

Postprint of Chinese Lake Microbiome Research

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Abstract

China possesses a large number of lakes with extensive distribution and diverse types, and is recognized as one of the countries with the richest lake ecological types worldwide. The diverse lake environments have fostered abundant microbial diversity and resources, yet our understanding of their structure and function remains largely limited to date. Through large-scale lake microbiome analysis and comparative genomic research, we can reveal the distribution patterns, formation mechanisms, and maintenance mechanisms of microbial diversity in China's lakes, elucidate the composition of functional genes in typical lake microorganisms, explore the adaptation mechanisms of lake microorganisms under extreme environmental conditions, discover novel microbial functions and metabolic pathways, and by comparing microbial functional genomes, environmental transcriptomes, and proteomes that drive global elemental cycling under different environmental conditions, uncover the processes, mechanisms, and environmental regulatory mechanisms by which microorganisms drive key elemental cycling in lake ecosystems. The acquisition of these research findings will not only facilitate a deeper understanding of lake ecosystem structure and function, promote interdisciplinary integration between geoscience and biology, and advance the sustained development of the discipline, but will also provide scientific and technological support for the conservation and sustainable utilization of China's lake microbial resources as well as the management and protection of lake ecological environments.

Full Text

Preamble

China Microbiome Initiatives

China Lake Microbiome Project*

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Abstract

China possesses a vast number of lakes with wide distribution and diverse types, making it one of the countries with the richest lake ecosystem diversity in the world. These diverse lake environments harbor abundant microbial diversity and resources, yet our understanding of their structure and function remains extremely limited. Through large-scale lake microbiome analyses and comparative genomic studies, we can reveal the distribution patterns, formation mechanisms, and maintenance processes of microbial diversity in Chinese lakes; elucidate the composition of functional genes in typical lake microbiomes; explore microbial adaptation mechanisms under extreme environmental conditions; and discover novel microbial functions and metabolic pathways. By comparing microbial functional genomes, environmental transcriptomes, and proteomes that drive elemental cycling under different environmental conditions, we can uncover the processes, mechanisms, and environmental regulatory principles by which microorganisms drive key elemental cycles in lake ecosystems. These research outcomes will not only deepen our understanding of lake ecosystem structure and function, promote interdisciplinary integration between earth sciences and biology, and drive continuous disciplinary development, but also provide scientific and technological support for the conservation and sustainable utilization of lake microbial resources and for the management and protection of lake ecological environments.

Keywords: lakes, eco-environment, microorganisms, microbiome, scientific program

Lakes represent important ecosystem types and natural complexes composed of lake basins, water bodies, and various substances contained within them. Lake ecosystems, formed through long-term interactions of various natural forces and continuously evolving, are geographic units of ecological systems. As components of the earth's surface system, lakes are distributed across all regions of the globe, exhibiting remarkable diversity in geomorphology, hydrophysics, climate, chemical composition, and biological community structure, which collectively give rise to highly diverse lake ecosystem types [1]. Lakes have distinct boundaries and function as relatively independent and complete ecosystems, primarily comprising environmental and biological subsystems that are inseparable, organically connected, and mutually interactive.

As a crucial component of the earth's surface system, lakes also exhibit openness, representing an important part of the terrestrial hydrosphere and participating in natural water and material cycles. Consequently, lakes not only provide water supply, flood regulation, climate moderation, fisheries, and tourism services, but also play vital roles in maintaining regional ecosystem balance and biodiversity conservation, offering high ecosystem service values that support economic and

social development [1]. Lakes serve as connection points where various spheres of the earth's surface system interact. At the watershed scale, various materials ultimately converge in lakes, where diverse physical, chemical, and biological processes occur. These processes significantly influence both lake environments and regional environmental ecology. Microorganisms are the key drivers of elemental cycling and ecosystem functioning in lakes and other aquatic systems, making microbiome research essential for revealing their structure and function and promising important scientific breakthroughs.

1 Significance of Lake Microbiome Research

1.1 Microorganisms as Key Drivers of Biogeochemical Cycles and Ecosystem Functioning in Lakes

Microorganisms constitute essential components of lake aquatic ecosystems and are the primary drivers of most elemental cycling processes, making them critical for understanding ecosystem structure and function [2]. Research on microbe-mediated elemental cycling processes has facilitated numerous major scientific discoveries. For example, studies on microbial iron reduction revealed that microorganisms can reduce solid metal oxides through extracellular electron transfer [3], which led to the development of microbial fuel cell technology based on electron transfer to external solid electrodes [4]. Iron-reducing microorganisms can produce filamentous nanowires with metallic conductivity [5] that transport electrons and, combined with inorganic metal substances and humus in sediments, form long-distance electron transfer channels [6-8]. Microorganisms in sediments thus utilize these media to form ordered electron transfer networks, generating bioelectric currents from their metabolic electrons. Through these bioelectric currents, spatially separated oxidation and reduction reactions can be synchronized, enabling microorganisms to efficiently participate in elemental cycling and pollutant decomposition in sediments. This provides scientific foundations for systematically regulating biogeochemical processes and pollutant transformation in aquatic systems through microbial extracellular electron transfer [9].

