



## A new finding of *Paraboreochlus okinawanus* Kobayashi et Kuranishi (Diptera: Chironomidae: Podonominae) in the Russian Far East, with the original description of the pupa and larva

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During a hydrobiological survey of a stream in the Terney Village in the Primorye Territory of the Russian Far East (spurs of the Eastern Sikhote-Alin Mountains), pupae and larvae of the rare chironomid genus *Paraboreochlus* Thienemann from the subfamily Podonominae were found in zoobenthos samples. This genus includes 3 species—*P. minutissimus* (Strobl), *P. okinawanus* Kobayashi et Kuranishi and *P. stahli* Coffman (Ashe & O'Connor 2009).

Study of the adult male hypopygium extracted from a mature pupa of our material allowed identification of the species *P. okinawanus*, for which the immature stages were unknown. This species was described from Okinawa Island of Japan (Kobayashi & Kuranishi 1999), and later recorded from the South Primorye of the Russian Far East (Makarchenko & Makarchenko 2003), Oriental China (Lin *et al.* 2013) and South Korea (Kang *et al.* 2017). Below we provide short redescription of pharate adult male and the original description of the pupa and larva of *P. okinawanus* from the Russian Far East.

### Material and methods

The pupae and larvae were preserved in 4% formalin and then transferred to 70% ethanol. Microscope slides were mounted in the polyvinyl lactophenol. The morphological terminology and abbreviations used below generally follow Sæther (1980) and Brundin (1966, 1983, 1986, 1989). The photographs were taken using an Axio Lab.A1 (Karl Zeiss) microscope with an AxioCam ERc5s digital camera and an Olympus SZX16 stereomicroscope with an Olympus DP74 digital camera, and then stacked using Helicon Focus software. The final illustrations were post-processed for contrast and brightness using Adobe®.

All material is deposited in the collection of the Federal Scientific Center of the East Asia Terrestrial Biodiversity, Far East Branch of the Russian Academy of Sciences, Vladivostok (FSCEATB FEB RAS).

