

The newly found imagoes of *Ochernova tshernovae* (Kazlauskas, 1963) from China validating its position in the family (Ephemeroptera: Neoephemeridae)

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Abstract

Previously, the male imago of the mayfly *Ochernova tshernovae* (Kazlauskas, 1963) was not reported. The nymphs and adults of it found in northwestern China in 2025 not only show that this species has a vaster distribution than previously known but also demonstrate it is close to the European species *Leucorhoenanthus maxima* (Joly, 1871). Both of them share smaller posterolateral projections of abdominal terga in nymphs and smaller compound eyes in males than other genera. Biogeographically, this new finding illustrates that three neoephemerid genera of China (*Ochernova*, *Potamanthellus*, and *Pulchephemera*) are connected. Therefore, both its nymphs and adults are described in detail herein, and the observed biology of it is also presented.

Key words: new record, phylogeny, morphology, biology, aquatic insect

Introduction

The family Neoephemeridae currently comprises approximately 17 extant species of five genera (Ma & Zhou 2021; Garces & Sartori 2022; Zheng & Chen, 2024; Vasanth, Kubendran & Subramanian 2025). Amongst them, the genus *Ochernova* Bae & McCafferty, 1998 is the latest one, which was established by Bae and McCafferty in 1998. However, unlike other genera in the family, its male characteristics are unclear although Kluge (2004) provided some information of it and Kluge (2022) described its subimaginal tarsal microtrichia.

Furthermore, historically, the nymphs of *Ochernova* were described vaguely. No researchers provided the exact structures of its gills I, or its gills III–VI. The mouthparts and abdominal tubercles in historical descriptions were not presented photographically.

The monospecific genus *Ochernova* was found in Central Asia (Turkmenistan, Kazakhstan, Tajikistan, and Uzbekistan) (Kluge 1995, 2004; Bae & McCafferty 1998). Type specimens of its single species *Ochernova tshernovae* (Kazlauskas, 1963) were collected almost a century ago (Kluge 1995). More fresh specimens from other localities will not only enlarge its geographical distribution but also provide vivid coloration and morphological details of it.

Upon both imaginal and nymphal materials of the species *Pulchephemera projecta* (Zhou & Zheng, 2001), Ma & Zhou (2021) reconstructed the phylogeny of Neoephemeridae with other related taxa, such as Fossoriae and Caenidae. However, due to the lack of imaginal information of the genus *Ochernova*, the relationship between these three genera (*Ochernova*, *Leucorhoenanthus* Lestage, 1931 and *Neoephemeridae* McDunnough, 1925) remained unsolved in that study.

The nymphs of *Pulchephemera* Zhou, 2021 (in Ma & Zhou 2021), *Potamanthellus* Lestage, 1931 and *Leucorhoenanthus* have flat and smooth body, resembling the nymphs of *Caenis* Stephens, 1835 in Caenidae. Reportedly, nymphs of *Ochernova* have slim bodies, long legs and caudal filaments (Kazlauskas 1963; Bae & McCafferty 1998; Kluge 2004), somewhat resembling the nymphs of *Brachycercus* Curtis, 1834 in Caenidae. Similarly, this kind of nymphs lives in dead plants in water (Kluge 2004). With further observations of their behavior and ecology, more biological details of them will be explored.

Early in 2001, a research group from Shanghai Ocean University (China) focusing on aquatic insects, collected some mayflies in Xinjiang Uygur Autonomous Region, Northwestern China. Surprisingly, among the specimens they sent us for identification, we found a broken unknown neoephemerid nymph, which triggered several following collections there. Eventually, in 2024, six nymphs were re-found, and in summer 2025, about ten adults of this species were reared from mature nymphs. These specimens confirm the species is the previously known *Ochernova tshernovae*, providing us an opportunity to show its real morphology and phylogeny.

Methods and materials

Materials examined in this study were collected by members of the mayfly research group at Nanjing Normal University (NNU), China. All specimens were preserved in 75–90% ethanol and are deposited in the mayfly collection of the College of Life Science (NNU).

Morphological examinations and imaging were conducted using a stereomicroscope (Nikon SMZ1000) with a visual system Mshot Image Analysis System (Mshot MZ81, Guangzhou, China) and DSLR cameras (Canon EOS 90D and Sony a7R). Fine structures, such as mouthparts, were observed and photographed using a microscope (Nikon Eclipse 50i). Final plates were prepared with Adobe Photoshop 2022.

Eggs and hindleg were dissected from the female subimago. All SEM samples were dehydrated in graded acetone solutions (70%, 80%, 90%, 2×98% and 2×100%) for 10 min each. Subsequently the specimens were immersed in 1-1.5 mL HMDS (1,1,1,3,3,3 hexamethyldisilazane; Merck-Suchardt, Darmstadt) in 20 mL glass vials. After a soak of 30 minutes, approximately 90% of the HMDS was removed and the vials were immediately transferred to a desiccator. The bottom of the desiccator was covered by silica gel beads (Merck-Suchardt, Darmstadt) and the desiccator itself was evacuated. The remaining HMDS was allowed to evaporate overnight under anhydrous conditions. Afterwards, the specimens were sputter coated with gold (BIO-RAD SC510, München) for 80 seconds. The samples were photographed by a Raman-Combined Field Emission Scanning Electron Microscope (FE-SEM) (TESCAN MAIA 3 GMU, TESCAN Company).

Result

Ochernova tshernovae (Kazlauskas, 1963) (new record of China)

Neoephemera tshernovae Kazlauskas, 1963: 582 (nymph), from Turkmenistan.

Neoephemera tshernovae: Kluge, 1995: 38 (type depository, Uzbekistan); Kluge, 1997: 179 (distribution).

Ochenova tshernovae: Bae & McCafferty 1998: 59 (nymph, Tadzhikistan, Uzbekistan); Kluge 2004: 272, 275 (nymph, female imago, male subimago, distribution and phylogeny); Kluge 2022: 171 (tarsal microtrichiae of subimago).

Distribution: Central Asia (Tadzhikistan, Uzbekistan, Turkmenistan, Kazakhstan); East Asia (China: Xinjiang).

Material examined. 1 nymph, Jiefang Bridge, Ili river, Xinjiang Uygur Autonomous Region, 2001-VIII-3, collected by Ruilei Zhang; 6 nymph, Jiefang Bridge, Ili river, Xinjiang Uygur Autonomous Region, 1640 m a.s.l., 2024-VI-6-9, collected by Xuhongyi Zheng and Dewen Gong; 1 nymph, County Road 142, Zhaosu County, Xinjiang Uygur Autonomous Region, 1637 m a.s.l., 2025-VI-27, collected by Mengyao Li and Xinhe Qiang; 3 male imagoes, 4 female imagoes, 1 male subimago, 1 female subimago, 23 nymphs, 8 nymphal exuviae, 2 subimaginal exuviae (1 male, 1 female), Zhaosu Wetland Park, Zhaosu County, Xinjiang Uygur Autonomous Region, 1595 m a.s.l., 2025-VII-1-9, collected by Mengyao Li and Xinhe Qiang.

