EVALUATION OF BACTERIAL AND HEAVY METAL STATUS OF WATER, SEDIMENT AND FISH SAMPLES FROM KAINJI AND JEBBA LAKES, NIGERIA

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ABSTRACT

Information on limnology and status of heavy metals of Kainji and Jebba Lakes are key in the development and management of the Lakes. These Lakes receive organic and inorganic wastes through animal husbandry operations, illegal mining activities, direct waste disposal and other human activities. Quantitative and qualitative analyses of bacterial isolates in water, sediment and fish samples (*Clarias gariepinus* and *Oreochromis niloticus*) from Kainji and Jebba Lakes were carried out using primary isolation media, microbact identification kits (12A and 12B MB1132A/Ausralia) and molecular analysis of some species. Concentrations of heavy metals: lead (Pb), copper (Cu), cadmium (Cd), chromium (Cr) and arsenic (As) in water, sediment and fish samples were determined using atomic absorption spectrophotometer (AA500). Physico-chemical parameters of the Lake water samples were determined. Biosorption potentials of bacterial isolates were also evaluated. Faecal coliform counts of water samples from Kainji Lake were below maximum permissible limit of 5.0×10^2 MPN/100 mL while sampling station 1 (580.83 MPN/100 ml) and 2 (700.83 MPN/100 ml) of Jebba Lake were above maximum permissible limit of 5.0×10^2 MPN/100 mL according to Federal Environmental Protection Agency (FEPA). Faecal coliform counts during wet season were higher than those obtained during the dry season in both Lakes. Bacterial species such as *Aeromonas aquatilis* strain AE 207, *Burkholderia pseudomallei, Bacillus lacus* AK 74, *Pseudomonas donghuensis* HYS, *Escherichia coli, Herbaspirillum aquaticum* strain IEH 4430, *Alcaligenes faecalis* strain Sihong 663-1, *Alcaligenes faecalis* strain HPRTAK198, *Oceanobacillus oncorhynchi* subsp. *incaldanensis* strain AM-75 were isolated from the water samples in both Lakes. *Vibrio alginolyticus*, *Moraxella* species, *Escherichia hermannii*, *Vibrio parahaemolyticus*, *Aeromonas hydrophila* were isolated from fish intestines and gills. Concentrations of Cu, Cr and As in water samples were below maximum permissible limit (2.0, 0.05 and 0.1 mg/L respectively). However, the concentrations of Cd were higher (0.09 and 0.17 mg/L for Kainji and Jebba Lakes respectively) than the maximum permissible limit of 0.01 mg/L according to Federal Environmental Protection Agency (FEPA). Concentrations of Pb in sample stations; Kl.2 (0.082 mg/L), Kl.3 (0.078 mg/L), Kl.4 (0.057 mg/L), Jl.1 (0.066 mg/L), Jl.2 (0.380 mg/L), Jl.3 (0.141 mg/L) and Jl.5 (0.064 mg/L) were above maximum permissible limit of 0.05 mg/ L. The concentrations of heavy metals in the water samples were higher during the wet season compared to the dry season. In fish samples, the concentrations of Pb in *Oreochromis niloticus* fish intestines (ONFI) and *Clarias gariepinus* fish intestines (CGFI) were above maximum permissible limit. Fish muscles of *Oreochromis niloticus* and *Clarias gariepinus* in both Lakes had low heavy metal concentrations. Heavy metal concentrations in sediment from the Lakes were found to be below threshold effect concentration (TEC). Physicochemical qualities of all the parameters examined in both Lakes were found to be within acceptable limit that is favourable for aquatic lives. Biosorption potentials of bacterial isolates revealed that *Bacillus lacus* strain AK 74 had high potential to biosorp Pb (99.0 %), Cu (98.0 %) and Cr (96.5 %) within 28 days at initial concentration of 1.0 parts per million. Kainji and Jebba Lakes are experiencing pollution due to increasing human activities. The results of this study showed high heavy metal concentrations in some of the sample stations; hence human activities such as mining in these stations should follow best practices.

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ABBREVIATIONS, GLOSSARIES AND SYMBOLS

µS	MicroSiemens
μScm^{-1} (µS/cm)	MicroSiemens per Centimetre
PPM	Part Per Million
>	Greater than
<	Less than
H^+	Hydrogen ion concentration
NCBI	National Centre for Biotechnology Information
VBC	Viable Bacterial Count
FC	Faecal Coliform
MPN	Most Probable Number
BLAST	Basic Local Allignment Search Tool
µm	Micrometre
µl	Microlitre
$	Dollar
SQGV	Sediment Quality Guideline Values
TEC	Threshold Effect Concentration
CFU	Colony Forming Unit
PCB	PolyChlorinated Biphenyl
Pb	Lead
Cu	Copper
Cd	Cadmium

Cr	Chromium
As	Arsenic
DO	Dissolved Oxygen
BOD	Biological Oxygen Demand
KL	Kainji Lake
JL	Jebba Lake
MPL	Maximum Permissible Limit

CHAPTER ONE

1.0 INTRODUCTION

1.1 Background to the Study

Microorganisms and heavy metals are common pollutants that originate from agricultural livestock areas, residential wastes, industries, mining zones, exhaust from automobiles and also natural factors such as weathering of soil and rocks. Aquatic pathogenic microorganisms constitute health-hazard to other aquatic lives and man. Most common pathogens include: *Salmonella, Shigella*, *Aeromonas, Leptospira* species, enteropathogenic *E.coli, Pasteurella, Vibrio* and *Mycobacterium* species. These organisms can be acquired by man and other animals through consumption of contaminated food and water sources. Fish exposed to certain contaminated food or water sources harbors different human pathogens and may carry these pathogens to clean streams and recreational areas. Direct faecal contamination by humans and other animals, wildlife, farm animals and farm runoffs are the major sources of microbial water pollution (Payment and Riley, 2002; Okafo *et al.*, 2003).

Metallic elements that have relatively high density of 5 g/cm^3 and above and are toxic or poisonous even at low concentration are referred to as heavy metals (Alloway and Ayres, 1993; Lenntech, 2004). Heavy metals such as cadmiun, aluminiun, arsenic, antimony, lead and mercury are classed as potentially toxic heavy metals. Nickel, vanadium and cobalt are classified as semi essential heavy metals while copper, zinc and selenium as essential heavy metals (Szentmihalyi and Then, 2007). The essential heavy metals are taken up from water, food or sediment for normal metabolism of fish (Canli and Atli, 2003). Excessive

intake of essential heavy metals results in toxic effect (Tuzen, 2003). Aquatic ecosystem is at the receiving end of these pollutants either through natural course (run off) or direct dumping of the waste into aquatic system by man.

Kainji Lake is the first man-made Lake in Nigeria with the main objective of providing hydro-power electricity generation for the country (Raji *et al*., 2011). However, it was realized that the Lake formation will have other impacts on the lives of the communities around it, particularly in the areas of agriculture – fisheries, wildlife and range; public health, human and infrastructural development of the fishing communities (with the experience from other parts of the world where similar man-made Lake have been established). Kainji Lake is a lentic water body on the Niger River, and it is located on the Niger and Kebbi State boarder in Northern Nigeria. The history of Kainji Lake dates back to the dam construction, which commenced in March 1964 and completed in December 1968 (Raji *et al*., 2011). The dam was built across the Niger River in Nigeria and is called Kainji Dam. Huge amount of electricity is generated in the dam and supplied to all the large cities in Nigeria and also sold to neighboring country, Niger. The formation of Kainji Lake is an epitome of history and also an important fish resources reservoir, contributing to the fish consumption and supplying important markets in Abuja, Lagos, Ibadan, Onitsha and Kano among others (Raji *et al*., 2011).

Niger River, which formed Kainji Lake, takes its source from the highlands of central Guinea located near the border between Guinea and Sierra Leone. The river flows Eastward between Timbuktu and Bourem and then takes a course in the Southeast direction through Gao in Mali and Niamey in Niger Republic, and finally to Kainji Lake in Nigeria. The river

forms part of the Niger-Benin border at the South of Niamey prior to entering Nigeria. Two distinct floods occur annually in the Lake. The first is the "black flood," which originates from Niger River (with its headwater from Guinea). The Niger River rise from the Fouta Djallon highlands in Central Guinea (an altitude of 1000 meter above sea level) and flows into Kainji Lake, a distance of 2737 km (Raji *et al*., 2011). As the water flows downstream, it drains the swamp of Timbuktu where it loses 65 percent of its water to evaporation and infiltration. Having deposited its silt in this swampy area, the water becomes relatively clean and appears black at a distant on its arrival at the Lake, hence, it is called 'black flood'. This flood occurs between December and March anually. The second flood is called "white flood" which originates from local runoffs around the catchments of Kainji Lake and this appears around August to November annually (with lot of silt and mud), thus, giving it the milky or whitish look from which it gets its name 'white flood'. The flood reaches its peak by September with higher volume than that of the black flood (Raji *et al*., 2011).

The construction of Kainji Lake led to the establishment of "Kainji Lake Research Project," which today is called "National Institute for Freshwater Fisheries Research, New-Bussa". The main function of Kainji Lake Research Project was to develop the natural resources of the Lake area in terms of fisheries, agriculture and socio-economic development. It also studied public health and vector of human diseases (Abiodun, 2002).

Jebba Lake is a watershed of Kainji Lake and is about 100 km from Kainji Dam to Jebba Dam. It is rich in fish biodiversity (Adelakun *et al*., 2017). Jebba Lake is located in North-Central Nigeria and is characterized mainly by two seasons- dry and rainy (Adelakun *et al*.,

2017). *Oreochromis niloticus* and *Clarias gariepinus* constitute some of the commercially important fish species in Jebba Lake (Oladipo *et al*., 2018) and Kainji Lake. Abiodun and Odunze (2011) observed 51 species of fish under 12 families from Jebba Lake. It is expected that Jebba Lake will habour many fish species but because only few research have been conducted on the Lake, about half the number of fish species in Kainji Lake have been reported for Jebba Lake (Abiodun and Odunze, 2011).

1.2 Statement of the Research Problem

According to World Health Organization (WHO), about 80% of ill-health in developing countries like Nigeria is water related (Cheesbrough, 2000). Cattle and other livestock are reared in the surroundings of Kainji and Jebba Lakes. These livestock drink and defecate into the Lake. Other anthropogenic factors like washing, bathing and electricity generation are regularly done in the Lakes. Illegal mining activities are also carried out around the Lakes. Undoubtedly, these activities may constitute the major sources of pollution to the Lakes (such as microbial pathogens and heavy metals). The direct or indirect consumption of these microbial pathogens and heavy metals by man through water intake and consumption of contaminated fish bring about varying diseases in man and animals. Various cases of water-borne diseases, involving the coliform organisms such as *Escherichia coli, Vibrio cholerae, Salmonella* species and *Shigella*, have been widely reported (Gugnani, 1999; Licence *et al.,* 2001). Pollution due to heavy metals in aquatic environments is a global concern because they are not utilizable by the organisms and most of them are toxic to organisms (ÖztÜrk *et al*., 2009). Heavy metals amongst other environmental pollutants, are of serious public and other animal health concern. This is due

to their potential toxicity and ability to bioaccumulate in aquatic ecosystems (Censi *et al*., 2006).

