

(RESEARCH ARTICLE)



Physicochemical attributes and bacteriome diversity of Lake Tunari, Taraba State, Nigeria

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International Journal of Science and Technology Research Archive, 2024, 06(02), 059–070

Publication history: Received on 05 March 2024; revised on 01 May 2024; accepted on 04 May 2024

Article DOI: <https://doi.org/10.53771/ijstra.2024.6.2.0043>

Abstract

Physicochemical parameters of aquatic habitats can play an influential part in shaping the microbial communities of water bodies. In this study, water samples from Lake Tunari were analysed for its microbiome and physicochemical attributes, using spectrophotometric and culture-dependent methodologies respectively.

The temperature of the lake ranged from 28.00 – 29.00 (°C), while the pH ranged from 6.50 – 7.20. Heavy metals such as Manganese (0.48 ppm), Lead (0.14 ppm) and Selenium (0.10 ppm) showed significantly high values in selected parts of the lake, surpassing limits set by the World Health Organisation. Microbiome analysis revealed the largest phylum across sample sites as Proteobacteria, Firmicutes, and Planctomycetota with peak relative abundance values of 64.13%, 45.29% and 16.11% respectively. Bacterial genera such as *Exiguobacterium*, *Methylocystis* and *Rhodofera* were detected from designated sites. The presence of these genera capable of tolerating, utilizing or degrading metal ions in water samples of the Lake might be indicative of chemical and metal trickles into the water body. In addition, relatively new and understudied bacterial phyla such as Gemmatimonadota, as well as unknown bacterial clades were identified from water samples of the lake.

This study has revealed that Lake Tunari is not impervious to pollution, caused by either anthropogenic activities or from adjoining water channels. Environmental health awareness programs can be introduced to enlighten the populace, while scheduled microbial and physicochemical surveillance can be adopted, to control microbial contamination and curtail toxic material inflow into the Lake.

Keywords: Lake Tunari; Bacteriome diversity; DADA2; Physicochemical analysis

1 Introduction

Lake Tunari is an isolated body of freshwater surrounded by land, like many other inland lakes. As a flowing lake, it receives a significant annual input from the River Donga, which starts on the Mambilla plateau and forms part of the international border that extends upward to the Republic of Cameroon, and discharges its overflow into the same river downstream, which connects to the River Benue [1]. The lake is an important component for the settlement of Tunari

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hamlet in Taraba state's Wukari Local Government Area. The vegetation on the terrain is mostly woody and herbaceous. The dry season is humid and partly cloudy, resulting in high temperatures throughout the year, whereas the rainy season is humid and overcast [1].

A few examples of human activities on and near the lake include farming, fishing, washing, and livestock rearing. A significant part of many freshwater lakes are aquatic vegetation. The water body is distinguished by a broad cover of floating macrophyte plants, including lettuce, hyacinth, lilies, duckweed, coontail, weeds, salvinia, and a variety of submerged plants [2].

The theory of Ecology now considers a lake to be a component of a drainage basin's bigger unit [3]. Common sources of pollution in lakes include pesticide production byproducts, animal excrement from roaming animals, and inflow runoff during rainy seasons that may contain waste from open defecation due to some people not digging pit latrines. Likewise, fermentation of dead organic remnants of fish species, insects, plants, and animals is a common visible microbial activity in situ [4].

Microbes are found in practically every part of the Earth, including soil, air and water [5]. On Earth, there are around 250,000 cubic kilometers of freshwater in the form of lakes, inland seas, and rivers, all of which have the potential to contain various microorganisms [6]. Freshwater bacteria are a dense assemblage of prokaryotic organisms with diverse morphology, physiology, and ecological preferences that are significant contributors to the transformation of complex organic compounds and minerals in freshwater sediments [7].

A significant portion of the entire biological diversity found on Earth is made up of microbes. Nearly 4 billion years of evolutionary development have resulted in the microbial diversity that we see today [8]. In addition to the different ecosystems found on Earth, freshwater lakes provide significant habitats for a variety of microbial communities. Despite having significant differences like salinity, average temperature, depth, and nutrient content, freshwater and marine settings both offer a wealth of fantastic habitat for microorganisms [9].

The idea that microorganisms living in terrestrial and aquatic environments were mostly alike due to the conventional culture-based techniques lasted until recently [10]. However, the development of modern technologies and methods, such as deep sequencing, allowed for previously unknown insight to the diversity and make-up of bacterial populations. This made it easier for researchers to distinguish between bacteria in various environments, revealing the unique physical and chemical properties of each species [11]. The microbial diversity of Lake Tunari was assessed using high-throughput sequencing to clearly identify various bacterial clades occupying this unique niche. In addition, the physicochemical characteristics of the lake water were also ascertained in order to establish the physical and chemical attributes of Lake Tunari.

1.1 Study Area and Sampling Sites

This research was conducted in Tunari village in Wukari Local Government Area of Taraba state, located to the far South of North-eastern Nigeria in the sub-tropical forest region. The village is inhabited by Jukum and a few Hausa/Fulani tribes of Kabawa extractions who are predominantly peasant farmers and fishermen [12]. Three sampling sites designated Abuja Area (AA), Gindin Dorawa (GD), and Dadin Wanka (DW) (Abuja Area - 7°53'32"N/10°3'45"E", Gindin Dorawa - 7°53'49"N/10°3'50"E", Dadin Wanka - 7°54'24"N/10°3'45"E) as named by the locals were selected for sampling and are depicted in Figure 1.0.