Description

Mature nymph (in ethanol): body length 10.5–12.0 mm, caudal filaments length 8.0–11.0 mm, body pale to light yellowish, with black stripes or markings. Head usually with longitudinal stripe along midline, thorax and abdomen with one pair of sublateral brown to black stripes (Figs 1A–D).

Head: generally light ochre, middle line and occipit washed with brown marking; dorsal surface of head with scattered stout setae (Figs 1A, B). Clypeus with a transverse row of club-like setae (Figs 2A, B). Scape and pedicel

of antennae brown to black, a few segments of basal flagella slightly brown, other portion pale; length of antennae ca. 2.0X head width; articulations with tiny hair-like setae (Fig. 3A).



FIGURE 1. Nymphal habitus of *Ochernova tshernovae*: **A**, male (dorsal view); **B**, female (dorsal view); **C**, female (ventral view); **D**, female (lateral view, arrow indicating the projection on prosternum and the tubercle on tergum X).

Mouthparts: Labrum: nearly rectangular, with smooth median emargination, dorsal surface covered with dense club-like setae and sparse hair-like setae; ventral surface with dense hair-like setae (Figs 2A, B). Mandibles: robust, slightly elongated; mesal margins nearby molar of both mandibles with a tuft of hair-like setae (Figs 2C, D). Outer incisor of left mandible with 4 blunt denticles, inner incisor with denticles, prostheca with a spur and a tuft of spines apically (Fig. 2C); outer incisor of right mandible with 3 blunt denticles, inner incisor with 2 denticles, apex of prostheca divided into a tuft of spines (Fig. 2D). Hypopharynx: lingua nearly rectangular, free margins with fine hair-like setae; outer margin of superlinguae nearly straight, with relatively long hair-like setae apically (Fig. 2E). Maxilla: galea-lacinia slightly elongated, with distinct 3 canines and 2 dentisetae; maxillary surface with sparse club-like setae; length ratio of three segments of maxillary palp= 1.0: 1.1: 0.5, segment I with club-like setae on both margins, segments II and III with hair-like setae, apex of segment III acute; cardo with club-like setae as well (Fig. 2F). Labium: glossae with blunt pointed apex, ventral surface densely covered with long hair-like setae; paraglossae with sharp apex, covered with dense long hair-like setae on both inner and outer surfaces; segment I of labial palp expanded, much broader than other segments; segment II broader than segment III; segment I and segment II subequal in length, segment III ca. 1/2 length of segment II; segment I with dense club-like setae, segment II with both club-like and hair-like setae on ventral surface and outer margin; segment III with 7–10 spine-like setae; mentum and submentum well-developed (Fig. 2G).

Thorax: generally light yellow, with a pair of sub-lateral brownish-black stripes along both sides. Pronotum expanded posteriorly, with distinct sub-antrolateral projections; mesonotum with waved lateral margins. Prosternum with a median spine-like projection at posterior margin (Fig. 1D). Thoracic surface with dense pale to white stout particle-like tubercles (Fig. 3B). Legs light ochre with brown longitudinal subapical band on femora; tibiae and tarsi of mid- and hindlegs darker than forelegs; inner margins of both tibiae and tarsi with bristles (Figs 3L–N). Length ratio of foreleg femur: tibia: tarsus = 2.1: 1.9: 1.0 (Fig. 3L); midleg femur: tibia: tarsus = 2.2: 2.2: 1.0 (Fig. 2M); hindleg femur: tibia: tarsus = 2.3: 2.7: 1.0 (Fig. 3N). Claws of all legs curved and smooth (Fig. 3K).

Abdomen: generally light ochre, with dense pale particles dorsally; terga with a pair of sub-lateral longitudinal stripes; sterna grey to black, usually progressively blacker from anterior to posterior, with a pair of indistinct sub-lateral markings too; tergum II with median projection, terga VI–VIII with median ridges, they extended into tubercles at posterior margin; tergum X with median ridge too, it expanded dorsally, forming an anteromedian tubercle (Fig. 1D); a semi-circular lobe-like projection between tergum X and terminal filament (=paracercus) (Fig. 3C); segments V–IX with inconspicuous posterolateral projections, progressively larger from anterior to posterior (Fig. 3C). Abdominal segment I without gills; dorsal plate of gills II operculate, with dense stout fine tubercles; two dorsal plates hinged together, each of them with a distinct nearly straight ridge (Figs 3D, E); ventral lamella small, divided into 4–5 long fringes (Fig. 3F). Gills III–V similar in structure, trapezoidal, with marginal fringes (Figs 3G–I); Gill VI single, long oval, with a row of fringes along free margin (Fig. 3J). Caudal filaments ca. 1.2x body length, with rows of spine-like setae at articulations, surface with slightly shorter setae (Fig. 3O).

Male imago (in ethanol): body length 8.5–10.0 mm, forewing 9.5–12.0 mm, hindwing 2.5–3.0 mm, caudal filaments 13.5–14.5 mm. Head and thorax generally brownish black, abdomen white, posterior end of abdomen reddish brown (Fig. 4A).

Head: compound eyes reddish-grey, distance between two eyes subequal to diameter of one compound eye in dorsal view. Frontal projection projected into spine-like structure. Length of antennae ca. 0.5x head width; scape pale, pedicel light reddish to reddish brown, flagella grey, base of flagella slightly widened; ocelli with reddish brown basal half but pale apical half, lateral ocelli slightly larger than median one (Fig. 5A).

Thorax: brownish-black, sutural ommatation indistinct (Figs 5A, C); furcasternal protuberances yellowish-brown to grey, widely separated (Figs 5B, D).

Wings: transparent, stigmatic area pigmented by grey. Rs of forewings forked at midpoint between base and fork of MA; MA forked at middle, MP forked at basal 1/8; crossveins distinct, including those between C and Sc, Sc and R₁ (Fig. 5E). Hind wings ca. 0.3X forewing length, with acute costal projection at base; MA single, MP forked near middle (Fig. 5F).

Legs: forelegs with grey femora, reddish brown tibiae, and reddish tarsi; mid- and hindlegs pale (Figs 5I–K). Length ratio of femora: tibiae: tarsi of forelegs = 1.4: 1.3: 1.0, tarsal segments arranged in descending order of length: 2, 3, 4, 5, 1 (Fig. 5I). Ratio of middle femora: tibiae: tarsi = 2.7: 2.9: 1.0, ratio of middle tarsal segments I: II: III: IV: V = 0.8: 1.0: 1.0: 1.0: 2.5 (Fig. 5J). Ratio of hind femora: tibiae: tarsi = 3.1: 3.6: 1.0, ratio of hind tarsal segments I: II: III: IV: V = 1.0: 1.4: 1.6: 1.4: 3.0 (Fig. 5K). Foreleg with two blunt claws; mid- and hind legs with one acute and one blunt claw (Figs 5L, M).