Heavy metals have great affinity for sulphur, and within enzymes, immobilize them. Heavy metals could also bind to cell membranes, thereby interfering with transportation processes in the body. Other vulnerable sites of attack include the protein, carboxylic acid (- COOH) and amino (- NH_3) groups (Adekola *et al*., 2010).

Severe health hazards are linked to the consumption of fish scarcely or greatly polluted by lead (Pb). Lead poisoning causes dagames to the central nervous system especially among children causing mental impairment. This condition affects oxygen transport in the body and leads to other digestive problems. Exposure of humans to lead for a long term can cause coma or death (Etim *et al*., 2013).

Cadmium (Cd), known to be a heavy metal, can cause cancer by interfering with the body metabolic processes. Cadmium being one of most harmful heavy metals is capable of causing renal, hepatic and testicular injury. Some adverse effects of acute cadmium toxicity include kidney damage, testicular tissue damage, high blood pressure and red blood cells destruction (Etim *et al*., 2013).

Weak immune system, likely disease of lung cancer, ulcer and liver damages may result from exposure and consumption of chromium (Cd)- containing substances. Long - term exposure to chromium can cause kidney, liver and nerve tissue damage. Many chromium compounds are also known to cause cancer, thus, making them carcinogenic (Etim *et al*., 2013).

1.3 Justification for the Study

Baseline data on the microbiological and heavy metal content of Jebba and Kainji Lakes are scanty. This information is essential for management of these inland water fisheries and utilization of the inland water bodies. Water quality in many Lakes and rivers are being impaired by high levels of pollutants. These constitutes health hazard to aquatic lives and also to humans that use the raw water for drinking, bathing, irrigation purposes and also consumption of fish from such polluted water bodies. About sixty seven million (6.7×10^7) Nigerians still rely exclusively on surface water source to meet their domestic needs, yet pollutant discharge into surface water by individuals and industries go on unmitigated, unregulated and unpunished (Longe *et al*., 2010). Large populations of human beings in the world today use fish as staple source of proteins and also as source of income. In recent time, fish is preferred more by people as a healthy alternative to red meat (Adebayo-Tayo *et al*., 2012). Fish is easily digestible. It contains less fats and high proportion of poly or mono unsaturated fatty acids (Omega – 3 fatty acid) and is thought to be associated with reducing the risk of human cardiovascular disease. Omega 3 fatty acids have been found to have cholesterol lowering properties as well as decreasing blood clotting activity (Browne, 1990).

Fish performs all their body functions in an aquatic environment; hence the quality of water is very important for their survival, productivity and quality. Fishes are major faunal components of aquatic environments and are usually used as excellent environmental bio-indicator of the health of aquatic system (Widianarko *et al*., 2000). These highlight the need to determine the bacteriological and heavy metal status of the water, sediment and fishes

from Kainji and Jebba Lakes, which are surrounded by various anthropogenic activities (including illegal mining around the Lakes), yet serving as sources of drinking water and fish supply for many communities around the Lakes. The quality of groundwater has been greatly affected by the rapid growth of urban areas due to overexploitation of resources and improper waste disposal practices (Ojutiku *et al*., 2013). Hence, the need for concern over the protection and management of surface water and groundwater quality is highly significant (Ojutiku *et al*., 2013). The results of this study would help to create public health awareness on the hazards associated with the use of polluted water bodies and also in the effective monitoring and management of the Lake water quality and health status of the aquatic organisms particularly the fish, through evidence based policy decisions.

1.4 Aim and Objectives of the Study

This study was aimed at evaluating the bacteriological and heavy metal status of water, sediments and fish samples from Kainji and Jebba Lakes, Niger State, Nigeria.

The objectives were to determine:

i. Bacteriological quality of water, sediments and fishes from Kainji and Jebba Lakes.

ii. Heavy metals (Pb, Cu, Cd, Cr, As) status of water, sediments and fishes from Kainji and Jebba Lakes.

iii. Physico-chemical qualities of water from Kainji and Jebba Lakes.

iv. Heavy metal biosorption potential of the bacterial isolates from the Lakes.

CHAPTER TWO

2.0 LITERATURE REVIEW

2.1 Water

Water is a chemical compound consisting of two hydrogen atoms and one oxygen atom. The name 'water' typically refers to the 'liquid state of the compound'. The solid phase is known as ice and the gaseous state is called steam. Water is a basic need for life, it freezes at 0^{o}C (Nduka, 2011).

2.1.1 Uses of water

Water is indispensable for man's activities. It has diverse uses, which vary from one culture to another. Some of its uses to man includes the following:

2.1.1.1 Composition of biological objects and domestic use

Water is a major component of the body of many living things. For instance, in human water accounts for about 92% of blood plasma, about 80% of muscle tissue, and about 60% of red blood cells. Most animals including humans are about 50 – 60% water by weight (Ukpong and Peter, 2012). This is as a result of the significant role water plays in most biochemical reactions taking place in human cells. Water is needed by all living things for their metabolic processes. A person may strive without food for a month, but can barely survive a week without water (Nduka, 2011). It is important to have water treated as to get rid of microorganisms and chemicals in order to get clean water for human consumption. Water is usually treated in some way to remove harmful microorganisms and chemicals; hence, the need to keep water sources clean is not only important but a necessity.

In our homes, water is used for drinking, cooking, washing and bathing, among many other uses.

2.1.1.2 Irrigation

Throughout the world, irrigation of crops and vegetables is an old tradition and use of water. Irrigation accounts for almost 60% of all the world's freshwater withdrawals. Irrigation is essential in large-scale farming to provide food for the world's large populations. Irrigation water can be from Lakes, rivers, reservoirs and wells. It would be impossible to cultivate crops in desert countries such as California and Israel without irrigation (Nduka, 2011).

2.1.1.3 Aquaculture

Another important agricultural use of water is for growing of fish in aquaculture. Wide variety of animal life inhabits surface water bodies. Water is the only habitat for fish where breathing, feeding, growth, reproduction and excretion take place. The quality of water determines the success or failure of aquaculture operations. Without good water quality, fish production will be a mirage (Nduka, 2011).

2.1.1.4 Power generation

Water is a source of energy for use in hydro-electric power generation. It is used as steam for driving turbines for the generation of electricity (Nduka, 2011).

2.1.1.5 Transportation

Water is one of the primary media used for the transport of heavy goods. Large quantities of raw materials and manufactured products are transported by huge ships plying the ocean from one region to another. Water transportation is indubitably one of the cheapest means of transportation of heavy goods (Nduka, 2011).

2.1.1.6 Recreation

Water is used sport and many other recreational activities, some of which include swimming, boating, fishing, and diving. Moreso, sport like ice hockey takes place on ice.

2.1.1.7 Human Affairs

Water has played a significant role in human affairs for centuries, and still does (Keeley, 2005).

Water is used for purification in most religions. It is considered as cleansing compound, for purifying both the body and soul, as well as representing the symbol of purity. As a result, water is used in a variety of religious and cultural ceremonies and traditions for ritual washing or immersion, as well as for cleansing the dead or certain parts of the body (Eran, 2012).

On the other hand, many religions revere particular bodies of water as being sacred or at least auspicious, for example, the River Ganges is held sacred in Hinduism. Water and rainfall are considered blessings among indigenous Igbos in Nigeria, rainfall during ceremony is believed to be good omen.

Some specific water bodies are also considered as deities or holy symbol, for example the Jordan River, the Ganges and various springs (Linton, 2010).

2.2 Pollutants

Pollutants are substances that when introduced into the environment cause harm to living things (plants and animals) in that environment. They interfere with man's use of his environment and interfere with the biota. Global Environmental Monitoring Systems (GEMS), states that agricultural waste, sewage, nutrients and toxic metals from industries are the main water pollutants, with organic matter in domestic sewage being the most

important. In developing countries, an estimated 25,000 deaths each day are induced either by direct consumption of polluted water or indirectly by contraction of diseases like malaria and bilharzia transmitted by vectors that live in polluted water (United Nation Environmental Protection, 1991).

The cases of water pollution has aggravated in most areas of Africa, Asia and Latin America since the 1990s (UNEP, 2016a). Water quality deterioration is expected to increase over the next decades which will pose serious threat to human health, the environment and sustainable development (Veolia/IFPRI, 2015). The most prevalent challenge of water quality across the world is nutrient loading, which is mostly connected with pathogen loading depending on the region (UNEP, 2016a).

2.2.1 Classification of water pollutants and their parameters

(A) Physical: These include the temperature, turbidity, colour, suspended solids and taste.

(B) Chemical: These include the pH, dissolved oxygen, ammonia, nitrate, nitrite, phosphorus, chlorides, sulphate, sulphide, detergents, organic solvents, pesticides, oil and grease and heavy metals, among others.

(C) Biological: These include the bacteria, viruses, nematodes, trematodes, worms and fungi.

Pollutants affect aquatic organisms and some of the effects produced include increase in osmotic pressure from chemical pollutants, decrease in oxygen content of water from organic pollutants, destruction of biota due to chemical and biological pollutants, toxic elements such as heavy metals, which may injure the gills of fish and other external structures, and these may cause death (Agarwal, 2005). Others include blocking of gills of fish from silt or other suspended materials. Pathogens may be introduced into the aquatic

environment by certain biological pollutants, which may result in death of aquatic food organisms such as fish and snails. Human beings can become infected with these pathogens when the food organisms are consumed (Agarwal, 2005). The guts of humans and animals are the main reservoirs of human water borne pathogens (Maynard *et al.,* 2005). Table 2.1 shows different types of pollutants, their sources and effects on aquatic lives and man.

2.3 Bacterial Pathogens Associated with Fish

Fishes like other animals, are exposed to various microorganisms of which some may be pathogenic, causing varying diseases. It is a well known fact that pathogenic bacteria (especially the coliforms) are commonly harbored by fishes from both fresh and brackish waters (Emikpe *et al.,* 2011). These pathogenic bacteria are dangerous pathogens for both cultured and wild fish and can cause mass losses in fish production (Taghreed, 2020). Numerous groups of bacteria (particularly the coliform group) enter into the fish gut through water, sediment and food (Dutta *et al.,* 2010). Detection of faecal coliforms in fish is an indication of the level of pollution in the aquatic environment since faecal coliforms are not normal flora of fish (Emikpe *et al*., 2011). Fish acts as an important food vehicle for the transmission of some zoonotic pathogens such as species of *Salmonella* and *Vibrio*. Transmission of infection through fish contaminated with pathogens is a major public health concern.

