

Original Research

Structure and Composition of Bacterial Communities in Water Bodies of Typical Polders in the Chaohu Lake Basin, China

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Abstract

As key drivers of aquatic ecological cycles, microorganisms serve as sensitive bioindicators for ecosystem monitoring. Unlike natural water systems, the hydrological characteristics of the polder area are artificially regulated through sluices, resulting in stagnant flows with limited self-purification capacity. Characterizing microbial communities in diverse polder water types (ditches, ponds, and drainage stations) is therefore crucial for understanding these unique engineered ecosystems. This research employed 16S rRNA gene high-throughput sequencing to analyze microbial communities in diverse water bodies across two Chaohu Lake polders. We examined spatial variations in physicochemical properties, bacterial diversity, community structure, dominant genera, and their environmental correlations, providing insights into polder aquatic ecosystems. Results indicated high bacterial similarity between the outer rivers (Nanfei and Dianbu Rivers). Dominant phyla in polder waters were Proteobacteria, Patescibacteria, Bacteroidota, and Actinobacteriota, with *Unclassified_f_Comamonadaceae*, *Magnetospirillum*, and *Azospirillum* as predominant genera. Notably, Patescibacteria and Verrucomicrobiota showed higher abundance in Chaohu polders than freshwater lakes, indicating distinct bacterial community characteristics in this basin. Proteobacteria and *Magnetospirillum* were correlated with phosphorus and nitrogen, respectively. RDA analysis indicated TP, TN, COD_{Mn}, and DO as the primary environmental factors influencing the microbial community composition. The present research aims to provide scientific support for polder water environment management.

Keywords: polder, bacterial community, structure and composition, environmental factor, Chaohu Lake basin

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Introduction

Microorganisms are the main carriers of metabolic activities in aquatic ecosystems and play an important role in pollutant decomposition and transformation [1, 2]. The response of bacterial communities to environmental changes is an important ecological issue because of their indicator ecological functions [3, 4]. The structure and distribution of bacterial communities change with changes in the aquatic environment, and their sensitivity to environmental changes is often used to characterize water quality changes in polluted ecosystems, with specific indicators including bacterial community biomass, diversity indices, and dominant species [5].

Hydraulic facilities such as gates and dams in polder areas weaken the water system connectivity and reduce the self-purification ability of the water body. In the Chaohu Lake Basin, the most common types of areas are the agricultural polder areas, aquaculture polder areas, and urban polder areas. Drainage from farmland, livestock and poultry wastewater, and domestic sewage are discharged into the Chaohu Lake unit volume to accept the amount of wastewater in China's freshwater lakes in the first place [6]; the water quality condition is poor [7, 8]. Chaohu Lake is the fifth-largest freshwater lake in China and is an important ecological reserve and economic development area [9]. The Chaohu Lake basin has more than 280 large and small polders, with a gross area of about 2,638 km², which accounts for about 20% of the basin area [10]. The formation of polders for flood storage in the basin is widely distributed in the Chaohu Lake Basin, which is the most common production unit with a dense water network and frequent agricultural activities. During the dry season, water is diverted from the river for irrigation, farming, and residential use in the polder; during the wet period, water is discharged outward from the polder through the drainage and irrigation station and flows into the receiving river and ultimately into Chaohu Lake [11]. The co-occurrence network at the lake-inlet of Chaohu Lake was constructed, and it was found that the highly connected microorganisms Gemmatimonadaceae and Nitrosomonadaceae were related to nitrogen transformation function [12]. It was explored that the dominant species of Chaohu Lake were Cyanobacteria, Bacteroidota, and Proteobacteria, and it was found that the spatial distribution of the water body can cause differences in the structure of bacterial communities [13]. The connectivity between dominant species of Cyanobacteria, Bacteroidetes, and Ascomycetes in Chaohu Lake based on the co-occurrence network was further explored, and the results revealed potential hosts of multiple antibiotic resistance genes (ARGs) in the water body [14]. By selecting Nanfei River, the main inflow river of Chaohu Lake, as the research object, it was found that differences in land types in different river sections would lead to differences in pollutant types, which in turn would affect the structure of the sediment bacterial community [15]. However, previous

research has mainly focused on Chaohu Lake and its main inflow rivers, and there is insufficient research on the water environment in the polder area, especially on the response of microorganisms to phosphorus and the correlation between the microbial community structure and the water environment factors. This paper analyzes the bacterial community characteristics and water quality of two types of typical polder waters in Chaohu Lake Basin, aiming to investigate (1) the bacterial community structure and distribution characteristics of polder waters and (2) the influence of environmental factors on the spatial differentiation of bacterial community structure. This research will provide a scientific basis for the improvement of the water ecological environment in the polder area.

Materials and Methods

Overview of the Research Area

The Chaohu Lake basin (N31°25'~31°43', E117°17'~117°51') has more than 280 large and small polders, with a total area of 2,638 km² [10]. The terrain of the watershed is dominated by hills and plains, and most of it belongs to the plain area along the river, with an average annual precipitation of 1031.6 mm, forming surface runoff of 250~280 mm, and a rural population of 6,424,300 people, accounting for about 81.3% of the total population [16]. The total area of Dongda Polder is about 23.5 km², of which 65,000 acres are cultivated. The land use type is mainly agricultural. It was historically known for rice and wheat cultivation and is now transformed into a base for economic crops such as grapes and vegetables. Zhaozhu Polder is a typical aquatic polder, located in Feidong County, Hefei City, which is located at the confluence of Dianbu River, Nanfei River, and Ershibu River. There are national and provincial highways and other traffic arteries, industrial enterprises, and logistics centers in the polder, which is an economically active area.

Sampling Methods

In this research, two typical polder areas in the Chaohu Lake basin, the Dongda polder and the Zhaozhu polder, were selected in July 2024, and samples of different types of water bodies in them were collected (Table 1 and Fig. 1). A total of 12 sampling points were set up to collect surface water samples from different types of water bodies in the polders (outer rivers, ditches, pits, and drainage stations). The sampling points were numbered according to the polder (D1~D7, Z1~Z5). A portable water sampler was used to collect 500 mL of surface water (0~30 cm) from each sampling point, which was frozen at -10°C in the freezer and then transported back to the laboratory in time for the next step of analysis.

Table 1. Introduction to sampling points.