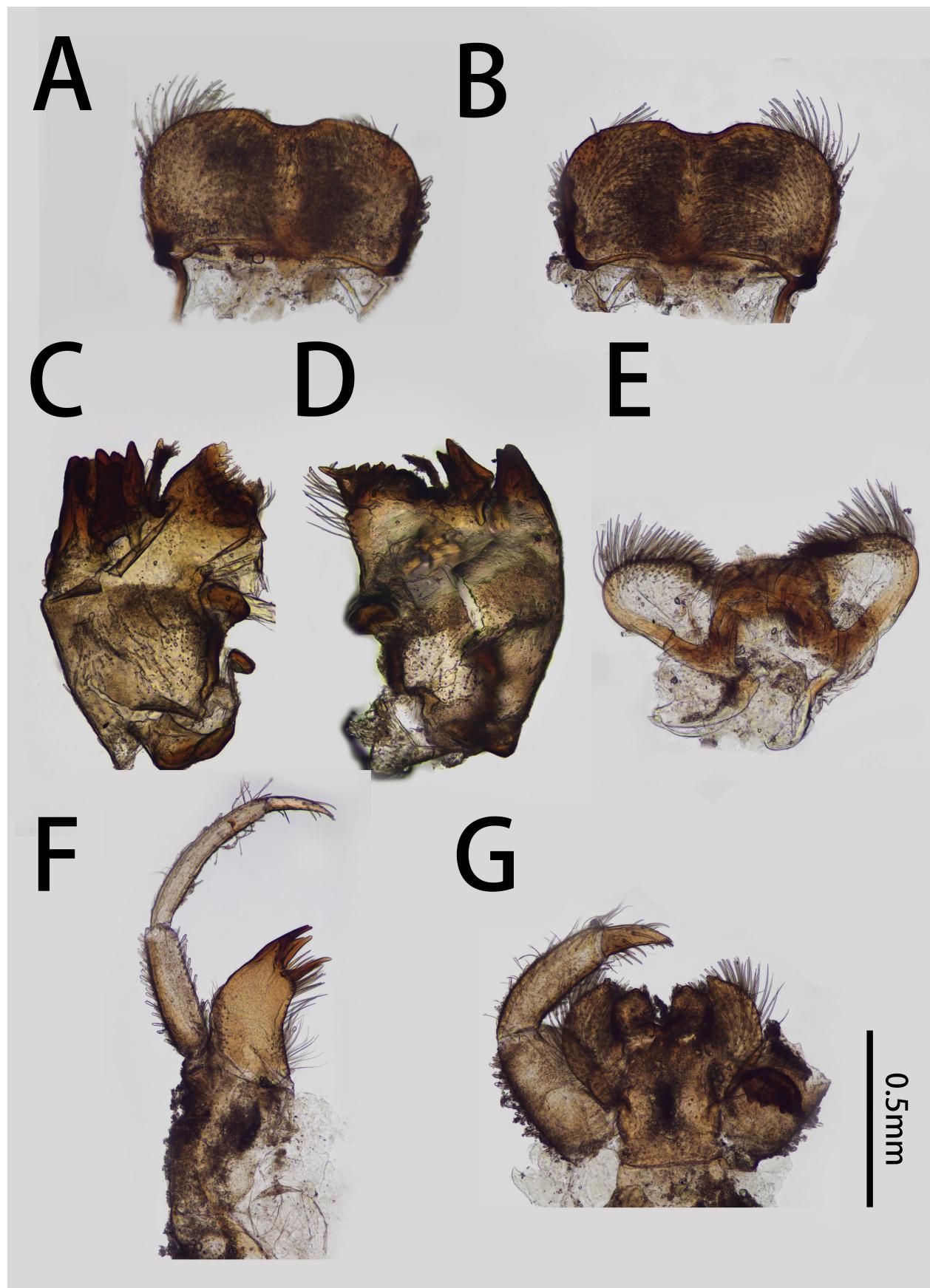


FIGURE 2. Mouthparts of *Ochernova tshernovae*: **A**, labrum (dorsal view); **B**, labrum (ventral view); **C**, left mandible; **D**, right mandible; **E**, hypopharynx; **F**, maxilla; **G**, labium.