Unfavorable environmental conditions such as poor water quality can cause various bacterial diseases in all fish. The susceptibility of fish to bacterial infections can be induced when there is inadequate oxygen level in the pond, which stresses the fish. Emikpe *et al*. (2011) notes that the common pathogens that contaminate fish samples from different

sources were total aerobic bacteria as well as enterobacteria. Bacterial count of less than 10^5 cfu per gram is estimated for fish with good quality as recommended by Food and Agricultural Organization and cited by Emikpe *et al*. (2011).

Table 2.1 Main Pollutants of Water Bodies, their Sources and Effects

	Pollutant	Source	Effects
1	Metals (Cd, Cr, Cu, Pb, As, Ni, Zn, Fe, Mn, Hg, Al)	Mining, petroleum; surface runoff, tanneries, textile metal/steel,	Destroys aquatic life and results in diverse diseases if man consumes aquatic biota that is contaminated.
2	Nutrients (Nitrogen and Phosphorus)	Sewage, abattoir, fertilizer, food processing industries, animal manure etc	Eutrophication of water bodies, algal blooms and oxygen depletion.
3	Organic matter	Sewage, effluents from tanneries paper mills textile, food processing breweries.	Introduction of pathogens into water bodies and oxygen depletion which is harmful to aquatic life.
4	Suspended matter (fiber, faeces, brewery residue, paper, saw dust)	Food processing, edible oil, paper/pulp, textile industries, tanneries, sewage, breweries.	Increases turbidity, limits photosynthesis and acts as carrier for pathogens and other pollutants.
5	Persistent Organic Pollutants (Pesticides, oil, grease, solvents)	Petroleum, textile, paper/pulp, steel and metal industries, Farm lands.	Causes irreversible damage to aquatic life.
6	Thermal Pollution	Cooling water from electricity generation and industrial effluents at high temperature	Reduces DO level, enhances microbial activity, denaturation of enzymes, kills organisms
7	Sediment	Natural erosion, agricultural runoff etc	Loss of soil resources and reduction of water quality due to increased sediment load, blocking of fish gills.

Source: Atiribom and Nnaji (2013)

Bacteria are common and widely distributed in the aquatic environments in various parts of the world with the water temperature having selective effect on their growth on fish. The psychrotrophic grows at low temperatures below 20^{o}C while mesophillic grows at a temperature between 20^{o}C to 45^{o}C. Non-indigenous bacteria consist of *Salmonella* species, *Shigella* species, *E. coli* and *Clostridium perfrengens*, which may be introduced from sewage and animal sources. These bacteria may find their way into the body of the fish through ingestion, gills, skin and wounds. Loss of appetite, fin, tail rot and pale gills are the various symptoms of bacterial infections in fish. Behavioural signs of diseases include weak or erratic swimming; floating on water with the belly up and crowding at the inlet while gaping mouth, cloudy or distended eyes, open sores, lesions, loss of scales, swollen, bloody or brownish gills, amongst others are the various physical signs (Valerie *et al*., 1994).

According to the Manual of Diagnostic Tests for Aquatic Animals (OIE, 2003), bacteriological analysis of fish is generally carried out at temperature range of 20 to 26°C. However, some bacteria are found to grow at 15°C, while bacteria isolated from warm water fish may be incubated at 30°C or 37°C to accelerate the diagnostic steps.

2.3.1 *Aeromonas hydrophila*

Aeromonas species are Gram negative, straight rod-shaped with rounded ends to coccoid cells measuring about 1.0-3.5µm long and 0.3 to 1.0 µm wide. They are commonly found in aquatic ecosystems worldwide with densities ranging from 1 to about 1,000 cells per ml of underground water, drinking water, rivers and lakes (Jatau and Yakubu, 2004). These bacteria grow in artificial medium over a wide range of incubation temperature (0-45^{o}C)

with human strains growing between 10-42^{o}C with 30^{o}C as optimum. *Aeromonas* reduces nitrates to nitrites and ferments D-glucose, maltose, sucrose and sorbitol to acid and gas. *Aeromonas* species are associated with a wide range of diseases in both warm and cold-blooded vertebrates including humans, fish, reptiles and birds. *Aeromonas* causes both intestinal and extra-intestinal disease. They have been known to cause septicaemia and infection of the eye, bones, joints and gastrointestinal tract. Cases of diarrhoea were reported after consumption of a ready to eat shrimp, with *Aeromonas hydrophila* being isolated from the incriminated food and from the stool of the patients (Jatau and Yakubu, 2004).

Freshwater fishes such as *Tilapia*, typically carry *Aeromonas hydrophila* counts of 10^{2}-10^{3} bacteria per square centimeters of skin surface (Jatau and Yakubu, 2004).

Studies (Jatau and Yakubu, 2004), have indicated that *Aeromonas* species can act as both infectious and enterotoxigenic pathogens. The motile mesophilic aeromonads, consisting of *A. hydrophila, A. sobria* and *A. caviae* are considered causative agents of human gastroenteritis, wound infections and septicaemia (Jomie and Maryanne, 2002). Ventura and Grizzle (1987) in their studies have shown that *Aeromonas hydrophila* infected internal organs of catfishes through the digestive tract or through uninjured skin under overcrowding (13.1g of Fish/L) and high temperature (34^{o}C) conditions. It is seen to be the cause of bacteraemia, tissue necrosis and clinical disease known as bacterial haemorrhagic septicaemia in fish. It also causes fin rot and scale oedema. In severe cases, the entire viscera may be bright red in colour and fibrinous adhesions may be present. The spleen will be visibly enlarged, round and cherry red. The enlarge kidney often undergo liquifactive

necrosis and when it is incised, thick necrotic fluids oozes out. *Aeromonas hydrophila* can be isolated from lesions and occasionally from internal organs (Jatau and Yakubu, 2004).

2.3.2 *Escherichia coli*

Escherichia coli (*E. coli)* are Gram negative thermo-tolerant coliforms that belong to the total coliform group. They are capable of fermenting lactose at 44.5 ± 0.2ºC. *Escherichia coli*, is the predominant faecal coliforms found in the intestinal tract, gills, muscles and skin of fish when sewage water is been used to rear fish (Ampofo and Clerk, 2010). *Escherichia coli* is likely to invade fish muscle if there is breakage of immunological barrier of fish by pathogens when fish are raised in an environment of *E coli* and *Salmonella* of greater than 10^3 cfu/ml (Guzman *et al*., 2004).

It is distinguished from other coliform bacteria by their ability to produce indole from tryptophan (Mwajuma, 2010). It is believed to be of faecal origin and has been found to be present in fresh faeces in concentrations as high as 10^9 colony forming units per gram (Onyuka *et al*., 2011). *E. coli* can be isolated from sewage, treated effluents, all natural waters and soils, which is subjected to recent faecal contamination, whether from humans, agriculture or wild animals and birds. It has been proposed that *E. coli* may be found or may even proliferate in tropical waters that are not subject to human faecal contamination. *Escherichia coli* and other members of the coliform group may be found where there has been contamination with faeces of warm-blooded animals (Chao *et al*., 2003).

2.3.3 *Edwardsiella tarda*

This is a Gram negative, straight rod (about 1 – 3µm), non spore forming and motile bacterium with peritrichous flagella. They have been isolated frequently from fresh water

fish and other cold blooded animals. It is said to be the causative agent of septicaemia of warm water fish particularly cat fish. Fish infected with *Edwardsiella tarda* when consumed by man is said to be the cause of gastroenteritis and other diseases of man. *Edwardsiella tarda* can be separated from *Edwardsiella ictaluri* by the former's salt tolerance, indole production, H_2S production on TSI and higher temperature tolerance. *Edwardsiella tarda* grows faster than *Edwardsiella ictaluri.* This bacterium is found in the muscles and intestinal organs of diseased fish, and so can be isolated using general purpose media like brain heart infusion agar (BHIA), tryptic soy agar (TSA) or nutrient agar (NA). *Edwardsiella tarda* is mainly isolated from the intestinal contents of carrier animals serving as its reservoir. The presence of *Edwardsiella tarda* in an aquatic environment is an indication of the presence of diseases. Although environmental stressors are not essential precussors to an *Edwardsiella tarda* infection, predisposing factors such as high temperature, poor water quality and crowding may contribute to the onset and severity of the disease (Valerie *et al*., 1994).

2.3.4 *Yersinia ruckeri*

This is of the genus *Yersinia*, within the family Enterobacteriaceae. It is a Gram negative, small straight rod to coccobacilli (0.5 – 0.8 x 1.3 µm). They are motile with peritrichous flagella. *Yersinia* is found in live and inanimate habitats. *Yersinia ruckeri* is an important primary pathogens of fish. It is the causative agent of enteric red mouth (ERM) disease of fish. The name "enteric red mouth" disease was used to distinguish it from pseudomonad and aeromonad infections with similar pathological signs. Enteric red mouth is much like other bacterial septicaemias (Valerie *et al*., 1994). Reddening at the base of fins and along

the lateral line as well as in the head region characterize haemorrhage on the body surface. Internally, there may be petechial haemorrhages on the liver, visceral fat and pyloric caeca.

Chronic infected fish may appear dark and lethargic with intermittent reversions to an apparently asymptomatic carrier state. Other pathological signs include the exophthalmos, reddening around the mouth and local or general darkning of the body. The spread of *Y. ruckeri* occurs from fish to fish through contaminated water. Environmental factors such as crowded conditions that may result in excess ammonia and metabolic waste products, high temperature and exposure to agents such as copper can act as predisposing factors (Valerie *et al*., 1994).

2.3.5 *Pseudomonas fluorescens*

These are Gram negative, straight or curved rods, with body measurement range of 0.5 – 1.0 x 1.5 – 5.0 µm. They are usually motile with one or more polar flagella and are strictly aerobic. Frequently they are isolated from the surface and intestines of fish and may cause disease. *Pseudomonas fluorescens* causes Pseudomonad septicaemia (haemorrhagic condition of fish). It can be isolated from the lesions of affected fish. It grows well on nutrient agar at 22 – 25^{o}C and produces diffusible yellow – green pigment. Almost all species of fish are vulnerable to pseudomonad septicaemia under adverse environmental conditions or when compromised by other factors. Though most pseudomonas species in aquatic habitats are found in fresh water, marine species do exist (Valerie *et al*., 1994).

2.3.6 *Vibrio* species

Vibrio species belong to the family Vibrionaceae. They are curved or straight Gram-negative bacilli, oxidase positive bacteria. They are non-spore forming and motile, having one (monotrichous) or more (amphitrichous or multitrichous) sheathed polar flagella. All species are facultative anaerobes, but not all species are pathogenic. Some particular strains within a species may be highly pathogenic; others may be innocuous or act only as secondary invaders. *Vibrio anguillarium*, *Vibrio alginolyticus* and *Vibrio salmonicida* are fish pathogens. All are associated with acute bacterial septicaemias or chronic focal lesions in fish. *Vibrio anguillarium* causes acute haemorrhagic condition in fish called 'red pest'. Chronically infected fishes have large granulating lesions deep in the muscles with gills very pale, reflecting severe anaemia (Valerie *et al*., 1994).