Sampling Points	Polder Name	Introduction	Longitudes	Latitude
D1	Dongda Polder	Ditch in Huanggang Village	117°24'58"	31°47'1.6"
D2		Nanfei River (Huanggang Village Section)	117°24'55"	31°47'9.7"
D3		Shangxin ridge drainage station	117°25'22"	31°45'58"
D4		Xiixin ridge drainage station	117°24'32"	31°45'25"
D5		Donglin Village Eco-pond	117°23'42"	31°43'58"
D6		Ditch in front of scenic gate	117°24'12"	31°44'55"
D7		Nanfei River (Xiaonan Village Section)	117°23'55"	31°44'11"
Z1	Zhaozhu Polder	Nanfei River (Zhaozhu Polder drainage station section)	117°25'26"	31°47'8.6"
Z2		Zhaozhu Polder drainage station	117°25'44"	31°47'14"
Z3		Aquaculture pond	117°26'56"	31°47'30"
Z4		Ditches adjacent to aquaculture pond	117°26'55"	31°47'26"
Z5		Dianbu River	117°27'10"	31°48'25"

Analytical Methods

The pretreatment and analytical testing of water samples were referred to the relevant national standard methods. A portable multi-parameter water quality meter was used to detect the pH and DO of the samples; continuous injection–gas phase molecular absorption spectrometry to determine the permanganate index (COD_{Mn}); the gravimetric method to determine the suspended substance (SS); and the acetone extraction–

spectrophotometric method to determine chlorophyll a (chl-a). Total phosphorus (TP), dissolved total phosphorus (DTP), soluble reactive phosphorus (SRP), total nitrogen (TN), ammonia nitrogen (NH_3-N), nitrite nitrogen (NO_2^-N) and nitrate nitrogen (NO_3^-N) were determined by potassium persulfate oxidation–molybdenum antimony anti-spectrophotometry (GB 11893-89), digestion after filtration–ammonium molybdate spectrophotometry (GB 11893-89), ion chromatography (HJ 669-2013), alkaline potassium

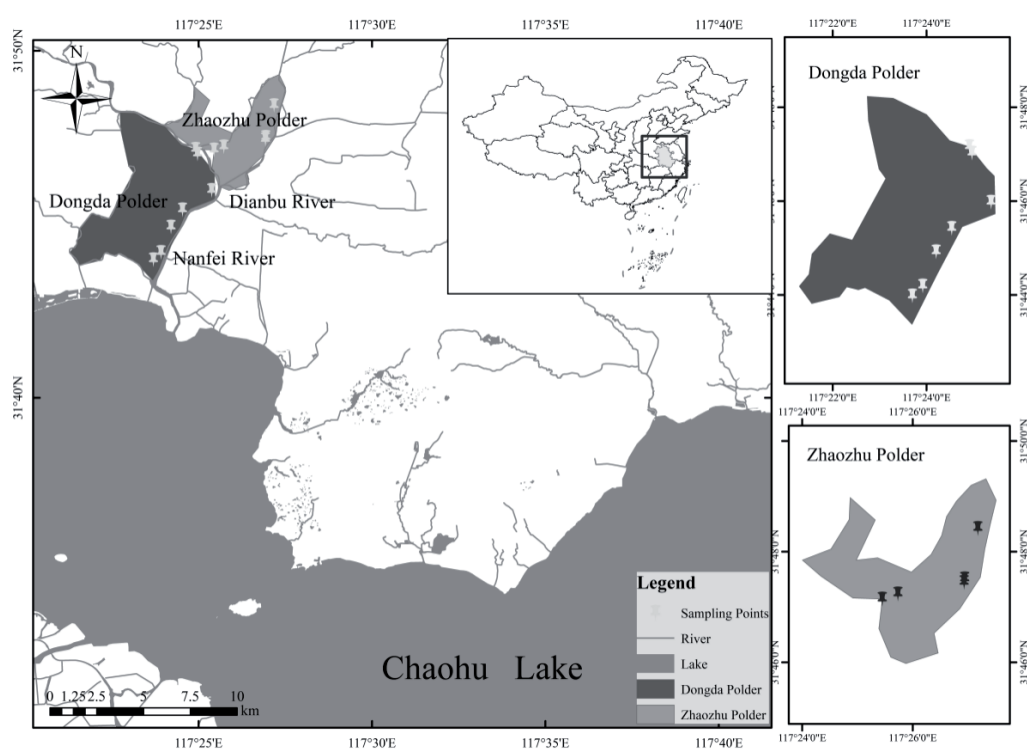


Fig. 1. Location of polder sampling points.

persulfate digestion-ultraviolet spectrophotometry (GB 11894-89), Nessler's reagent spectrophotometry (HJ 535-2009), and spectrophotometric method with phenol disulfonic acid (GB 7480-87) [17]. To ensure the accuracy of the data, three groups were taken in parallel, and the measured results were expressed as the average value. The experimental data were analyzed and plotted using Origin 2021, SPSS 27, R 4.5.0, and ArcGIS PRO 2.5.

DNA Extraction, PCR Amplification, and Illumina MiSeq Sequencing

Genomic DNA was extracted using a DNA extraction kit, and its concentration and purity were measured with a NanoPropOne instrument [18, 19]. Polymerase Chain Reaction (PCR) amplification was conducted using genomic DNA as the template, with barcoded primers and Premix Taq (TaKaRa). The PCR products were quantified by GeneTools Analysis Software (Version 4.03.05.0, SynGene) and pooled based on the equal mass principle. The mixed PCR products were purified using the E.Z.N.A.®GelExtractionKit, and target DNA fragments were recovered by elution with TE buffer. A sequencing library was constructed following the standard protocol of the NEBNext®UltraDNA Library Prep Kit for Illumina®. Finally, high-throughput sequencing was performed on an Illumina MiSeq PE300 platform at Megger Genetics Technology Co., Ltd. (Guangzhou, China), to obtain OTU information and species identification.

Data Analysis Methods

One-way ANOVA was used to analyze the variability of the physical and chemical properties of the water bodies in the polder area of Chaohu Lake across different points. Spearman's correlation coefficient was used to test the diversity index and the correlation between the dominant bacteria and the environmental factors. Origin 2021 was used to calculate the relative abundance. Redundancy analysis was performed by R (version 4.5.0) (RDA). Pearson analysis was performed using SPSS 27. The visualization of network analysis is realized by Gephi (0.9.2).