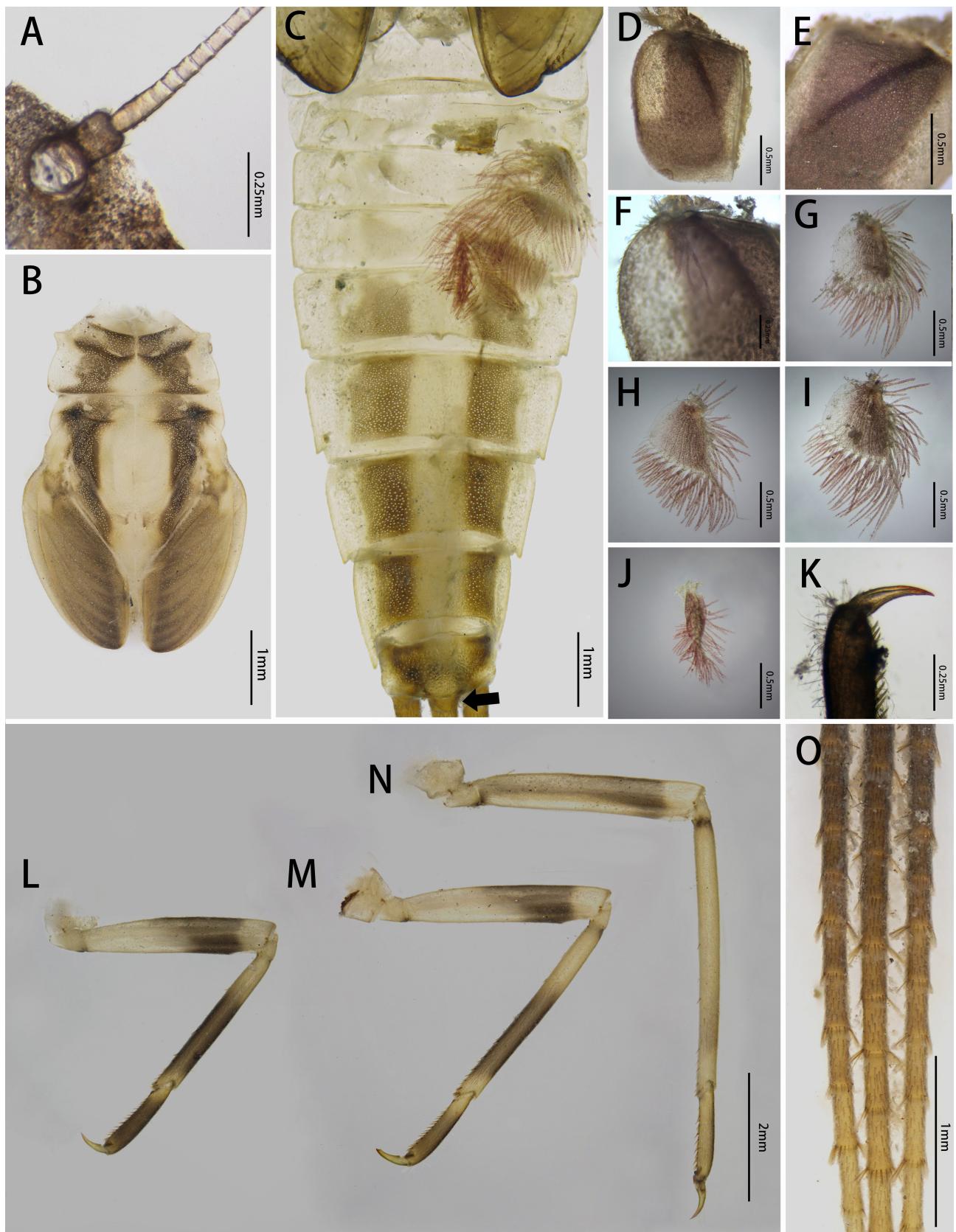


FIGURE 3. Structures of nymphal *Ochernova tshernovae*: **A**, antenna; **B**, thorax; **C**, abdominal terga (arrow indicating the projection behind tergum X); **D**, operculate gill II (dorsal view); **E**, operculate gill II (enlarged, dorsal view); **F**, filiform lamellae of gill II (ventral view); **G**, gill III; **H**, gill IV; **I**, gill V; **J**, gill VI; **K**, claw; **L**, foreleg; **M**, middle leg; **N**, hind leg; **O**, caudal filaments.

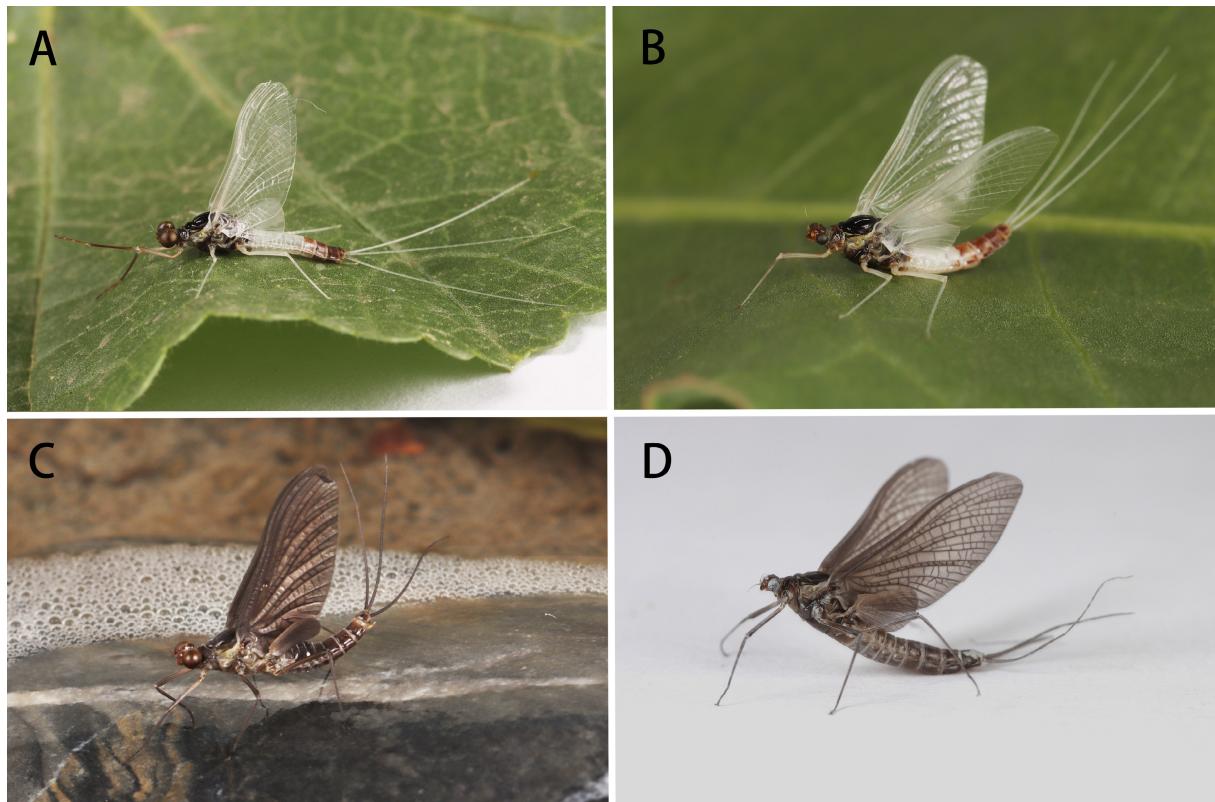


FIGURE 4. Imaginal habitus of *Ochernova tshernovae*: **A**, male imago; **B**, female imago; **C**, male subimago; **D**, female subimago.

Abdomen: generally translucent, tergum I obscure, terga II–V white, terga VI–X reddish-brown. Terga VII–IX with weak posterolateral projections, those of tergum IX longer (Fig. 5A). Sterna similar to terga in color, each with a pair of short dark lateral stripes; sterna IX–X darker (Fig. 5B). Genitalia: posterior margin of styliger plate concave; forceps pale ochre; segment I of forceps slightly broader than others, with distinct inner apical projection; segment II ca. 1.5x length of segment I, bending inward; segments III–IV ca. 0.15x length of segment I respectively, incompletely separated (Fig. 5G). Penes fused, bearing a distinct median notch (Fig. 5H). Caudal filaments with tiny setae on surfaces, terminal filament slightly longer than cerci (Fig. 4A).

Female imago (in ethanol): body length 10.0–11.0 mm, forewing 9.0–11.0 mm, hindwing 2.5–3.0 mm, caudal filament 9.0–11.0 mm. Coloration pattern similar to male imago (Fig. 4B). Wings similar to male (Figs 6A, B). All legs pale, length ratio of femora: tibiae: tarsi of forelegs = 3.7: 2.8: 1.0; length ratio of femora: tibiae: tarsi of middle legs = 2.7: 3.0: 1.0; length ratio of femora: tibiae: tarsi of hind legs = 3.4: 2.5: 1.0; all legs with one blunt and one sharp claw. Abdominal coloration similar to male, subgenital plate slightly expanded posteriorly, posterior margin of subanal plate nearly straight (Figs 6C, D).