1.2 Sampling site

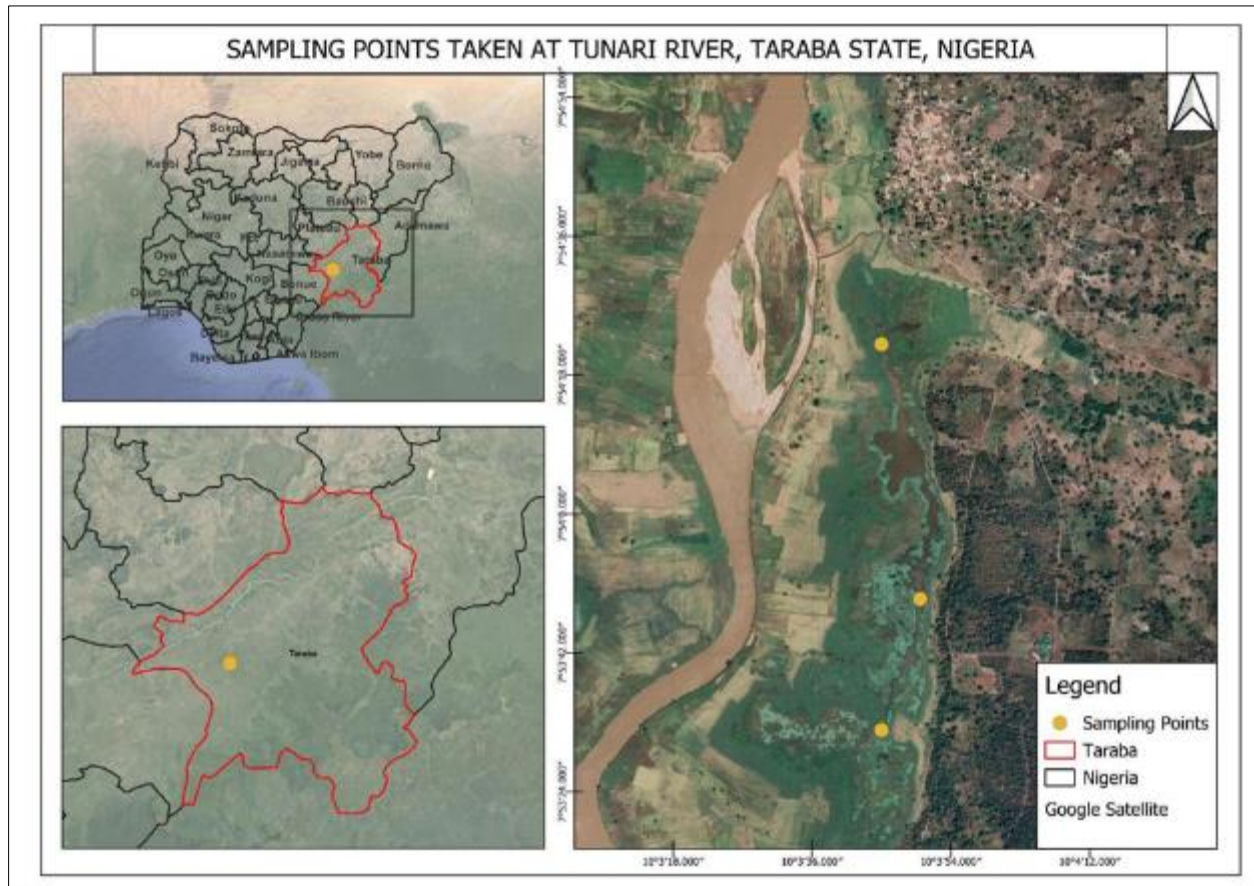


Figure 1 Map of the study area, and its sampling sites where water samples were collected

2 Materials and methods

2.1 Sample Collection

In May 2022, water samples were collected aseptically from three selected sites of the lake designated Abuja Area (AA), Gindin Dorawa (GD) and Dadin Wanka (DW). The location of all sample sites was recorded as Abuja Area - $7^{\circ}53'32''\text{N}/10^{\circ}3'45''\text{E}$, Gindin Dorawa - $7^{\circ}53'49''\text{N}/10^{\circ}3'50''\text{E}$, Dadin Wanka - $7^{\circ}54'24''\text{N}/10^{\circ}3'45''\text{E}$.

Briefly, sample bottles (1 Litre) were dipped into the Lake, by lowering them 20cm deep into the water body. After collection, sample bottles were immediately covered and stored in a cooler box containing ice packs and transported to the Laboratory. All samples were collected in duplicates for both physicochemical and microbiome analysis resulting in a total of 12 water samples. *In-situ* physicochemical (temperature and pH) analysis was done and recorded on the sampling site, using a mercury glass thermometer and a pH meter (MeterTech) respectively.

2.2 Physicochemical examination of water samples

Water samples for physicochemical analysis were transported in ice packs within 24 to 48 hours of collection, to the Soil Microbiology Laboratory at the International Institute of Tropical Agriculture, Ibadan, Oyo State, Nigeria. Twenty-two physicochemical parameters (in addition to temperature and pH) were considered in this study. They include Total Dissolved Solids, Electrical Conductivity, Turbidity, cations and anions such as Calcium, Magnesium, Potassium, Sodium, Manganese, Iron, Zinc, Lead, Nickel, Chromium, Cobalt, Cadmium, Phosphorus, Nitrates and Sulphates using spectrophotometric, colorimetric and titrimetric assays [13].

2.3 Microbiome Analysis

2.3.1 DNA Extraction

Total genomic DNA extraction was done as previously described by [14]. Briefly, water samples were pumped through a 0.2µm membrane filter using a vacuum pump, to concentrate bacterial cells onto the membrane filter. Following this, membrane filters were subjected to DNA extraction using ZymoBIOMICS MiniPrep Kit (Zymo Research, United States), following the manufacturer's instructions.

2.3.2 Library Preparation, Amplification and Full Length 16s Metagenomic Sequencing

Bacterial communities present in Lake Tunari water samples were analyzed using the PacBio Single-Molecule Real-Time platform (Pacific Biosciences, USA). Forward and reverse primers employed in the full-length amplification process were as follows: 27F (5'- AGRGTTYGATYMTGGCTCAG -3') and 1492R (5'- RGYTACCTTGTTACGACTT -3'). SMRTbell libraries were created from the amplified DNA using the "Procedure & Checklist - Full-Length 16S Amplification SMRTbell® Library Preparation and Sequencing procedure- Part Number 101-599-700 Version 02" according to the manufacturer's instruction (Pacific Biosciences, CA, USA) as detailed in supplementary document 1.

2.3.3 Bioinformatics Analysis

Raw sequences were processed using the DADA2 workflow designed for PacBio long-read amplicons [15]. The following parameters were used for quality control and filtering: minQ set to 3, minLen set at 1000, maxLen fixed to 1600 and maxN set to 0. Thereafter, chimeras were removed, subjected to error-model learning and denoised to generate Amplicon Sequence Variants (ASVs) using default DADA2 parameters. Assignment of ASVs to various taxonomies was computed using the SILVA 16s rRNA database (version 132) [16]. Abundance and taxa tables produced by DADA2 were thereafter imported into a phyloseq object (v1.28.0) [17] where further downstream analysis, such as, alpha diversity computation, taxonomic compositions bar plots and Venn diagrams were plotted [18, 19].

2.4 Statistical Analysis

Generated results were subjected to analysis of variance, while all means were separated using Duncan's Multiple Range Test at a 5% level of significance, using the SPSS version 25 statistical software. For generated physicochemical data, all means were separated using Duncan's Multiple Range Test at a 5% level of significance using SPSS (version 25.0). In addition, alpha diversity statistics were computed (using Shannon's, Inverse Simpsons Phylogenetic Diversity and Observed Species Index), to further examine the microbiome composition available per sample site. The non-parametric Kruskal-Wallis rank sum test was implemented to evaluate alpha diversity variations per sample site.

2.5 Data Availability

All raw PacBio HiFi fastq files generated in this study have been deposited into NCBI Sequence Read Archive, with SRA numbers SAMN36949871, SAMN36949872, SAMN36949873 under the Bioproject ID: PRJNA1004677, titled Bacteriome Diversity of Lake Tunari, Taraba State.