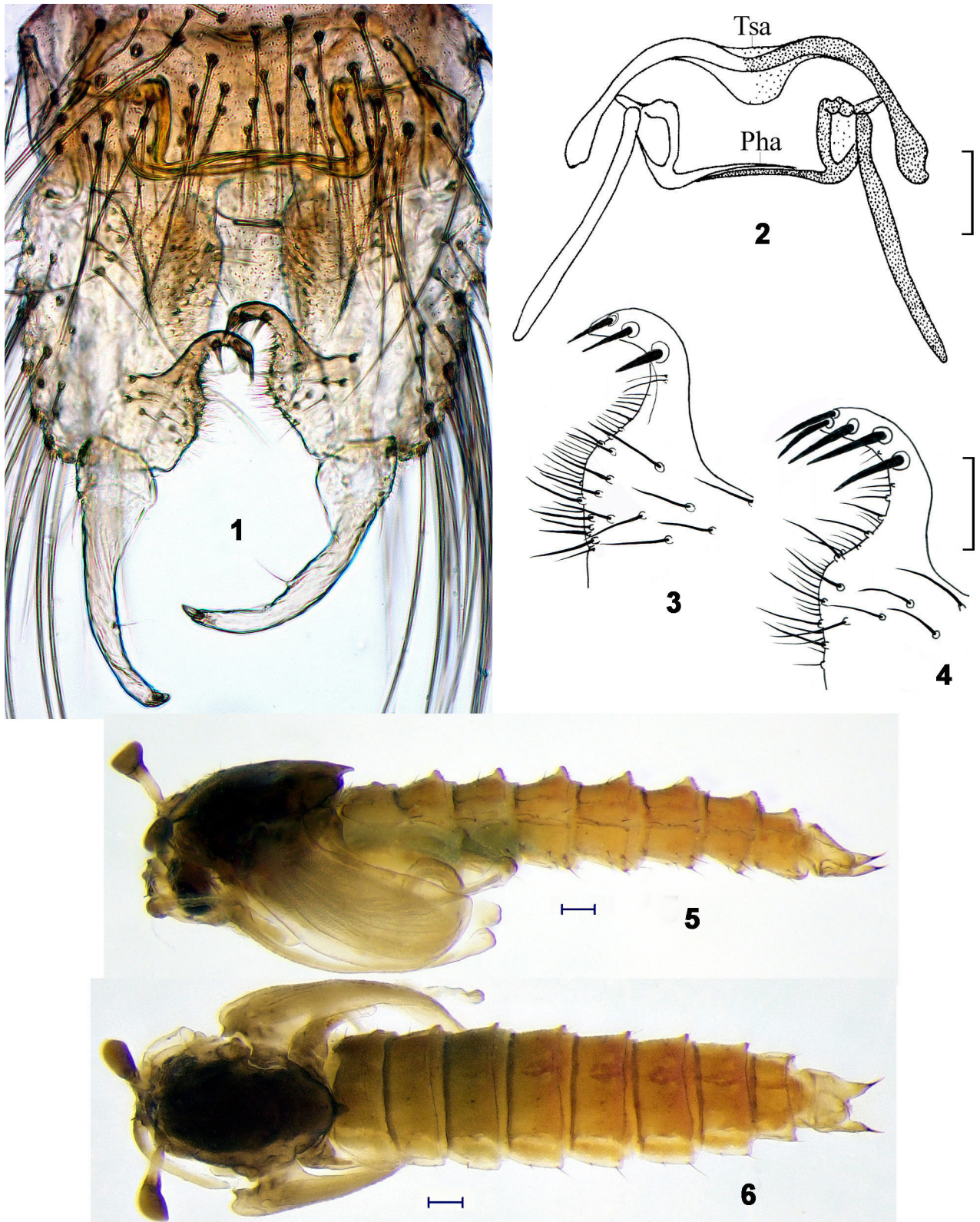
### Taxonomy

#### *Paraboreochlus okinawanus* Kobayashi et Kuranishi

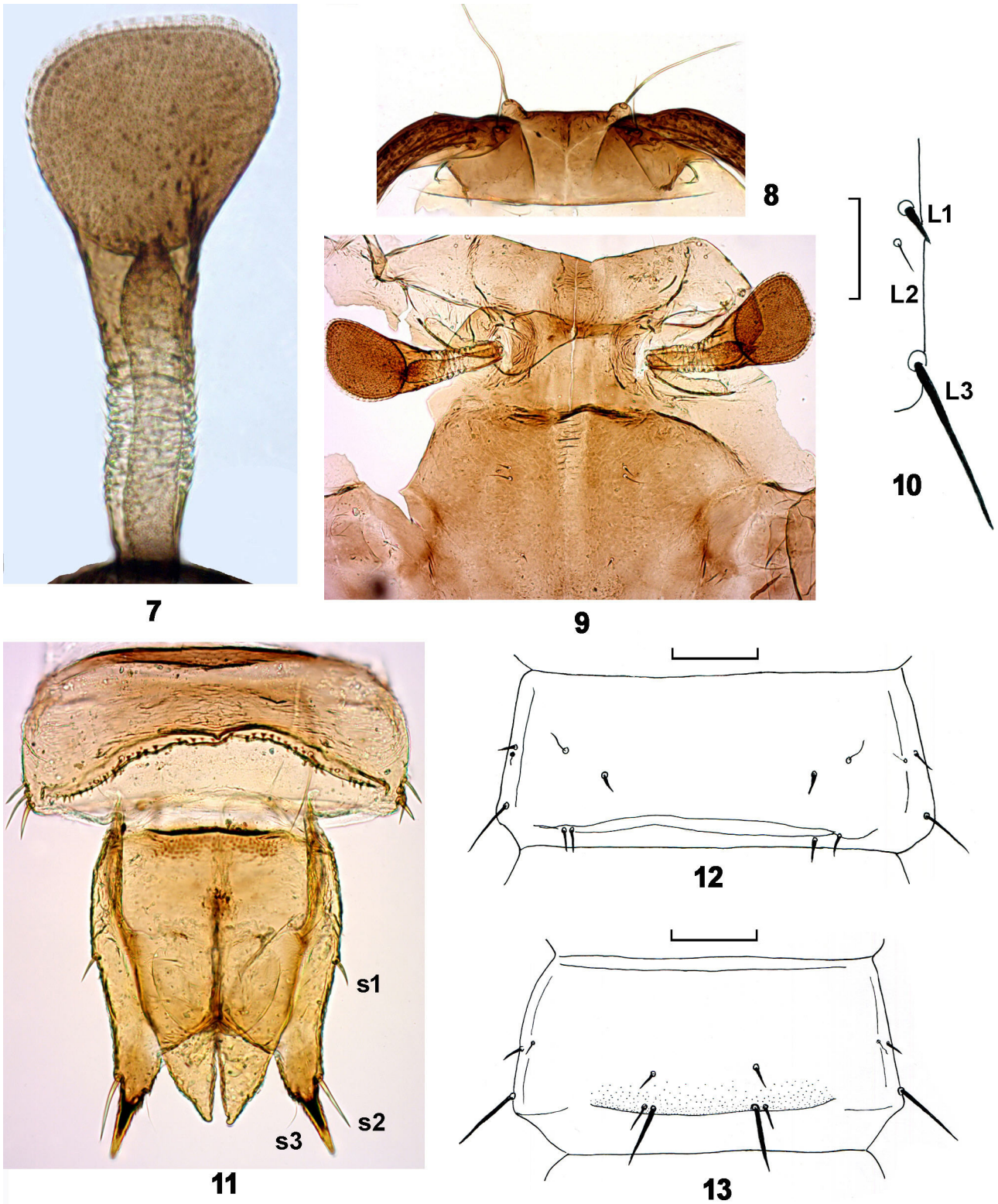
(Figs 1–20)