Results

Chaohu Lake's Typical Polder Water Physicochemical Properties

Based on the "surface water environmental quality standards" (GB3838-2002), Chaohu Lake's typical polder sample points are between Class III and inferior Class V water bodies, of which the Dongda polder water quality conditions are relatively better than those of the Zhaozhu Polder (Table 2 and Fig. 2). Dongda Polder sample points (D1 ~ D7) pH, DO, TN, NH₃-N, TP, and SS concentration range of 7.17~8.30, 2.32~9.34 mg/L, 0.80~3.10 mg/L, 0.17~1.04 mg/L, 0.05~0.26 mg/L, and 2~43 mg/L, belong to the Class III~Class IV type of water bodies. Zhaozhu Polder sampling points (Z1~Z5) pH, DO, TN, NH₃-N, TP, and SS concentration range of 7.67~8.50, 1.58~16.1 mg/L, 1.46~4.76 mg/L, 0.65~1.64 mg/L, 0.15~0.68 mg/L, and 2~22 mg/L, belong to the Class IV~inferior Class V water bodies.

DO concentration is highest at Z3 of the Zhaozhu Polder aquaculture pond. The reason may be that the aquaculture pond is covered with aeration and oxygenation devices. The pH value of each sample point has little difference and stabilizes between 7.60 and 8.30. The high value of COD_{Mn} appears at Z2 of the Zhaozhu Polder drainage station and Z4 of the ditch next to the aquaculture pond due to the closure of the gates and dams, and the organic matter carried by domestic sewage is enriched at the drainage station, and the organic matter content is higher in the aquaculture wastewater and humus. The maximum value of SS was found in Nanfei River (Huanggang Village section), which may be due to the high sediment content of the outer river water body. Chl-a was significantly higher than other sample points in Zhaozhu Polder aquaculture pond Z3, which may be related to the higher content of nitrogen and phosphorus in the bait and fish excreta in the breeding pond and eutrophication of the water body.

The maximum value of TN concentration appeared in Nanfei River (Zhaozhu Polder drainage station section) Z1, which may be related to the area of domestic sewage discharge. The minimum value appeared in Donglin Village ecological pond D5, which is equipped with ecological floating islands and aquatic plants planted on the slope of the bank, indicating that the aquatic plants have a better nitrogen removal effect. The trend of DTN change is basically the same as that of TN.

Table 2. Physicochemical properties of water in polders.

Index	pH	DO/(mg/L)	SS/(mg/L)	TN/(mg/L)	NH ₃ -N/(mg/L)	TP/(mg/L)
Dongda Polder	7.17~8.30	2.32~9.34	2~43	0.80~3.10	0.17~1.04	0.05~0.26
Zhaozhu Polder	7.66~8.49	1.58~16.1	2~22	1.45~4.76	0.65~1.64	0.15~0.68
Range	7.17~8.50	1.58~16.10	2~43	0.80~4.76	0.17~1.64	0.05~0.68
Mean Value	7.87	6.48	13	2.16	0.81	0.28

The highest values of TP, DTP, and SRP concentrations appeared in the Zhaozhu Polder drainage station Z2, which may be related to the interception of gates and dams, where the self-purification capacity of the water body is limited. The lowest value appeared in the ecological ponds in Donglin Village D5, which indicates that aquatic plants also have a good purification effect on phosphorus. The analysis results show that there is a certain similarity between the distribution patterns of phosphorus and nitrogen.

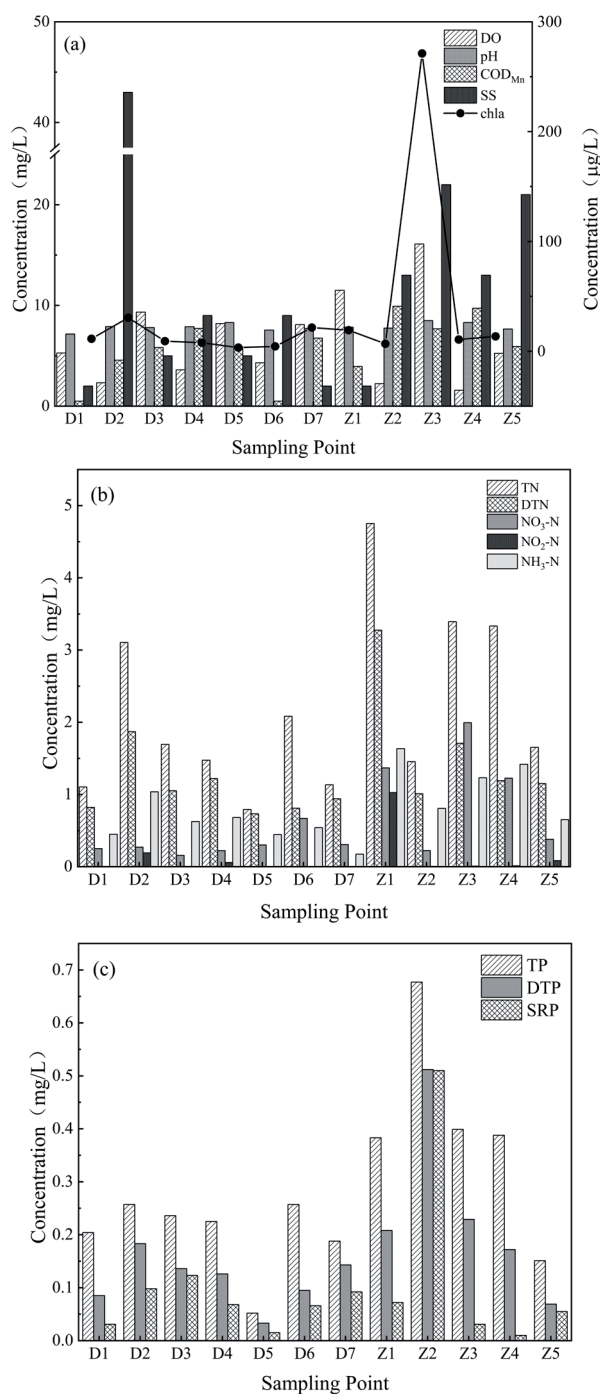


Fig. 2. Physicochemical properties of water bodies in the polder area of Chaohu Lake. a) physical water quality factors; b) nitrogen concentration; c) phosphorus concentration.

Bacterial Community Characteristics

Bacterial Community Structure

In the 12 sample points collected in this survey, a total of 1,036,965 valid sequences of information on bacteria were obtained, and the sequence information of a single sample site ranged from 68,057 to 108,598. Through taxonomic analysis with the database, a total of 6931 OTUs were obtained, which belonged to 59 phyla and 778 genera. There were a total of 12 dominant phyla (relative abundance >1%) in the polder (Fig. 3a), in order of relative abundance: Proteobacteria (20%-64%), Patescibacteria (2%-39%), Bacteroidota (3%-29%), Chloroflexi (1%-15%), Verrucomicrobiota (1%-18%), Actinobacteriota (2%-20%), Acidobacteriota (0%-12%), Firmicutes (1%-10%), etc. The top 3 species groups with the highest abundance accounted for 42.15%~81.10% of each sample community. The relative abundance of Proteobacteria was higher at D1, D4, D7, Z2, and Z4, while the relative abundance of Patescibacteria was higher at D6, Z1, and Z4.