Bacterial pathogen (*Vibirio cholera*) can produce a serious acute intestinal disease called cholera, which is characterized by sudden diarrhoea with profused watery stools, vomiting, rapid dehydration, fall of blood pressure (hypotension), subnormal temperature (hypothermia) and complete collapse. Death may occur within a few hours on onset of the disease unless prompt medical treatment is given to the patient (APHA 1992). The spread of cholera may be through person to person contact, ingestion of food contaminated by processing water or food handling and drinking of polluted water. The viability of *Vibrio cholerae* in surface waters varies from 1 hour to 13 days. Autoclaving of this water increases their survival possibly because heat produces a breakdown of the suspended organic matter, thereby killing some bacterial competitors and increasing the water

alkalinity to a more favourable pH range between 8.2 - 8.7. *Vibrio cholerae* viability is brief in water of pH 5.6 (Ivanova, *et al*., 2001).

Although *Vibrio cholerae* persists for a short time in a polluted aquatic environment, faecal contamination from victims and carriers may continue to re-enforce their population in water.

Chlorination (2.0 to 3.0 mg/liter for 10 min. contact time) of turbid, heavily polluted waters without any prior treatment has not produced a potable water supply free of cholera *Vibrios* or *Salmonella* (Ivanova, *et al*., 2001). It is assumed that the pathogenic bacteria in raw water may be protected in particle clumps from exposure to chlorine during disinfection period.

2.4 Fish

2.4.1 Fish as source of protein

Fish serves as important source of protein and other elements required for the development and maintenance of a healthy body (Emikpe *et al*., 2011). Fish is a staple animal protein and it is a very cheap source of protein. Fish and fish products form an important part in the international trade; more than 50 billion fish are eaten annually indicating increasing consumer interests in the commodity (Wafaa *et al*., 2011).

In Africa, as much as 5 % of the population depends partly or wholly on fish for their livelihood. Egbere *et al*. (2008) stated that fishes are responsible for about 55 % of protein intake of Nigerians. Most of these are obtained through naturally occurring fish from the wild and partly through fish importation. Generally, fish serves as good sources of fluorine and iodine, which are needed for development of strong teeth and the prevention of goiter in man. Important vitamins found in fish include vitamin B12 and V6 is also a good source

of fluorine and iodine (Adebayo-Tayo *et al*., 2012). Fish products serve as important nutritional factors and also as an item of international trade and foreign exchange earnings for a number of countries across the world (Adebayo-Tayo *et al*., 2012).

2.4.2 Biology of fish samples - *Clarias gariepinus* and *Oreochromis niloticus*

African catfish (*Clarias gariepinus*) is a major commercial fish species in the country because of its high resistance to adverse environmental conditions. It inhabits the bottom/benthic level of the aquatic environment. It is a scaleless, bony, elongated body fish with long dorsal and anal fins (Plate I). At the dorsal part, the colour varies from dark to light brown while the underside is pale cream to white (Skelton, 2001). It can grow very large to a maximum weight of 60 kg (Robbins *et. al*., 1991). It is an omnivorous species displaying both scavenging and predatory behavior thus feeds on fish seeds, insects, small mammals and plankton. *Clarias gariepinus* is widely tolerant of many different habitats and conditions, but it is considered to be a freshwater species. It is highly resistant to handling and stress. The head is flattened and the pelvic fin normally has six soft trays (Gertjan and Johannes, 1996).

Nile tilapia (*Oreochromis niloticus*) is a deep bodied fish with cycloid scales. Silver in colour with olive/grey/black body bars (Plate II). It often flushes red during the breeding season (Picker and Griffiths, 2011). It is a tropical species that prefers to live in shallow water. It inhabits the surface and sub–surface layer of an aquatic environment. The lower and upper lethal temperatures are 11^{o}C and 42^{o}C respectively. Its preferred temperature ranges from 31^{o}C to 36^{o}C. It is an omnivorous grazer that feeds on phytoplankton, periphyton aquatic plants, small invertebrates, benthic fauna, detritus and bacterial films

(Rakocy, 2005). Sexual maturity is reached at 5 – 6 months. The body is compressed, caudal peduncle depth equal to length. First gill arch has 27 to 33 gillrakers. Dorsal fin has 16 to 17 spines and 11 to 15 soft rays. Anal fin has 3 spines and 10 to 11 rays. Caudal fin is truncated with numerous black bars (Rakocy, 2005).

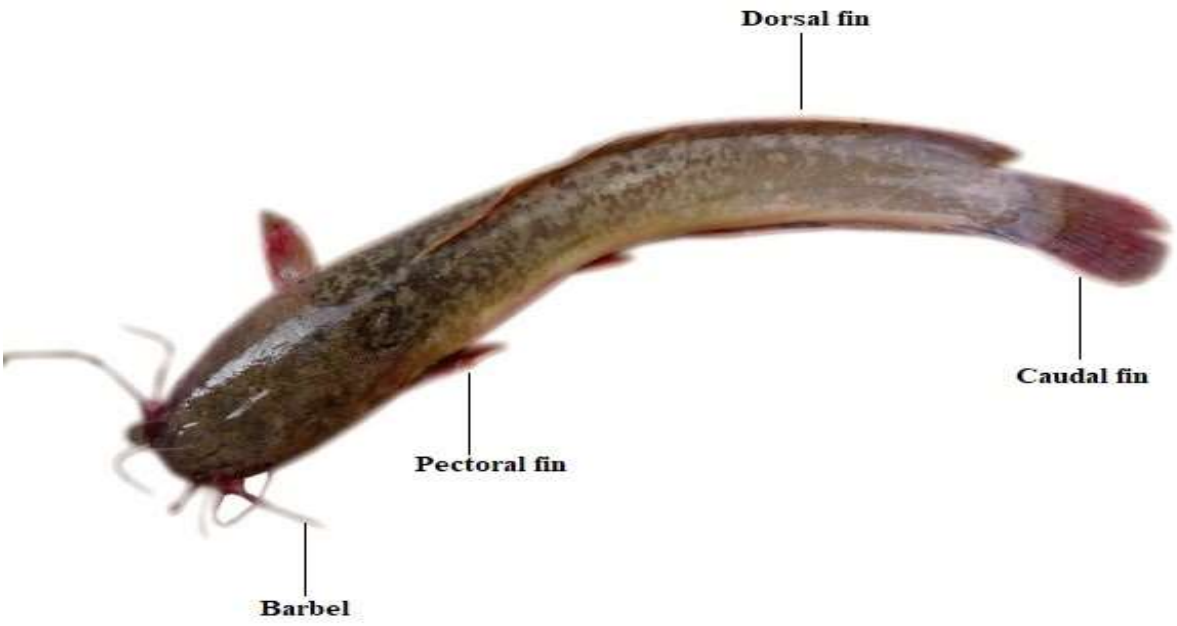

Plate I: ***Clarias gariepinus***

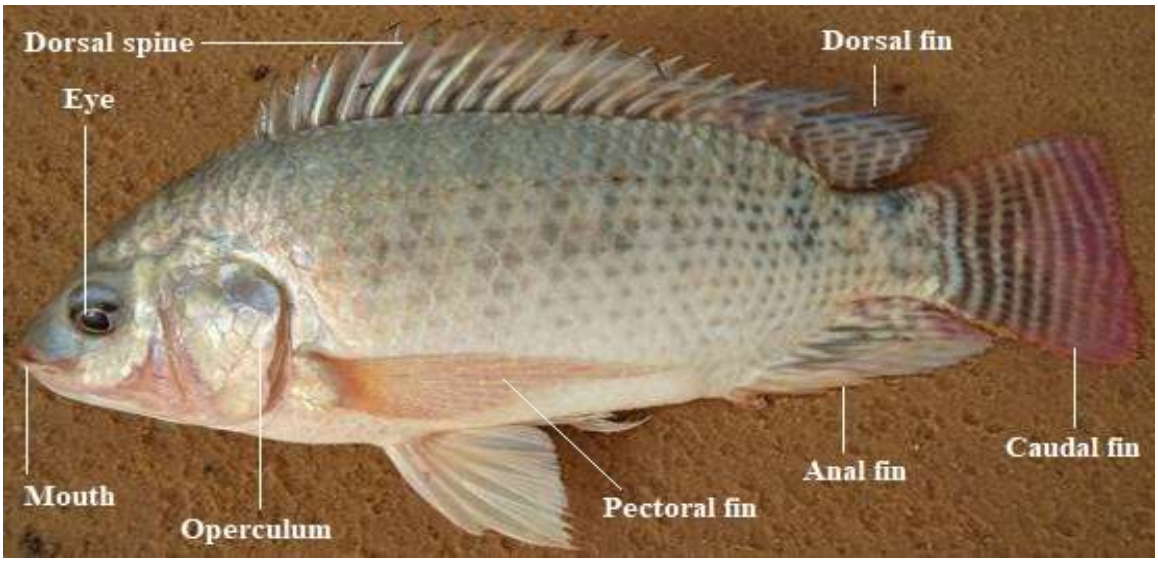

Plate II: ***Oreochromis niloticus***

2.5 Sediment

These are the loose sand, clay, silt, and other soil particles or the decomposition of plants and animals remains that are washed from a site by runoff water that eventually settles at the bottom of lakes, streams, rivers and ponds. As water flows from land and sites, the runoff collects and transports soil as sediment, pet waste, salt, pesticides, fertilizer, oil and grease, litter and other potentially toxic pollutants to surface water bodies. Sediment is the most common pollutant in our waterways. While natural soil erosion produces about 30 percent of waterway sedimentation, accelerated erosion from human modifications of the land accounts for the remaining 70 percent.

Sediment can cause severe degradation of water quality for drinking, fish and wildlife habitat, recreation in the form of swimming, fishing, and boating. Excess sediment can also cause flooding, severe stream bank erosion, and undesirable physical and chemical changes to our Lakes and Ponds. It increases the cost of treating our drinking water. Sediment disrupts the natural food chain by destroying the habitat of the smallest stream organisms

and causing massive declines in fish populations. Sediment clog fish gills and reduces resistance to disease, lowers growth rates, and affects fish egg and larvae development. Sediment pollution causes an estimated $16 billion in environmental damage each year in the US (United States Environmental Protection Agency, 2004).

2.5.1 Classification of sediment

The Wentworth scale classifies sediment by grain size. Clay sediments are the smallest with a grain diameter of less than 0.004 mm and boulders are the largest with grain diameters of 256 mm or larger. Sediments are also classified by origin. According to Mazzulo *et al.* (1988), sediments can be classified into four types: lithogenous, hydrogenous, biogenous and cosmogenous.

Lithogenous sediment that comes from land via rivers, ice, wind and other processes.

Biogenous sediment that comes from organisms like planktons (when their exoskeletons break down).

