Temporal and spatial variations in the bacterial community composition in Lake Bosten, a large, brackish lake in China

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The bacteria inhabiting brackish lake environments in arid or semi-arid regions have not been thoroughly identified. In this study, the 454 pyrosequencing method was used to study the sedimentary bacterial community composition (BCC) and diversity in Lake Bosten, which is located in the arid regions of northwestern China. A total of 210,233 high-quality sequence reads and 8,427 operational taxonomic units (OTUs) were successfully obtained from 20 selected sediment samples. The samples were quantitatively dominated by members of Proteobacteria (34.1% ± 11.0%), Firmicutes (21.8% ± 21.9%) and Chloroflexi (13.8% ± 5.2%), which accounted for more than 69% of the bacterial sequences. The results showed that (i) Lake Bosten had significant spatial heterogeneity, and TOC (total organic carbon), TN (total nitrogen) and TP (total phosphorus) were the most important contributors to bacterial diversity; (ii) there was lower taxonomic richness in Lake Bosten, which is located in an arid region, than in reference lakes in eutrophic floodplains and marine systems; and (iii) there was a low percentage of dominant species in the BCC and a high percentage of unidentified bacteria. Our data help to better describe the diversity and distribution of bacterial communities in contaminated brackish lakes in arid regions and how microbes respond to environmental changes in these stable inland waters in arid or semi-arid regions.

Heterotrophic bacteria are major constituents of pelagic aquatic ecosystems, where they play a prominent role in the breakdown of organic compounds and the remineralization of nutrients as well as in trophic coupling to eukaryote predators1. Recently, lakes, especially lakes in arid and semi-arid regions, have been described as early indicators of both regional and global environmental change. Microorganisms are some of the most important factors in the nutrient cycling and decomposition of organic matter in lake sediments2, and bacteria are thought to be a sensitive sentinel of those environmental changes3.

Sediments are some of the most diverse microbial habitats and play a key role in the biogeochemical cycles of organic matter decomposition and of major elements, including carbon, nitrogen and phosphorus. Sediment is now an major topic in the investigation of bacterial communities because analysis of the community in sediments can provide clues to understanding benthic ecosystem processes, particularly in saline and eutrophic aquatic environments. Sediment bacteria, due to their high population density (> 108 cells/g), play an important ecological role in the biogeochemical cycle in lake ecosystems4. The biomass and community structure of sediment bacteria change due to changes in the physical, chemical and biological factors in water and sediment, so they can also be considered representative organisms indicating environmental changes5. The combination of abiotic factors and microorganisms in sediments has an important effect on nutrient balance in water.

Arid and semi-arid regions account for almost one-third of the world's land area6, and lakes in these areas provide sparse but valuable water resources for fragile environments and humans. However, these water resources are also inevitably affected by human activities. Studies have shown that many water resources in arid and semi-arid regions have been contaminated by eutrophication or salinity7–9. Lake Bosten is the largest inland brackish lake in the arid regions of northwestern China, and it plays an important role in the surrounding terrestrial ecosystems. Surprisingly, the lake was a freshwater lake before the 1960s, but climate change and human activities over the past 50 years have developed it into a brackish lake with moderate nutrient levels10,11. Salinization and

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eutrophication continue to affect Lake Bosten. Therefore, Lake Bosten provides us with an exclusive opportunity to carefully study the potential response mechanisms of bacterial communities in the early stages of salinization and eutrophication. At the same time, due to anthropogenic agricultural and tourism activities, it also forms a nutritional gradient from malnutrition to eutrophication\(^\text{12}\), and this complex ecosystem is an ideal model for studying the response of bacterial communities to salinity and nutrient gradients\(^\text{13}\). This work is particularly important for the restoration of polluted dry lakes, as changes in microbial communities may be some of the more rapid agents of biological change.

