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Original Article

First reports on the genetic diversity and phylogenetic relationships of bathynellids (Malacostraca: Bathynellidae) from Lake Baikal

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ABSTRACT. Within the fauna of Lake Baikal, two species of bathynellids are known: *Bathynella baicalensis* Bazikalova, 1954 and *Baicalobathynella magna* (Bazikalova, 1954), both originally described from the southern basin of the lake. The authors' findings indicate that bathynellids are prevalent across all three basins of the lake, occurring on habitats such as methane seeps, mud volcanoes and hydrothermal vents. A comprehensive analysis of the COI (mtDNA) and 18S rRNA (nuclear DNA) gene fragments from bathynellids collected at tectonic fault sites in the northern basin of the lake revealed the absence of any representatives of the genus *Bathynella* Vejdovsky, 1882 among them. Instead, individuals identified as *Baicalobathynella* cf. *magna* comprise two genetic lineages at the species level, which diverged from a common ancestor approximately 7 Ma. The genus *Altainella* Camacho, 2020, whose representatives inhabit the interstitial hyporheic zone of the Onon river in Mongolia and Aktru mountain river of Altai, is designated as the closest relative of the Baikal bathynellids studied.

Keywords: *Baicalobathynella*, COI, 18S rRNA, Lake Baikal, phylogenetic relationship, divergence time

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1. Introduction

The order Bathynelacea represents one of the oldest crustacean lineages within the class Malacostraca. These organisms belong to highly specialized groups of stygobionts and interstitiobionts, mostly inhabiting cave, interstitial, ground, capillary, and phreatic waters of all continents except Antarctica, as well as the Canary Islands, Fiji, and New Caledonia. Due to their unique habitats, Bathynelacea are typically eyeless, small, and possess transparent, worm-like bodies with short, weak limbs covered in long, sensitive bristles.

Bathynelacea includes three families: Bathynellidae Grobben, 1905; Parabathynellidae Noodt, 1965; and Leptobathynellidae Noodt, 1965, comprising over 330 valid species and subspecies (Camacho et al., 2021). The family Bathynellidae is widespread throughout the world and includes 36 genera and 109 species (Camacho et al., 2021). In Russia and adjacent countries (ex USSR), 18 species and subspecies from five genera of the family Bathynellidae have been recorded: *Antrobathynella* Serban, 1966; *Baicalobathynella* Birstein & Ljovuschkin, 1967; *Bathynella* Vejdovsky, 1882; *Altainella* Camacho, 2020; and *Tianschanobathynella* Serban, 1993 (Birstein and Ljovuschkin, 1967; Turbanov, 2016; Camacho et al., 2021). In mid-century research, two species of bathynellids were discovered and described in Lake Baikal: *Bathynella baicalensis* Bazikalova, 1954 and *Baicalobathynella magna* (Bazikalova, 1954), differing in size: the length of *B. baicalensis* is 1.5–2 mm, and in *B. magna*, the largest representative of Bathynelacea, it varies from 2.1 to 3.4 mm; the number of setae on the pleopods (up to 12 in *B. magna* and 4–5 in *B. baikalensis*); a complete reduction of exopod of the antenna and the presence of two pairs of pleopods in *B. magna* (Birstein and Ljovuschkin, 1967). A unique feature of *B. magna* is that the morphological characters of this species do not fully correspond to any of the three families, and these characters are plesiomorphic for Bathynellidae (Birstein and Ljovuschkin, 1967; Drewes and Schminke, 2011; Camacho et al., 2021).

Bazikalova A.Ya. (1949; 1954) reports that the crustaceans she described were found in Listvenichny Bay in depth zones of 200–250 and 140–180 m, as well as at depths of 0.2 and 1440 m in the southern basin of

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Lake Baikal, but does not mention their cohabitation. Later, bathynellids were found in several areas of the deep-water zone (Takhteev et al., 1993) and interstitial at depths of up to 0.6 m in Bolshiye Koty Bay (south-western coast) (Weinberg and Kamaltynov, 1998; Timoshkin et al., 2011).

In the Lake Baikal basin, bathynellids have been repeatedly found in the upper reaches of rivers that are tributaries of the southern part of the lake (Chernaya, Bolshiye Koty, Sennaya, Goloustnaya) and in nearby pits filled with water. The crustaceans have been found in the Barguzin and Angara river basins, as well as in the mountain river Bely Irkut (Eastern Sayan, northern slope of the Munku-Sardyk ridge on the border with Mongolia) (Takhteev et al., 2000).

The diversity of bathynellid species in Lake Baikal and its basin likely exceeds what is currently known. However, due to their small size and reclusive lifestyle, gathering material is labor-intensive, leaving their fauna underexplored not only in Eastern Siberia but also in Central Asia as a whole.