*Paraboreochlus okinawanus* Kobayashi et Kuranishi, 1999: 602; Makarchenko & Makarchenko 2003: 217; Ashe & O'Connor 2009: 86; Lin *et al.* 2013: 74.

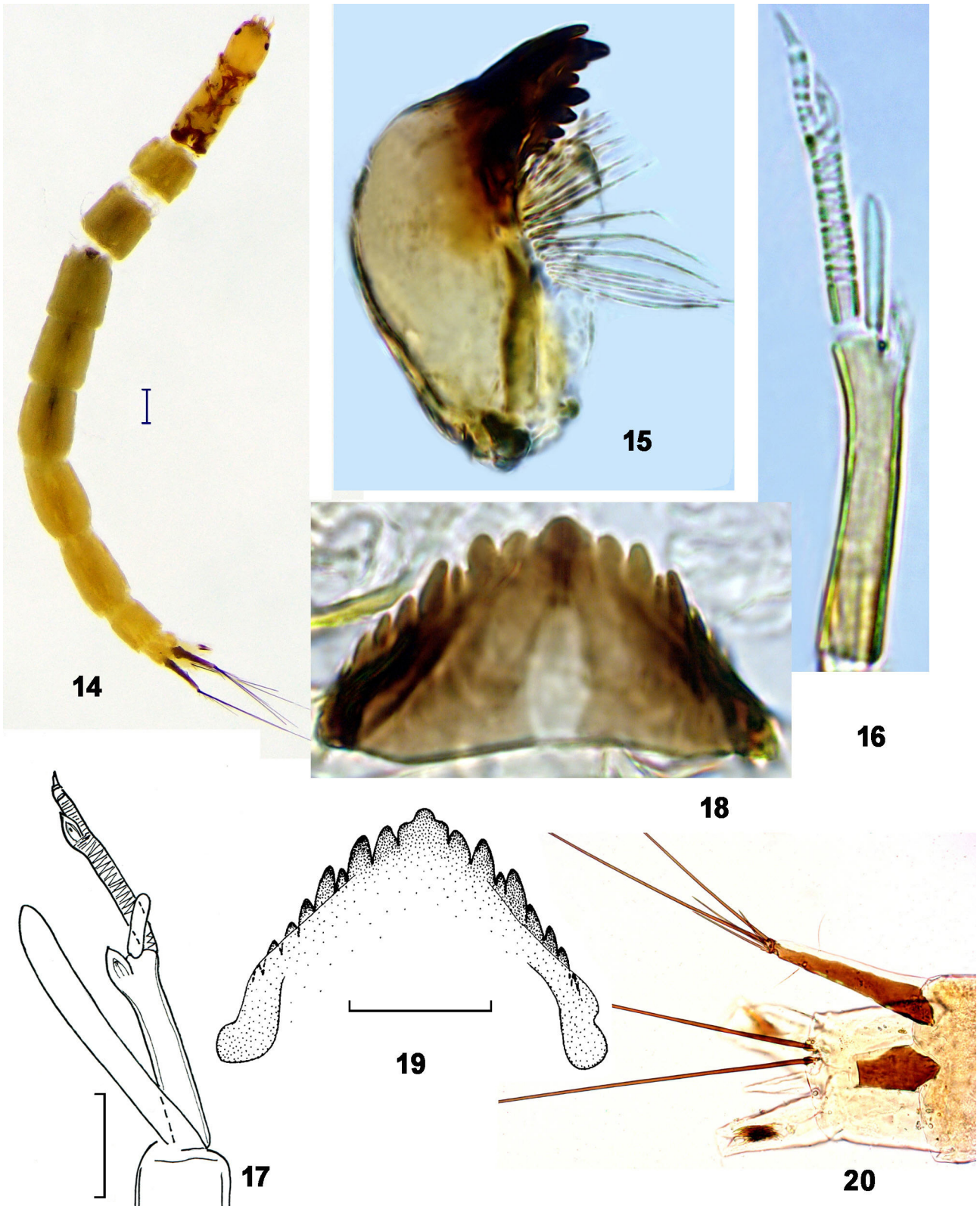
**Material examined.** RUSSIA: 3 pharate adult males, extracted from mature pupae, 215 pupae, 26 larvae, Primorye Territory, Terney District, Terney Village, Mashnikovsky Stream, tributary of the Vilka River (Serebrianka River basin), spurs of the Eastern Sikhote-Alin Mountains, 21.VI.2024, 45.048184 N, 136.609847E, leg. E. Potikha and A. Kozlova.