Although a small number of studies have reported temporal changes in community composition in the sediments of the Lake Bosten, these studies have concentrated on a single sample from a single season\(^\text{14}\). There is little research on the variations over the four seasons across the whole lake. In this study, we used high-throughput sequencing technology and redundancy analysis to explore the association between spatial-temporal changes in Lake Bosten bacterial communities and environmental factors. The research objectives are to identify (1) Whether there is spatial-temporal heterogeneity in the bacterial communities of lake sediments under the dual effects of eutrophication and salinity and (2) Which environmental factors control the assembly of bacterial communities in the sediments of Lake Bosten.

**Materials and Methods**

**Study area and sampling.** Lake Bosten (86°19′~87°28′E, 41°46′~42°08′N, 1046 m above sea level) is located in the southern part of Bohu County, Xinjiang Province, in the lower reaches of the Kaidu River. The basin is located in the center of Eurasia. The combination of sunlight and heat, the annual average precipitation is 64.3 mm, and the annual average evaporation is 1,881.2 mm, resulting in an inland desert climate\(^\text{15}\). The lake covers an area of approximately 1,100 km\(^2\)\(^\text{16}\) with an average water depth of 7 meters and a maximum depth of more than 16 meters\(^\text{17}\). The annual average temperature is about 7.9 °C, and the coldest temperature occurs in January (−12.7 °C). There is a four-month freeze season at Lake Bosten from December to March every year\(^\text{15}\). Mountain Tian in Xinjiang, which is the lowest point of the Basin Yan. Additionally, the lake covers a vast area of more than 1,100 km\(^2\) and has a maximum water depth of more than 16 m.Originating from the snow-covered Mountain Tian, the Kaidu River, with an annual average runoff volume of 34 × 10\(^6\) m\(^3\), supplies 85% of the water volume of Lake Bosten\(^\text{18}\). Moreover, as Lake Bosten is located in the centre of Eurasia, a good level of light and heat provide extremely favourable natural conditions for the development of the lake, allowing the basin to be rich in animal and plant resources. The Kaidu River is divided into two branches at the Baolang Sumu Water Diversion Project, which are respectively injected into two lakes of Lake Bosten. The Large Lake Area is 55 km from east to west and 20 km wide from north to south. At an altitude of 1,048.75 m, the water surface area is 1,002.4 km\(^2\), volume is 88 × 10\(^6\) m\(^3\), average water depth is 7.38 m, the maximum depth is 16 m. The centre of lake area (HX) is located in the centre of the Large Lake Area and is highly disturbed by hydrodynamic forces, but is less affected by external influences. Huangshuigou area (HS) located in the northwest of the Large Lake Area and is enriched with nutrients (nitrogen and phosphorus) and salinity from agricultural irrigation and industrial wastewater discharged from the surrounding rivers. There are the highest nutrients and salinity in the area of Lake Bosten\(^\text{17}\). The Large Lake Area is regularly monitored by the Environmental Protection Bureau of Bayinguoleng Mongolian Autonomous Prefecture and has been continuously monitored for over ten years. Small Lake Area, also known as macrophyte-dominated area (SC), covers an area of about 300 km\(^2\), and is an important reed production base in China.

From 8 May 2013 to 18 January 2014, 20 samples were collected from the centre of Lake area (HX, 87°08′00″E, 42°00′00″N), Huangshuigou area (HS, 86°50′30″E, 42°06′00″N), estuary of the Kaidu River (KD, 86°44′30″E, 41°53′40″N) and its nearby area (KF, 86°46′20″E, 41°57′00″N) and macrophyte-dominated area (SC, 86°38′5′E, 41°47′24″N). We avoided extreme weather when sampling and sampled when it was fine. Undisturbed duplicate sediments (0–5 cm deep) were collected seasonally using a Petersen grab sampler from 8 May 2013 to 18 January 2014 (Fig. 1). The sediment samples were transferred into sterile plastic containers, and immediately frozen at −80 °C for storage until analysis.
Physicochemical analysis. The sediment samples were dried to constant weight using a freeze dryer (Alpha 1–2 LD, Martin Christ Instrument Co, Germany). Total nitrogen (TN), total phosphorus (TP) and total organic carbon (TOC) content in the sediments were determined according to standard methods (Jin and Tu 1990). The temperature, salinity, dissolved oxygen (DO), Salinity (TDS), and pH of the lowest water layer covering the sediment were measured using a multi-parameter water quality detector (YSI 6600V2, USA). After DAPI (4′, 6-diamidino-2-phenylindole) staining direct counting, the bacterial abundance in the deposited samples was determined by epifluorescence microscopy12,15. We used one-way ANOVA to test the correlation between environmental factors.