3 Results

3.1 Physical and chemical characteristics of water samples from different sites of Lake Tunari

Table 1 illustrates the physical and chemical characteristics of water samples collected at three different points of Lake Tunari.

Considering the physical qualities of the Lake, the mean values of pH, temperature, turbidity, Total Dissolved Solids and Electrical Conductivity were assessed. There was a significant difference ($P < 0.05$) between pH values recorded across all sampling points (AA, DW and GD) within Lake Tunari. However, site DW recorded the highest mean value at 7.20. The temperature range of the Lake was quite narrow, recording the lowest temperature at 28.00°C at sampling points AA and GD, and the highest temperature at 29.00°C at DW. These values obtained across these sites were, however, statistically insignificant ($P < 0.05$). Turbidity mean values ranged from 4.33 FNU at GD – 13.30 FNU at DW, and these values were significantly different ($P < 0.05$) across all sites. The electrical conductivity of water samples from different points of the lake depicted significantly different values ($P < 0.05$), with GD recording the highest mean value of 73.00 S/m. Mean values for Total Dissolved Solids in AA and DW were uniform (0.016 ppm), while the GD site recorded a value of 0.006 ppm.

Chemical qualities of water samples collected from the lake showed that mean values of Chromium (0.01 – 0.03 ppm) and Cadmium (0.01 ppm) across all sampled sites were not statistically significant ($P < 0.05$). In addition, Lead (0.07 ppm), Nickel (0.03 ppm), Cobalt (0.01 ppm) and Selenium (0.05 ppm) depicted similar values in sites DW and GD. Furthermore, site AA recorded the highest mean values of Calcium (200.59 ppm), Magnesium (5.04 ppm), Zinc (0.19 ppm), Sulphates (10.15 ppm) and Phosphorus (0.003 ppm). Potassium, Manganese, Iron, Copper and Nitrates were highest at site DW with statistically significant mean values of 89.43 ppm, 0.48 ppm, 5.73 ppm, 0.23 ppm and 1.78 ppm, respectively, while site GD recorded the highest value of Sodium (2.15 ppm).

Table 1 Mean physicochemical values of Lake Tunari water samples

Parameter	Sample Sites				WHO Limits (2011)
	AA	DW	GD	Significance	
Ca (ppm)	200.59 ^a	109.86 ^b	59.46 ^c	*	200.00
Mg (ppm)	5.049 ^a	3.09 ^b	0.68 ^c	*	200.00
K (ppm)	67.42 ^b	89.43 ^a	30.93 ^c	*	30.00
Na (ppm)	1.43 ^b	1.14 ^c	2.15 ^a	*	200.00
Mn (ppm)	0.27 ^b	0.48 ^a	0.07 ^c	*	0.05
Fe (ppm)	2.00 ^b	5.73 ^a	0.31 ^c	*	0.50 - 50.00
Cu (ppm)	0.09 ^b	0.23 ^a	0.07 ^c	*	2.00
Zn (ppm)	0.1 ^{9a}	0.09 ^b	0.05 ^c	*	5.00
Pb (ppm)	0.14 ^a	0.07 ^b	0.07 ^b	*	0.05
Ni (ppm)	0.06 ^a	0.03 ^b	0.03 ^b	*	0.10
Cr (ppm)	0.01 ^a	0.03 ^a	0.01 ^a	NS	0.05
Co (ppm)	0.03 ^a	0.01 ^b	0.01 ^b	*	0.01
Cd (ppm)	0.01 ^a	0.01 ^a	0.01 ^a	NS	0.05
Se (ppm)	0.10 ^a	0.05 ^b	0.05 ^b	*	0.04
NO ₃ (ppm)	1.17 ^b	1.78 ^a	0.27 ^c	*	<50.00
SO ₄ (ppm)	10.15 ^a	4.63 ^c	8.30 ^b	*	250.00
P (ppm)	0.003 ^a	0.002 ^b	0.001 ^c	*	0.001
TDS (ppm)	0.016 ^a	0.016 ^a	0.006 ^b	*	600.00
EC (S/m)	65.00 ^b	61.01 ^c	73.00 ^a	*	<1000.00
Turbidity (NTU)	12.60 ^b	13.30 ^a	4.33 ^c	*	5.00
Temperature (°C)	28.00 ^a	29.00 ^a	28.00 ^a	NS	30
pH	6.50 ^c	7.20 ^a	6.80 ^b	*	6.50 – 8.50

AA - Abuja Area, GD - Gindin Dorawa, DW - Dadin Wanka, Ca - Calcium, Mg - Magnesium, K - Potassium, Na - Sodium, Mn - Manganese, Fe - Iron, Cu - Copper, Zn - Zinc, Pb - Lead, Ni - Nickel, Cr - Chromium, Co - Cobalt, Cd - Cadmium, Se - Selenium, NO₃ - Nitrates, SO₄ - Sulphates, P - Phosphorus, TDS - Total Dissolved Solids. Mean values with similar letter(s) across rows are not significantly different at 5% level of significance by Duncan's Multiple Range Test (DMRT). NS- Not significant; * - Significant

3.2 Bacterial distribution of reads, composition, and diversity of Lake Tunari water samples

Table 2 shows the distribution of reads, taxa and alpha diversity values of bacterial communities present in three different sampling points of Lake Tunari. A total of 21,774 genomic reads were obtained. The lowest reads were obtained from site DW (5125 reads), being 23% of total reads, while the highest reads were observed to belong to site GD (8959) which was 40% of all reads, and site AA had 7990 reads constituting 37% of total genomic reads. Alpha diversity indices such as Shannon, Inverse Simpson, observed species, and Phylogenetic Diversity were computed. These were computed to scrutinize the microbial structure present in different sampling points of Lake Tunari, in terms

of phylogenetic relatedness, evenness and richness of species. Statistics for Shannon indices varied from 7.61 to 7.88, while Inverse Simpson's values varied considerably from 239.64 to 1700.32. Observed species values ranged from 2353 to 3527, and Phylogenetic diversity values ranged from 153.25 to 185.24. Bacterial consortia in site AA were represented by 15 phyla, 27 classes, 76 orders, 111 families, 188 genera and 204 species. Site GD comprised of 16 bacterial phyla, 28 classes, 68 orders, 106 families, 199 genera and 206 bacterial species. Lastly, the bacterial population in site DW was characterized by 17 phyla, 30 classes, 75 orders, 123 families, 217 genera and 174 bacterial species.