In recent decades, comprehensive studies of the world fauna of Bathynellacea have been conducted using the molecular genetic markers 18S rRNA and COI (Camacho et al., 2013; 2020; 2021; Ji, 2024, etc.), which makes it possible to compare Baikal bathynellids with representatives of other faunas, as well as to obtain a more complete pattern of their biodiversity and evolutionary history. The 18S gene is most often used to solve systematic problems at the high taxa level (genera, families) due to its high conservatism and, generally, low genetic differentiation between species (Tang et al., 2012). The molecular genetic marker COI is widely used to separate species, including cryptic ones, and for DNA barcoding (Antil et al., 2023; Kabus et al., 2023).

This study aims to investigate the genetic polymorphism and analyze phylogenetic relationships of Baikal bathynellids with related taxa through comparison of nucleotide sequences of COI (mtDNA) and 18S rRNA (nuclear DNA) gene fragments, contributing to a deeper understanding of this intriguing and enigmatic crustacean group.

2. Material and methods

Bathynellids were collected in 25 meiobenthic samples in the summer periods from 2009 to 2023 using a dredge, a bottom grab, a box grab and a remotely operated underwater vehicle (RovBuilder) (Table 1, Fig. 1A) in places of tectonic faults, including areas with gas and thermal water outlets.

The animals were photographed under an Olympus BX53 light microscope using an Olympus SC180 digital camera.

Some of the material was used for molecular genetic studies. DNA was extracted from the entire animal body according to the protocol described by Doyle and Dikson (1987). The amplification conditions and primer structure are presented in Table 2. The amplicons were analyzed by electrophoresis in 1% agarose gel. The bands of the expected size were excised and purified according to T. Maniatis (1982). Sequencing was performed on a NANOFOR 05 genetic analyzer using the Brilliant Dye Terminator (v.3.1) Sequencing kit (NimaGene, Holland). The nucleotide sequences were edited and aligned using the BioEdit program (Hall, 2011).

A median-joining network between COI haplotypes was constructed using the Network v.10 (Fluxus Technology Ltd. 2008), which is freely available on the website: https://www.fluxus-engineering.com/sharenet.htm.

Phylogenetic analysis based on the 18S rRNA gene fragment was performed using the web version of the IQ-TREE v.1.6.8 program (Nguyen et al., 2015), available at: http://iqtree.cibiv.univie.ac.at/. The most suitable model of molecular evolution was selected using the Model Finder module integrated into IQ-TREE (Kalyaanamoorthy et al., 2017). Branch supports were estimated using the ultrafast bootstrap approximation (Minh et al., 2013), as well as the approximate Bayes test (Anisimova et al., 2011). For phylogenetic analysis and calculation of genetic distances, 17 COI sequences and 28 18S gene fragment sequences belonging to different species of the Bathynellidae and one species of the Leptobathynellidae, which we used as an out-