DNA extraction, purification and PCR amplification. Total DNA was extracted from twenty 0.25 g freeze-dried pellets using the Soil Rapid DNA Rotation Kit (MP Biomedicals, Fountain Pkwy. Solon, OH, USA). Genomic DNA concentration and purity were determined by an ultraviolet spectrophotometer. According to the concentration test results, the integrity of the DNA sample was detected by 0.8% agarose gel electrophoresis. The test conditions were as follows: voltage 120 V, electrophoresis time was about 20 minutes.

The extracted DNA stock was diluted to 20 ng·μl⁻¹ and used as a PCR template. The V1–V3 region of the bacterial 16S rRNA gene was amplified using PCR amplification of universal primers 8 F (5′-AGAGTTTGATCCTGGCTCAG-3′) and 533 R (5′-TTACCGCGGCTGCTGGCAC-3′)18. PCR amplification was carried out using a 50 μl reaction system under the following conditions: pre-denaturation at 94 °C for 5 minutes, and denaturation at 50 °C. Anneal at 94 °C for 30 seconds, 55 °C for 45 seconds, 72 °C for 1 minute, 27 cycles, 72 °C for 8 minutes, and stored at 4 °C. The experiment was repeated 3 times. The PCR product was purified and concentrated using E.Z.N.A.® Cycle-Pure Kit. The PCR products from each sample were then mixed in a single tube in equimolar ratios.

454 pyrosequencing and data analysis. Amplicon pyrosequencing was performed on a Roche Genome Sequencer GS FLX Titanium platform using a 454/Roche A sequencing primer kit. Each read file produced by a pyrosequencing run is associated with one quality file, which contains the quality score for each base. After sequencing, the QIIME software package17 was used to exclude sequences with a length of less than 200 bases, an average mass fraction of less than 25, or no primers and barcode sequences. The rest of the sequences were trimmed and compared against the Bacterial Silva database (SILVA version 106; http://www.arb-silva.de/documentation/background/release-106/) via QIIME. Similar sequences were clustered into Operational Taxonomic Units (OTUs) using a minimum identity of 97% by UCLUST software20. Good’s coverage, abundance-based coverage estimator (ACE), Chao 1 richness estimator, Shannon and Simpson diversity indices were also calculated by QIIME pipeline.

Results

Physical and chemical factors. The environmental characteristics of the studied lake are presented in Fig. S1. The overlying water temperatures showed a characteristic annual cycle, with a higher temperature in the HS sample (26.9 °C, summer) and lower temperatures in the KF and SC samples (1.0 °C, winter). In the overlying water, the concentration of TDS increased from 278 mg L⁻¹ in the KD sample (winter) to 2,238 mg L⁻¹ in the HS sample (spring). The average concentrations of TN and TP in the seasonal sediment samples remained stable at approximately 1.74 (KD, winter) to 4.08 (SC, winter) mg·g⁻¹, and 0.15 (KD, winter) to 0.52 (HS, winter) mg·g⁻¹, respectively. The average concentration of TOC among the seasonal sediment samples ranged from 3.07 (HX, summer) to 12.4 (HS, summer) mg·g⁻¹.