Additionally, research has shown that syntrophic microorganisms can transmit molecular signals through flagella [10], and horizontal gene transfer may occur between syntrophic microorganisms, potentially playing an extremely important role in microbial evolution. These discoveries have enriched our understanding of microorganisms' critical driving role in elemental cycling in lakes and other aquatic systems, promoted interdisciplinary integration between earth sciences and biology, advanced relevant disciplinary fields, and developed microbial-based pollutant reduction and enhanced degradation technologies.

1.2 Microbiome Research is Crucial for Revealing Lake Ecosystem Structure and Function and Promises Major Breakthroughs

Despite important discoveries in aquatic microbial research in recent years, lake microorganisms have long been treated as a “black box” due to methodological limitations. Overall, our understanding of lake microbial diversity, structure, and function has been extremely limited. It was not until the early 21st century that scientists basically recognized that lake microorganisms have unique flora and compositions [11]. However, to date, conservative estimates suggest that over 95% of lake microorganisms cannot be cultured in laboratories, and their physiological, biochemical, genetic, metabolic, and ecological functions remain unknown [12]. For instance, ammonia-oxidizing archaea discovered in the early 21st century are widely distributed in lake and river sediments, yet only a few strains have been cultured to date [13]. Moreover, microorganisms isolated under laboratory conditions differ substantially from in situ lake microbial communities.

Since the vast majority of lake microorganisms cannot be isolated and cultured, revealing their structure and function through new technologies has become a central challenge in lake microbiology research. Microbiome technology represents an important approach for studying environmental microbial structure and function without cultivation. The microbiome refers to the sum of all microorganisms and their genetic information in a specific environment. Sequencing and analyzing all genetic information (genes) of microorganisms in a particular environment can reveal not only microbial community structure but also predict functional gene composition and metabolic potential, facilitating the discovery of new microbial genes and metabolic pathways and promoting profound understanding of material transformation and cycling in the environment [14]. In 2004, Venter et al. [15] discovered through microbiome research that the number of new genes in the Sargasso Sea exceeded the total number of genes in international gene databases at that time, and subsequently achieved a series of important discoveries in marine systems using microbiome approaches.

Over the past decade, the continuous development of high-throughput sequencing technologies and declining sequencing costs have provided tremendous support for applying microbiome techniques to study lake microbial structure and function. For example, in surveys of microbial diversity in Tibetan lakes, bacterial diversity based on 16S rRNA gene sequencing of prokaryotic microorganisms was found to be extremely rich, with approximately 30% of sequences unable to be assigned to known taxonomic positions in databases, and their functions remain unknown [16]. Currently, lake microbial metagenomic studies are extremely rare worldwide, with relevant research conducted only in some cold salt lakes in Antarctica, yielding important advances in the biogeography of psychrophilic and halophilic archaea, mechanisms of microbial sympatric speciation, and microbial participation in carbon, nitrogen, and sulfur metabolism under low-temperature and high-salt conditions [17-19].

Overall, current understanding of lake microbiomes is extremely limited, lagging far behind marine microbiome and human microbiome research. The new microbial species and metabolic pathways involved represent only the tip of the iceberg of lake microbial resources, and our knowledge of microorganism-driven biogeochemical cycling in lakes is very limited. Clearly, revealing the mysteries of lake microbiomes and their roles in lake elemental cycling will be a tremendous challenge for lake science for a long time to come, while also representing a field with great potential for scientific breakthroughs.

2 Current Status and Strategies for China' s Lake Microbiome Research

Compared with terrestrial and marine ecosystems, lakes are ecosystems with relatively clear boundaries and stable structures, and different lakes have relative environmental specificity due to geographic isolation. China has numerous lakes (Figure 1 [Figure 1: see original paper]), with 2,693 natural lakes larger than 1.0 km² distributed across 28 provincial-level administrative regions, covering a total area of 81,414.6 km², approximately 0.9% of China' s total land area [20].

Chinese lakes exhibit tremendous diversity in morphology, distribution region, environmental heterogeneity, chemical composition, pollution level, and biological community structure. For example, salinity ranges from 0.1% to approximately 40%; pH ranges from 2 to 13; chlorophyll-a concentration, an important indicator of primary productivity, ranges from near zero to several thousand g/L; and lake elevation ranges from the East China Sea coast to glacial relics at approximately 6,000 m on the Tibetan Plateau (Figure 1). Therefore, China is considered to have the most complex and diverse lake types worldwide [20]. This extremely high environmental heterogeneity likely influences microbial evolution and differentiation, forming highly diverse microbial compositions and functions, many of which are likely novel or unknown. Consequently, conducting lake microbiome research in China has distinctive regional characteristics and promises important scientific discoveries and breakthroughs.

