



<http://dx.doi.org/10.11646/zootaxa.3872.4.2>

<http://zoobank.org/urn:lsid:zoobank.org:pub:8B19A492-A820-4E19-9F33-2DA9E5AEDB76>

Morphological redescription and DNA barcoding of *Linevitshia prima* Makarchenko, 1987 (Diptera: Chironomidae: Diamesinae) from Amur River basin (Russian Far East), with notes on systematics of the genus

EUGENYI A. MAKARCHENKO^{1,3} & ALEXANDER A. SEMENCHENKO²

¹Institute of Biology and Soil Science, Far East Branch of the Russian Academy of Sciences, 100 let Vladivostoku 159, 690022 Vladivostok, Russia. E-mail: makarchenko@biosoil.ru

²Far Eastern Federal University, Suhanova St. 8, 690950 Vladivostok, Russia. E-mail: semenchenko_alexander@mail.ru

³Corresponding author

Abstract

Additions and corrections to the diagnosis of the genus *Linevitshia* for male adult, pupa and larva are given, and systematic position of the genus is discussed. Illustrated redescription of adult male and first description of 4th instar larva of *L. prima* Makarchenko from Amur River basin are provided. Comparison of data based on a new material with those of *L. yezoensis* Endo showed that the latter name is a junior synonym of *L. prima*. The species-specificity of *L. prima* COI sequences is analyzed and the sequences are presented as diagnostic characters—molecular markers of *L. prima*.

Key words: Diptera, Chironomidae, *Linevitshia*, taxonomy, redescription, DNA barcoding, Russian Far East

Introduction

The genus *Linevitshia* Makarchenko was established for *L. prima* Makarchenko, the species described from the southern part of Russian Far East (Makarchenko 1987). First, the genus was placed in the subfamily Podonominae, partly due to the incompletely developed wing vein R₂₊₃ in the freshly emerged specimens originally described. Later, however, K. Endo collected very similar midges in Hokkaido with distinct R₂₊₃. The male genitalia of these Japanese specimens differ slightly from *L. prima*, thus were described as *L. yezoensis* Endo, 2007, and consequently, the genus *Linevitshia* was provisionally transferred from the Podonominae to the Diamesinae (Endo *et al.* 2007). It was shown also that the genus *Linevitshia* and *Protanypus* Kieffer are closely related based on adult morphology, namely by complex of setae on pronotum (median and lateral Aps present) and mesonotum—Ac and Dc stripes connected in base, setae present on PAII and EII, and the presence of setae on alula of wing in adult males. This opinion was supported after examination of pupal morphology of *L. yezoensis* later (Makarchenko & Endo 2009).

In April 2014 we have got new materials consisted of adult males, pupae and larvae of *L. prima* from Amur River basin, including some males and larvae treated in DNA barcoding analysis. Characters of the *Linevitshia* larva, here described for the first time in the genus, confirm close relations to those of *Protanypus*, as was shown earlier for pupae and adults (Makarchenko & Endo 2009). Comparison of data based on a new material with those of *L. yezoensis* showed that the latter must be synonymized with *L. prima*. Additions and corrections to diagnoses for male adults, pupa and larva of the genus *Linevitshia* are also given, and systematics of the genus is discussed. The species-specificity of *L. prima* COI sequences is shown. These sequences could be used in future as diagnostic characters—molecular markers of *L. prima*.

TABLE 1. Lengths (in μm) and proportions of leg segments of *Linevitshia prima* Makarchenko, male (n = 4).

	fe	ti	ta ₁	ta ₂	ta ₃
P ₁	1168–1336	1376–1520	960–1069	451–518	301–351
P ₂	1152–1269	1269–1403	560–618	304–359	217–251
P ₃	1376–1503	1664–1787	832–935	464–534	272–301

continued.

	ta ₄	ta ₅	LR	BV	SV
P ₁	200–288	150–160	0.68–0.70	2.74–3.28	2.65–2.76
P ₂	142–150	134–150	0.44–0.46	3.67–4.12	4.20–4.34
P ₃	167–184	150–167	0.50–0.52	3.56–3.61	3.52–3.65

Results of DNA barcoding

The final alignment of the COI gene yielded 686 bp for 4 individuals of *L. prima* that were 4 haplotypes. The nucleotide composition of the studied sequences of *L. prima* COI gene fragments deviated from an equilibrium one, comprising 25.8 % of A, 36.6 % of T, 20.0 % of C, and 17.6 % of G. Total pairwise sequence divergence within *L. prima* ranged from 0.0015 to 0.0029, which is based on three nucleotide substitutions. All the substitutions were synonymous transitions (A-G) and observed only in the third codon positions. The HKY (Hasegawa, Kishino, & Yano 1985) model was the best-fit in describing the pattern of nucleotide variation in the *L. prima* COI sequences.

Average intergenus P-distance between *L. prima* and other genera of the subfamily Diamesinae showed the following results: *Potthastia* Kieffer (JF287767)—15.9%, *Pseudodiamesa* Goetghebuer (JF764760, JF764764, JF764771)—16.7%, *Pagastia* Oliver (JF287653)—17.2%, *Diamesa* Meigen (AB704934)—17.4%, *Sympotthastia* Pagast (JF288065)—18.2%. High differences between the groups can argue genus independence of *Linevitshia* (Ekrem *et al.* 2007).

