

## Microbial communities in gradient zones of Lake Baikal

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*Motivation and Aim:* Lake Baikal, the oldest lake on the planet (> 25 million years), is of tectonic origin, the bottom sediments of which are about 9 km thick. Distinctive features of Baikal hydrology and hydrochemistry (namely, its great depth, long ice-covered period, low temperatures of surface waters even in the summer, stable temperatures near the bottom, high oxygen concentrations at all depths, and low concentrations of nutrients) make this lake a very peculiar environment for its inhabitants [1], including microbial communities. Microorganisms populating these extreme conditions need to have a flexible metabolism and the enzymatic systems suitable for the specific conditions of Lake Baikal. This study aims to assess the diversity and metabolic potential of prokaryotes inhabiting ecotopes with gradients of oxygen, methane, temperature, light, and sources of carbon and energy.

*Methods and Algorithms:* Water and bottom sediments were sampled based on geological and geochemical data. A comprehensive approach was used for their study, including the analysis of lithological composition of bottom sediments, elemental composition and concentrations of major ions, methane, intensity of fluid flows, DNA analysis of native samples of water column and sediments, functional genes and genomes, culture media with various substrates and energy sources as well as analysis of bacterial and archaeal metabolism.

*Results:* To date, we have obtained a large dataset on the diversity and structure of microbial communities in the water column and bottom sediments of Lake Baikal, taking into account specific geochemical parameters (light, oxygen concentration, methane of different genesis, temperature, concentrations of major ions, elemental composition of pore water, intensity of fluid flows, and sources of carbon and energy), and determined dominant taxa of bacteria and archaea inhabiting different ecotopes. We expect to present the results of the study of microorganisms inhabiting gradients of temperatures, methane and some ions used as electron acceptors. Methane concentration and nitrogen sources determine the diversity and distribution of aerobic methanotrophs, and the rates of methane oxidation are comparable to those observed in other freshwater ecosystems [2]. The results of cultivation and analysis of functional genes and genomes indicated that Baikal methanotrophs differed from known species. Bacteria and archaea involved in anaerobic oxidation of methane (AOM) through metabolic pathways typical of freshwater bodies were present in the sedimentary strata of Lake Baikal. A comparison of the integral rates of aerobic and anaerobic methane oxidation (MO) in the sediments of Lake Baikal revealed that their values were comparable, and in some areas, AOM was much higher than the MO rate in the aerobic zone. Archaea of the family *Methanoperedenacea* and bacteria of the phylum *Methylomirabilota* that carry out AOM

through the denitrifying pathway are diverse among these microorganisms. Analysis of key anaerobic methanotrophy (*mcrA* and *pmoA* genes) indicated a wide diversity of groups of microorganisms involved in these processes in the sediments of Lake Baikal. In particular, we suggested an active role of the members of the class *Bathyarchaeia*, in whose genomes we detected the genes of the methane cycle and which have a wide range of phylotypes in Lake Baikal. Currently, we are conducting laboratory experiments with enrichment cultures of these archaea. Deciphering the genomes and identifying the metabolism characteristics in these microorganisms will significantly elucidate their influence on the composition of discharged gases and their likely participation in the processes of methane oxidation in the bottom sediments of Lake Baikal. In our study, special attention was paid to the assessment of the diversity and spatial distribution of methanogenic archaea in the sediments of Lake Baikal. We detected groups of microorganisms capable of methane generation through four different biochemical pathways. Their ability to produce methane from substrates such as H<sub>2</sub>/CO<sub>2</sub>, acetate and methylated (C<sub>1</sub>) compounds was experimentally shown. We found that hydrogenotrophic methanogens and ‘free-living’ members of the order *Methanomassiliicoccales*, which have the genetic potential to produce methane by reduction of methyl compounds and dihydrogen oxidation, showed the highest relative abundance. Analysis of communities from the aerobic and anaerobic zones of deep sediments and from the coastal zone in gradients of T, O<sub>2</sub> and H<sub>2</sub>S revealed the mass development of colourless sulfur bacteria of two genera, *Thioploca* and *Thiotrix*, represented by a wide range of ecotypes. These bacteria are a basis for the development of a wide range of microorganism species that form a natural biofilter that prevents the influx of hydrogen sulfide, methane and other compounds into near-bottom water from bottom sediments. The area of distribution of bacteria of the genus *Thiostrix* in Lake Baikal is very wide: they form massive fouling in the water of hot springs and in the littoral zone of the lake within the temperature range of +33 to +4 °C and are also part of epibiotic consortiums formed on the surface of the bodies of benthic animals (ostracods and amphipods) living in psychrophilic conditions. Lake Baikal, as a rift lake, is a promising object for studying the deep biosphere. Based on experimental and genomic data, we described for the first time the facultative lithotrophic thermophilic bacteria of the genus *Thermaerobacter* with metabolism atypical for this genus and evolutionarily distant from the type species. The acquired genomic abilities of the investigated strain allow it to function chemolithoautotrophically in deep sediments using inorganic donors and acceptors of electrons with the formation of oxidized compounds of sulfur, carbon and hydrogen necessary for the redox processes carried out by other members of microbial community in the conditions of the low-mineralized bottom sediments of Lake Baikal.

**Conclusion:** The microbiome of Lake Baikal is a rich source of new microbes, enzymes and bioactive compounds, differing from other microbes by the ability to manage their physiology during their survival under psychrophilic conditions with extremely small number of vital elements, sources of carbon (Corg and CH<sub>4</sub>) and energy (SO<sub>4</sub>, NO<sub>3</sub>, Fe<sup>+2</sup>, and Mn<sup>+2</sup>). Studying the characteristics of their metabolism using different approaches can become a key to understanding the role of individual taxa in the ecosystem of Lake Baikal and will provide a more targeted search for strains to use them in the future for biotechnological purposes.

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## References

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