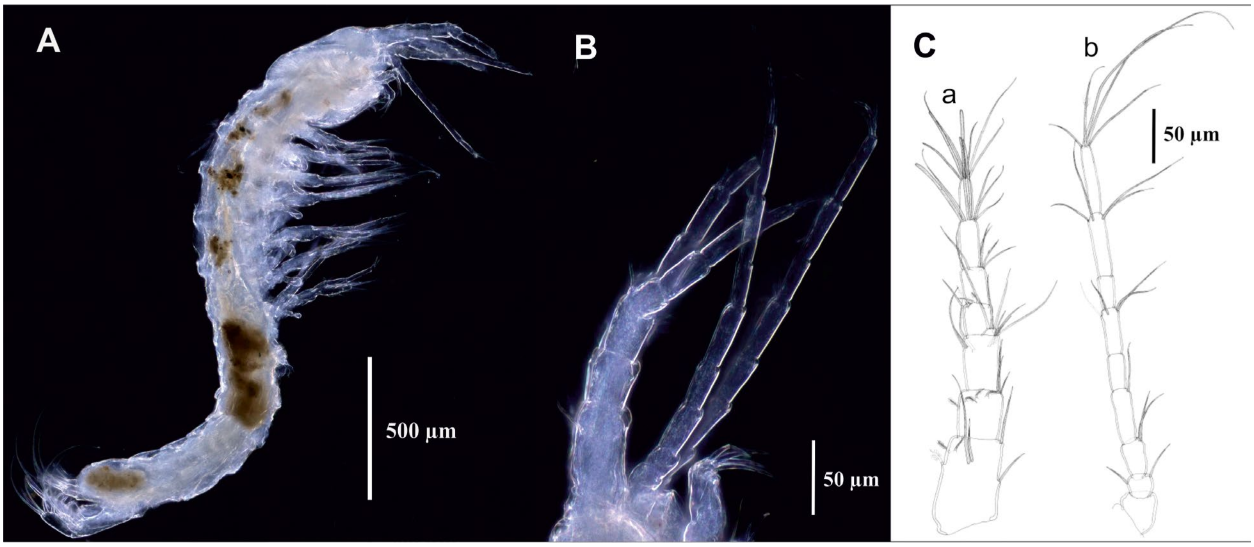

Fig.1. A – photo of *Baicalobathynella* cf. *magna* (Bazikalova, 1954); B – photo of antennae of *B.* cf. *magna* (**A** and **B** – photo by Enushchenko I.V.); **C** – drawing of antennae (a – antennula, b – antenna), adapted from Drewes and Schminke (2011).

Table 1. Sampling locations of Bathynellinae specimens in Lake Baikal.

Date of sample	Место сбора	Depth, m; sampler	Coordinates, N.L., E.L.	Тип грунта
27.09.2009	**Krasniy Yar,** methane seep, 1 km from the coast	340 m, bottom grab "Ocean"	52°24,5496 N, 109°53,4354 E	Oxidized brown silt, small pebbles, crushed rock and large stones, sand. Dump.
02.10.2009	**Elokhin-Davsha** section (center)	857 m, bottom grab "Ocean"	54°44,697 N, 109°05,857 E	Oxidized brown silt, layer about 7 cm, contains inclusions of hydrotroilite.
03.10.2009	**Academician Ridge,** mud volcanoes	389 m, bottom grab "Ocean"	53°62994 N, 108°12172 E	Oxidized silt, 25 cm thick.
17.06.2010	**Bolshoye Goloustnoye**	93 m, bottom grab "Ocean"	52°00,2084 N, 105°21,6354 E	Reduced silt.
03.07.2015	**Mud volcano Malenkiy (Bolshoye Goloustnoye area)**	1393 m, box corer	51°92,3788 N, 105°63,1519 E	Oxidized silt 2-3 cm thick, iron-manganese concretions, diatomaceous (from senerda) detritus.
03.07.2015	**Posolskaya Banka**, methane seep	500 m, box corer	52°03,5767 N, 105°84,3775 E	Oxidized silt 0.5 cm, blue clay without inclusions or layering.
04.07.2015	**Academician Ridge,** near gas hydrate sediment	531 m, box corer	53°400148 N, 107°891083 E	Oxidized silt layer 1.5 cm, then very soft floating blue silt, many veins of hydrotroilite.
05.07.2015	**Academician Ridge**	536 m, box corer	53°399337 N, 107°891467 E	Oxidized silt layer 3 cm, then very soft floating blue silt, there are layers of hydrotroilite, iron-manganese concretions.
29.06.2016.	**Kukuysky Canyon,** mud volcano K-2	939 m, box corer	52°589665 N, 106°770625 E	Oxidized silt layer 3 cm, restored blue silt without inclusions.
29.06.2016.	**Kukuysky Canyon,** mud volcano K-2	939 m, box corer	52°589613 N, 106°770332 E	Oxidized silt layer 5 cm, reduced blue silt and iron-manganese concretions.
01.07.2016	**Gorevoy Utyos Cape,** oil-methane seep	885 m, box corer	53°304265 N, 108°391662 E	Oxidized silt layer 1 cm, in the lower layers of restored silt, oil seepage.
03.07.2016	**Begul**	1169 m, box corer	52°728098 N, 106°586247 E	Small oxidized layer, gray sulfur crystals.
03.07.2016	**Begul**	1169 m, box corer	52°728902 N, 106°587093 E	Iron-manganese concretions, smell of hydrogen sulfide.
30.06.2017	Seaward of **Izhimey** Cape	1630 m, box corer	53°16542 N, 107°993352 E	Oxidized silt layer 1-1.5 cm, restored blue silt with iron-manganese concretions and sulfur crystals.
30.06.2017	Seaward of **Izhimey** Cape	1630 m, box corer	53°275108 N, 107°953110 E	Oxidized silt layer 3 cm, restored blue silt with iron-manganese concretions.
04.07.2017	Hydrothermal vent **Frolikha**	330–374 m, dredge	55°517062 N, 109°804481 E → 55°520457 N, 109°792553 E	Sand, brown-orange diatomaceous silt, plant-wood detritus.
04.07.2017	Hydrothermal vent **Frolikha**	413 m, box corer	55°521705 N, 109°776540 E	Dark brown oxidized silt layer 1-2 mm, then homogeneous greenish silt.
18.07.2018	Near the **Babushkin** town, bubble gas discharge	39 m, box corer	51°749587 N, 105°833133 E	Coarse and small sand.
20.07.2018	Near the **Baikalsk** town	375 m, box corer	51°538720 N, 104°197605 E	Oxidized silt layer 2-3 cm, restored silt is grey-black, with mica and clay crusts, smell of hydrogen sulfide.
19.06.2021	**Academician** Ridge	513 m, Ekman-Burge bottom grab	53°26,260 N, 107°53,812 E	Much oxidized silt, reduced blue silt with layers of hydrotroilite.