Distribution and biology. East Palaearctic species, known from Japan, China and the Russian Far East (Kunashir Island, Primorye Territory, Amur River basin) (Wang 2000, Endo *et al.* 2007, Ashe & Connor 2009). Pupae and larvae from Amur River basin were collected in springs with water temperature 5–7°C, from gravel and stones covered with moss *Brachythecium rivulare* Bruch *et al.* Larvae in Kunashir Island were collected from small stones covered with moss in spring with water temperature 2°C.

Acknowledgments

We are grateful to Dmitry Palatov from Moscow University for making material available to us and to Valery Loktionov for help in preparing selected photographs.

Investigation was partly supported by grants from the Presidium Far East Branch of the Russian Academy of Sciences N 14-III-D-06-001 and N 12-I-P30-01.

References

- Ashe, P. & O'Connor, J.P. (2009) *A World Catalogue of Chironomidae (Diptera). Part 1. Buchonomyiinae, Chilenomyiinae, Podonominae, Aphroteniinae, Tanypodinae, Usambaromyiinae, Diamesinae, Prodiamesinae and Telmatogetoninae.* Irish Biogeographical Society & National Museum of Ireland, Dublin, 445 pp.
- Brundin, L. (1989) 4. The adult males of Podonominae—Keys and diagnoses. *Entomologica scandinavica*, 34 (Supplement), 23–36.
- Ekrem, T., Willassen, E. & Stur, E. (2007) A comprehensive DNA sequence library is essential for identification with DNA

- barcodes. *Molecular Phylogenetics and Evolution*, 43, 530–542.
<http://dx.doi.org/10.1016/j.ympev.2006.11.021>
- Endo, K., Makarchenko, E.A. & Willassen, E. (2007) On the systematics of *Linevitshia* Makarchenko, 1987 (Diptera, Chironomidae, Diamesinae), with the description of *L. yezoensis* Endo, new species. In: Andersen, T. (Ed.), *Contributions to the systematics and ecology of aquatic Diptera. A tribute to Ole A. Sæther*. Caddis Press, Columbus, pp. 93–98.
- Folmer, O., Black, M., Hoeh, W., Lutz, R. & Vrijenhoek, R. (1994) DNA primers for amplification of mitochondrial cytochrome c oxidase subunit I from diverse metazoan invertebrates. *Molecular Marine Biology and Biotechnology*, 3, 294–299.
- Hasegawa, M., Kishino, H. & Yano, T. (1985) Dating of the human-ape splitting by a molecular clock of mitochondrial DNA. *Journal of Molecular Evolution*, 22, 160–174.
<http://dx.doi.org/10.1007/bf02101694>
- Hebert, P.D.N., Cywinska, A., Ball, S.L. & deWaard, J.R. (2003) Biological identifications through DNA barcodes. *Proceedings of the Royal Society of London, B* 270, 313–321.
<http://dx.doi.org/10.1098/rspb.2002.2218>
- Makarchenko, E.A. (1985) *Chironomids of the Soviet Far East. Subfamilies Podonominae, Diamesinae and Prodiamesinae (Diptera, Chironomidae)*. DVNC AN SSSR Press, Vladivostok, 208 pp. [in Russian]
- Makarchenko, E.A. (1987) New or little known chironomids of Podonominae and Diamesinae (Diptera, Chironomidae) from the USSR. In: Sæther, O.A. (Ed.), *A conspectus of contemporary studies in Chironomidae (Diptera). Contributions from the IXth International Symposium on Chironomidae, Bergen, Norway, 1985. Entomologica scandinavica*, 29 (Supplement), pp. 205–209.
- Makarchenko, E.A. & Endo, K. (2009) The description of immature stages of *Linevitshia* Makarchenko and *Sasayusurika* Makarchenko (Diptera, Chironomidae, Diamesinae), with some remarks on taxonomy and systematics of these genera. *Euroasian Entomological Journal*, 8 (Supplement 1), 64–70.
- Oliver, D.R. & Roussel, M.E. (1983) *The genera of larval midges of Canada. Diptera, Chironomidae. Insects and Arachnids of Canada, pt. II*. Research Branch, Agriculture Canada, Ottawa, 263 pp.
- Sæther, O.A. (1980) Glossary of chironomid morphology terminology (Chironomidae, Diptera). *Entomologica scandinavica*, Supplement 14, 1–51.
- Sæther, O.A., Ashe, P. & Murray, D.A. (2000) Family Chironomidae. In: Papp, L. & Darvas, B. (Eds.), *Contributions to a Manual of Palaearctic Diptera (with special reference to the flies of economic importance). Vol. 4. Appendix A. 6*. Science Herald, Budapest, pp. 113–334.
- Tamura, K., Peterson, D., Peterson, N., Stecher, G., Nei, M. & Kumar, S. (2011) MEGA5: Molecular Evolutionary Genetics Analysis using Maximum Likelihood, Evolutionary Distance, and Maximum Parsimony Methods. *Molecular Biology and Evolution*, 28, 2731–2739.
<http://dx.doi.org/10.1093/molbev/msr121>
- Wang, X. (2000) A revised checklist of chironomids from China (Diptera) In: Hoffrichter, O. (Ed.), *Late 20th Century Research on Chironomidae: an Anthology from the 13th International Symposium on Chironomidae*. Shaker Verlag, Aachen, pp. 629–652.