Male subimago: body length 9.0 mm, forewing 9.5 mm, hindwing 2.5 mm, caudal filaments missing; coloration brownish-black, thorax darker (Fig. 4C). Wings brownish-grey. Legs dark grey; claws of forelegs all blunt (Fig. 5O), mid- and hindlegs bear one blunt and one sharp claw; all tarsal segments covered by microtrichiae (Figs 7A, B). Abdomen generally brown, tergum I obscure, other segments progressively darkening from light brown to brownish-black (terga VII–X darkest); tergum II with vestigial posteromedial tubercle (Fig. 5N); terga II–VI with pair of white submedian dots near anterior margin respectively, lateral margins of terga pale; sterna II–VIII with two pairs of submedian dots, anterior pair slightly longer than posterior pair. Caudal filaments covered with dense setae.

Female subimago: body length 11.0 mm, forewing 10.5 mm, hindwing 3.0 mm, caudal filament 9.0 mm (ca 0.8X body length). Coloration similar to male imago but paler (Fig. 4D). All legs bear one blunt and one sharp claw; all tarsal segments covered with microtrichiae (Figs 7A, B). Caudal filaments covered with dense setae; terminal filament slightly shorter than cerci.

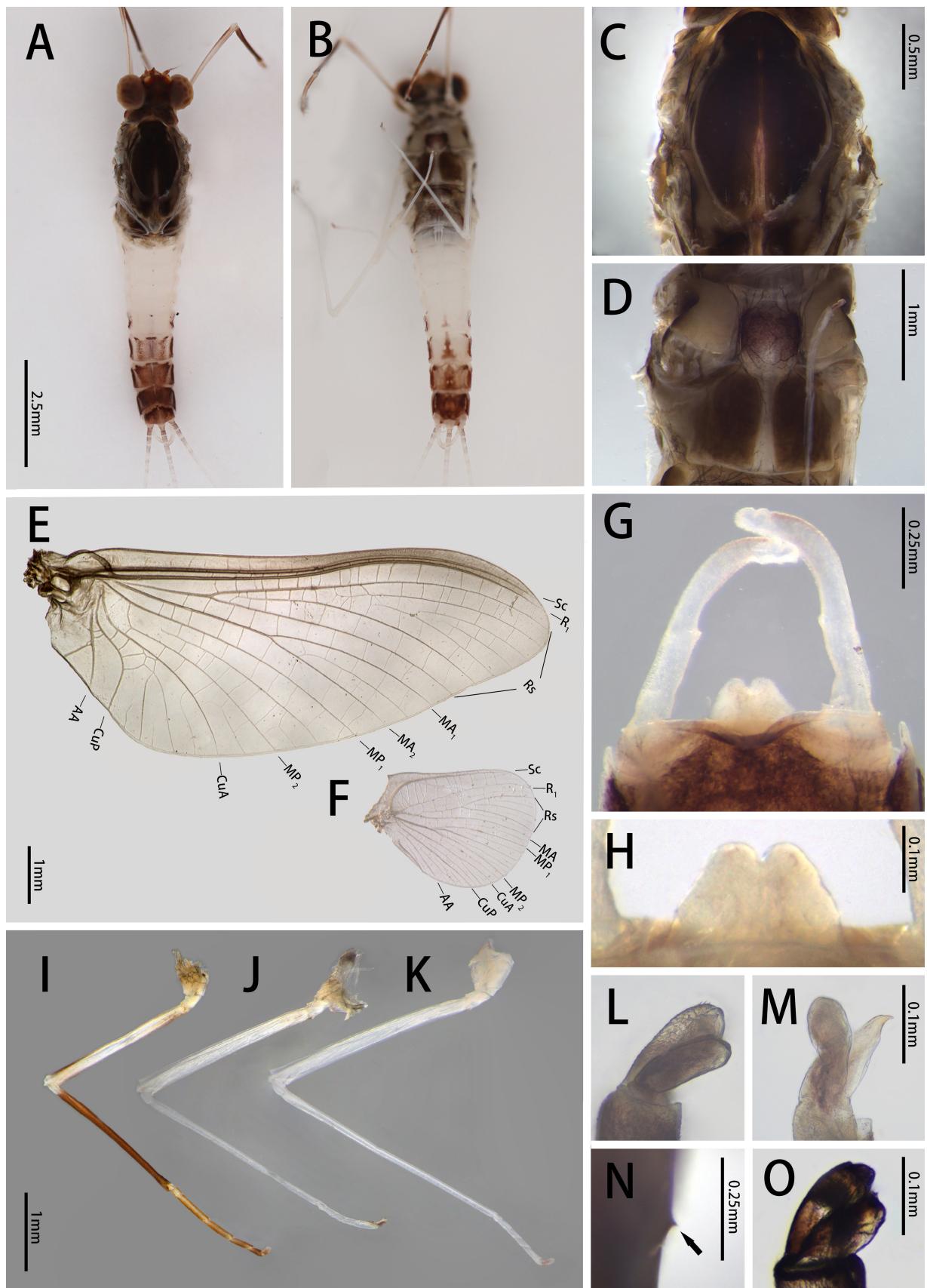


FIGURE 5. Structures of male imago and subimago of *Ochernova tshernovae*: **A–M** imago: **A**, dorsal view; **B**, ventral view; **C**, thorax (dorsal view); **D**, thorax (ventral view); **E**, forewing; **F**, hindwing; **G**, genitalia (ventral view); **H**, penis (dorsal view); **I**, foreleg; **J**, middle leg; **K**, hindleg; **L**, claw (foreleg); **M**, claw (middle leg); **N**, posteromedial tubercle on tergum II of male subimago (arrow indicating); **O**, claw of male subimago (foreleg).