Bacterial diversity. The seasonal variation in the bacterial community structure in Lake Bosten was determined using 454 pyrosequencing technology. From all sediment samples, sequencing analysis yielded a total of 210,233 high-quality sequence reads with an average length of approximately 400 bp effective sequence, 8,427 OTUs, and 97% sequence identity threshold. The effective sequence length for each sediment sample ranged from 8,729 to 12,909, and the number of OTUs ranged from 1,319 to 2,209 (Table 1). The sparse curve with a cluster distance of 0.03 was still not saturated and increased (Fig. 2), indicating that new bacterial populations may continue to appear after 10,000 reads and sequencing. However, the values of the Shannon index (4.68–6.64) for all sediment samples in this study stabilized, suggesting that more sequencing may be required, but most of the bacterial diversity of the sample has been captured. In addition, good reports showed that these libraries represent the majority of bacterial 16S rRNA sequences present in each sediment sample, ranging from 0.902 to 0.951 (Table 1).

Bacterial community composition. In the filtered high-quality sequences, the dominant bacterial phylum or sub-phylum in each sediment sample was identified by the SILVA database using the QIIME algorithm (the top 10 most abundant phyla or sub-phyla in each sample). The most abundant bacterial phyla were Proteobacteria (34.1 ± 11.0%), Firmicutes (21.8 ± 21.9%) and Chloroflexi (13.8 ± 5.2%), representing more than 69.0% of the bacterial sequences (Fig. 3). In addition, a large number of bacteria were detected at lower numbers (<4% relative abundance per phyla) in the sediment samples, such as Planctomycetes, TA06, Candidate division WS3, Bacteroidetes, Spirochaetes, Chlorobi, OD1, OP8 and Deferribacteres (Fig. 3). Firmicutes (10.9–65.7%) dominated in the spring and summer samples. In contrast, the autumn and winter samples were mainly dominated by Deltaproteobacteria (0.19%–27.2%) and Chloroflexi (0.18%–22.1%) (Fig. 3).

Figure 4 shows the 56 dominant genera in the sediment samples. At the genus level, the differences between the sediment samples are more pronounced. Microorganisms from the genera Uncultured Neisseriaceae (21.62%), Geobacter (22.63%) and Lysinibacillus (22.63%) predominated in the BS1 and BS2 samples but were less abundant in the other samples. The genus Clostridium (27.94%) and unclassified Peptostreptococcaceae (16.47%) had higher proportions in the HS (summer) sample. Unclassified Anaerolineaceae (2.42%–16.38%) and Sva0485 (0.77%–16.33%) were
more abundant in SC (spring) and KD (summer) samples. Therefore, the main bacterial populations in the twenty sediment samples are slightly different.

**Redundancy analysis of sediment bacteria and environmental factors.** Microorganisms are highly sensitive to environmental changes, and the relationship between different environmental factors and the microflora structure can be determined according to the size of the angle between bacterial and environmental
Figure 3. Relative abundances of top 10 phyla or sub-phyla in each sample (a total of 15 phyla or classes for all twenty sedimentary samples) from Lake Bosten.

Figure 4. Comparison of percentage of the sequences affiliated with the frequently identified genera to the total number of sequences from twenty sedimentary samples in Lake Bosten.
factors and the length of the connection in a redundancy analysis (Fig. 5). Betaproteobacteria, Firmicutes, and Gammaproteobacteria were negatively correlated with TDS and DO, whereas Chloroflexi and Bacteroidetes were positively correlated with TDS and DO. Deltaproteobacteria was positively correlated with each indicator. In the RDA analysis diagram, combining the sediments showed that the levels of the physical and chemical indicators phylum flora structure, TN, TOC and TP were the main factors that influenced the growth of bacteria, but other environmental factors also had certain effects.