Date of sample	Место сбора	Depth, m; sampler	Coordinates, N.L., E.L.	Тип грунта
*10.07.2021	**Academician Ridge** (near Bolshoy Ushkany Island)	517 m, Ekman-Burge bottom grab	53°26,260 N, 107°53,812 E	Oxidized silt, fine sand.
*02.07.2023	North of **Shartlay** Cape, a ledge near a cliff	58,5 m RovBuilder RB-300	53°56,408 N, 108°11,775 E	Silted gray coarse-grained sand.
*08.07.2023	**Sosnovskaya Banka** (the northernmost point of the **Academician Ridge**)	449 m, Ekman-Burge bottom grab	54°08,836 N, 109°25,046 E	Oxidized silt and reduced silt, some sand.
*08.07.2023	**Academician Ridge** (the bank between the Upper Head of Svyatoy Nos peninsula and Ushkany Islands)	341–349 m, dredge	53°55,409 N, 108°53,135 E → 53°55,272 N, 108°52,866 E	Oxidized silt, a lot of copepod-diatom detritus.
*09.07.2023	**Academician Ridge** near Khoboy Cape	210-208 m, dredge	53°26,235 N, 107°48,546 E → 53°26,174 N, 107°48,345 E	Very fine, heavily silted sand, a lot of mica.

Note: * – sampling locations of bathynellids for which molecular data were obtained.

group, were additionally retrieved from the GenBank (Table 3).

The matrix of *p*-distance between 18S and COI nucleotide sequences was constructed using the Mega 11 (Tamura et al., 2021).

The divergence time of Baikal bathynellids was calculated using BEAST v.10.5.0 (Suchard et al., 2018). The analysis was performed using the Monte Carlo chain (MCMC) with length 50,000,000 steps. The convergence of the results was assessed using the Tracer 1.7 (Rambaut et al., 2018). Since there are no data on fossil Bathynellacea to calibrate the molecular clock, mean COI nucleotide substitution rate was utilized, calculated on the highest - 1.38% per Ma (Wares and Cunningham, 2001), and the lowest - 0.7% per Ma (Knowlton and Weigt, 1998) rates available for this marker, published for other crustaceans. The ucld.mean was set as a normal distribution with mean (Mean) = 0.0104 with standard deviation (Stdev) = 0.0016925. The ucld.Stdev was set as an exponential distribution with mean = 1.5.

3. Results

Bathynellids were found in all three basins of Lake Baikal, particularly in areas of methane bubble and jet discharge (Posolskaya Banka, Bolshoe Goloustnoye, Krasny Yar, Babushkin), on the oil-methane seep Gorevoy Utes, on active and extinct mud volcanoes (Malenkiy, Academician Ridge, Kukuysky Canyon), and also on the hydrothermal vent Frolikha. Remarkably, specimens were also discovered in the area of the maximum depths of the lake (1630 m) near Cape Izhimey (Table 1). The collected bathynellids inhabited soft oxidized bottom sediments of light brown and brown color with a thickness of 0.5 to 5 cm with an admixture of sand, pebbles, often ferromanganese crusts, detritus and layers of hydrotroilite. The underlying layer is usually represented by reduced gray and blue clays.

The collected bathynellids were identified as *Baicalobathynella* cf. *magna*, based on morphological features (body size from 2.0 to 2.5 mm and antenna structure) (Fig. 2). Most bathynellids lacked body coloration, exept for pink specimens found in the waters of Cape Izhimei, and red specimens found on the Academician Ridge (depth 517 m).

Nucleotide sequences of COI of 661 bp long and 18S rRNA of 1748 bp long were obtained from 11 individuals (nine colorless and two red) and deposited in GenBank under accession numbers PQ476139–PQ476149 for COI and PQ476288–PQ476298 for 18S. Analysis revealed six unique haplotypes for the COI

Table 2. Amplification conditions and primers used in this study.