Table 2 Summary of bacterial diversity, reads, and composition of Lake Tunari water samples

	AA	GD	DW
Total number of reads	7990	8659	5125
Percentage distribution of reads	37	40	23
Shannon	7.61	7.89	7.69
Inverse Simpson	239.64	990.94	1700.32
Observed species	3244.00	3527.00	2353.00
Phylogenetic Diversity	163.65	185.24	153.252
Number of Phyla	13	17	17
Number of Class	27	28	30
Number of Order	76	68	75
Number of Family	111	106	123
Number of Genus	126	136	169
Number of Species	204	206	174

Among thirteen phyla identified in the AA sample, the most prevalent bacterial phyla were Firmicutes (45.39%), Proteobacteria (33.69%), Planctomycetota (10.40%) and Actinobacteriota (6.78%). In the DW sample, seventeen bacterial phyla were recovered, with six of them being the most abundant, namely, Proteobacteria (64.13%), Actinobacteriota (8.74%), Bacteroidota (7.8%), Planctomycetes (5.58%), Verrucomicrobiota (3.96%) and Cyanobacteria (3.82%). Seventeen bacterial phyla were identified from GD sample, however, Proteobacteria (62.98%), Planctomycetota (16.11%), Bacteroidota (4.64%), Gemmatimonadota (4.37%), Verrucomicrobiota (2.80%), Acidobacteriota (2.72%), Chloroflexi (2.12%) and Actinobacteriota (1.73%) were the most abundant. Furthermore, Proteobacteria and Planctomycetota were the only phyla that were present in large abundance across all sample sites (Figure 2A).

Figure 2B shows the relative abundance (%) of genera identified from all designated sites. Site AA showed that *Exiguobacterium*, Unknown, *Methylocystis*, *Bacillus*, *Clostridium sensu stricto* 13, *Fictibacillus*, *Noviherbaspirillum* and *Mycobacterium* were the most abundant with a relative abundance of 30.85%, 18.85%, 5.22%, 3.55%, 3.35%, 2.34%, 2.20% and 2.03% respectively. Other identified bacterial genera with $\leq 1\%$ relative abundance were 118 genera. DW site depicted unknown (38.36%), *Methylocystis* (3.79%), *Mycobacterium* (2.96%), *Rhodofera* (2.51%) and *Limnohabitans* (2.00%) as the four most abundant genera present in the site. Other genera such as *Terrimonas*, *Paucibacter*, *Pelomonas*, in addition to 161 other genera were also detected, but they had relative abundance that was $\leq 1\%$. Lastly, Unknown (32.49%), *Lysobacter* (14.05%), *Schlesneria* (5.42%), *Paulibacter* (3.42%), *Novosphingobium* (2.63%), *Curvibacter* (2.12%) and *Gemmatimonas* (2.06%) were ranked as the most abundant bacterial genera recovered from GD site. Other genera that were $\leq 1\%$ were 129, including *Duganella* and *Aquisphaera*.

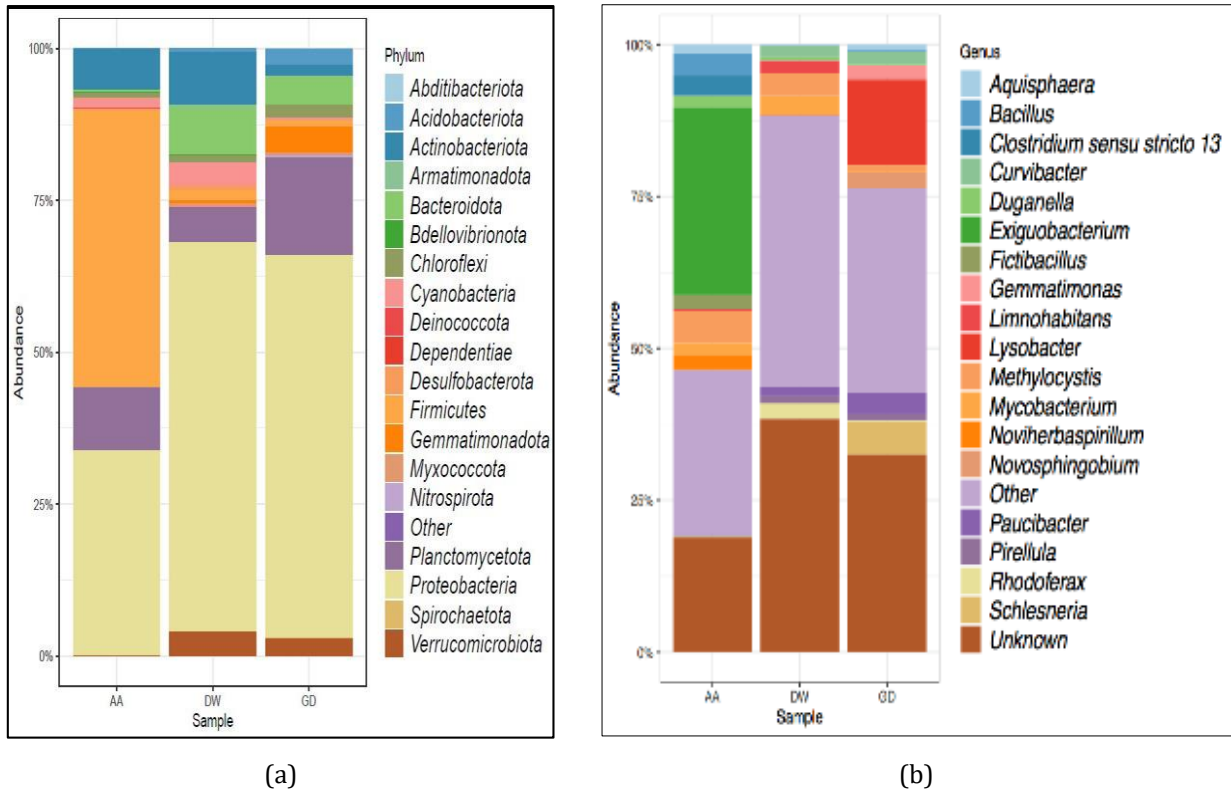
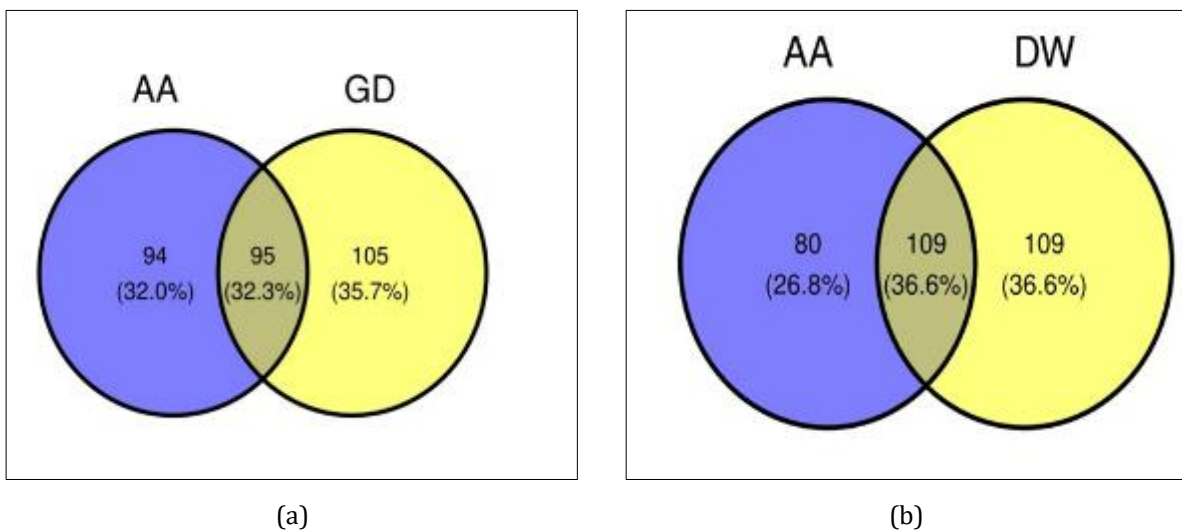