There were 15 dominant bacterial genera (relative abundance >1%) in the bacterial community of typical polders in the Chaohu Lake basin (Fig. 3b), in order of relative abundance: unclassified_f_Comamonadaceae (1%-64%), *Magnetospirillum* (1%-32%), *Azospirillum* (1%-66%), unclassified_o_Candidatus_Kaiserbacteria (1%-25%), unclassified_f_Xanthobacteraceae (1%-47%), unclassified_c_Alphaproteobacteria (3%-25%), unclassified_f_env.OPS_17 (1%-74%), etc. The dominant bacterial genus varied greatly among the sample points, among which unclassified_f_Comamonadaceae accounted for the highest percentage at D1, D4, and Z2; unclassified_f_env.OPS_17 accounted for the highest percentage of 73.99% at D3; *Azospirillum* had a significantly higher relative abundance at D7 than at the other sample points.

Bacterial Diversity and Abundance

Alpha diversity indices, such as the Shannon index and Simpson index, and species richness indices, such as the Chao 1 index and ACE index, are commonly used to describe microbial diversity [20]. The Shannon index was lowest at D6 and highest at D5 (Fig. 4a). The Simpson index of bacterial community was highest at D7 and significantly lower than other sample points at D5 and Z1 ($p < 0.05$) (Fig. 4b), i.e., it indicated that the diversity of bacterial community species was significantly higher at D5 and Z1 than other sample points. The Chao1 index was lowest at D4 and significantly higher than other sample points at D2, Z1, and Z3 ($p < 0.05$) (Fig. 4c). The ACE index was relatively high at D2 and Z1 and lower than other sample points at D4, D6, and D7 (Fig. 4d). For two different land-use polder areas (agriculture and aquaculture), the OTUs number, ACE index and Chao1 index exhibited similar trends [18]. For Dongda Polder, the areas with the lowest bacterial community richness

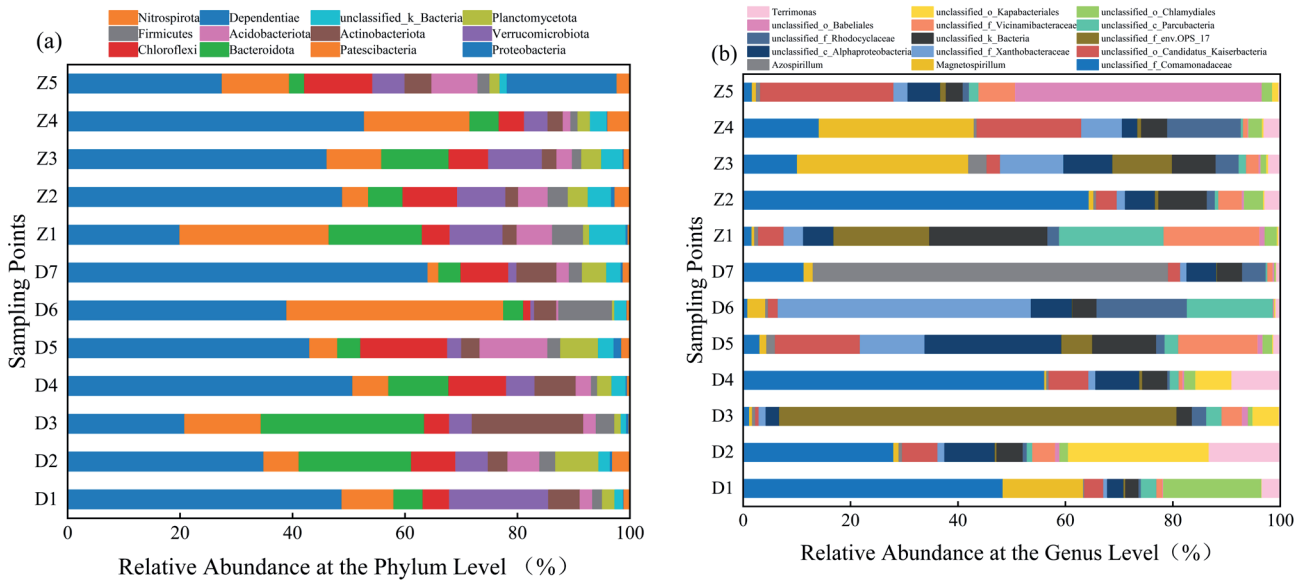


Fig. 3. Bacterial community structure at a) the phylum level and b) genus level in water bodies of typical polder areas.

were Shangxin Ridge Drainage Station, Xiaxin Ridge Drainage Station, the ditch in front of the scenic gate, and Nanfei River (Xiaonan village section), and the highest richness was the ecological pond in Donglin village. As for Zhaozhu Polder, the difference in bacterial community richness was not significant and overall showed high species diversity. According to the data from the sampling points, the trend of the change of

alpha diversity and richness of bacteria among the points was consistent. Simpson's index was significantly lower than the other sample sites at D5 and Z1, and significantly different from the lowest diversity at D3, D6, and D7. The ACE index and Chao1 index were relatively high at Z1, Z3, and D2, while lower at Z5 and D4, indicating higher microbial community richness in Nanfei River and aquaculture ponds, with relatively lower richness in

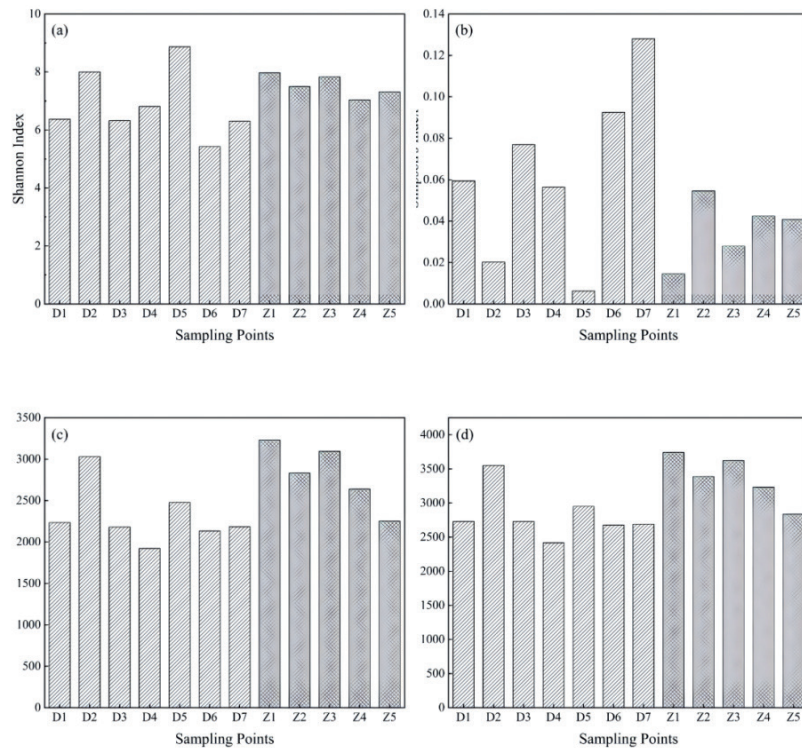


Fig. 4. a) Shannon index, b) Simpson's index, c) Chao1 index, and d) ACE index reflect the diversity and abundance of bacterial communities in water bodies of polder areas.