Hydrogenous sediment that comes from chemical reactions in the water.

Cosmogenous sediment that comes from space, filtering in through the atmosphere or carried to earth on meteorites.

Douglas *et al*. (2003) classified sediments into:

(i) Clastic sediments: Clastic sediments are composed of discrete grains of generally allocthonous (quartz grains, volcanic ash) origin deposited by physical processes of sedimentation (air fall, sub-aqueous currents). These are classified by designating a principal name describing the major textural component of the sediment (example, sand, silt and clay).

(ii) Chemical sediment: Chemical sediments are composed of inorganic materials formed within the Lake (autochthonous) by inorganic or biologically mediated chemical processes such as precipitation from solution, or recrystallization of detrital evaporites and siliceous, calcareous, or carbonaceous biogenic debris (example, calcite, halite, pyrite and gypsum). The category of sediment refers to sediments that are composed primarily of authigenic and diagenetic minerals formed by inorganic precipitation within the water column or post-depositionally in the sediment column (Douglas *et al*, 2003).

(iii) Biogenic sediments: Biogenic sediments comprise a variety of mineralogies (e.g., CHO, CaCO, SiO) but all are essentially the fossil remains of former living organisms (ostracodes, diatom frustules, mollusks or amorphous algal organic matter). Examples of biogenic sediment include the peat, coal, diatomites, and shelly hashes or coquinas. These are derived from the accumulation of organic matter in lacustrine basins. Lacustrine organic matter may be derive from organisms inhabiting the Lake (cyanobacterial mats, macro- and microphytes, phytoplankton, zooplankton, benthic organisms or feces from aquatic or terrestrial organisms; or from organic matter introduced into the lake from the surrounding drainage basin (terrestrial herbaceous and woody plants) (Douglas *et al*, 2003).

2.5.2 Contaminants of sediment

The accumulation of toxic and hazardous materials in water bodies, especially at the bottom of water bodies has been reported to cause adverse effects on the environment and human health. (USEPA, 1998). The contaminants may be adsorbed to sediments, organic materials, soil or other materials and deposited from air, washed by water, erode from river beds, or from the breakdown of underwater and the buildup of minerals (USEPA, 1998).

Contaminated sediment may be found in lakes, rivers, streams, reservoirs, or wetlands. The contaminants may enter the water bodies *via* anthropogenic or natural sources. Both organic compounds, including sulphides and heavy metals have contaminated sediments. These contaminants accumulate for decades, non-degraded. The contaminants may also accumulate from lower trophic level with the organisms at the higher level more affected. Some xenobiotics are considered to be biopersistent, examples include methyl mercury, pesticides, heavy metals and polychlorinated biphenyls (PCBs).

2.5.3 Sources of contaminants in sediment

The sources of contaminants in sediments may occur from (i) soil erosion, especially from soil having the contaminants such as sewage from homes and industries, (ii) air emissions arising from pesticide, incinerators, power plants. This could find its way to water bodies via direct deposition or precipitation, (iii) spillage of chemicals into water bodies and (iv) seepages or upwelling into water body of contaminated ground water or non-aqueous phase liquids (NAPL) (Douglas *et al*, 2003).

2.5.4 Treatment process of contaminated sediment

Treatment process in sediment sites is more complex than sites with soil or ground water contamination alone. This cleanup process may be *Ex-situ* or *In-situ*.

The *Ex-situ* approach include: dredging, treatment of dredged sediment and backfill of dredged area if needed or its disposal (USEPA, 2005).

In-situ approach according to USEPA (2005) involves:

(a) *In-situ* caping: Single-layer granular caps, multi-layer granular caps, combination granular or geotextile caps.

(b) Hybrid approach: This involve the placement of thin layer of sand or other material to enhance recovery via natural deposition (USEPA, 2005).

(c) Monitored natural recovery: This involves physical isolation, chemical sequestration and biological transformation/sequestration.

(d) *In-situ* Treatment: This involves reactive caps, additives/enhanced biodegradation.

In-situ sediment treatment involves applying or mixing of an amendment into sediments. Mixing may be achieved either passively, through natural biological processes such as bioturbation, or actively through mechanical means using augers. If amendments are added to sediment, it inhibits biological processes that would normally cause contaminants to be transformed into more toxic forms under existing conditions. For example, applying nitrate can inhibit the release of methyl mercury (USEPA, 2005).

Bioaugmentation is the introduction of new cultured microorganisms or addition of existing microorganisms directly into the sediment to degrade and transform specific contaminants. For example, although not common for *in-situ* sediment treatment, KB-1 is a commercially available culture microorganism for treatment of certain chlorinated solvents (USEPA, 2005).

Biostimulation is the enhancement of rate-limiting sediment conditions in order to stimulate the indigenous microorganisms to enhance the rate at which the contaminants are degraded and transformed (USEPA, 2005).

2.6 Heavy Metals and Their Sources

Heavy metal is any metal or metalloids with atomic density which is greater than 5 g/cm^3. Heavy metals occur naturally in rocks and ore minerals, hence, there is a range of normal concentrations of these elements in soils, sediments, waters and living organisms. Mining areas are the chief areas where heavy metal pollution readily occurs, in which the surrounding land receives it first (Salaudeen *et al*., 2016), and later the surface water bodies through runoff or direct washing. It is the concentration of the metals relative to the normal background concentration which determine whether or not an item is polluted (Bio-ellite, 2000). Some of these metals (Cu, Mn, Co, Fe and Zn) are beneficial to most living organisms as they are required in small concentrations for enzymatic activity and healthy growth. Excess concentrations of these essential metals can, however, cause toxicity (Alloway and Ayres, 1993). Some heavy metals have no known essential biochemical functions and are toxic at low concentrations (examples are As, Cd, Hg, Pb). Excess concentration of these metals cause some toxic effects including competition for sites with essential metabolites, replacement of essential ions, reactions with – SH groups, damage to cell membranes and reactions with the phosphate groups of adenosine diphosphate (ADP) and adenosine triphosphate (ATP) (Biney *et al.*, 1994; Bio-ellite, 2000). Arsenic has its major source from pesticides, fungicides and metal smelting. It causes bronchitis and dermatitis in man (Alluri *et al*., 2007). The presence but low level arsenic concentration in commercially-available inorganic fertilizers in Nigeria has also been reported (Benson *et al.*, 2014). Apart from natural sources, mining related activities are major sources of arsenic contamination of surface waters (Atobatele and Olutona, 2015).

2.6.1 Pedogenic sources

This is a primary or natural source of heavy metals, which involves weathering of mineral, erosion and volcanic eruption. Heavy metals form part of the elements, which are referred to as `trace elements' in geology and they form less than 1% of the rocks in earth crust. Trace elements exist as impurities in primary minerals found in igneous rocks, which are formed from molten magma, (Biney *et al.*, 1994). In sedimentary rocks, trace elements occur as sorbed to the secondary minerals.

2.6.2 Anthropogenic sources

Anthropogenic sources include the following:

a. Mining

Ores from the source of metals utilized in manufacturing and from gold mining zones. Tailings, which are finely divided or milled fragments of rock left behind after extraction of metals, or continued weathering of ore minerals in historical and abandoned mining sites are important sources of heavy metals.

b. Agricultural materials

These are non-point sources and they include fertilizers, pesticides and herbicides that are applied in farm lands.

c. Metallurgical industries

Heavy metals are used in the manufacturing of steel and alloys and their disposal can lead to environmental pollution.

d. Fossil fuel

A number of heavy metals in fossil fuel are either emitted into the atmosphere during combustion or remain as ash, which may be leached in situ or transported. For example, most gasoline contains Pb additives and their combustion give rise to large amount of Pb particles (Alloway and Ayres, 1993).

2.7 Heavy Metals in Aquatic Environment

In the aquatic environment, heavy metals can be found in its different compartments: water, suspended solids, sediments and biota. The distribution is governed by dilution, advection, dispersion, sedimentation and adsorption/desorption (Biney *et al*., 1994).

2.7.1 Heavy metals in Lake waters

Surface water pollution by heavy metals in recent times has become a problem of increasing public concern due to their gradual increase in excess of natural background resulting to deterioration in water quality for consumption as well as threatens aquatic life including fish. This situation arises as a result of anthropogenic source including mining activities, industrial and domestic effluents, sewage disposal and petroleum contamination (Santos *et al*., 2005).

Heavy metals in water can be found in the following forms:

(i) Solution.

(ii) Adsorbed on the minerals of the suspended load.

(iii) Associated with organic material.

Lakes and reservoirs are natural sinks for nutrients, metals and organic matter, which enter through run-off from agricultural land, controlled waste discharges, feeder streams and industrial effluent (Hayes *et al*., 1998). The biological availability and toxic effects of

metals once in a water body are closely related to their chemical forms. The forms in which the metals exist also determine their availability and distribution (Harrison and Laren, 1980). Whether they exist in suspension or associated with bottom sediment, it will in turn find its way to man either through public water supplies or by consumption of aquatic organisms.

2.7.2 Heavy metals in sediments

Accumulation of metals in waters (Lakes) goes together with its accumulation in sediments as a result of suspended solids sedimentation (Gonzalez *et al.,* 2000). According to Awadallah *et al.* (1994), distribution of trace metals in mud sediments of the Aswam high Lake depends on:

(i) Adsorption or co-precipitation on organic and detritus organic particles,

(ii) Preferential accumulation of trace metals by benthic organisms.

The importance of metals bound to sediments with respect to the overall concentration and mass transport of metals in an environment has been well established (Presley, 1992). Metal concentration in sediments can be influenced by factors such as salinity (Coakly *et al.,* 1993), water discharge (Forstner and Whittmann, 1981) and flow rate (Schoelhammer, 1995).

2.7.3 Heavy metals in fish

One of the significant effects of metal pollution in lakes or rivers is that aquatic organisms can absorb and accumulate concentrations in their tissues. Among the aquatic organisms,

fish has been reported to have a high tendency to accumulate heavy metals because they are the most common aquatic organisms at higher trophic level (Mansour, 2002). Therefore, bioaccumulation of metals in fish can be considered as an index of metal pollution in aquatic ecosystems (Forstner & Wittmann (1981). Heavy metals bioaccumulate in fish tissues and the accumulation of these heavy metals over time in fish leads to the suppression of fish immunity, hence, allowing the normal flora to cause ulceration and possible septicaemia (Mutuku, 2010).

The concentrations of heavy metal in fish depend on different factors such as ecological needs, size and age of individuals, their life cycle and life history feeding habits, season of capture, and physico-chemical parameters of water and sediment (Etim *et al*., 2013). Fish and other aquatic organisms have the tendency of accumulating heavy metals in their living cells to a very high concentration, much higher than those present in water, sediment and microflora in their environment (Forstner and Wittmann, 1981; Jonathan and Maina, 2009).