**FIGURES 1–6.** Adult male (1–4) and pupa (5–6) *Paraboreochlus okinawanus* Kobayashi et Kuranishi. 1, hypopygium in dorsal view; 2, transverse sternapodeme and phallapodeme; 3–4, inferior volsellae; 5, total view, from side; 6, total view, from above. Tsa—transverse sternapodeme, Pha—phallapodeme. Scale bar for Figs 2–4—20  $\mu\text{m}$ , for Figs 5–6—100  $\mu\text{m}$ .



**FIGURES 7–13.** Pupa of *Paraboreochlus okinawanus* Kobayashi et Kuranishi. **7**, thoracic horn; **8**, frontal setae; **9**, part of thorax; **10**, lateral setae ( $L_1$ – $L_3$ ) of segment IV; **11**, tergite VIII and anal segment; **12**, tergite V; **13**, sternite V. s1–s3, lateral setae of anal segment after Brundin 1966. Scale bar for Fig. 10—50  $\mu$ m, for Figs 12–13—100  $\mu$ m.



**FIGURES 14–20.** Fourth instar larva of *Paraboreochlus okinawanus* Kobayashi et Kuranishi. **14**, total view, from above; **15**, mandible; **16–17**, flagellum of antenna; **18–19**, mentum; **20**, anal segment with procercus, supra-anal setae, posterior parapods and anal tubulus. Scale bar for Fig. 14—100  $\mu\text{m}$ , for Figs 17, 19—20  $\mu\text{m}$ .

## Description

**Pharate adult male** (n = 3), extracted from mature pupae.

Coloration. Head, thorax and abdomen dark brown.

Head. Eyes bare, with moderate dorsomedial extension. Temporal setae including 4–7 inner verticals and 11–13 outer verticals. Clypeus swollen, with 8–12 setae. Antenna with 14 flagellomeres and developed plume; terminal flagellomere with 2 subapical setae, 40 µm long; penultimate flagellomere 2.7 times length of the ultimate; flagellomeres 13 and 14 completely separated. Length of 1–14 flagellomeres (µm): 44, 24, 24, 24, 24, 24, 24, 28, 24, 28, 28, 28, 152, 56; AR 0.59–0.61. Palpomere lengths (in µm): 16; 28; 36; 24; 52. Head width/palp length 1.6.