**Discussion**

The five sampling points selected from the Large Lake area and Small Lake area in Lake Bosten are representative areas for studying the sediment bacterial community structure of Lake Bosten. Based on the results of the physical and chemical analyses, the physical and chemical indicators of the 20 samples showed certain similarities and differences due to factors such as time and geographical location. There was no significant change in the pH value among the 20 samples, but the other indicators had significant differences (Fig. S1). Most obviously, the values of TDS at the Huangshuigou and central lake areas were significantly higher than those at the other sampling points ($P < 0.05$). According to previous research, agricultural production around Lake Bosten discharges a large amount of farmland drainage with high salinity that includes chloride ions, sulfate ions, ammonia nitrogen and so on. This discharge was the main reason for the increased salt content in Lake Bosten, as 71% and 29% of the lake water flows into the Large Lake Area and Small Lake Area, respectively. In addition, the TDS of the summer and autumn samples from the estuary of Lake Bosten and nearby areas was higher than the TDS in other seasons. Weberscannell et al. noted that changes in the concentrations of TDS in natural waters are often attributed to the discharge of industrial effluent, increased precipitation, or saltwater intrusion. In the summer and autumn, the Kaidu River glaciers melted and created abundant snow and ice water resources, which caused the water volume to increase rapidly, leading to a significant increase in TDS content. However, the specific causes of this difference in TDS could be complex, and other potential reasons for this difference still deserve further investigation.

Interestingly, the contents of TD, TP and TOC in the Huangshuigou and macrophyte-dominated area were higher in winter than in summer (Fig. S1), which may be because the continuous low temperature in winter inhibits microbial activity and reduces the effect of microorganisms on the removal of nitrogen and phosphorus. On the other hand, TN and TP concentrations were reduced due to dilution and biological blocking. In addition, a large number of aquatic plants in the aquatic grass area can enrich N and P and desalinize water by adsorbing, decomposing, oxidizing and precipitating the water, salt, and N and P. This also explained why the value of pollution indicators in the macrophyte-dominated area was higher than that in the Huangshuigou area.

A large number of studies have shown that temperature is an important factor affecting the structure of bacterial communities. High temperatures can increase the metabolic rate, thus causing the cycle of organic matter to increase in speed; a drop in temperature will lower the growth rate of bacteria and lower productivity. This trend has also been confirmed in the laboratory. Therefore, the bacterial community composition of the samples in autumn and winter was more complicated than that in the samples in spring and summer. This may be due to the relative abundance of the dominant bacteria in spring and summer decreasing greatly under the influence of temperature. Temperature changes improve the competitive advantage of other bacteria and increase the relative abundance of other bacteria, leading to a higher diversity of bacteria in autumn and winter. The competitive advantage between microorganisms changes under conditions such as temperature changes, and some studies on bacteria also illustrate this point. In aquatic ecosystems, in addition to temperature, the most relevant factor to bacterial growth and reproduction is inorganic salt nutrition (such as TN, TP) and algae biomass. Shah’s study also suggests that in the Chesapeake at temperatures below 20°C, temperature, more than nutrient levels, functions as the main control factor of planktonic bacteria. At temperatures above 20°C, the nutrient control became dominant.
In the whole bacterial community, the three most abundant phyla were Firmicutes (21.8% ± 21.9%), Proteobacteria (Deltaproteobacteria and Gammaproteobacteria) (34.1% ± 11.0%) and Chloroflexi (13.8% ± 5.2%), accounting for more than 69.0% of the bacterial community. This community structure is largely in accordance with those found in other sediments and soils worldwide.63,64

Firmicutes (10.9%–65.7%) dominated sediment samples in spring and summer but was present at less than 1% abundance in autumn and winter. The RDA suggested that the members of Firmicutes were not particularly dependent on the nutrients in the seasonal samples (Fig. 5). This finding was consistent with that from a previous study in Lake Dongping, a shallow lake.65 Under anaerobic conditions, nitrate can be used to achieve denitrification.65,66 In spring and summer was lower than that in autumn and winter, suggesting that the increase in DO would restrain the growth of Firmicutes. Related studies have shown that DO controls the Firmicutes community structure more than other main factors, which is consistent with the results of RDA analysis (Fig. 5). The genus Clostridium are specialized anaerobic bacteria that mainly exist in anaerobic environments and have strong degradation ability and high metabolic activity.67,68 The genus Bacillus was the primary component of Firmicutes, and the group was abundant in the spring and summer samples (Fig. 4). A previous study observed significant variations in the composition of the genus Bacillus in different estuary sediments from Lake Taihu69, and their presence caused the production of high amounts of ammonia, nitrite and other hazardous substances.70 The genus Lysinibacillus, which includes facultative bacteria, can reduce macromolecular organic matter to low molecular weight acids, dissolve nutrients in the soil sediment,71,72 degrade crude compounds,73,74 fix nitrogen from the air and prevent plant diseases and insect pests,75,76 and therefore has certain environmental benefits.