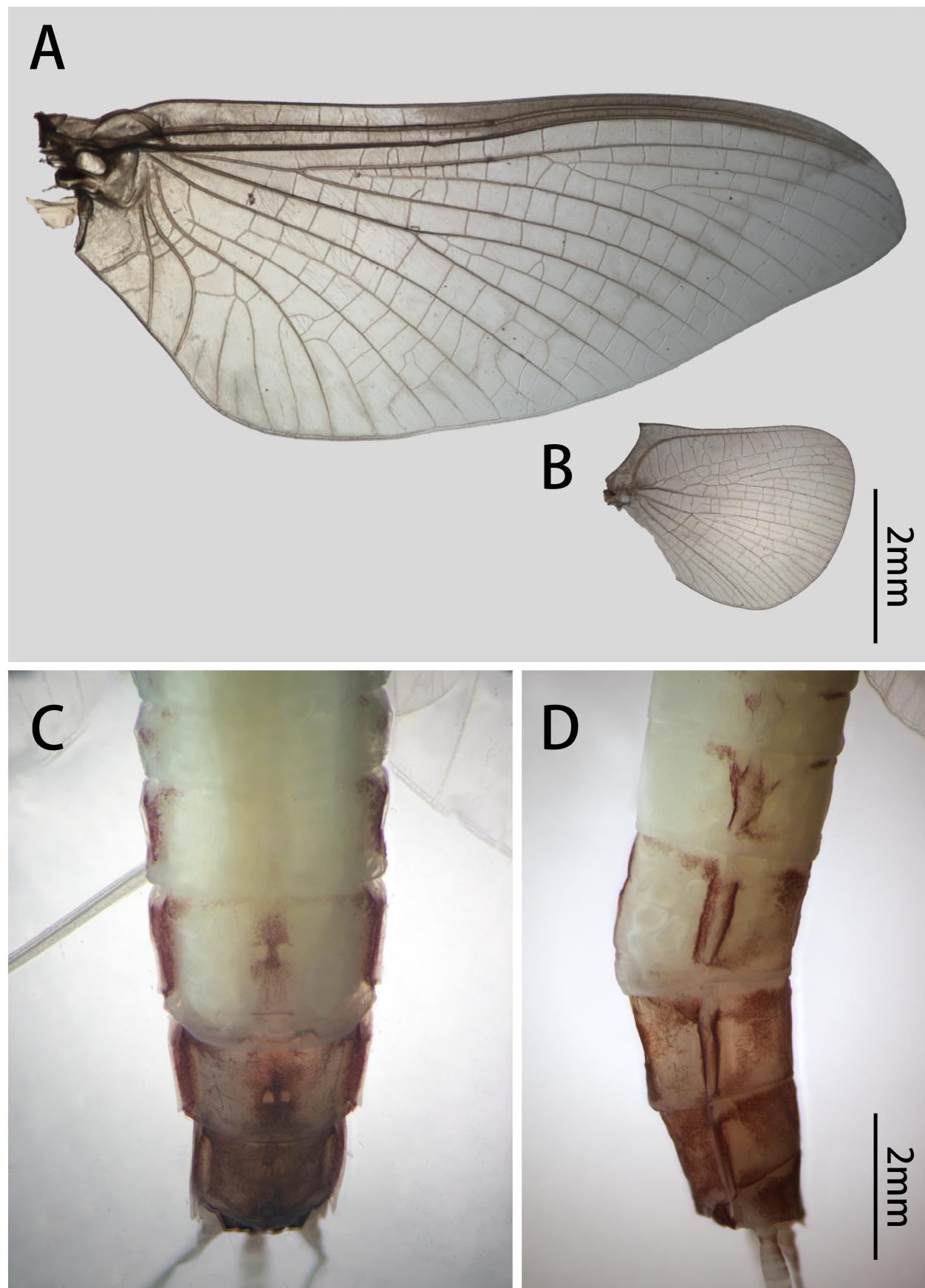


FIGURE 6. Structures of female imago of *Ochernova tshernovae*: **A**, forewing; **B**, hindwing; **C**, sterna V–IX (ventral view); **D**, sterna V–IX (lateral view).

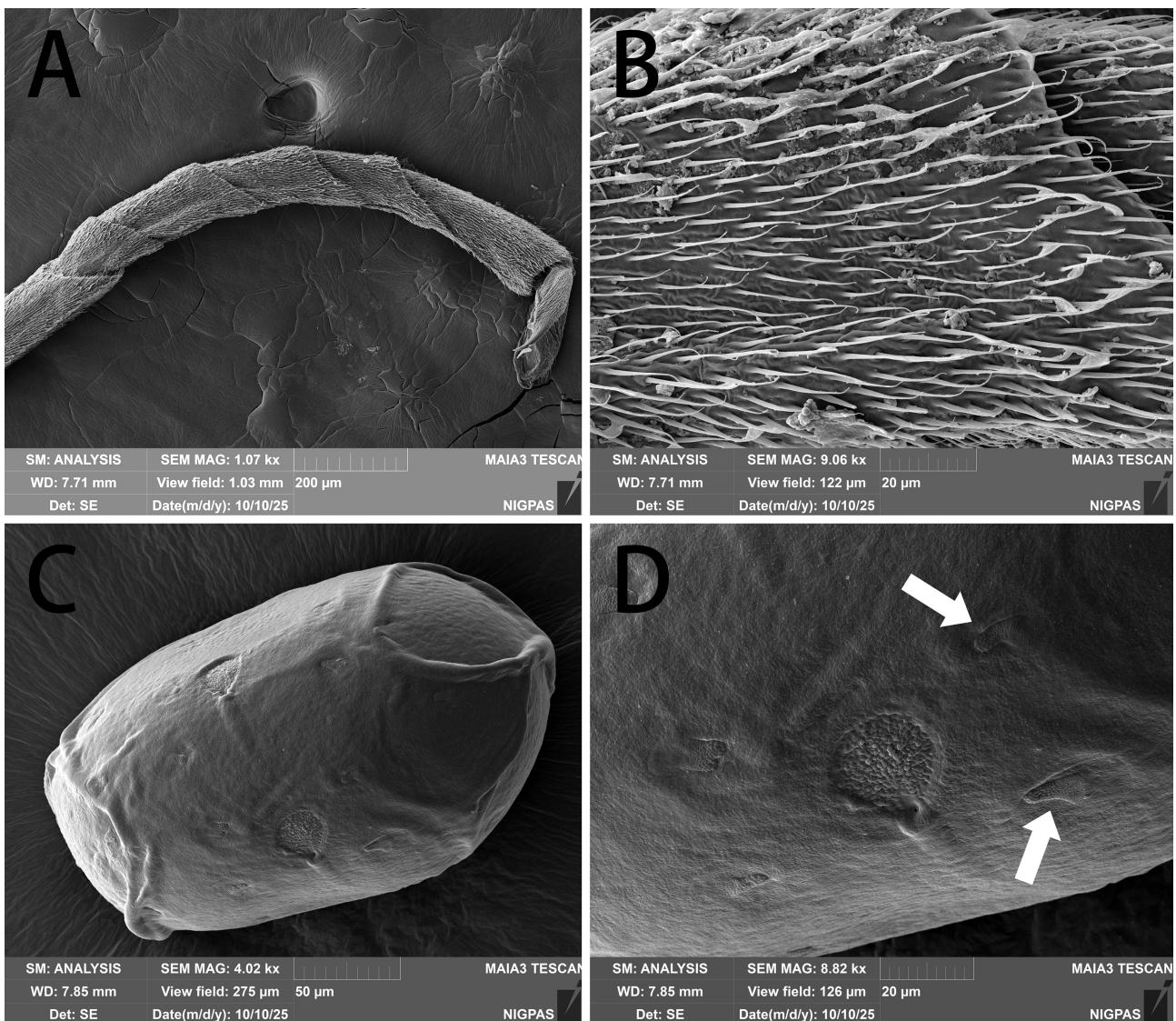


FIGURE 7. Subimaginal tarsi and eggs of *Ochernova tshernovae*: **A**, hindtarsus; **B**, microtrichiae of tarsal segment II of hindleg (female subimago); **C**, egg; **D**, detail view of surface of eggs (arrows indicating the finger-like projections).

