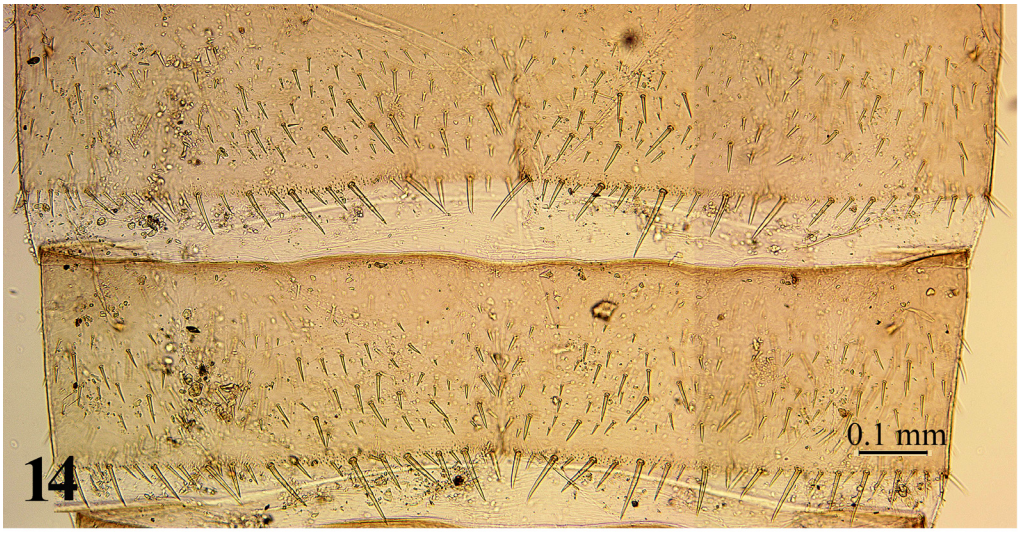
FIGURES 8–9. Larva of *Nemoura sirotskii* **sp. n.**, habitus, dorsal view, not to scale. Photo by VT.