Dianbu River. The above results demonstrate significant spatial and temporal variations in species diversity and richness among typical water bodies in the polder areas [21].

Analysis of the Significance of Differences in Bacterial Community Structure between Groups

Pearson correlation heat map analysis of the community composition of the sample points (Fig. 5) showed that D1, D2, D5, and D6 showed highly significant positive correlation ($p < 0.01$); Z1, Z2, and Z4

showed highly significant positive correlation ($p < 0.01$); and the community composition of Z3 and D3 showed lower correlation with the rest of the sample points. In terms of water body type, there was a highly significant positive correlation ($p < 0.01$) between Z1, Z5, D2, and D7 in the outer river of the Chaohu Lake basin; D1 and D6 in the inner ditch of the Dongda Polder were highly significantly positively correlated ($p < 0.01$), and there was also some correlation with Z4 of the ditch of the Zhaozhu Polder ($p < 0.05$). Drainage stations D4 and Z2 showed a highly significant positive correlation ($p < 0.01$), while the correlation between Shangxin Ridge

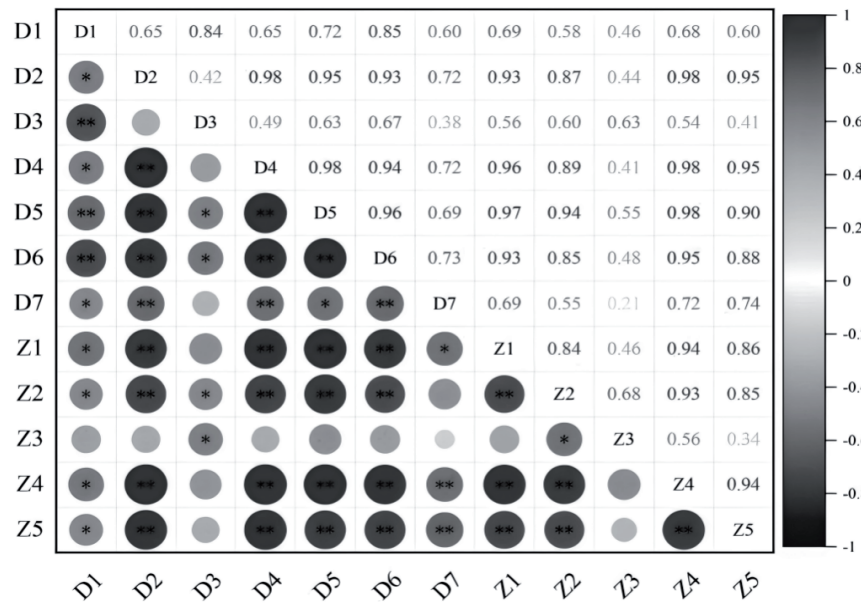


Fig. 5. Differences in bacterial community structure between groups. *indicates $p \leq 0.05$, **indicates $p \leq 0.01$.

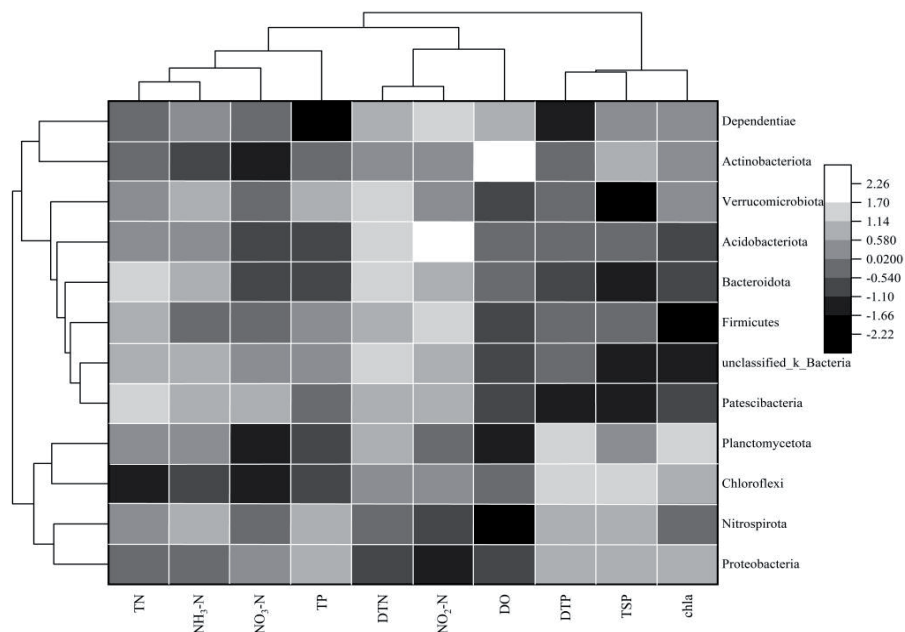


Fig. 6. The correlation between bacteria and environmental factors at the phylum level.

Drainage Station D3 and the rest of the sampled points was not significant. Generally speaking, the correlation between points (D1~D7) in Dongda Polder is higher than that between points (Z1~Z5) in Zhaozhu Polder.

Correlation of Bacterial Community Characteristics with Environmental Factors

Correlation analysis based on environmental factors and the top 12 bacterial phyla in terms of relative abundance (Fig. 6), TN, DTN, NO_3^- -N, and NO_2^- -N all showed significant positive correlation with unclassified_k_Bacteria. TP, DTP, and SRP all showed significant positive correlation with Proteobacteria and Nitrospirota. TP showed a significant negative correlation with Dependistia. DO showed a significant negative correlation with Nitrospirota, and chl-a showed a significant negative correlation with Firmicutes. Among the dominant bacterial phyla, Patiscibacteria showed a significant positive correlation ($p<0.05$) with the content of TN, Bacteroidota showed a significant positive correlation ($p<0.05$) with the content of DTN, and Proteobacteria showed a significant positive

correlation ($p<0.05$) with the content of phosphorus (TP, DTP, and SRP).