Egg: oval, length 243–264 μm ; width 136–152 μm , finger-like projections 8–15 μm , scattered on surface, these projections represented as tiny tubercles. Micropyle near equator, diameter 18–21 μm , sperm guide round (Figs 7C, D).

Biology

The nymphs of this species were observed living in the bed of river that flow through sandy deserts with moderate water flow (0.3–1.0 m deep, ca. 1500–1650 m altitude), where the bottom is usually composed of gravel and fine sand (Fig. 8A). The nymphs have weak swimming ability, whose bodies are usually covered with dense muddy debris, and they like to cling to the plant debris or dead branches in the water (Fig. 8B).

The observed emergence occurs from late June to early July. The nymphs emerged between 12:30 and 13:30 local time. After the emergence, they fly onto the water surface and drift downstream with the water flow. During indoor rearing, it was observed that the subimagoes molted into imagoes 3 to 5 hours after the emergence. The entire molting process lasts about 10 to 13 minutes, and then the imagoes die between 19:00 and 21:30 in the same evening.

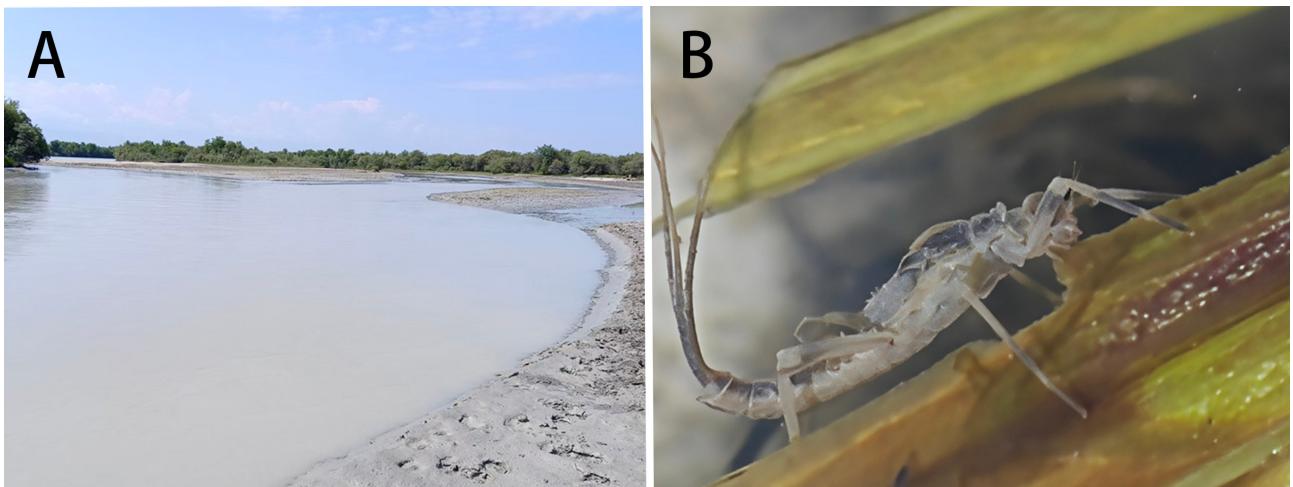


FIGURE 8. Habitat and alive nymph of *Ochernova tshernovae*: **A**, living river; **B**, feeding nymph.

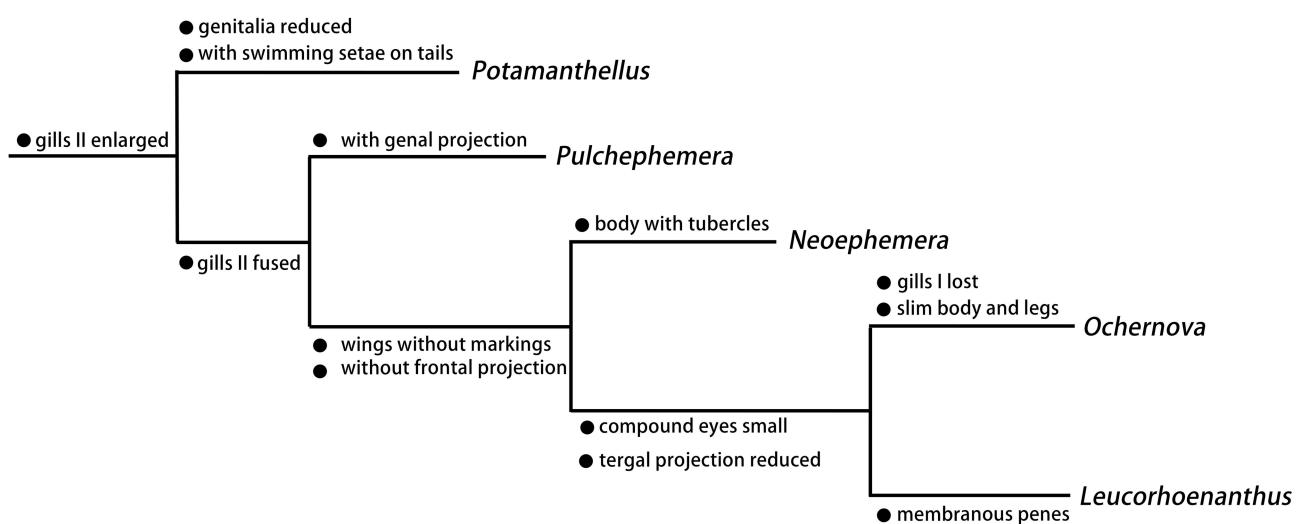


FIGURE 9. The phylogenetic relationship of five genera in Neoephemeridae (synapomorphies or automorphies of each clade are listed; modified from Ma & Zhou, 2021).

Diagnosis

The nymphs of *Ochernova tshernovae* can be easily recognized by their slim body and legs (Figs 1A–D, 8B), long caudal filaments (longer than body) and absence of gills I. They do not have any tubercles on dorsal head or thorax (in contrast to *Neoephemera*) (Fig. 3B) but with a spine-like structure on prosternum (Fig. 1D), with very small posterolateral projections of abdominal terga (Fig. 3C) (in contrast to *Neoephemera*, *Pulchephemera* and *Leucorhoenanthus*), possessing hinged gills II (in contrast to *Potamanthellus*, which has two separated gills II).