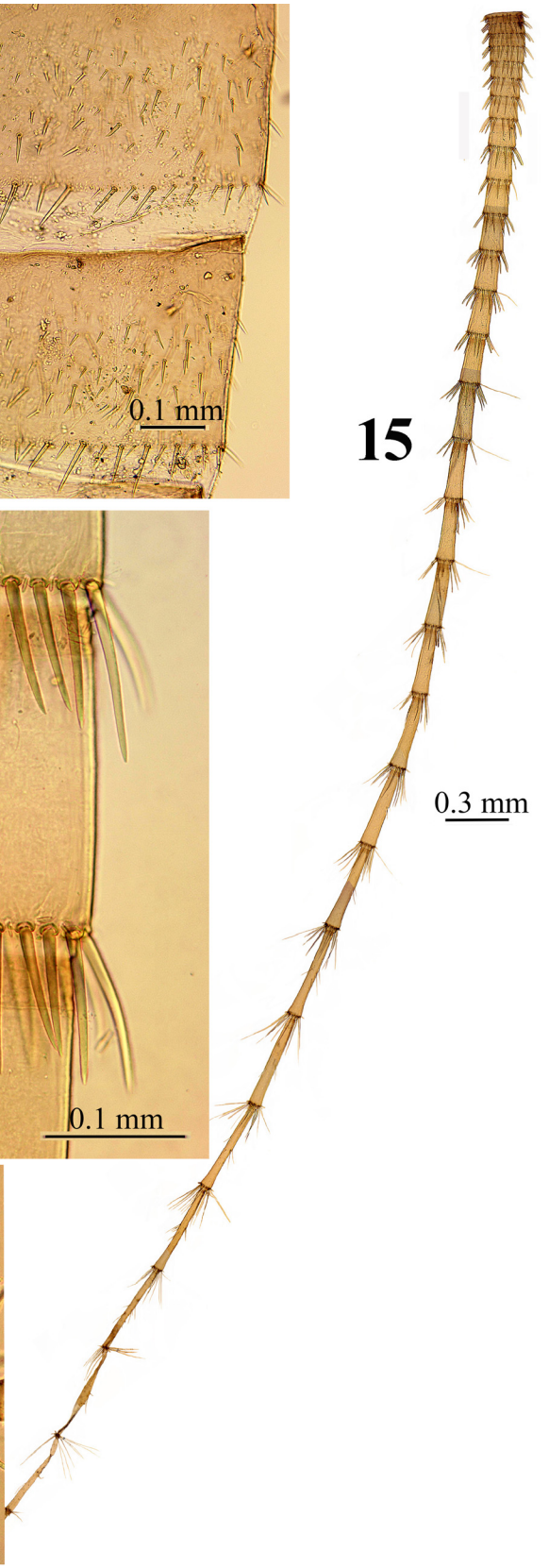


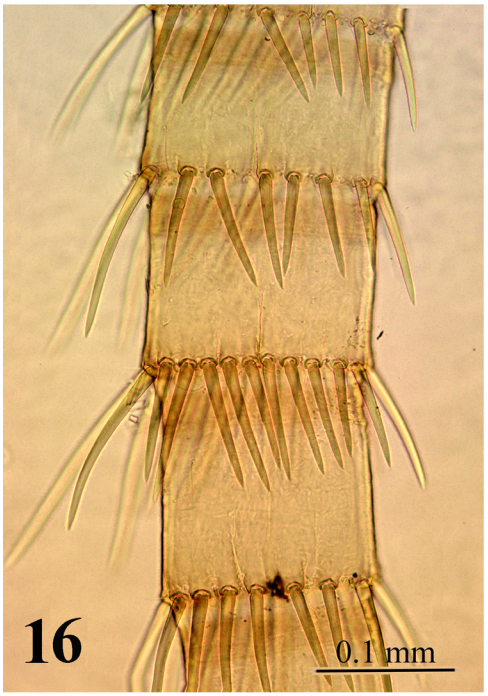
FIGURES 10–13. Larva of *Nemoura sirotskii* **sp. n.** 10. Right anterior pronotal corner with pointed bristles, dorsal. 11. Chaetotaxy on pronotal disc, dorsal. 12. Outer wing pad margin covered with acute bristles, long and tiny acute spine-like setae in the lines on wing pads. 13. Right foreleg. Photos by VT.

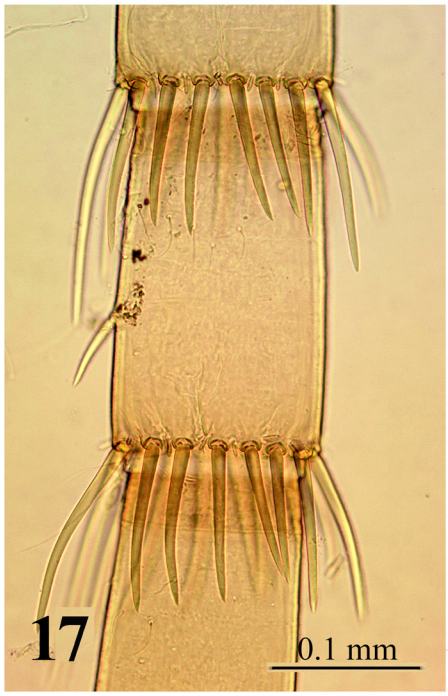
A first indication of taxonomic complexity is the description of a subspecies of *N. arctica*, *N. a. mongolica* Zhiltzova, 1972. Further evidence comes from preliminary analyses of DNA sequences. The mitochondrial marker cytochrome oxidase I, the so-called DNA barcode region, shows several deeply divergent but sympatric haploclades in both North America and Scandinavia. This information is available from the Barcode of Life Data System (BOLD) website (Ratnasingham & Hebert 2007), and in part from our work on the Scandinavian *Nemoura* fauna (Boumans & Brittain 2012, Boumans 2014: 10), with specimens identified as either '*N. arctica'*, '*N. trispinosa'* or '*Nemoura* sp.'. Since the mitochondrial genome does not recombine, there are scenarios that can explain within-species deep haplotype divergence even within an interbreeding population (Boumans & Tierno de Figueroa 2016). However, we also collected two sequences of the nuclear marker 28S (GenBank accession nrs. MH407653 and MH407652) from two specimens representing distinct Scandinavian '*N. arctica*' haploclades