Among the top 15 bacteria in relative abundance at the genus level (Fig. 7), TN was highly significantly negatively correlated with unclassified_o_chlamydiales ($p<0.01$). NH_3 -N was highly significantly negatively correlated with *Azospirillum* ($p<0.01$) and was significantly positively correlated with unclassified_o_Candidatus_Kaiserbacteria, unclassified_f_Rhodocyclaceae, and *Magnetospirillum* ($p<0.05$). DTN was significantly positively correlated with Candidatus Kapabacteria, and unclassified_f_Vicinamibacteraceae were significantly positively correlated ($p<0.05$). NO_3^- -N was significantly positively correlated with unclassified_f_Xanthobacteraceae, *Magnetospirillum*, and unclassified_f_Rhodocyclaceae ($p<0.05$). TP, DTP, and SRP were significantly positively correlated with unclassified_o_chlamydiales and unclassified_f_Comamonadaceae ($p<0.05$). DO was significantly positively correlated with *Terrimonas*, Candidatus Kapabacteria, unclassified_f_Rhodocyclaceae, and unclassified_o_Candidatus_Kaiserbacteria were significantly negatively correlated ($p<0.05$), and

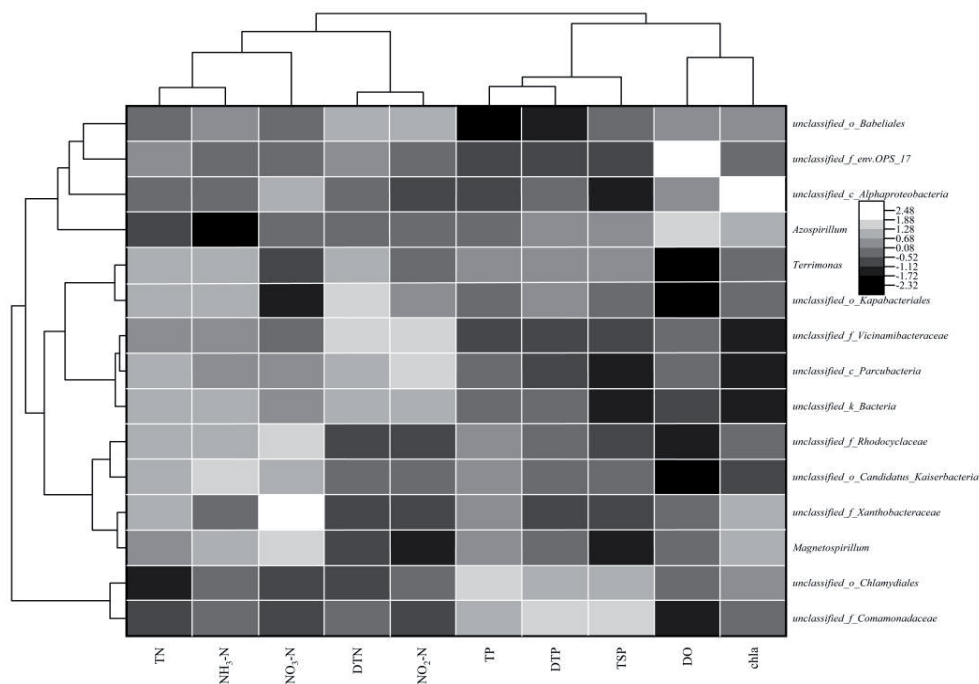


Fig. 7. The correlation between bacteria and environmental factors at the genus level.

Table 3. Topological parameters of the co-occurrence network of bacterioplankton communities in the research area.

Research area	Nodes	Edges	Average degree	Average weighted degree	Positive correlation	Negative correlation	Density	Modularity factor	Average cluster coefficient	Average path length
Dongda Polder	514	563	2.19	4.17	84.9%	15.1%	0.004	0.681	1	1
Zhaozhu Polder	937	4596	9.81	9.81	60.3%	39.7%	0.010	0.970	1	1

significantly positively correlated with unclassified_f_env.OPS_17, Alphaproteobacteria and *Azospirillum* ($p < 0.05$). Chl-a was significantly positively correlated with unclassified_c_Alphaproteobacteria ($p < 0.05$) and significantly negatively correlated with unclassified_f_Vicinamibacteraceae and unclassified_c_Parcubacteria ($p < 0.05$).

The correlation analysis of abundance and diversity indices with environmental factors found that TN, DTN, and $\text{NH}_3\text{-N}$ were simultaneously and highly significantly positively correlated ($p < 0.01$) with both the Chao1 index and the ACE index. $\text{NH}_3\text{-N}$ was significantly and negatively correlated ($p < 0.05$) with the Shannon index in a typical polder in the Chaohu Lake basin. Phosphorus was not significantly correlated with α -diversity, and TP was significantly positively correlated with the ACE index ($p < 0.05$).

Potential interactions between planktonic bacteria in different types of polders were revealed by constructing

a co-occurrence network based on OTUs with a relative abundance of more than 0.1%. As shown in Fig. 8 and Table 3, the phylum of bacterioplankton had the most nodes occupied by Proteobacteria, Patescibacteria, and Bacteroidota. The number of edges in the community co-occurrence network was Zhaozhu Polder (4596) > Dongda Polder (563). The graph density was Zhaozhu Polder (0.010) > Dongda Polder (0.004). The modularity order of the network was Zhaozhu Polder (0.970) > Dongda Polder (0.681); both have some degree of modularity. The average degree is Zhaozhu Polder (9.81) > Dongda Polder (2.19). The higher degree of modularity and average degree in the co-occurrence network indicated that its network clustering was more obvious, and the higher degree of interactions. The above results showed that the interactions between bacterioplankton communities were more complex in Zhaozhu Polder (aquaculture polder) and simpler in Dongda Polder (agricultural polder). The number of