Water polluted with heavy metals adversely affects fish production. Metals may enter the systems of aquatic organisms (example, fish) via three main path ways, namely:

(i) Free metal ions got absorbed through respiratory surface (gills) and readily diffused into the blood stream

(ii) Free metal ions that are adsorbed onto body surfaces are passively diffused into the blood stream

(iii) Metals that are sorbet onto food and particulates may be ingested as well as free ions ingested with water (Bio-ellite, 2000).

Heavy metals exert a range of effects, from metabolic and physiological to behavioral and ecological effect on fishes (Forstner and Wittmann, 1981). Heavy metals are known to cause delayed embryonic development, malformation, and reduced growth rate of fishes, poor swimming performance, change in biochemistry such as enzyme activity and blood chemistry as well as changes in reproduction (Biney *et al*., 1994; Bio-ellite, 2000).

2.7.3.1 Effects of heavy metals on fish

(a) Lead: It has been reported by Olademeji and Offem (1989) that lead ions combine with mucus on the gills of fish and this interfers with its respiration, causing death due to suffocation. It is also reported that prolonged exposure of fish to lead causes changes in the liver, spleen and blood (Dawson, 1985).

(b) Copper: Copper when absorbed through the gills causes tissue damage and becomes fatal to the life of fish (Agarwal, 2005). Concentration of copper at the range of 0.8 ppm to 1.0 ppm in water causes severe toxic effect on many fish species and the physiological death appears as a result of interruption in gas exchange at the gills (Gautam, 1989).

(c) Cadmium: Eisler (1985) reported that fish exposed to 25 ppb of cadmium in an aquatic environment shows damage to testicular tissues and low reproductive capacity. They also reported enhanced lymphocyte count, slight anemia and changes in the concentration of potassium and magnesium in the blood, chronic depletion of glycogen in the liver and muscles of fish. The central nervous system and parenchymatous organs of fish is damaged due to acute toxic exposure.

(d) Chromium: Chromium does not accumulate in the bodies of fish, although high concentration of chromium resulting from frequent disposal of metal products in surface waters can cause damages to the gills of fish that swim near the point of disposal (Abioye *et al.,* 2015).

(e) Arsenic: Large proportion of arsenic in fish is the non toxic organic arsenic. The toxic inorganic form causes damage in the liver and other internal tissues. The main sources of human exposure to arsenic poisoning are consumption of fish and drinking water; while water is an important source of the more toxic inorganic arsenic, food fish is an important source of exposure to the less toxic organic arsenic (WHO, 2008b).

2.8 Biosorption of Heavy Metals

Biosorption is the use of biological materials as sorbents for the uptake of metal ions from aqueous solutions. It could also be said as the accumulation of metal ions from solution by microbial or plant material (Abioye *et al.,* 2015). This process utilizes inexpensive biomass for selective sequesteration of toxic heavy metals and is particularly useful for the removal of heavy metal contaminants from water and sediments. Various methods have been employed for the treatment of heavy metal-bearing industrial effluents, which usually include precipitation, adsorption, ion exchange, membrane and electrochemical technologies (Wierzba, 2010). Large quantities of metals can be accumulated by a variety of processes dependent and independent on metabolism.

However, these techniques are expensive, not environmentally friendly and usually dependent on the concentration of the waste which is ineffective in much diluted solutions (Volesky and Naja, 2007). The search for efficient, eco-friendly and cost-effective remedies

for wastewater treatment has been initiated (Olukanni and Kokumo, 2013). Of the different biological methods, biosorption has been identified and demonstrated to possess good potential to replace conventional methods such as reverse osmosis, electro dialysis, ultra filtration, ion-exchange and chemical precipitation for the removal of heavy metals (Malik, 2004).

Biosorption is very effective and can be easily adopted in low cost to remove heavy metals from large amount of industrial wastewaters. Examples of biosorbents include the following categories: bacteria, fungi, yeast, algae, industrial wastes, agricultural wastes and other polysaccharide materials (Vijayaraghavan and Yun, 2008). These biosorbents possess metal-sequestering property and can be used to decrease the concentration of heavy metal ions in solution from ppm to ppb level (Wang and Chen, 2009). Biosorbent behavior for metallic ions is a function of the chemical make-up of the microbial cells of which it consists. Mechanisms responsible for biosorption, although understood to a limited extent, may be one or combination of ion exchange, complexation, coordination, adsorption, electrostatic interaction, chelation and microprecipitation (Vijayaraghavan and Yun, 2008).

Bacteria are the most abundant and versatile of microorganisms and constitute a significant fraction of the entire living terrestrial biomass of 10^{18}g (Mann, 1990). Early 1980, some microorganisms were found to accumulate metallic elements with high capacity (Vijayaraghavan and Yun, 2008). Bacteria were used as biosorbents because of their small size, ubiquitous nature, ability to grow under controlled conditions and their resilience to a wide range of environmental conditions. Bacteria may either possess the ability for biosorption of more than one element or may be element specific depending on the species.

Biosorption process involves mainly cell surface sequestration; hence, the modification of cell wall can greatly alter the binding of metal ions. A number of methods have been employed for cell wall modification of microbial cells in order to enhance the metal binding capacity of biomass and to elucidate the mechanism of biosorption (Wang and Chen, 2009). The physical treatments include heating/boiling, freezing/thawing, drying and lyophilization. The various chemical treatments used for biomass modification include washing the biomass with detergents, cross-linking with organic solvents, and alkali or acid treatment. The pretreatments could modify the surface characteristics/groups either by removing or masking the groups or by exposing more metal binding site (Wang and Chen, 2009). Chemical modification methods can increase/activate the binding sites on the biomass surface and they include pretreatment, binding site enhancement, binding site modification and polymerization (Vijayaraghavan and Yun, 2008).

Biosorption involves a solid phase (sorbent or biosorbent; usually a biological material) and a liquid phase (solvent, normally water) containing a dissolved species to be sorbed (sorbate, a metal ion). Due to higher affinity of the sorbent for the sorbate species the latter is attracted and bound with different mechanisms. The process continues till equilibrium is established between the amount of solid-bound sorbate species and its portion remaining in the solution. While there is a preponderance of solute (sorbate) molecules in the solution, there are none in the sorbent particle to start with (Alluri *et al*., 2007). This imbalance between the two environments creates a driving force for the solute species. The sorption potential of any biomass depends on various factors such as the number of sites in the biosorbent material, the accessibility of the sites, the chemical state of the site (i.e.,

availability) and affinity between site and metal (i.e., binding strength) (Regine and Volesky, 2000).

Regeneration of the biosorbent is also important for keeping the process cost down and in opening the possibility of recovering the metals extracted from the liquid phase. For this purpose, it is desirable to desorb the sorbed metals and to regenerate the biosorbent material for another cycle of application. The desorption process should yield the metals in a concentrated form, restore the biosorbent close to the original state for effective reuse with undiminished metal uptake and no physical changes or damages to the biomass. Dilute mineral acids (HCl, H_2SO_4, HNO_3) have been used for the removal of metals from biomass (Zhou and Kiff, 1991; Puranik and Paknikar, 1997) and also organic acids (Citric, acetic, lactic) and complexing agents (EDTA, thiosulphate) can be used for metal elution without affecting the biosorbent (Mattuschka *et al*., 1993) as cited by Alluri *et al*. (2007).

Batch experiments are often used to evaluate the required fundamental information, such as biosorbent efficiency, optimum experimental conditions and biosorption rate, although most industrial applications prefer a continuous mode of operation (Omran and Mosstafa, 2015).

Biosorption have a lot of advantages over conventional methods and these include: high selectivity, more efficient, easy to operate, short operation time, reusability of biomass and hence, cost-effective for treatment of large volume of wastewaters (Abioye *et al.*, 2015).

CHAPTER THREE

3.0 MATERIALS AND METHODS

3.1 Study Area

Kainji Lake is located along River Niger in Northwestern Nigeria. It was impounded in 1968 along the Guinea savannah vegetation zone. This water body is located between longitude 9^o 50' and 10^o 55' East and Latitude 4^o 20' and 4^o 45' North. Kainji Lake is the largest manmade lake in Nigeria (Adekola *et al*., 2010). It has a length of 134 km, maximum width of 21.1 km, maximum depth of 60 meters and a surface area of 1270 km^2. The Lake with its large sizes after impoundment, attracted many fishermen both within and outside the country who settles along the bank with their families and depends on the lake fishery for their livelihood. Other activities like navigation, irrigation and washings are also carried out in addition to the generation of hydroelectric power. Figure 3.1 shows the map of Kainji Lake.

Jebba Lake is located in North Central Nigeria. It is a man-made Lake created in 1983 by the damming of River Niger below Kainji Lake, for the purpose of hydroelectric power generation, but, with opportunities for fishing, irrigated farming and navigation (Abiodun, 2002). The Lake is about 100 km long and about 12 km at its widest point. It is situated between latitudes 9^o 35' and 9^o 50' North and longitudes 4^o 30' and 5^o 00' East. The lake has an estimated surface area of approximately 303 km^2 (varying with seasonal fluctuations), and a volume of 3.31 x 109 m^2 with a mean depth of 3.3 m (max 32.5 m). It has a shoreline length of 74 km with catchments area of 0.3 x 106 km^2 (Ita *et al.*, 1983; Abiodun, 2002). The Lake gets its major bulk of water supply from Kainji Lake while tributaries like Rivers Oli and Eku also discharge into the lake. About 60 fishing villages

are situated around the Lake and the Lake supports about 1,160 people working directly as fishers (both fishing input owners and their assistants), using over 772 wooden planked canoes with 6 different types of fishing gears, gillnets, castnets, longlines, driftnets, liftnets and traps (Abiodun and Odunze, 2011). The Lake was primarily impounded for hydroelectric power generation, however, it also offers opportunities for developmental projects like fisheries, irrigation and navigation. Figure 3.2 shows the map of Jebba Lake and Plate III shows the aerial view of Kainji and Jebba Lakes.