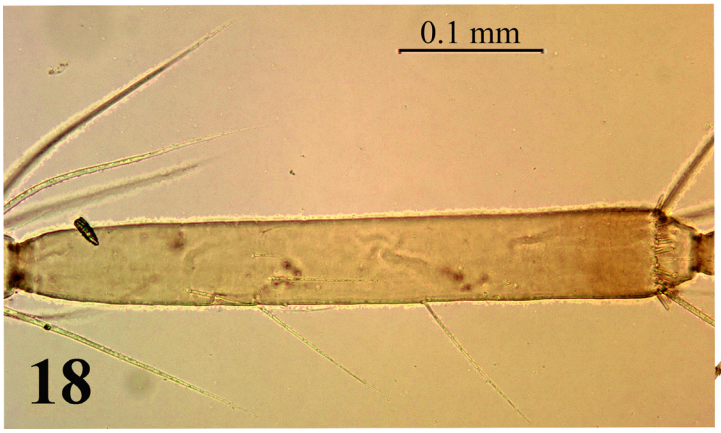
(accession nos. JX905511 and MH407226, 8.4% uncorrected genetic distance), and these, admittedly are very limited, nuclear data, but also suggest that they may be non-interbreeding, cryptic species.



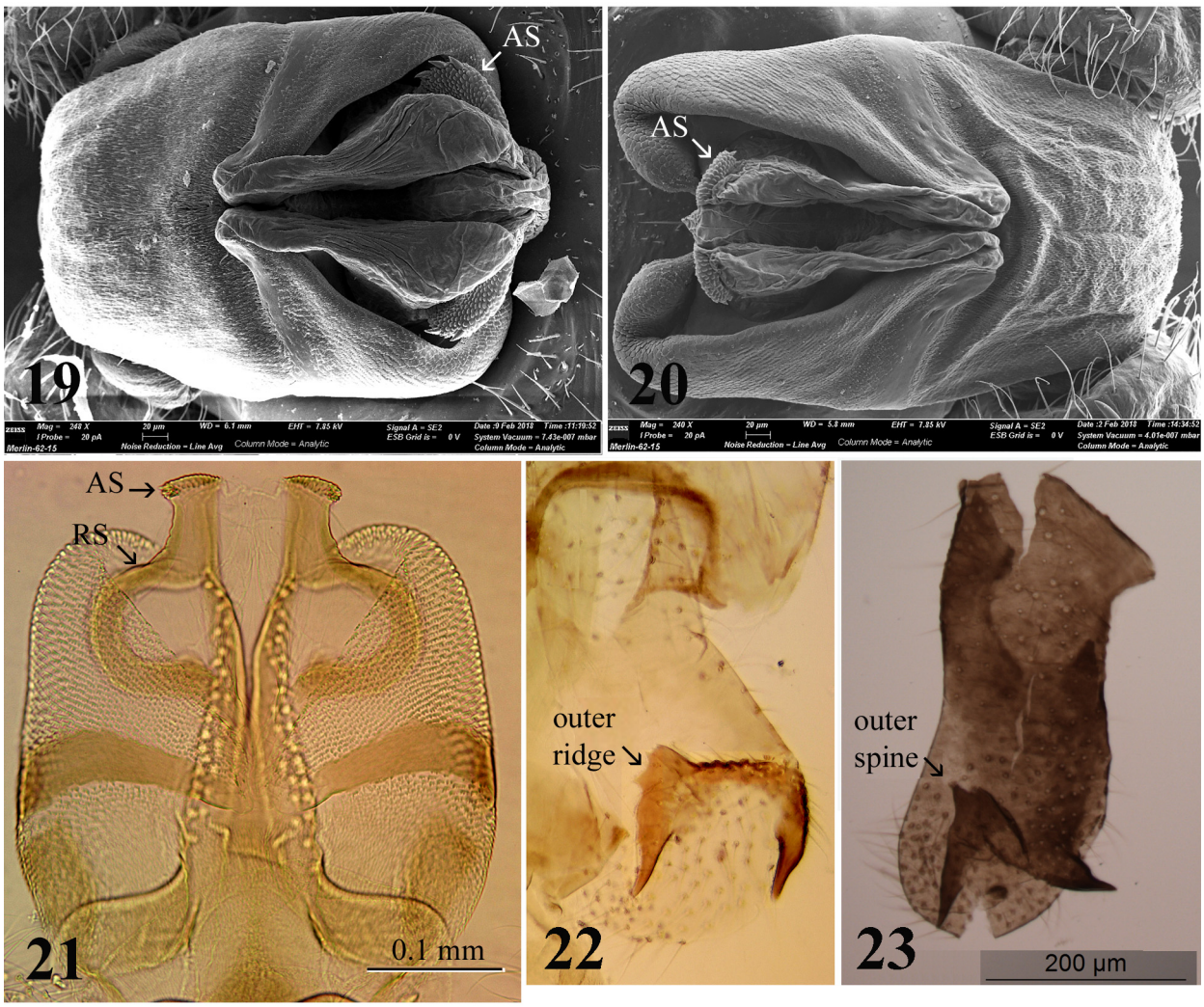








FIGURES 14–18. Larva of *Nemoura sirotskii* **sp. n.** 14. Chaetotaxy of terga 5–6. 15. Left cercus. 16. Cercus segments 8−10 with acute bristles. 17. Cercus segments 15 & 16 with acute bristles, and sparse intercalary setae. 18. Apical cercus segment with long intercalary setae. Photos by VT.



FIGURES 19–23. 19. Male of *Nemoura sirotskii* **sp. n.**, epiproct, dorsal view. 20–22. Male of *Nemoura arctica* from Shirokovskaya River. 20, 21. Epiproct, dorsal view. 22. Cercus, distal view, arrow points to outer spine. Photos by VT. 23. Right cercus of *Nemoura arctica* from Kautokeino, Norway, arrow pointing to outer spine. Photo by LB. AS—apical sclerite, RS—ring sclerite.

FIGURES 24–25. Habitats of *Nemoura sirotskii* **sp. n.**. 24. Izhak River, the Zeya Reservoir Basin (Amur River Basin) the type locality. 25. Shirokovskaya River, Zeya Reservoir. Photos by M. Tiunov.