Gene	Amplification conditions, 30 cycles	Primers	References
COI	DNA denaturation at 95°C – 40 sec (5 minutes on the first cycle), primer annealing at 50°C – 60 sec, nucleotide chain elongation at 72°C – 60 sec (10 minutes on the last cycle)	LCO1490 (f) 5’-GGT CAA CAA ATC ATA AAG ATA TTG G-3’ HCO2198 (r) 5’-TAA ACT TCA GGG TGA CCA AAA AAT CA-3’	Folmer et al. (1994)
18S	DNA denaturation at 95°C – 60 sec (5 minutes on the first cycle), primer annealing at 55°C – 60 sec, nucleotide chain elongation at 72°C – 60 sec (10 minutes on the last cycle)	SWAM18S (f) 5’-GAA TGG CTC ATT AAA TCA GTC GAG GTT CCT TAG ATG ATC CAA ATC-3’ SWAM18S (r) 5’-ATC CTC GTT AAA GGG TTT AAA GTG TAC TC ATT CCA ATT ACG GAG C-3’	Palumbi et al. (1996)

Table 3. List of taxa used for phylogenetic analysis and calculation of genetic distances with GenBank accession numbers and references.

Species name	18S GB#	COI GB#	References
Bathynella sp.	MN262083, MN262082, MN262079	MN258521	Camacho et al., 2020
Bathynella sp.	MF094716, MF094715	MF114308, MF114309	Camacho et al., 2018b
B. ruffoi	MF436214, MF436212	MF443327, MF443329	Camacho et al., 2020
B. cf. *ruffoi*	MF436213	MF443328	Camacho et al., 2020
Altainella calcarata	MN262081, MN262080	MN258522, MN258523	Camacho et al., 2020
A. mongoliensis	PQ037633, PQ037634		Ji, 2024
Vejdovskybathynella sp.	MF094713	MF114306	Camacho et al., 2018b
Vejdovskybathynella sp.	KC469515	KC469534	Camacho et al., 2013
V. caroloi	KC469525	KC469538	Camacho et al., 2013
V. vasconica	KC469521, KC469520	KC469535, KC469536	Camacho et al., 2013
Antrobathynella stammeri	MF094714		Camacho et al., 2018b
Gallobathynella sp.	KP999756		Camacho et al., 2017
G. coiffaiti	KP999760		Camacho et al., 2017
G. boui	KP999758	KP974147, KP974146	Camacho et al., 2020
G. tarissei	KP999753		Camacho et al., 2017
Hobbsinella sp.	MN262078, MN262077	MN258519	Camacho et al., 2020
H. edwardensis		MF443323	Camacho et al., 2018a
Paradoxiclamousella sp.	JX121235		
P. fideli	KC469524		Camacho et al., 2013
Parvulobathynella distincta (Leptobathynellidae)	MF436218		Camacho et al., 2020
Baicalobathynella cf. *magna*	PQ476288–PQ476298	PQ476139–PQ476149;	This study

gene fragment, whereas only two haplotypes were identified for the 18S gene fragment.

The median-joining network derived from COI mtDNA demonstrates distinct genetic structuring among bathynellids, forming two groups (Fig. 1B). The first group consists of colorless individuals of *B.* cf. *magna* from Cape Shartlay collected at a depth of 58.5 m, while the second group includes colorless and red specimens from the Academician Ridge near Cape Khoboy, Bolshoy Ushkany Island, the upper head of Svyatoy Nos, and Sosnovskaya Banka collected at depths ranging from 341 to 513 m. Average genetic distances between these groups were 8.8% to 9.6% nucleotide substitutions, with intragroup distances being negligible.

The phylogenetic tree based on 18S rRNA, with statistical supports for branch topology, is illustrated in Figure 3. Small clade supports within larger clusters are omitted for clarity. On the phylogenetic tree, Baikal bathynellids form a distinct clade within the subfamily Bathynellinae, clustering as a sister group to the European representatives of *Bathynella* Vejdovsky, 1882, and *Altainella* Camacho, 2020, from Altai and Mongolia.

Mean genetic *p*-distances distances for the family Bathynellidae, as shown in Table 4, indicate intergeneric distances based on the COI fragment ranged from 16.3% to 23.0%, whereas 18S distances were lower, between 1.7% and 6.7%. Intrageneric COI varied from 0.1% to 21%, and for 18S, from 0.1 to 1.9%. According to molecular data available in GenBank, the closest relatives of the Baikal bathynellids are representatives of the *Altainella* inhabiting Aktru mountain river (Altai Krai, Russia), including *Altainella calcarata* Camacho, 2019 (Camacho et al., 2020), and *Altainella mongoliensis* Ji, Camacho, Lee, and Min, 2024 found in the interstitial hyporheic zone of the Onon River basin (Mongolia) (Ji, 2024). The average *p*-distance between species of the genera *Baicalobathynella* and *Altainella* was 20.3% for COI and 3.1% for 18S molecular markers.