Figure 2 Taxonomic compositional bar plots illustrating the percentage relative abundance of top 19 (a) Bacterial phyla and (b) Bacterial genera, recovered from sampling sites AA, DW and GD of Lake Tunari

Figure 3A depicts sample sites AA as having 94 unique genera, and 105 genera, exclusive to GD only. It also showed that they had 95 genera that were common to both sites, accounting for 32.3% of all genera identified across both sites. Furthermore, the Venn diagram showed that site GD had more unique genera (35.7%) than site AA (32.0%). Figure 3B shows common and unique phyla of sites AA and DW. It depicted AA and DW as sites in possession of 80 (26.8%) and 109 (36.6%) unique genera. It also recorded common genera between these sites as 36.6% of all observed genera. Figure 3C describes shared genera between sites GD and DW as 37.0% of total genera observed among these sites. It also illustrated that 28.5% of the total genera were unique to site GD, and 34.4% were unique to site DW. Figure 3D depicts common genera observed among all sites and among 2 different sites simultaneously. It also showed genera distinct from individual sample sites. AA, GD and DW were observed to have 20.3% of genera common to their locations. Additionally, sites AA, GD and DW, possessed 16.2%, 18.1% and 19.2% of taxa unique to them respectively.



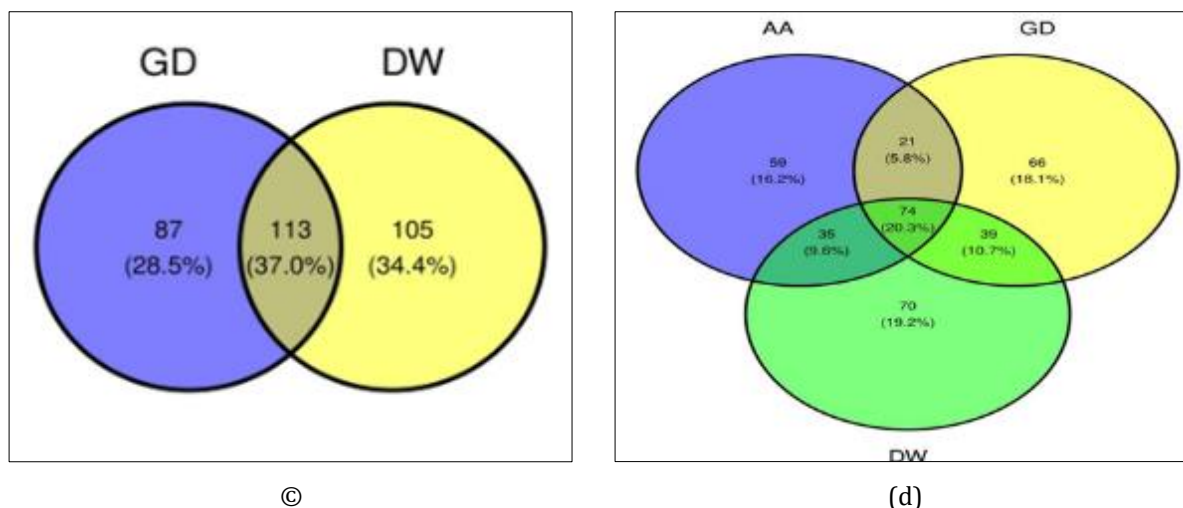


Figure 3 Venn diagram indicating shared genera as intersecting areas, and unique genera between (A) sites AA and GD (B) sites AA and DW (C) sites GD and DW and (D) sites AA, GD and DW

4 Discussion

Surface water bodies such as springs, rivers, streams and lakes are vital sources of freshwater [20]. The shape, connectivity and sizes of surface water bodies can produce significant effects on their ecosystems, as well as their ecological communities [21]. The percentage of freshwater bodies on earth has been estimated to be 2.5%, of which lakes account for about 0.3% of total freshwater bodies. Lakes have been accounted to be one of the most crucial sources of water supply, as they are being used for human consumption, such as drinking, and for meeting other anthropogenic needs [20]. The quality of these water bodies can however be accessed via three parameters: biological, physical and chemical parameters [22]. Water is considered safe if these parameters are within the range of standards set by recognised regulatory bodies concerned with aquatic systems, such as World Health Organization, Standard Organisation of Nigeria, etc. [23]. Twenty-two physicochemical parameters were analyzed to determine the quality of water samples from lake Tunari. These parameters included: Chemical parameters such as cations, anions, and some heavy metals, while, physical parameters considered were, temperature, pH, Total dissolved solids (TDS), turbidity and electrical conductivity. Water bodies assessment also include biological monitoring of aquatic systems. They are one of the foremost concerns of water quality. This usually involves plants, animals or microbial surveillance of water bodies [24]. This study, therefore, serves as a baseline for unraveling the microbial diversity and physicochemical attributes of Lake Tunari in Taraba state.

Microorganisms play a significant role in the functionality of aquatic ecosystems. They act as decomposers of organic matter, producers and recyclers of nutrients in aquatic environments [25, 26]. Bacterial diversity studies of natural water bodies are crucial methods to evaluate the evolution and ecology of bacteria and can also be used to support risk assessment studies [27]. It has been widely established that culture-independent methodologies provide a more robust view of naturally occurring microbiomes than culture-dependent analysis [28]. Hence, they are being used more intensively in microbial diversity studies, than conventional methods, such as isolation of pure cultures, chemical assays, and morphological identification [14].