Proteobacteria (13.5%–55.0%) were the dominant bacteria in all sediment samples. However, in samples from different seasons, the relative abundances of different kinds of Proteobacteria were very different (Fig. 3). In this study, Deltaproteobacteria were the main group in Gammaproteobacteria and Betaproteobacteria (Fig. 3). In the sediment samples from autumn and winter, Deltaproteobacteria dominated. In freshwater sediments, Deltaproteobacteria were also abundant62,63. Deltaproteobacteria was mainly represented by uncultured Desulfarculaceae in the present study (Fig. 4). As sulfate-reducing bacteria, the family Desulfarculaceae has been detected as being dominant in the salt marsh sediments of the North Atlantic and on the East Coast of the US. These bacteria are important participants in the sulfur biogeochemical cycle43,44 and can degrade organic matter. The resultant compensatory nutrition and energy sources play an important ecological role, especially in the anaerobic degradation of organic matter and the conversion process45,46. In the past 20 years, the chemicals in Lake Bosten have come to include high levels of sodium sulfate;77 a high concentration of SO2−

and TDS provide good nutrient conditions, thereby promoting the growth of bacteria. Therefore, its dominant position in the sample may be related to nutrients in the lake.

As a second group of Proteobacteria,65,66 Gammaproteobacteria play an important role in the degradation and absorption of organic compounds, ammonia,78 and sulfide79,80. The relative abundance of Gammaproteobacteria (1.27–16.40%) in the estuary and its vicinity was significantly higher than that in the other samples. Several previous studies have revealed that Gammaproteobacteria usually exist in sediments in areas contaminated with agricultural pollution or organic matter.53,54. The agricultural activities around Lake Bosten have increased the amounts of organic pollutants in the estuary55, and the organic content of the sediments has increased.56 Therefore, it was not surprising that Gammaproteobacteria were abundant in the estuarine sediments of Lake Bosten, and a high relative abundance of Gammaproteobacteria has also been observed in other aquatic systems.57,58. Additionally, the genus Pseudomonas was found to be representative of Gammaproteobacteria. The group appeared as the dominant genus in the estuarine sediment samples, especially in the spring sample (Fig. 4). A high relative abundance of the genus Pseudomonas has been observed in the estuarine sediments of Lake Poyang.59 In addition, the genus Acinetobacter was a minor proportion of the Gammaproteobacteria (Fig. 4). A previous study indicated that the genus Acinetobacter is commonly detected as phosphorus-accumulating microorganisms in sediments60 and that the presence of the genus Acinetobacter might explain the phosphorus-accumulating microorganism concentration in the phosphorus-containing sediment samples from summer (Fig. S1)59.

In this study, Betaproteobacteria appeared as the predominant phylum in all sediment samples (Fig. 3). This finding was consistent with that of a previous study in which Betaproteobacteria was found to be the predominant group in the upper sediments of Lake Taihu64. Betaproteobacteria (0.87–40.56%) are a dominant bacterial group widely distributed in freshwater lakes around the world. They are present in surface waters and are one of the most studied groups to date65, but they are rarely found on the open seas or in salt lakes.66,67. The presence and success of Betaproteobacteria in freshwater lakes are related to their ability to respond quickly to changes in bioavailable nutrient content, e.g., they have a rapid response to dissolved organic carbon68. However, the results of the RDA analysis in this study showed that nutrients such as TOC, TP and TN did not exhibit positive correlations with the relative abundance of Betaproteobacteria (Fig. 5). Interestingly, the most abundant OTU in Betaproteobacteria was the genus Thiobacillus. The genus Thiobacillus is a dominant genus in habitats polluted with mine wastewater, and they are very abundant in all four seasons. Thiobacillus was the dominant genus in the HS samples. This indicates that there may be substantial mineral pollution in the HS area.