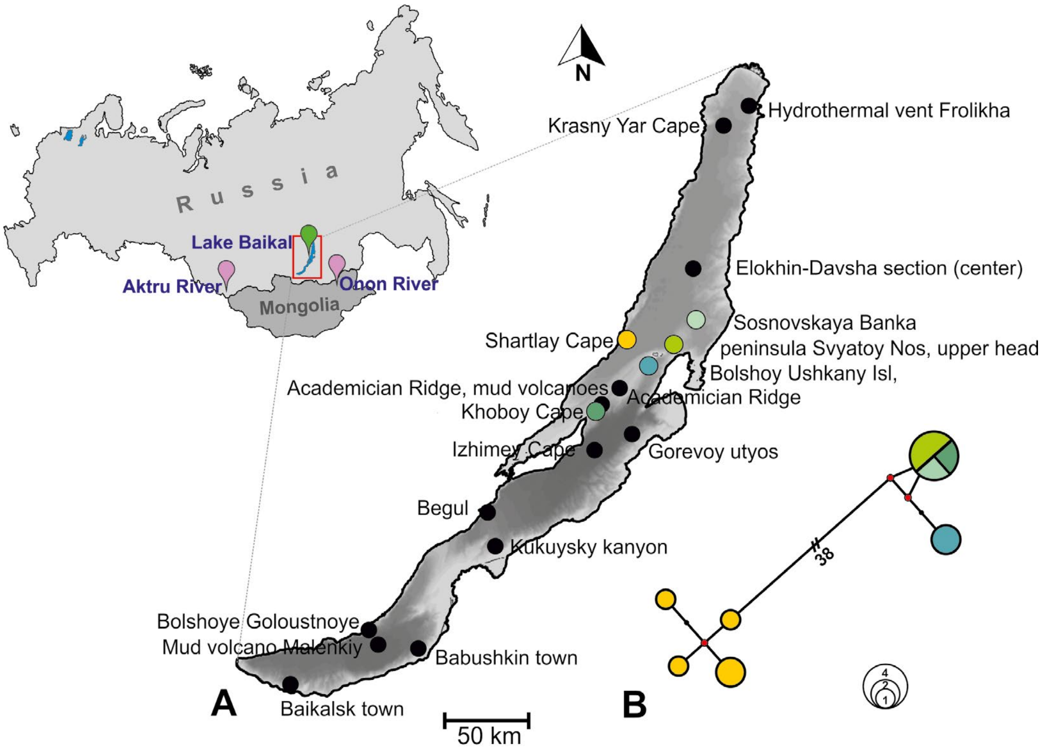

Fig.2. **A** – Map of sample collection. **B** – Median joining network constructed based on COI gene fragment sequences. Colors correspond to collection points. The size of the circle is proportional to the number of nucleotide sequences included in it.

The genus *Baicalobathynella* originated in the late Oligocene–early Miocene (around 20.2 Ma), while the divergence time estimated for the two genetic lineages of *B.* cf. *magna* falls on the late Miocene–early Pliocene (on average 7.2 Ma) (Fig. 4).

4. Discussion

The data obtained on locations of bathynellid finds indicate a mosaic distribution across the three basins of Lake Baikal. It was found that these organisms methane seeps, mud volcanoes, and hydrothermal vents within zones of active tectonic faults varying in length and orientation (Seminsky et al., 2022). Some sites, even without detected methane “flare” discharges, such as the waters of Academician Ridge devoid of mud volcanoes (Khlystov et al., 2000), Begul and Izhimey Capes (Bukharov and Fialkov, 1996), Shartlay Cape (Lunina et al., 2024), and Sosnovka Bay (Kazmin et al., 1995), are also associated with tectonic faults.

Analysis of the nucleotide sequences of mtDNA and 18S rRNA COI gene fragments from bathynellids in northern Lake Baikal fault zones revealed the absence of any representatives of the genus *Bathynella*. Instead, individuals attributed to *B.* cf. *magna* exhibited polymorphism in their COI gene sequences, forming two genetic lineages: “Shartlay” and “Academician Ridge and Sosnovka.” It should also be noted that the genetic differences between colorless and red individuals of this group do not exceed three substitutions per the COI gene fragment, suggesting intraspecific variability. Probably, mosaic distribution implies that active tectonic faults might influence the genetic structure of Baikal bathynellid.