The concentration of cations such as magnesium, sodium and copper from all sample sites was very low when compared to permissible WHO limits. However, the concentration of calcium was relatively high, exceeding permissible standards, in site AA. Likewise, excess concentrations of potassium ions, which were observed from all sample sites surpassed acceptable standards of WHO, 2011 (Table 1). Elevated concentrations of potassium in natural water bodies may limit the proliferation of some species of algae, for instance, the Chryophytes. This form of algae plays an important role in the aquatic food chain, as a primary producer, and also contributes to the production of nutrients and oxygen [29]. Excessive cations such as potassium observed in Lake Tunari may be due to runoff of synthetic fertilizers from nearby farmlands. A similar type of runoff is described by [30].

Heavy metals are considered deleterious to human health, as well as aquatic life [31]. They are distributed in almost every ecological niche due to their use in industrial, medical, agricultural and even domestic applications [32]. Analysis of heavy metals such as zinc, nickel, chromium, cadmium, copper and Iron all fell within the acceptable range of the WHO. In contrast, other metals such as manganese, lead, cobalt and selenium, exceeded the acceptable range at different

locations of the lake. The presence of these heavy metals in the lake may signify that certain anthropogenic activities and domestic effluents may be contributing factors. Likewise, these metallic ions might have been deposited to the lake over a while, from other adjoining river bodies, such as River Donga. Their concentrations in high levels might indicate that the lake might pose a risk to individuals who consume aquatic foods in high proportions harvested from the lake. It can also be considered dangerous to aquatic life, including plants.

The presence of anions such as nitrates, sulfates and phosphorus from various points of the lake indicated that at site AA and DW, phosphorus content had surpassed the accepted limit. As much as phosphorus is fundamental to the growth of microorganisms, it is used for DNA synthesis and repair. However, excess phosphorus in water bodies would inadvertently reduce primary productivity by increasing eutrophication [33]. Some anthropogenic activities observed around the lake included domestic activities such as washing of clothes and farming tools. This could have contributed to the slight increment in phosphorus in sites AA and DW. Turbidity is a measure of the relative clarity or cloudiness of water [34]. Turbidity in aquatic systems can be induced by the presence of sediments generated from erosion, algal growth, urban runoffs and matter from decomposing vegetation [35]. It is known to have a strong effect on aquatic life in terms of sunlight penetration and can also provide a medium for microbial growth [36]. The high values obtained in this study area (4.33 NTU – 13.30 NTU) are strong indicators of pollution in Lake Tunari. In this study, it appeared there was a positive link between total dissolved solids and turbidity. Sites AA and DW expressed the highest turbidity values, while the highest TDS value (0.016 ppm) was also observed in both sites. Furthermore, site GD, with the lowest turbidity value (4.33 NTU) equally showed the lowest mean value for TDS (0.006 ppm).

In this study, Firmicutes were the most dominant phylum recovered from site AA (%). Firmicutes have been described to contain arrays of carbohydrate-active enzymes (CAZymes) responsible for the breakdown of complex polysaccharides in plant cell walls, such as cellulose, hemicellulose and lignin [36]. A wide variety of assorted bacterial genera were recovered from this site, such as *Bacillus*, *Methylocystis* sp., *Clostridium sensu stricto* 13 and the most abundant genera, *Exiguobacterium* (Figure 2B). *Exiguobacterium* are Gram-positive, facultative anaerobes, with various morphologies, ranging from short rods to cocci, belonging to the Firmicutes phylum [37]. They have been isolated from several environmental niches, such as freshwaters, soil, plant rhizospheres, marine sediment and seawater [38]. They have also been isolated from extreme environments, such as glaciers and salt lakes [39]. Genomic insights of some species of *Exiguobacterium* have revealed the presence of multiple stress-responsive genes, such as resistance to toxic compounds including chromium, cadmium, zinc mercury, cobalt, lead and antibiotics, high salt tolerance [40, 41]. The presence of bacterial consortia such as *Exiguobacterium* sp., *Bacillus* sp. and *Methylocystis* sp. (Figure 2B) with unique biodegradative capabilities, and elevated tolerance to salts is not surprising, as physicochemical studies of site AA have indicated the occurrence of heavy metals (lead, cobalt, selenium and manganese) and some cations (calcium, phosphorus and potassium) in elevated concentration. There is therefore a strong indication that the lake might be receiving effluents containing toxic materials during seasonal flooding from external sources. For instance, the inflow of inorganic debris from River Donga, and the possible activity of community dwellers dumping household items into nearby canals during a flooding event, might ultimately be fed into the lake.

The proportion of the phylum Proteobacteria was the highest across GD and DW sample sites (Figure 2A). This is not surprising as several researchers have proven the dominance of this bacterial phylum in several lakes [42, 43, 44].

Site DW recorded high concentrations of selenium and lead, and also showed the largest concentration of potassium, manganese, iron, copper, zinc, nitrates and unclassified sequences among the three sampled sites. Our microbiome study showed that this site harbored interesting bacterial genera involved in bioremediation, metal and carbon cycling. Some of such genera detected were *Methylocystis* sp., a methanotroph, equipped with the mechanism of accruing polyhydroxybutyrate (PHB) [45]. It has also been documented to be involved in facilitating methanotrophic bioremediation [46]. Another striking genus detected from site DW was *Rhodoferrax* sp., a Gram-negative, motile rod, frequently discovered in stagnant aquatic niches such as lakes [47]. Some species of *Rhodoferrax* have been reported as being capable of chemolithoautotrophic growth under limited aerobic conditions, by oxidizing iron, hydrogen and thiosulfate. Another report [48] stated that *Rhodoferrax ferrireducens* is capable of utilizing manganese(IV) oxide, nitrate and fumarate in a bid to thrive under anaerobic conditions. The discovery of these microbes is not unexpected in Lake Tunari, since members of the *Rhodoferrax* and *Methylocystis* genera are commonly found in aquatic environments contaminated with chemical compounds such as herbicides and hydrocarbons [49].

Site GD is not alien to cattle and human disturbances, as it was recorded to be a preferable spot for livestock watering, yet physicochemical studies show that a larger proportion of parameters examined were within the WHO standard range. Also, it is noteworthy that this site recorded the lowest value in turbidity and total dissolved solids, some cations and anions but was in concentrations exceeding this standard in terms of selenium, lead and manganese. This location harboured a wide array of bacteria, some of which include: *Gemmatimonas* of the phylum Gemmatimonadetes,

Schlesneria of the phylum Planctomycetota, and a consortium of bacteria belonging to the large Proteobacteria phylum. According to [50], the bacterial phylum Gemmatimonadetes are among the understudied group of bacteria found in natural habitats. Studies based on 16S rRNA genes have shown that these phyla can adapt in oligotrophic conditions [51], similar to what was observed in site GD. Metagenomic studies have shown the ubiquity of these phyla in soil, lakes and sediments [52].