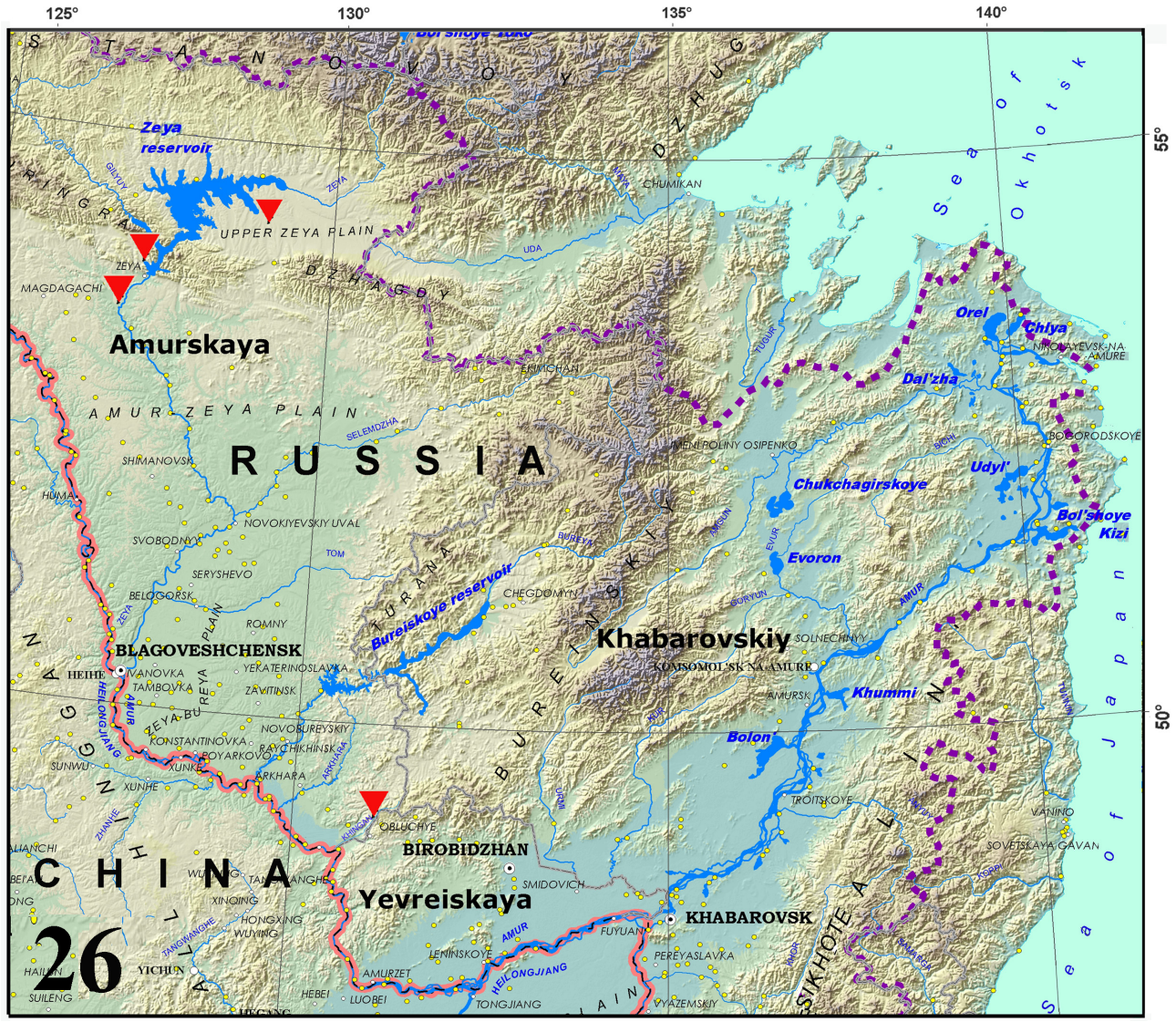


FIGURE 26. Distributional map of *Nemoura sirotskii* **sp. n.** in Amur River Basin.

Possibly, salient morphological characteristics will be found later to distinguish genetically distinct lineages of *N. arctica* sensu lato. Unfortunately, we have not succeeded in locating the type specimens of *N. arctica* after searching in Oslo (Boumans 2011) and requesting collection curators at other museums in Norway and Denmark. Grubbs *et al*. (2018) studied the holotype of *N. trispinosa*, but do not include images of the terminalia of the holotype.

A discussion of possible cryptic species of '*N. arctica*' and revaluation of the distinction between the two subspecies of *N. arctica* requires additional research and is beyond the scope of this paper. However, herein, we wish to emphasize that taxonomic experts so far have not been able to find diagnostic morphological characters for any possible cryptic species that currently are determined as *N. arctica* (Grubbs *et al*. 2018), whereas *N. sirotskii* is similar but clearly appears distinct. For this reason, we decided not to postpone the description of *N. sirotskii* until the uncertainty about '*N. arctica*' senso lato is resolved. The syntopic occurrence of *N. sirotskii* and *N. arctica* is a further supports the species status of the new species.

Acknowledgements

The first author thanks T.A. Eroshenko, T.Yu. Gorpenchenko,V.M. Kazarin and M.P. Tiunov (all are from FSC EATB FEB RAS, Vladivostok) for drawings and preparation of SEM pictures and photos; T.M. Tiunova for

collecting material, A. Semenchenko (Far Eastern Federal University, Vladivostok) for discussion of data from the BOLD and R. Baumann (Provo, Utah, U.S.A) for comments and suggestions. The authors are very grateful to P. Zwick (Schlitz, Germany) and B. Kondratieff (Fort Collins, Colorado, USA) for sending material for comparison, comments, suggestions, and linguistic revision of an earlier version of the manuscript. Curators H. Enghoff (Natural History Museum of Denmark, Copenhagen), Thomas Simonsen (Natural History Museum Aarhus), Vladimir Gusarov (Natural History Museum of Oslo), Steffen Roth (University Museum of Bergen), Torbjørn Ekrem (NTNU University Museum in Trondheim), and Robert Bergersen and Jostein Kjærandsen (both Tromsø University Museum) kindly helped in our fruitless search for the type specimens of *N. arctica* in Denmark and Norway.

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