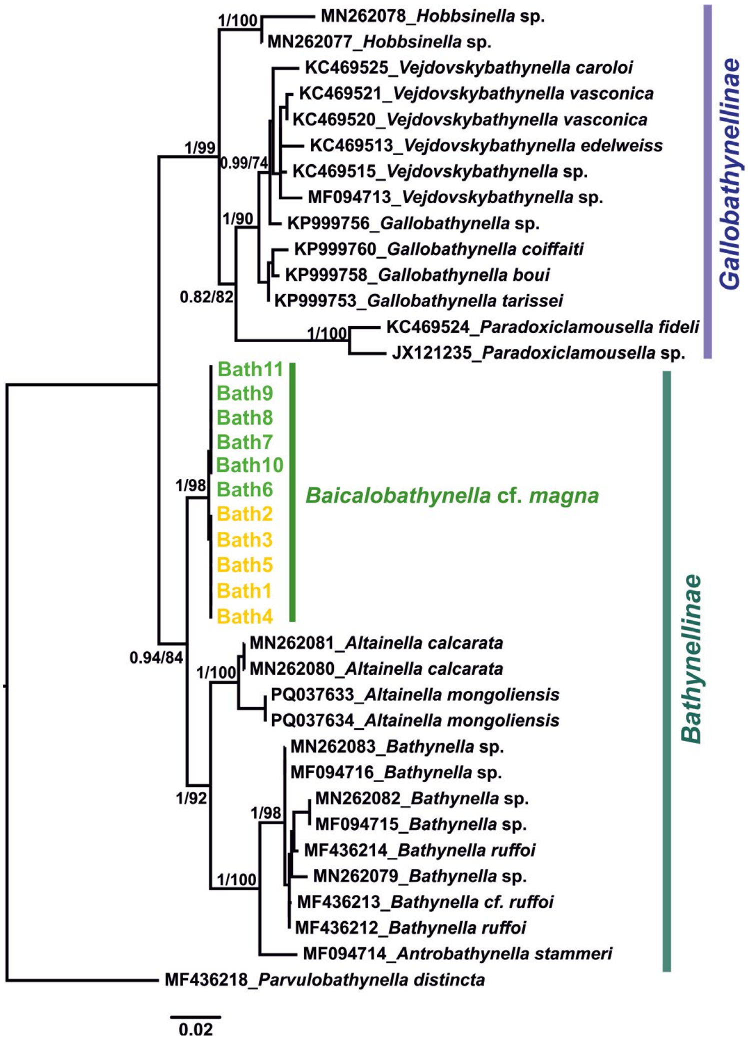

Fig.3. Phylogenetic tree based on 18S nucleotide sequences constructed for the family Bathynellidae. Sequences obtained during this study are highlighted in bright green (“Academician Ridge and Sosnovka”) and yellow (“Shartlay”). Bayesian/bootstrap supports are given at tree nodes.

Table 4. Matrix of mean pairwise *p*-distances between different genera of Bathynellidae. 18S and COI genetic distances are given above and below diagonal, respectively. Mean intragenus pairwise *p*-distances for the COI/18S gene fragment are in italics and shown on the diagonal. Minimal genetic distances are shown in bold.

	1.	2.	3.	4.	5.	6.
1. ***Baicalobathynella***	*5,0/0,1*	4,4	**3,1**	5,4	4,6	4,0
2. *Gallobathynella*	22,2	*0,1/1,0*	5,7	3,8	1,7	6,2
3. *Altainella*	**20,1**	19,7	*15^{*}/0,8*	6,3	6,0	4,0
4. *Hobbsinella*	23,0	21,0	20,5	*21,0/1,9*	4,2	6,6
5. *Vejdovskybathynella*	21,2	16,4	18,5	20,7	*14,0/1,2*	6,7
6. *Bathynella*	20,8	19,7	16,3	20,9	20,4	*13,0/0,6*

Note: * Since the nucleotide sequences for the species *A. mongoliensis* are not yet publicly available in GenBank, COI genetic distances for the genus *Altainella* were taken from Ji et al., 2024.

Molecular genetic data indicate two genetic lineages of *B.* cf. *magna* represent two independent species, with a significant COI genetic distance of 9.2% between them. Determining whether one corresponds to *B. magna* and the other represents a new species requires further study.

The divergence time of these lineages (or species) dated to the late Miocene–early Pliocene (Fig. 4), and probably aligns with the geological development of Lake Baikal, marked by partial geographical separation of the Northern and Central basins and the development of a single deep-water reservoir around 6–7 million years ago (Kuzmin et al., 2001; Mats et al., 2011). This assumption is based on the fact that the first lineage resides in the Northern Basin near Cape Shartlay, while the second is found in various parts of the Academician Ridge, which serves as the boundary between the Central and Northern Basins.