In male imagoes, the species *Ochernova tshernovae* can be differentiated from other neophtemerids by hyaline wings (Figs 5E, F) (in contrast to *Pulchephemera* and *Potamanthellus*, both of them have pigmented wings and body), well-developed genitalia (including penes and forceps) (Figs 5G, H) (in contrast to *Potamanthellus*, which has shortened forceps and penes and *Leucorhoenanthus*, with membranous penes), terminal filament subequal to cerci (Figs 4A, B) (in contrast to *Leucorhoenanthus*, which has reduced terminal filament) and relatively smaller compound eyes (distance between eyes subequal to diameter of eye) (Figs 5A, B) (in contrast to *Neoephemera*, *Pulchephemera* and *Potamanthellus*, whose eyes nearly contiguous) (Table 1).

TABLE 1. Comparison of five genera of Neoephemeridae (male imago, modified from Ma & Zhou 2021).

Characters	<i>Ochernova</i>	<i>Leucorhoenanthus</i>	<i>Neoephemera</i>	<i>Pulchephemera</i>	<i>Potamanthellus</i>
Ratio of distance between compound eyes/diameter of compound eyes	1.00	1.15	0.15-0.5	0.1	0.04-0.15
Forewing length (mm)	10.3-14	8-11	8-17	18-20	6-10
Coloration of wings	Without markings	Without markings	Without markings	With distinct markings	With distinct markings
Basal C-Sc crossveins of forewings	Not reduced	Reduced	Reduced	Not reduced	Not reduced
Shape of costal projection of hindwings	Acute	Acute	Acute	Rounded	Rounded
Claws of forelegs	Two blunt	Two blunt	One sharp, one blunt	Two blunt	Two blunt
Forceps	Well developed, 4-segmented	Well developed, 4-segmented	Well developed, 4-segmented	Well developed, 4-segmented	Vestigial, 3-segmented
Penis	Well developed	Membranous	Well developed	Well developed	Membranous
Median incision of penis	Small	Wide	Small	Small	Wide
Median caudal filament	Well developed	Vestigial	Well developed	Well developed	Vestigial
Length ratio of cerci/body	1.4-1.5	2.4	1.0-1.5	2.3-2.5	2.2-4.5

The eggs of the species *Ochernova tshernovae* have fewer, more indistinct finger-like projections on surface than counterparts of *Pulchephemera*, *Neoephemera*, or *Leucorhoenanthus* (Bauernfeind & Soldán 2013; Hollard *et al.* 2016; Ma & Zhou 2021) but it has more projections than *Potamanthellus* (unpublished data) (Figs 7C, D).

Phylogeny

Based on male *Ochernova tshernovae* adults described in this study and its nymphs and females, the genus *Ochernova* is regarded as the sister group of European genus *Leucorhoenanthus* (Fig. 9). Among five genera of the family Neoephemeridae, the genus *Potamanthellus* left away first due to its vestigial male forceps and penes. On the other hand, it has a series of plesiomorphies, such as pigmented wings, smooth nymph body and swimming tails, and the most importantly, its two independent gills II. In the remaining four genera, the *Pulchephemera*, which bears the most plesiomorphies (like colorful body and wings in adults, larger cephalic frons and abdominal posterolateral projections than others), is the basal one. The three other genera (*Neoephemera*, *Leucorhoenanthus* and *Ochernova*) form a clade because of their clean and transparent wings, well-developed male forceps, acute costal projection of hindwings, and smaller posterolateral projection of nymphal abdomen. In addition, the genus *Ochernova* seems close to the *Leucorhoenanthus* because both of them have relatively smaller abdominal median projections (Fig. 3C), smoother and flatter nymphal body than *Neoephemera* (Figs 1A–D). In imaginal stage, their relatively smaller eyes can group them together (Fig. 5A).

The genus *Ochernova* has at least five autapomorphies. (1) slim nymphal body and legs (Figs 1A–D); (2) long caudal filaments with spine-like setae but without hair-like setae (Fig. 3O); (3) acute maxillary and labial palpi (Figs 2F, G); (4) absence of gills I; (5) very small posterolateral projections of abdominal terga (Fig. 3C).

The species *Ochernova tshernovae* remains some plesiomorphies in both adults and nymphs. (1) distinct male genitalia (Figs 5G, H); (2) posterior end of imaginal abdomen with clear reddish pigments (Figs 5A, B); (3) in nymph, body of them is smooth (Figs 1A–D); (4) body is flat (Figs 1A–D); (5) terminal filament subequal to or slightly longer than cerci in both imaginal and nymphal stages. Those characters are similar to *Pulchephemera*. Geographically, it is also close to *Pulchephemera*. However, its nymphs definitely adapt to sandy and muddy rapid currents (Fig. 8A), and evolved to *Brachycercus*-like morphology.

Discussion

In most mayflies, the mid- and hindtarsi usually fused with tibiae. In *Neoephemera* and *Pulchephemera*, the basal tarsal segment is partially fused with tibiae in mid- and hindlegs (see Holland *et al.* 2016; Ma & Zhou 2021). The *Ochernova tshernovae* adults remain distinct five-segmented tarsi of all legs but the basal one is immovable from tibiae (Figs 5I–K). Kluge (2004) stated this condition is a “secondarily separated from tibia”. However, it is believed here that this character can be a plesiomorphy because most neoephemerids have this condition, including the plesiomorphic *Pulchephemera*.

The family Neoephemeridae is considered as a relative of Fossoriae by Ma & Zhou (2021), which is supported by the expanded frons and abdominal posterolateral projections of *Pulchephemera*. The *Ochernova tshernovae* nymphs, which are very specialized to cling on aquatic plants, still have this kind of character (Fig. 8B). In addition, the genitalia of both *Pulchephemera* and *Ochernova* are very similar to some *Ephemera* Linnaeus, 1758 species, such as *Ephemera shengmi* Hsu, 1937 (see Lei & Zhou 2024).

In most recent articles, the family Neoephemeridae is grouped with the Caenidae because of their similar gills of nymphs. The re-described nymphs and newly found male imagoes of *Ochernova* show that both families have some similar evolutionary trends, such as reduction or loss of gills I, nymph living in silt habitus or vegetations in water, and degenerated genitalia. Generally, the Caenidae can be roughly divided into two groups: *Caenis*-like and *Brachycercus*-like. Parallelly, in Neoephemeridae, the *Ochernova* nymphs slightly resemble to *Brachycercus*, and those of *Potamanthellus* live in similar habitus of *Caenis* and morphologically look like a big *Caenis*.

Previously, the genera *Potamanthellus*, *Leucorhoenanthus*, *Ochernova* and *Neoephemera* are found distributing disjunct. The report of *Pulchephemera* in 2011 and 2021 narrowed the *Potamanthellus* and *Ochernova* geographically. Furthermore, our present findings on *Ochernova* in China show that three neoephemerid genera in China (*Potamanthellus*, *Ochernova*, *Pulchephemera*) are connected in geographic distribution.

Because we did not find any molecular data of *Leucorhoenanthus* in GenBank, the molecular phylogeny of Neoephemeridae and its genera is not reconstructed here. With more information from Europe, the exact position of this family and its genera will be revealed from morphological and molecular evidences.

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