5 Conclusion

This pilot study has presented the bacterial diversity occupying the unique Tunari Lake, using high throughput sequencing methodology. It has also showcased both the physical and chemical attributes of the lake. This lake undoubtedly contains bacterial consortia and physicochemical values indicative of pollution, and this study has highlighted possible factors that might be responsible. Lake Tunari seemed to be a potential haven for microbial communities that could be harnessed for bioremediation studies. Lastly, this research can serve as a baseline study for future bacteriome and ecological studies of the lake. To the best of our knowledge, this is the first microbiome and physicochemical report of Lake Tunari, situated in Taraba state, Nigeria.

Compliance with ethical standards

Disclosure of conflict of interest

The authors declare no conflict of interest regarding the publication of this paper.

Supplementary Data

<https://sciesjournals.com/ijstra/sites/default/files/Supplementary-File-IJSTRA-2024-0043.pdf>

References

- [1] Bernard BM, Iyiola T. Assessment of Woody Species Diversity in different Ecological Zones of Taraba State Nigeria: A strategy for conservation. *Asian Journal of Research in Agriculture and Forestry*, 2018;1(4):1-12.
- [2] Lacoul P. Aquatic Macrophyte Distribution in response to physical and chemical environment of the lakes along an altitudinal gradients in Himalayas, Nepal,(Ph.D. dissertation), Dalhousie University, Halifax; 2004
- [3] Crump BC, Adams HE, Hobbie JE, Kling GW. Biogeography of bacterioplankton in lakes and streams of an arctic tundra catchment. *Ecology*. 2007 Jun;88(6):1365-78.
- [4] Anhwange BA, Agbaji EB, Gimba EC. Impact assessment of human activities and seasonal variation on River Benue, within Makurdi Metropolis. *International journal of Science and Technology*. 2012 May;2(5):248-254.
- [5] Kumar M, Yadav AN, Tiwari R, Prasanna R, Saxena AK. Evaluating the diversity of culturable thermotolerant bacteria from four hot springs of India. *J Biodivers Biopros Dev*. 2014;1(127):2.
- [6] Joshi P, Pande V, Joshi P. Microbial diversity of aquatic ecosystem and its industrial potential. *J. Bacteriol. Mycol. Open Access*. 2016;3:177-9.
- [7] Jurgens G, Glöckner FO, Amann R, Saano A, Montonen L, Likolammi M, Münster U. Identification of novel Archaea in bacterioplankton of a boreal forest lake by phylogenetic analysis and fluorescent in situ hybridization. *FEMS Microbiology Ecology*. 2000 Oct 1;34(1):45-56.
- [8] Bertrand JC, Brochier-Armanet C, Gouy M, Westall F. For three billion years, microorganisms were the only inhabitants of the earth. *Environmental Microbiology: Fundamentals and Applications: Microbial Ecology*. 2015:75-106.
- [9] Sandrin TR, Dowd SE, Herman DC, Maier RM. Aquatic environments. In *Environmental microbiology 2009* Jan 1 (pp. 103-122). Academic Press.
- [10] Lozupone CA, Knight R. Global patterns in bacterial diversity. *Proceedings of the National Academy of Sciences*. 2007 Jul 3;104(27):11436-40.
- [11] Zwart G, van Hannen EJ, Kamst-van Agterveld MP, Van der Gucht K, Lindström ES, Van Wichelen J, Lauridsen T, Crump BC, Han SK, Declerck S. Rapid screening for freshwater bacterial groups by using reverse line blot hybridization. *Applied and Environmental Microbiology*. 2003 Oct;69(10):5875-83.

- [12] Blench R. An atlas of Nigerian languages. 4th ed. Oxford: Kay Williamson Educational Foundation; 2012.
- [13] Albakaa AR, Ameen DS, Abed NK, Jabbar ZA, Musaa LA. Quantification of Ca, K, Mg, Zn and Fe elements in grape leaves from different regions of Iraq by atomic absorption spectroscopy. In *Journal of Physics: Conference Series* 2021 Mar 1 (Vol. 1853, No. 1, p. 012018). IOP Publishing.
- [14] Adedire DE, Jimoh AO, Kashim-Bello Z, Shuaibu BA, Popoola OA, Pate KI, Uzor OS, Etingwa E, Joda JF, Opaleye OO, Ogunlowo VA. Microbiome diversity analysis of the bacterial community in idah river, Kogi state, Nigeria. *Advances in Microbiology*. 2022 May 9;12(5):343-62.
- [15] Callahan BJ, Wong J, Heiner C, Oh S, Theriot CM, Gulati AS, McGill SK, Dougherty MK. High-throughput amplicon sequencing of the full-length 16S rRNA gene with single-nucleotide resolution. *Nucleic acids research*. 2019 Oct 10;47(18):pp.e103-e103.
- [16] Quast C, Pruesse E, Yilmaz P, Gerken J, Schweer T, Yarza P, Peplies J, Glöckner FO. The SILVA ribosomal RNA gene database project: improved data processing and web-based tools. *Nucleic acids research*. 2012 Nov 27;41(D1):D590-6.
- [17] McMurdie PJ, Holmes S. phyloseq: an R package for reproducible interactive analysis and graphics of microbiome census data. *PloS one*. 2013 Apr 22;8(4):e61217.
- [18] Shetty SA, Lahti L. *Microbiomeutilities: Utilities for Microbiome Analytics*. 2020
- [19] Gao CH, Yu G, Cai P. ggVennDiagram: an intuitive, easy-to-use, and highly customizable R package to generate Venn diagram. *Frontiers in Genetics*. 2021 Sep 7;12:706907.
- [20] Vasistha P, Ganguly R. Water quality assessment of natural lakes and its importance: An overview. *Materials Today: Proceedings*. 2020 Jan 1;32:544-52.
- [21] Steele, M.K., & Heffernan, J.B. (2014). Morphological characteristics of urban water bodies: mechanisms of change and implications for ecosystem function. *Ecological Applications*, 24(5), 1070-1084.
- [22] Ben-Coker MO. Effect of Slaughter house discharge on Water Quality of Ikpoba River, Nigeria. *Bioscene Technology*. 2012;52(1):1-4.
- [23] Onyegeme-Okerenta BM, Obia C, Wegwu MO. Physicochemical properties of water quality of Imeh, Edegelem and Chokocho communities located along Otamiri-oché River in Etche Ethnic Nationality of Rivers State, Nigeria. *Journal of Applied Sciences and Environmental Management*. 2016;20(1):113-9.
- [24] Boudeffa K, Fekrache F, Bouchareb N. Physicochemical and biological water quality assessment of the Guebli River, northeastern Algeria. *Rasayan J. Chem*. 2020 Jan 1;13(1):168-76.
- [25] Tang X, Xie G, Shao K, Tian W, Gao G, Qin B. Aquatic bacterial diversity, community composition and assembly in the semi-arid inner mongolia plateau: combined effects of salinity and nutrient levels. *Microorganisms*. 2021 Jan 20;9(2):208.
- [26] Custodio M, Espinoza C, Peñaloza R, Peralta-Ortiz T, Sánchez-Suárez H, Ordinola-Zapata A, Vieyra-Peña E. Microbial diversity in intensively farmed lake sediment contaminated by heavy metals and identification of microbial taxa bioindicators of environmental quality. *Scientific Reports*. 2022 Jan 7;12(1):80.
- [27] Vaz-Moreira I, Egas C, Nunes OC, Manaia CM. Culture-dependent and culture-independent diversity surveys target different bacteria: a case study in a freshwater sample. *Antonie Van Leeuwenhoek*. 2011 Aug;100:245-57.
- [28] Fasesan D, Dawkins K, Ramirez R, Rasheed-Jada H, Onilude A, Nash O, Esiobu N. Analysis of a tropical warm spring microbiota using 16S rRNA metabarcoding. *Advances in Microbiology*. 2020 Apr 14;10(4):145-65.
- [29] Moriarty DJ. The role of microorganisms in aquaculture ponds. *Aquaculture*. 1997 May 15;151(1-4):333-49.
- [30] Khare MJ, Warke A. Selection of significant input parameters for water quality prediction-a comparative approach. *Int J Res Advent Technol*. 2014;2(03):81-90.
- [31] Tchounwou PB, Yedjou CG, Patlolla AK, Sutton DJ. Heavy metal toxicity and the environment. *Molecular, clinical and environmental toxicology: volume 3: Environmental toxicology*. 2012:133-64.
- [32] He ZL, Yang XE, Stoffella PJ. Trace elements in agroecosystems and impacts on the environment. *Journal of Trace elements in Medicine and Biology*. 2005 Dec 2;19(2-3):125-40.
- [33] Reeder BC. Primary productivity limitations in relatively low alkalinity, high phosphorus, oligotrophic Kentucky reservoirs. *Ecological engineering*. 2017 Nov 1;108:477-81.