Representatives of the genus *Altainella* from Altai and Mongolia are identified as the closest relatives to the Baikal bathynellids studied based on 18S rRNA sequences from GenBank. The genetic distance between the Altai and Mongolian species of *Altainella* is 15% of nucleotide substitutions, whereas the distance between the Baikal genetic lineages “Shartlay” and “Academician Ridge and Sosnovka” is 9% nucleotide substitutions. It is important to note that the minimum geographical distance between Cape Shartlay and the Academician Ridge near Bolshoy Ushkany Island is approximately 25 km, which is nearly 100 times less than the distance between the habitats of two closely related *Altainella* species.

The estimated time of existence for the common ancestor of *Altainella* and Baikal bathynellids suggests their widespread distribution across southern Siberia and northern Mongolia until the late Oligocene–early Miocene (Fig. 4). This period was characterized by the onset of significant global changes within the Baikal rift zone (Logachev, 2003; Mats et al., 2011).

Thus, the bathynellids fauna of Lake Baikal is ancient in origin, and exceeds the two species described in the middle of the last century, is distributed in three

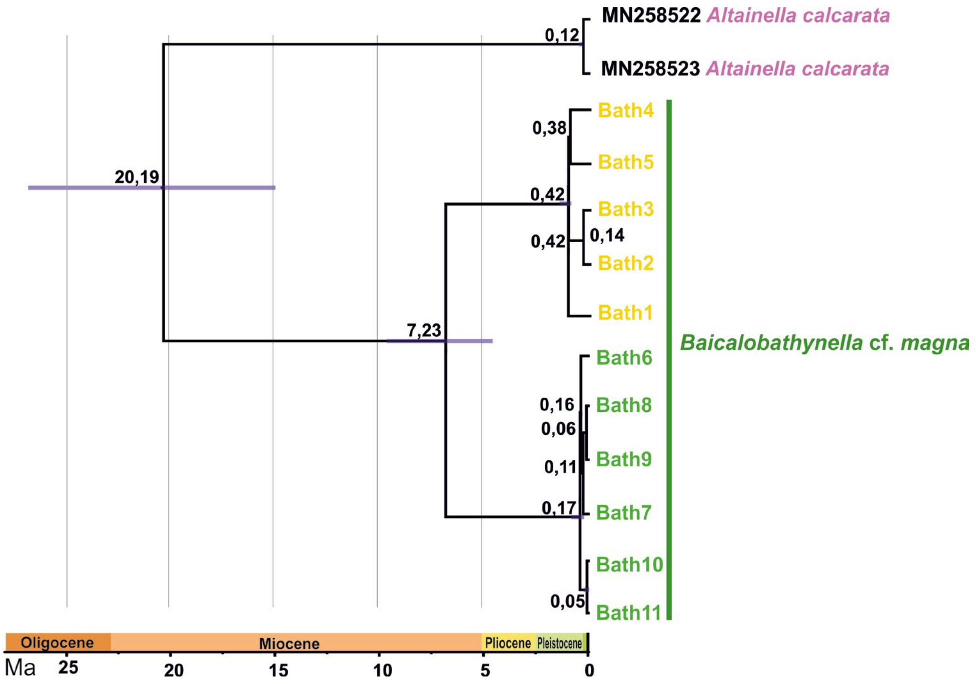

Fig.4. Divergence times derived from a relaxed molecular clock analysis based on COI sequences for *Baicalobathynella* and *Altainella*. Genetic lineage “Academician Ridge and Sosnovka” is bright green, and genetic lineage “Shartlay” is yellow. Blue bars at nodes indicate the 95% highest posterior density (HPD) intervals.

basins of the lake from the interstitial to maximum depths, including regions with gas and thermal water outlets. Future studies should focus on accurately characterizing the species composition of bathynellids in both interstitial and open waters of Baikal and its tributaries, employing a comprehensive approach integrating morphological and molecular genetics data.

5. Conclusions

The data on the locations of bathynellid finds indicate their mosaic distribution in three basins of Lake Baikal. Bathynellids sampled from tectonic fault sites have been identified as *Baicalobathynella* cf. *magna*. Molecular phylogenetic analysis suggests that their closest relative is the genus *Altainella* Camacho, 2020. Representatives of this genus are found in the interstitial water of the Aktru mountain river in Altai and the Onon river in Mongolia. The study revealed the presence of two distinct lineages of *B.* cf. *magna*; the average genetic distance between them, measured at 9.2% of nucleotide substitutions, is indicative of divergence at the species level. The evolution of genetic lineages aligns with the geological period of partial geographical separation between the Northern and Central basins of Lake Baikal during the formation of a single deep-water reservoir, occurring in the late Miocene–early Pliocene.

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Conflict of interest

The authors declare no conflict of interest

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