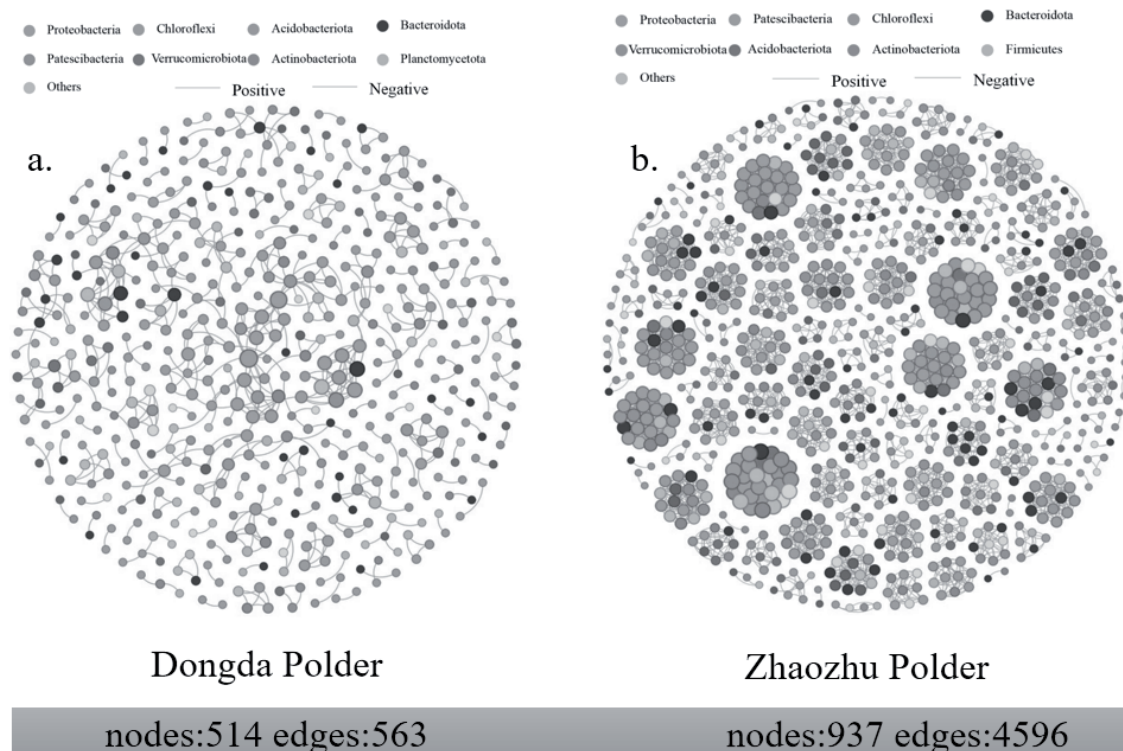


Fig. 8. Co-occurrence network of bacterioplankton of a) Dongda Polder and b) Zhaozhu Polder.

Table 4. Pearson correlation between bacterial diversity and environmental factors.

Category	TN	DTN	$\text{NO}_3\text{-N}$	$\text{NO}_2\text{-N}$	$\text{NH}_3\text{-N}$	TP	DTP	SRP
Shannon	-0.443	-0.483	-0.34	-0.578	-0.613*	-0.064	-0.038	0.121
Simpson	0.269	0.409	0.233	0.47	0.418	0.055	0.163	0.021
Chao1	0.758**	0.727**	0.617*	0.544	0.780**	0.547	0.52	0.209
ACE	0.771**	0.716**	0.627*	0.522	0.800**	0.576*	0.538	0.236

*indicates $p < 0.05$, **indicates $p < 0.01$.

[13]. The dominant phyla in surface sediments of Nanfei River are Proteobacteria, Chloroflexi, and Bacteroidota [15]. Different pollutant contribution levels of guest water in Nanfei River show that the TN of overlying water in the harbor discharge section is significantly lower than that in the tributary discharge and agricultural non-point source section [16]. The nitrogen and phosphorus concentrations in the tailwater discharge and the tributary discharge are higher, and the distribution of chl-a is higher in each tributary flow [22]. It has been shown that there are spatial differences in microbial community structure in freshwater lakes, and most of them are concentrated in the phylum of Proteobacteria, Bacteroidota, and Actinobacteriota [23-26]. Compared with the above phyla, the dominant bacteria in the typical polder area of Chaohu Lake Basin have been added to the Patescibacteria, Acidobacteriota, and so on. The results showed that the bacterial community of the polder water body has both similarity and uniqueness with the water bodies of lakes and lake inlets. The dominant bacteria in Dongda Polder are Proteobacteria, Patescibacteria, and Bacteroidota. The dominant bacteria in Zhaozhu Polder are Proteobacteria, Patescibacteria, and Verrucomicrobiota. Considering that the Dongda Polder is a typical agricultural polder, dominated by cultivation, while the Zhaozhu Polder is dominated by aquaculture, the difference in dominant populations may be related to the different land use types in the polder.

The bacterial diversity of Nanfei River in the polder sample points was significantly higher than that in the rest of the sample points, indicating the spatial variability of the water environment [27], probably because Nanfei River flows through the urban area, the endogenous pollution characteristics are obvious [28]: the level of eutrophication is high, the water body contains a large number of nutrients such as nitrogen and phosphorus, and at the same time provides important nutrient factors used for Cyanobacteria to grow and multiply. With the difference in the sampling time, the higher temperature, and the Cyanobacteria expansion and multiplication rate accelerated [29]. The Proteobacteria showed strong decomposition of organic matter [30], and the Bacteroidota could further settle and decompose organic matter [31].

Bacterial Diversity Characteristics and Environmental Factors

In different types of water bodies in each typical polder, the trends of bacterial α -diversity and abundance were consistent, while their respective peaks appeared at different sample points. It has been shown that there are significant spatial differences in the abundance and diversity of bacterioplankton communities in freshwater lakes [23, 24, 32, 33]. The lowest bacterial abundance at D3 and D4 at Shangxin Ridges Drainage Stations and Xiixin Ridges Drainage Stations could be attributed to the fact that the lifting and lowering of the gates and

dams blocked the continuous flow of the water body. And when the gates and dams were closed, the water flow was slow, which suppressed the bio-accumulation and self-purification ability of the water body [34]. At the same time, the content of TP, DTP, and SRP near the drainage station was relatively high, and the level of DO was low in the Dongda Polder (Fig. 1), which showed that the presence of the gates and dams affected the water quality conditions and microbial community richness in the water body [35]. In addition, higher levels of microbial richness were found at Z3 and Z4 in the aquaculture ponds and their adjacent ditches, probably due to the fact that the water body was provided with more habitat and microenvironment by the ample feed, organic matter, aquatic grass clumps, and sediments in this environment [36]. At Z1 and D2, different sections of Nanfei River showed higher bacterial community richness, while all forms of nitrogen and phosphorus were significantly higher here than at the points in the same polders, which coincided with the environmental characteristics of Nanfei River flowing through the city [37], where domestic sewage and industrial wastewater carry large amounts of nitrogen and phosphorus nutrient inputs.