Chloroflexi (3.94–22.13%) was evenly distributed among the various sediment samples. A previous study indicated that members of Chloroflexi are active not only in deep subsurface sediments on the ocean floor but also in various wastewater treatment systems.86 In this study, the RDA showed a positive correlation between the concentration of TOC and the relative abundance of Chloroflexi (Fig. 5). This finding was consistent with that of a previous study that reported on riparian sedimentary ecosystems.84 Chloroflexi can biodegrade organic pollutants,85 giving it a better growth advantage in eutrophic environments.86 In addition, Chloroflexi efficiently degrades chlorides. Zanaroli et al.70 found that the genus does not produce O2 by photosynthesis and cannot fix nitrogen but has a dechlorination function. Unclassified Anaerolineaceae was dominant in Chloroflexi, and the group dominated in all seasonal sediment samples (Fig. 4). The family Anaerolineaceae has previously been found
to be dominant in the estuarine sediments of Lake Taihu69, and members of this family are often involved in the anaerobic degradation process of alkanes71,72, as well as in the biodegradation process of organic pollutants73,74.

Bacteroidetes were dominant in samples from all seasons (Fig. 3). A previous study revealed that Bacteroidetes has the capacity to decompose complex molecules in freshwater sediments75. Furthermore, our results indicated that the genus Flavobacterium and unclassified vadinHA17 were the two primary components of Bacteroidetes, of which the genus Flavobacterium was the most abundant OTU. A similar study revealed that the genus Flavobacterium functions in heterotrophic nitrification and metabolizes refractory organic compounds75. Therefore, the enrichment of the genus Flavobacterium may indicate widespread organic pollution in Lake Bosten.

It is noteworthy that there was a substantial percentage of unclassified bacterial groups detected in these seasonal samples (Fig. 4). In addition to those mentioned above, there were still several dominant unclassified bacteria found in the samples, such as unclassified Caldilineaceae, unclassified KD4–96, and some incertae cedis bacteria (Fig. 4). Numerous studies have observed the abundance of unclassified bacteria in various lake systems76,77, especially in sulfur-rich and anoxic environments8,78. The existence of abundant unclassified species in the estuarine sediments of Lake Bosten indicates that new bacterial communities inhabit Lake Bosten and that the bacterial diversity, taxonomy and phylogenetics of the location deserve further attention.

**Conclusion**

In this study, 454 pyrosequencing technology was used to study the bacterial community structure of the sediments in Lake Bosten. We found that the bacterial community of Lake Bosten sediments is spatially specific. For example, the genus *Thiobacillus*, which is mainly found in mine wastewater and had a relatively high abundance in other areas, was found in the HS area; enrichment in the HS area may indicate that the area is rich in minerals. In addition, there are obvious seasonal changes in the bacterial community of sediments. For example, the relative abundance of the Firmicutes in the SC samples in the spring sampling was 53.3%, but only 0.14% in winter. The discharge of agricultural sewage caused the TN and TP in the HS area to be higher than those of other samples, and the adsorption and decomposition of aquatic plants led to the enrichment of pollutants in the SC area. Furthermore, temperature and nutrient status are key factors driving the bacterial community structure in sediments. We have discovered nitrifying bacteria with a large amount of degradable pollutants in Bosten Lake, and our research provides a reference for preventing and remediating lake pollution.

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L.Z.: wrote the manuscript. T.T.S.: provided materials. Y.C.: proof read the manuscript. T.T.Z.: data analysis. L.L.: laboratory analysis. F.P.Q.: collection of the sediments samples. All authors read and approved the final manuscript.

Competing interests
The authors declare no competing interests.

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