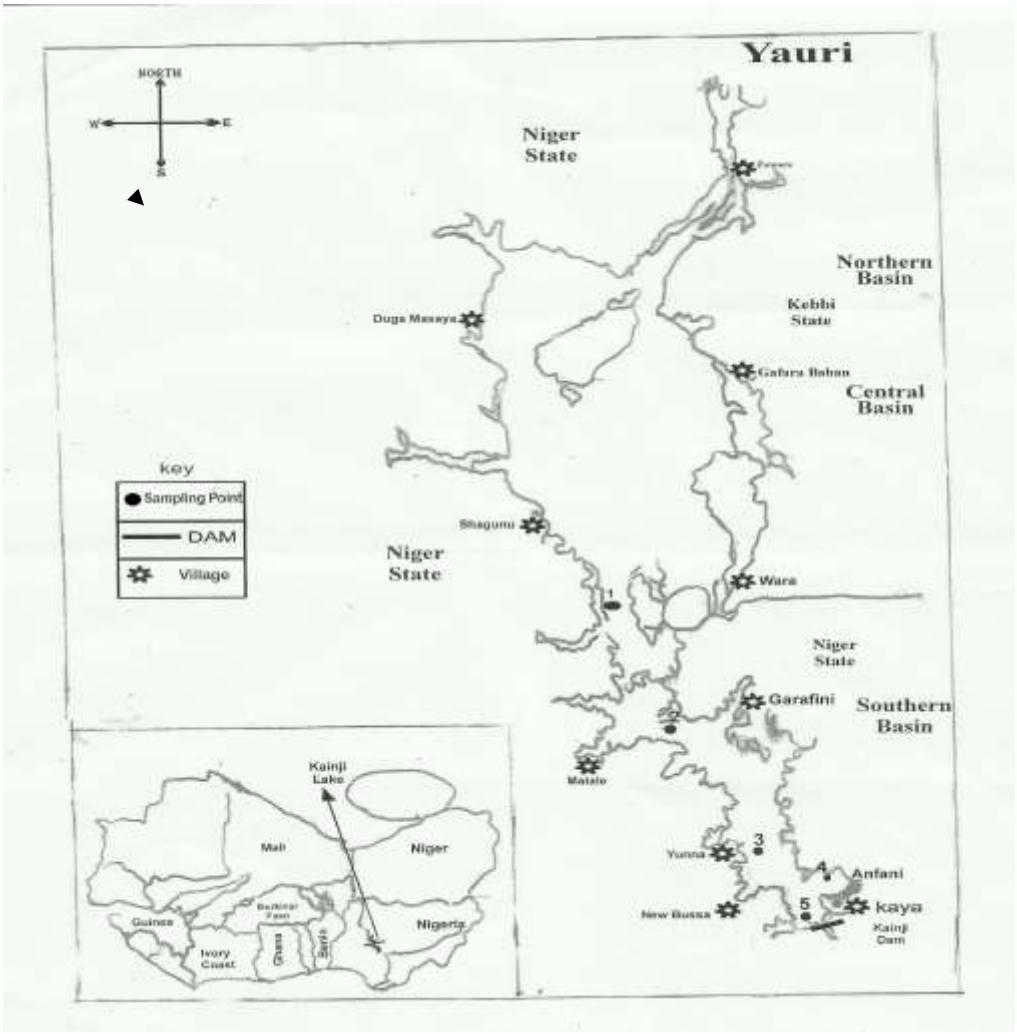

Figure 3.1: Kainji Lake
Source: Abiodun (2002)

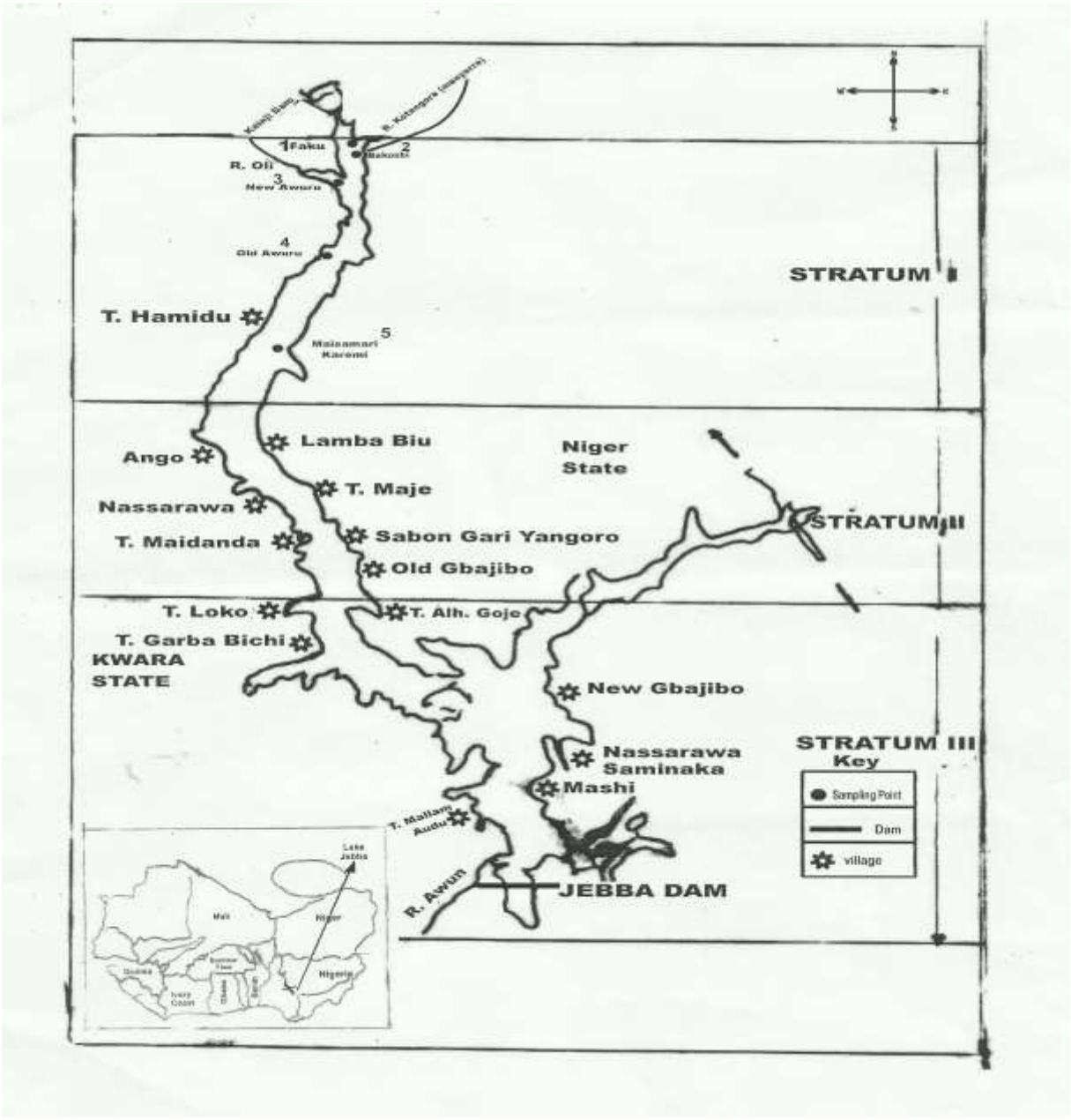

Figure 3.2: Jebba Lake
Source: Abiodun (2002)

Plate III: Aerial view of Kainji and Jebba Lakes, Nigeria

3.2 Description of Sampling Stations

Samples (water and sediment) were collected from 5 different stations each from the southern and northern basin of Kainji and Jebba Lakes respectively. Sampling Stations were at a distance of 7-10 km apart from each other. The selection of the sample Stations was based on different human activities being carried out around the station (such as agricultural farms, cattle husbandry operations, artisanal gold mining, domestic washings, irrigation and power generation).

Kainji Lake Station 1 (Kl.1) is characterized with much agricultural farms, domestic washings and high animal husbandry operations with low mining activities. It lies adjacent to Warra market and it is also the route in which boats come in and out of Warra town. Station 2 (Kl.2) is at the lower course of Warra and is characterized by high mining activities and agricultural farms. This Station has low animal husbandry operation. Station 3 **(**Kl.3) has heavy animal husbandry operations and much domestic activities (washing, bathing and fishing). It has high mining activities going on around the Lake. Station 4 (Kl.4) is at the lower course of Yunawa, it also has minimal mining activities and animal husbandry operation going on around the station. Station 5 (Kl.5) has minimal animal husbandry operation and less domestic activities (such as washings and bathing, without any mining activities). In some of these Stations, the water is used raw for drinking purposes.

Jebba Lake, Station 1 (JL.1) is at Faku with minimal animal husbandry operation but much human activities such as domestic washings and high fishing activities. It is also the Station that is directly below power generation Station. The water in this Station is used raw for drinking purposes by the surrounding villagers.

Station 2 (Jl.2) is at Manyarrah which is characterized by high irrigation activities, high mining activities, high animal husbandry operations, fishing activities, among others. It is at the lower course of Faku. Station 3 (Jl.3) is at New Awuru where human activities is reduced; however it forms a tributaries where river Oli with high mining activities flows into Jebba Lake. It is at the lower course of Manyarrah. Station 4 (Jl.4) is at Old awuru with much domestic washings and animal husbandry operations. It lies adjacent to Awuru market and is at the lower course of New Awuru. Station 5 (Jl.5) is at Maisamari Karemi, the lower course of Old Awuru with minimal human activities. Table 3.1 shows various coordinates of the sampling stations.

Table 3.1: Coordinates of Sample Stations in Kainji and Jebba Lakes

Sampling station	**Coordinates**		**Lake**
Kl. 1 (Warra)	N10^{o} 13.845’	E 004^{o} 36.951’	Kainji
Kl. 2 (Garafini)	N10^{o} 02.624’	E 004^{o} 36.713’	Kainji
Kl. 3 (Yunawa)	N 09^{o} 56.035’	E 004^{o} 36.163’	Kainji
Kl. 4 (Anfani)	N 09^{o} 54.556’	E 004^{o} 37.709’	Kainji
Kl. 5 (Kaya)	N 09^{o} 52.309’	E 004^{o} 37.123’	Kainji
Jl. 1 (Faku)	N 09^{o} 51.296’	E 004^{o} 36.890’	Jebba
Jl. 2 (Manyarrah)	N 09^{o} 49.771’	E 004^{o} 37.699’	Jebba
Jl. 3 (New Awuru)	N 09^{o} 46.347’	E 004^{o} 37.858’	Jebba
Jl. 4 (Old Awuru)	N 09^{o} 43.166’	E 004^{o} 37.373’	Jebba
Jl.5 (Maisamari Kerimi)	N 09^{o} 41.241’	E 004^{o} 37.185’	Jebba

Keys: Kl = Kainji Lake, Jl = Jebba Lake

3.3 Sample Collection

3.3.1 Collection of water samples for microbiological and physico-chemical analyses

Water samples were collected from five (5) different sampling Stations in each of the Lakes. During the sampling exercise, a global positioning system (GPS) device (Model 12 XL : S/N 84567093) was used to take coordinates at each sampling station. This was carried out once every month (monthly) for a period of twelve months (January 2017 – December 2017). With the help of a boat, water samples were collected at 150 m away from the bank and at a depth of 30 cm below the surface using van-dorn water sampler (Plate IV). The water was collected directly into 250 mL sterilized plastic bottles and corked as described by the American Public Health Association (APHA, 1985). The bottle containing each water sample was labeled and stored in portable ice box at temperature of not more than $5^{o}C$ so as to stop or reduce microbial activity. These samples were transported to microbiology laboratory and chemistry laboratory at central laboratory unit of National Institute for Freshwater Fisheries Research, New Bussa, Niger State for analyses.