The Shannon index of bacterial diversity was the lowest in the hardened ditch in the scenic area, but the highest in the ecological pond adjacent to it, which may be due to the fact that the ecological pond is planted with a variety of aquatic plants, such as calamus and cattail, supplemented with ecological floating islands and aeration devices. In addition to directly absorbing nutrients such as nitrogen and phosphorus from the water environment, aquatic plants can also control algal reproduction and metabolism through the release of chemosensory substances and create a good environment for microbial growth and reproduction [38]. At the same time, DO plays an important role in the growth and reproduction of microorganisms [39], and high bacterial diversity was observed in D5, Z1, and Z3, where the DO content was high. Overall, the Shannon index was higher at each sample point (Z1~Z5) in the Zhaozhu polder, and its mean value was higher than that of the Dongda polder (D1~D7). And the bacterial diversity index varied greatly among different sample points in the Dongda polder. As a typical aquaculture polder, the Zhaozhu polder is rich in nutrients such as nitrogen and phosphorus, mainly animal excreta and aquaculture flushing water [40], which provides sufficient nutrients for the growth and reproduction of bacteria, thus making the Zhaozhu Polder water body have high microbial abundance and diversity. There is a significant decline in bacterial diversity in the sampling points D2, D7, and scenic hardened ditch D6 and outer rivers of Dongda Polder, whose incoming water is greatly affected by human activities, and it is considered that changes in water quality due to human activities have a certain degree of screening of bacterial communities, and with the continuous influx of sewage and wastewater, the vulnerable microflora are gradually eliminated, thus

resulting in a significant reduction in the diversity of the bacterial community [41].

Bacterial community diversity and abundance were lower in the surrounding water bodies of the areas affected by human activities, while bacterial community diversity and abundance were higher in the areas rich in organic matter and aquatic grasses, such as ecological ponds, natural ditches, etc. (Fig. 1 and Fig. 3), which indicated that geographic location differences could affect the bacterial community structure by influencing the distribution of nutrients such as nitrogen and phosphorus and environmental factors such as heavy metals [42].

Distribution of Dominant Bacteria and Environmental Factors

In this research, the dominant bacteria in Chaohu Lake polder waters were identified as Proteobacteria, Patescibacteria, Bacteroidota, Chloroflexi, Verrucomicrobiota, Actinobacteriota, and Acidobacteriota, consistent with findings in ecological ditches [43] and natural water bodies [13]. In the typical polder sample points of Chaohu Lake, Proteobacteria showed significant positive correlations with TP, DTP, and SRP (Fig. 6), suggesting their role in decomposing organic phosphorus into soluble phosphate under phosphorus-limited conditions, thereby increasing aquatic phosphorus levels. Fig. 7 demonstrates unclassified_c_Alphaproteobacteria's strong positive correlation with soluble phosphate and DO levels, though its relative abundance decreased with DO [44]. Patescibacteria, a superphylum widely distributed in aquatic environments [45], requires symbiotic relationships with other microorganisms. Its nitrogen correlation (Fig. 6) aligns with its established role in carbon and nitrogen cycling [46]. Bacteroidota, associated with eutrophic conditions, participate in denitrification and complex compound degradation, showing positive correlations with TN and NO_3^- -N [47, 48]. Actinobacteriota and Proteobacteria serve as indicators of anthropogenic impacts [49], with Actinobacteriota correlating positively with DO and SRP but negatively with NH_3 -N and NO_3^- -N. Verrucomicrobiota's abundance (up to 18%) exceeded typical freshwater levels (1%-11.5%) [50-52], potentially due to polder eutrophication, showing positive correlations with DTN, NH_3 -N, and TP. Chloroflexi, capable of anaerobic photosynthesis [53], correlated positively with chl-a and negatively with DO.

Several bacterial genera, including unclassified_f_Comamonadaceae, *Magnetospirillum*, *Azospirillum*, unclassified_o_Candidatus_Kaiserbacteria, unclassified_f_Xanthobacteraceae, and unclassified_c_Alphaproteobacteria, exhibited high abundance in Chaohu Lake polders, though their dominance varied spatially. Unclassified_f_Comamonadaceae emerged as the predominant genus across all drainage stations. As β -proteobacteria, Comamonadaceae contribute

to carbon/nitrogen cycling and phosphorus removal [54]. These strictly aerobic gram-negative bacteria mediate denitrification by converting ammonia to NO_2^- -N and ultimately N_2 [55], while also demonstrating exceptional PAHs degradation capacity [56]. Due to dam-induced water stagnation, oil slicks were observed at the flooding station sampling site, which explains the dominance of Comamonadaceae given its remarkable tolerance to such polluted conditions. *Magnetospirillum*, participating in iron, nitrogen, sulfur, and carbon cycling [57], was particularly abundant at high-nitrogen sites Z3 and Z4. α -Proteobacteria - including phototrophs, plant symbionts, and animal-associated bacteria [58] showed elevated abundance in plant-rich site D5 and oxygenated aquaculture pond Z3. *Nitrospira* also demonstrated a significant presence in these nitrogen-rich environments. Importantly, genus-level distributions correlated strongly with environmental parameters (chl-a, PP, pH, and DO, etc.), indicating bacterial community structure results from complex interactions with aquatic environmental factors.

Conclusions

This research utilizes 16S rRNA gene high-throughput sequencing to analyze the bacterial diversity, community structure, dominant genera, and their environmental correlations. The results can be concluded that:

1. the polder water bodies were dominated by Proteobacteria, Patescibacteria, Bacteroidota, Chloroflexi, Verrucomicrobiota, and Actinobacteriota at the phylum level, with Unclassified_f_Comamonadaceae, *Magnetospirillum*, and *Azospirillum* as dominant genera. Notably, Patescibacteria and Verrucomicrobiota represented unique dominant phyla in polders compared to other Chaohu Lake Basin water systems.

2. bacterial community richness and diversity were significantly lower in ditches and drainage stations, likely due to low dissolved oxygen conditions and water stagnation caused by sluice operations. In contrast, ecological ponds and aquaculture ponds with higher oxygen levels exhibited greater microbial diversity.

Finally, nutrient analysis revealed Proteobacteria's positive correlation with phosphorus but negative correlation with total nitrogen, suggesting preferential nutrient utilization. Microbial community structure was shaped by multiple interacting factors rather than single environmental variables. At the genus level, temporal variations in bacterial communities showed significant environmental correlations. RDA analysis indicated TP, TN, COD_{Mn} , and DO as primary drivers of microbial community composition.

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Conflict of Interest

The authors declare that this article has no conflict of interest.

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