2.1 Scientific Objectives

The overall scientific objectives of China' s Lake Microbiome Project are to comprehensively elucidate the distribution patterns, formation mechanisms, and maintenance processes of microbial diversity in Chinese lakes; understand the distribution characteristics of microbial functional genes; reveal adaptation mechanisms of lake microorganisms under special environments (high salinity, low temperature, strong radiation, extreme pollution, etc.); discover novel microbial functions and metabolic pathways; and reveal the influence and mechanisms of environmental factors on microorganism-driven carbon, nitrogen, and phosphorus cycling in lakes by comparing functional genomes, transcriptomes, and proteomes under different environmental conditions. Additionally, based on in-depth understanding of lake microbiome structure and function and their

response and feedback mechanisms to lake pollution, we aim to explore new lake water quality and ecological improvement technologies from a microbial perspective. This will become another important scientific objective of China's Lake Microbiome Project.

2.2 Frontier Scientific Questions and Recommended Future Research Directions

2.2.1 Distribution Patterns and Formation Mechanisms of Microbial Diversity in Chinese Lakes Based on the distribution, origin, water environment, resource occurrence, and hydrological characteristics of Chinese lakes, combined with the macro-geomorphic feature of high elevation in the west and low elevation in the east and the climatic condition of humidity in the south and aridity in the north, we will focus on microbial diversity and distribution patterns in the five major lake regions of China (Figure 1). We will employ high-throughput sequencing and omics technologies to investigate the diversity, community structure, and distribution patterns of microorganisms in typical lakes, with emphasis on analyzing the relative roles of environmental filtering mechanisms and stochastic processes in lake microbial community assembly to reveal the formation and maintenance mechanisms of microbial diversity in Chinese lakes.

Based on revealed distribution patterns of lake microbial diversity, we will conduct studies on functional gene composition and microbial functional diversity in typical lakes using metagenomics and other techniques according to differences in microbial community composition and existing environmental gradients. We will analyze microbiome variations along environmental gradients, particularly to understand the core microbiome in lake ecosystems and the variable microbiome that changes along environmental factor gradients, and focus on analyzing the intrinsic relationships between these functional gene composition changes and environmental factor variations.

2.2.2 Mechanisms of Microbial Adaptation to Extreme Lake Environments and Novel Metabolic Pathways Some Chinese lakes, particularly those on the Tibetan Plateau, have extreme environmental conditions such as extremely high salinity, low temperature, low pH, strong radiation, and extreme pollution. Microorganisms in these habitats possess unique environmental adaptation and metabolic mechanisms that harbor valuable information for exploring the occurrence and evolution of microbial metabolic diversity. Using microbiome technologies, we can gain in-depth understanding of the mechanisms underlying microbial community formation and their interactions with the environment in special lake habitats, reveal the molecular mechanisms of energy metabolism and the driving of organic synthesis and carbon, nitrogen, and sulfur cycling by key functional microorganisms (or microbial communities), and explore the physiological, ecological, and genetic mechanisms of microbial adaptation to extreme lake environmental conditions. Based on these insights, we can discover new

microbial functional genes or metabolic pathways to promote the exploitation and utilization of lake microbial resources.

2.2.3 Processes, Mechanisms, and Environmental Regulation of Microbially Driven Elemental Cycling in Lakes Lakes are major sites for carbon and nitrogen cycling in the earth's surface system, with microorganisms being the key drivers of these processes. Through studies on the composition and distribution characteristics of microbial functional genes in Chinese lakes, we will analyze the distribution and abundance of functional genes related to carbon and nitrogen metabolism and their relationships with environmental factors. Using gene chips, genomics, transcriptomics, proteomics, and ecological simulation technologies, we will elucidate the biogeochemical processes, coupling mechanisms, and ecological effects of microbe-mediated carbon and nitrogen cycling in lakes. We will reveal the mechanisms of microbial community formation and their interactions with the environment in typical lake habitats, discover novel microbial metabolic mechanisms driving carbon and nitrogen transformation and cycling, and gain in-depth understanding of the processes, mechanisms, and environmental regulation of microbe-driven elemental cycling in lake ecosystems.

2.2.4 Microbial Metabolic Mechanisms in Extremely Polluted Water and Water Quality Improvement Technologies With intensifying lake pollution and eutrophication, large amounts of organic matter and nitrogen and sulfur compounds accumulate in lake water and sediments. Under microbial decomposition, these easily cause environmental hypoxia, generating large quantities of metal sulfides and sulfur-containing amino acids as blackening and odor precursors, causing water and sediments to turn black and smelly. This "black and odorous water" phenomenon has become a major water environmental problem in China. Microorganisms are the key drivers of black and odorous water phenomena. Microbiome research will help reveal the microbial metabolic networks and key microbial pathways for black substance decomposition and transformation in black and odorous water and sediments, elucidate the response characteristics of microbial metabolic network structures in black and odorous water and sediments under enhanced extracellular electron transfer conditions, and develop efficient bioremediation technologies for black and odorous water and sediments based on the regulation of microbial community metabolic pathways.

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