Thorax. Anteprenotum with 5–10 lateral setae. Acrostichals 44 (in 2 rows); dorsocentrals 25–36 (in 2 rows); prealars 17–20; scutellars: 15–16 long setae in 1 row and 8–9 shorter setae in group anteriorly; postnotals 5.

Hypopygium (Figs 1–4). Tergite IX with 37–48 long setae, laterosternite IX with 2–4 long setae. Gonocoxite straight, 88–112 µm long. Inferior volsellae broad basally, with 10–12 long setae, and narrow curved distally, with a row of 3–5 large spine-like setae, 14–16 µm long (Figs 1, 3–4). Gonostylus 80–104 µm long, swollen and with 2–3 setae at base, with 2 short setae in distal part; megaseta 8 µm long. Two central portions of phallopodemes well-sclerotized, 72–80 µm long and together forming transverse straight bar as shown in Fig. 2. Transverse sternapodeme 128 µm long. HR 1.02–1.08.

**Pupa** (n=5). Total length 1.9–2.4 mm. Coloration of fixed pupa: cephalothorax dark brown, first three abdominal segments are brownish-brown, the rest are yellowish-brown (Figs 5–6). Exuviae brownish.

Cephalothorax. Frontal setae on tubercles, anterior ones 140–192 µm long, posterior—60–104 µm long (Fig. 8). Thoracic horn with large plastron, 200–248 µm long, 88–112 µm wide in distal part and 24–36 µm long at base. Precorneal setae 3, lengths (µm): Pc<sub>1</sub>—36 (hair-like, Pc<sub>2,3</sub>—6–8 (spine-like) (Figs 7, 9). Anteprenotum with 2 median and 1 lateral anteprenotals. Mesonotum with 3 pairs of dorsocentrals, first and second dorsocentrals close together, third located 96 µm behind second dorsocentral seta (Fig. 9). Wing sheath lacking nose.

Abdomen. Tergites II–VIII with posterior margin distinct row of denticles, which in number maximum (ca 50–60) on tergite VIII. The same on sternites. Segments I with 2 pairs of lateral hair-like setae, 28–32 µm long. Segments II–VII with spine-like setae L<sub>1</sub>, 16–24 µm long and L<sub>3</sub>, 68–80 µm long; L<sub>2</sub> hair-like, 8–10 µm long (Fig. 10). Segment VIII with 3 spine-like lateral setae in posterolateral corner, 12–36 µm long (Fig. 11). Anal lobe with 3 lateral setae which Brundin (1966) called s1–s3 setae (Fig. 11), length of these ones (µm): s1—20–24; s2—44–48; s3—28–32. Tergites I–VIII with 4 pairs of short dorsal setae from which 3 pair setae are spine-like and 1—hair-like (Fig. 12). Sternites I–II without ventral setae; sternite III with 2 pairs of ventral setae at the posterior edge, 20 µm and 72 µm long; sternites IV–VII with 1 pair of ventral setae in middle (20–24 µm long) and 2 pairs at the posterior edge (29–24 µm and 50–69 µm long) (Fig. 13); sternite VIII with 2 pairs of ventral setae at the posterior edge (64–68 µm long). Male genital sac not extended beyond anal lobe (Fig. 11).