- [34] Health Canada Guidelines for Canadian drinking water quality: Guideline technical document—Turbidity. Water and Air Quality Bureau, Healthy Environments and Consumer Safety Branch; Health Canada: Ottawa, ON, Canada. 2012
- [35] Otieno AA, Kitur EL, Gathuru G. Physico-Chemical Properties of River Kisat, Lake Victoria Catchment, Kisumu County, Kenya. *Environment Pollution and Climate Change*. 2017;1(4):1-4.
- [36] Gavande PV, Basak A, Sen S, Lepcha K, Murmu N, Rai V, Mazumdar D, Saha SP, Das V, Ghosh S. Functional characterization of thermotolerant microbial consortium for lignocellulolytic enzymes with central role of Firmicutes in rice straw depolymerization. *Scientific reports*. 2021 Feb 4;11(1):3032.
- [37] López MC, Galán B, Carmona M, Navarro Llorens JM, Peretó J, Porcar M, Getino L, Olivera ER, Luengo JM, Castro L, García JL. Xerotolerance: a new property in *Exiguobacterium* genus. *Microorganisms*. 2021 Nov 28;9(12):2455.
- [38] Zhang J, Ding X, Guan R, Zhu C, Xu C, Zhu B, Zhang H, Xiong Z, Xue Y, Tu J, Lu Z. Evaluation of different 16S rRNA gene V regions for exploring bacterial diversity in a eutrophic freshwater lake. *Science of the Total Environment*. 2018 Mar 15;618:1254-67.
- [39] Jiang X, Xue Y, Wang L, Yu B, Ma Y. Genome sequence of a novel polymer-grade l-lactate-producing alkaliphile, *Exiguobacterium* sp. strain 8-11-1. *Genome announcements*. 2013 Aug 29;1(4):10-128.
- [40] Bian G, Zhang Y, Qin S, Xing K, Xie H, Jiang J. Isolation and biodiversity of heavy metal tolerant endophytic bacteria from halotolerant plant species located in coastal shoal of Nantong. *Wei Sheng wu xue bao= Acta Microbiologica Sinica*. 2011 Nov 1;51(11):1538-47.
- [41] Kasana RC, Pandey CB. *Exiguobacterium*: an overview of a versatile genus with potential in industry and agriculture. *Critical reviews in biotechnology*. 2018 Jan 2;38(1):141-56.
- [42] Newton RJ, Jones SE, Eiler A, McMahon KD, Bertilsson S. A guide to the natural history of freshwater lake bacteria. *Microbiology and molecular biology reviews*. 2011 Mar;75(1):14-49.
- [43] Zhang D, Zhu Z, Li Y, Li X, Guan Z, Zheng J. Comparative genomics of *Exiguobacterium* reveals what makes a cosmopolitan bacterium. *Msystems*. 2021 Aug 31;6(4):10-128.
- [44] Betiku OC, Sarjeant KC, Ngatia LW, Aghimien MO, Odewumi CO, Latinwo LM. Evaluation of microbial diversity of three recreational water bodies using 16S rRNA metagenomic approach. *Science of The Total Environment*. 2021 Jun 1; 771:144773.
- [45] Naizabekov S, Hyun SW, Na JG, Yoon S, Lee OK, Lee EY. Comparative genomic analysis of *Methylocystis* sp. MJC1 as a platform strain for polyhydroxybutyrate biosynthesis. *Plos one*. 2023 May 10;18(5):e0284846.
- [46] Im J, Semrau JD. Pollutant degradation by a *Methylocystis* strain SB2 grown on ethanol: bioremediation via facultative methanotrophy. *FEMS microbiology letters*. 2011 May 1;318(2):137-42.
- [47] Park M, Song J, Nam GG, Cho JC. *Rhodoferrax lacus* sp. nov., isolated from a large freshwater lake. *International Journal of Systematic and Evolutionary Microbiology*. 2019 Oct;69(10):3135-40.
- [48] Risso C, Sun J, Zhuang K, Mahadevan R, DeBoy R, Ismail W, Shrivastava S, Huot H, Kothari S, Daugherty S, Bui O. Genome-scale comparison and constraint-based metabolic reconstruction of the facultative anaerobic Fe (III)-reducer *Rhodoferrax ferrireducens*. *BMC genomics*. 2009 Dec;10:1-9.
- [49] Kasanke CP, Collins RE, Leigh MB. Identification and characterization of a dominant sulfolane-degrading *Rhodoferrax* sp. via stable isotope probing combined with metagenomics. *Scientific Reports*. 2019 Feb 28;9(1):3121.
- [50] Zeng Y, Baumbach J, Barbosa EG, Azevedo V, Zhang C, Koblížek M. Metagenomic evidence for the presence of phototrophic Gemmatimonadetes bacteria in diverse environments. *Environmental microbiology reports*. 2016 Feb;8(1):139-49.
- [51] Hanada S, Sekiguchi Y. The phylum gemmatimonadetes. *The prokaryotes*. 2014:677-81.