**Fourth instar larva** (n=5). Coloration of fixed larvae: head yellow, thoracic segments yellowish brown, abdominal segments yellowish (Fig. 14). Total length 2.8–3.4 mm. Head capsule 230–246 µm long and 180–197 µm wide. Labral setae typical for genus (Brundin 1983). Antenna 5-segmented, length of segments (µm): 71–80, 36–38, 31–33, 3–4, 4–5. Antennal blade is divided; larger projection 51 µm long, reaches middle of 3<sup>rd</sup> segment, smaller projection 26 µm long; segments 2 and 3 with Lauterborn organs; segment 3 annulated; large ring organ with a diameter of 4–7 µm is located at distance of 33 µm from the base of basal segment (Figs 16–17); AR 0.92–1.05. Mandible dark brown, black in distal part, with 8 teeth; seta subdentalis reaches top of fourth tooth; seta interna with 16–18 branches (Fig. 15). Mentum with 1 median and 7–8 pairs of lateral teeth; 8<sup>th</sup> lateral tooth is very small and often invisible or absent; median tooth 3 times wider than the first lateral tooth; ventromental plates small (Figs 18–19). Procercus hyaline anteriorly, blackish posteriorly, 196–212 µm long, 40–48 µm wide at base, bearing 8 dark brown or black strong anal setae varies length, longest of which 490 µm long, shortest 36 µm long; 2 hair-like lateral seta, 32–56 µm long. Posterior parapods 120–160 µm long. Dorsal and ventral pairs of anal tubuli 72–80 µm long. Two black supra-anal setae, 590–607 µm long, just before anal tubuli (Fig. 20).

**Remarks.** The adult male of *P. okinawanus* from the Eastern Sikhote-Alin Mountains slightly differs in some features from males described earlier from Japan, China and the southern Primorye of Russia (Table 1). Since the pupa and larva of *P. minutissimus* are not described fully enough, a comparison of *P. okinawanus* can only be made with *P. stahli*, pupa of which on the mesonotum with 4 dorsocentral setae, tergites I–VII with 5 dorsal setae, sternites IV–VII with 4 ventral setae, segments II–VII with 4 lateral setae. Total length of larva 4.4 mm, antenna 4-segmented, AR 1.14, seta interna of mandible with 20–25 branches, median tooth of mentum 2.5 times wider than the first lateral tooth. Pupa of *P. okinawanus* on the mesonotum with 3 dorsocentral setae, tergites I–VII with 4 dorsal setae, sternites IV–VII with 3 ventral setae,

segments II–VII with 3 lateral setae. Total length of larva 2.8–3.4 mm, antenna 5-segmented, AR 0.92–1.05, seta interna of mandible with 16–18 branches, median tooth of mentum 3 times wider than the first lateral tooth.

**TABLE 1.** Comparison of some features of *Paraboreochlus okinawanus* Kobayashi et Kuranishi males from different regions of East Asia.

Character	Eastern Sikhote- Alin Mountains (n=3)	Japan; Kobayashi & Kuranishi 1999 (n=5)	China; Lin <i>et al.</i> 2013 (n=1)	South Primorye; Makarchenko & Makarchenko 2003 (n=1)
Clypeals	8–12	7–10	8	8
AR	0.59–0.61	0.48–0.51	0.64	0.72–0.73
Penultimate flagellomere length / ultimate flagellomere length	2.7	2.38–3.69	3.7	4.1–4.3
Acrostichals	44	34–35	15	42
Dorsocentrals	25–36	29–41	29	38–40
Prealars	17–20	17–19	13	17–21
Scutellars	23–25	12–17	10	22
Postnotals	5	4–5	-	4
Number of apical spines in inferior volsella	3–5	4	3	3

It should be noted that at all stages of development the three species of the *Paraboreochlus* are very close and in this regard we join the opinion of colleagues (Lin *et al.* 2013) about the need for DNA barcoding of known species of this genus to clarify their taxonomic status.

**Ecology.** Pupae and larvae were collected in cold stream on gravel-pebble soil at a depth of 14 cm, with a current speed of 0.89 m/sec and a water temperature of 7.5 °C. At the time of sampling on June 21, 2024, the number of pupae and larvae reached 4016 ind./m<sup>2</sup>.

**Distribution.** *P. okinawanus* has an eastern Asian distribution—Oriental China, Japan, South Korea and Russian Far East.

## Acknowledgements

The authors are much grateful to Dr. E.V. Potikha (The Abramov K.G. Sikhote-Alin Biosphere Nature Reserve, Terney Village, Primorye Territory) for making material available for us as well as to Dr. V.M. Loktionov (Federal Scientific Center of the East Asia Terrestrial Biodiversity, Vladivostok) for help with preparing of some microphotographs.

The research was carried out within the state assignment of Ministry of Science and Higher Education of the Russian Federation (theme No. 124012400285-7).

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