Water samples for the determination of physico-chemical parameters were collected into a clean two-litre capacity bottle. It was collected from each sample station for the determination of dissolved oxygen (DO), temperature, pH, biological oxygen demand (BOD), conductivity, nitrate, total alkalinity and phosphate. Water samples for BOD were covered immediately with thick black coloured plastic bag while the other set for dissolved oxygen (DO) was fixed immediately with 2 mL each of Winkler's reagents [Winkler's I ($MgCl_2$) and Winkler's II (KOH + KI)] reagents before it was taken to the laboratory for analysis.

Plate IV: Vandorn water sampler

3.3.2 Collection of water samples for the determination of heavy metals

Water samples were collected from all the sample stations of the Lakes. Two litre capacity polyethylene sampling bottles were used to collect the water samples (4 times) at each sampling station. The pre-cleaned sampling bottle was immersed below the surface to collect the water sample. These water samples collected at each sampling site were mixed in a plastic bucket and a representative sample of one litre was transferred into the polyethylene bottle. Samples were acidified with 2 cm^3 of 10 % HNO_3. This was placed in an ice box in order to stabilize the metal ions and prevent volatility of the constituents

(APHA, 1992). This was transported to the laboratory for further analysis according to the procedure described by ÖztÜrk *et al*. (2009).

3.3.3 Collection of fish samples for bacteriological analysis

The fish species of African catfish (*Clarias gariepinus*) and Nile tilapia (*Oreochromis niloticus*) were sampled because of their abundance and also been the most common fish caught from the Lakes. The fish species were properly identified by fish biologists from Artesinal Fisheries Division of the National Institute for Freshwater Fisheries Research, New-Bussa, Niger state, Nigeria.

Samples of freshly caught *Oreochromis niloticus* and *Clarias gariepinus* were collected from the fishermen, killed by decapitation (severance of the spinal cord behind the head), rinsed three times with distilled water and dropped into a clean sterilized plastic container of 6.0 liters capacity. This was then kept in an ice chest box and transported to the laboratory for bacteriological analysis according to the procedure described by Ogbondeminu *et al*. (1991).

3.3.4 Collection of fish samples for heavy metal analysis

Fish samples were collected from the local fishermen, rinsed properly with distilled water to get rid of the debris and other external adherents. They were properly weighed, wrapped in aluminium foil paper and then kept in an ice box before being transported to the central services laboratory of National Cereals Research Institute, Badeggi, Niger State, Nigeria. It was frozen at – 10 ^{o}C prior to its analysis. With the use of sterilized dissecting set, the gills, muscles and intestines were removed. These different organs were subjected to drying in hot air oven, IKA HB 10, Germany, and dried to a constant weight at 80 ^{o}C. Porcelain

mortar was used to grind the dried samples to fine powder with an electric blender, Microbiology international, EZ-Mix 400, USA. These powders of the different organs/tissues were further digested for heavy metal analysis.

3.3.5 Collection of sediment samples for bacteriological analysis

Sediments were collected from the five (5) sampling stations (each from Kainji and Jebba Lakes) for twelve (12) months (January 2017 – December 2017). An Ekman grab (Plate V) was used to collect these samples from the bottom of the Lakes into a steriled container for bacteriological analysis. The sediments were kept in an ice box and taken to the microbiology laboratory at central laboratory unit of National Institute for Freshwater Fisheries Research, New Bussa, Niger State for analysis according to the procedure of APHA (1992).

3.3.6 Collection of sediment samples for heavy metal analysis

Bottom sediments were collected from each of the sample stations into pre-cleaned polythene bags using Ekman grab. Sediments were collected from the five (5) different points in each station and these were pulled together to make a composite sample. The pre-cleaned bag containing the sample was sealed properly to prevent any interference. This was kept in an ice chest box to reduce further microbial activity (APHA, 1992). It was air-dried and sieved with 200 mm mesh screen to remove larger particles.

Plate V: Ekman grab

3.4 Bacterial Enumeration

3.4.1 Determination of faecal coliforms using most probable number (MPN) technique

Faecal coliform count was carried out using most probable number technique (APHA, 2003). In the presumptive test, a three series of five tubes each containing 10 mL, 1mL and 0.1 mL portions of the water sample were inoculated in sterilized macConkey broth. Sterilized macConkey broth was inoculated with sterile distilled water to serve as a control. Inoculated tubes were then incubated at 37 °C for 24 hours. Inoculum was transferred from

all tubes showing acid and gas production (an indication of the presence of total coliform) to tryptose bile broth (using a sterile wire loop) and incubated at 44.5 °C for 24 hours (for faecal coliforms). Gas production in a fermentation tube within 24 hour was an indication of a positive reaction for faecal coliforms. The estimated number of faecal coliforms, present in 100 ml was read from a tabulated probability table using corresponding results of various combinations of positive and negative reactions from each of the three batches (APHA, 2003). In the confirmatory test, two loopful of broth from the positive tubes above were transferred to brilliant green lactose bile broth (BGLB) and incubated at 44.5 ^{o}C. Gas formation after 24 hours is an indication of a positive confirmed test (Nduka, 2011).

In the completed test, with the help of a sterilized wire loop, positive growth from BGLB was streak on eosin methylene blue (EMB) agar and incubated at 44.5 ^{o}C for 24 hours. After 24 hours, colonies showing metallic sheen colouration and Gram negative rods (on staining) were indications of positive completed test (Nduka, 2011).

3.4.2 Enumeration of viable bacterial count (VBC) in water samples

A 1.0 mL of water sample was serially diluted into 9.0 mL steriled distilled water. After dilution, 1.0 mL of each serially diluted sample was inoculated onto sterilized plate count agar and spread evenly on the surface of the media using sterilized bent glass rod (standard plate count technique) according to the method of Ijah *et al.* (2008). These inoculated plates were then incubated at 35 ^{o}C ± 2 ^{o}C for 24 hours. After incubation, the number of colonies were counted using colony counter, SCIQUIP Ltd. Scan 100, UK. Colony forming units (CFU) of bacteria in the water sample were then calculated and recorded as colony forming units (CFU) = colony count x dilution factor / inoculum volume.

3.5 Preparation of Starch Ampicillin Agar (SAA) for Isolation of *Aeromonas*

Starch ampicillin agar (SAA) was prepared based on the method of Palumbo *et al*. (1985) for primary isolation of *Aeromonas*. This medium consisted of nutrient agar as the base medium, starch and ampicillin were added as differential and selective agents respectively. Ten (10) grams of soluble starch and 24 grams of nutrient agar were dissolved in one litre of distilled water. This was autoclaved at 121 ^{o}C for 15 minutes and cooled to 50 ^{o}C and ampicillin was added to achieve a concentration of 10 mg/litre. The medium was poured into sterile Petri dishes and left on the bench to solidify. After it has solidified, the plates were packed, ready for inoculation of samples.

3.6 Bacterial Identification

Bacterial identification was carried out following standard procedures of Clemens *et al.* (1999), Lejeune *et al.* (2001), Cheesbrough (2000), Johnson *et al*. (2003), Louis (2003) and Bergey's Manual of Systematic Bacteriology (Krieg and Holt, 1994).

3.6.1 Bacterial identification from water and sediment samples

Enrichment media (Selenite F broth, tryptic soy broth (TSB) and alkaline peptone water (APW) were inoculated with samples (water and sediment samples) and incubated at 37 ^{o}C. After 8 hours, the broth cultures were re-inoculated into appropriate media (Nutrient agar, MacConkey agar, Sorbitol MacConkey agar, Eosin Methylene blue agar, Starch Ampicillin agar, Bismuth sulphite, Thiosulfate Citrate Bile Salt Sucrose agar) for primary isolation. These inoculated plates were incubated at 37 ^{o}C for 24 hours using Thermo scientific incubator, 51028063, USA. After incubation, colonies that differ in size, shape and colour were picked, re-inoculated into fresh media (Nutrient agar, MacConkey agar, Sorbitol

MacConkey agar, Eosin Methylene blue agar, Starch Ampicillin agar, Bismuth sulphite, Thiosulfate Citrate Bile Salt Sucrose agar) and incubated at 37 ^{o}C to obtain pure colonies. The pure colonies were sub-cultured into nutrient agar slants and stored in the refrigerator for further tests.

A gram staining, oxidase test and motility tests were conducted on the pure colonies prior to the use of Microbact Identification Kits.

3.6.1.1 Gram staining

Gram staining was carried out following the procedures of Cheesbrough (2000).

A smear of a colony from the isolate was made on a clean glass slide, air dried and was heat fixed.The fixed smear was stained with crystal violet stain for 1 minute and washed off with distilled water.The smear was floaded with lugol's iodine for 1 minute and was washed off with distilled water. This was decolourized rapidly with acetone and washed immediately with distilled water. Then, smear was counter stained with neutral red for 1 minute and washed off with distilled water. The back of the slide was wiped clean and place in a draining rack to air-dry. The stained slide was examined using oil immersion objective. Bacterial cells were recorded as Gram positive or gram negative.

3.6.1.2 Oxidase test

An oxidase strip was placed in a clean Petri dish. Inoculum of the test organism was picked and smeared on the filter paper. A positive test was recorded if colour changed to purple blue within 5 seconds.

3.6.1.3 Motility test

The test organism was inoculated into motility medium in a tube by making a fine stab with a sterilized inoculation pin. The inoculated tube was incubated at 35 ^{o}C for 24 hours. Motility test was recorded as positive if faint line from the stab and cloudy medium were observed. The bacterial isolates were further screened biochemically using Microbact Identification Kits, 12A and 12B (MB1132A/ Australia) and the identies were confirmed using molecular analysis.

3.6.1.4 Biochemical analysis of the bacterial isolates using microbact identification kits

(a) Preparation of strip

Microbact incubation tray was prepared and the strip was placed in it.

(b) Preparation of inoculum

Fresh isolates (24 hours old) culture was used for this inoculation. A colony was picked using a sterilized inoculation wire loop and this was emulsified to obtain a homogenous bacterial suspension in a tube containing 5 mL of sterilized saline (0.85 %) water.

(c) Inoculation of the strip

With the help of a sterile Pasteur pipette, four (4) drops (approximately 100 µL) of the bacterial suspension was filled into the wells. Using a sterile dropper, wells 1, 2, 3 and 8 for 12A and well 12 for 12B were overlayed with sterile mineral oil. Well 8 for 12B was not overlay with oil for oxidase positive bacteria. Anaerobic condition was created in the tests lysine decarboxylase (LDC), ornithine decarboxylase (ODC), hydrogen sulphide production (H_2S), Indole production from tryptophan (Ind) and arginine dihydrolase (ADH) respectively. The strip was then resealed and incubated at 37 ^{o}C ±2 ^{o}C for 24 hours.

(d) Reading of strip

Upon incubation for a period of 24 hours, the strips were examined and all spontaneous reactions (+/-) after being compared with colour chart were recorded on the result sheet. Additional reagent was added to tests such as: Tryptophane deaminase (TDA) of which a drop of TDA reagent was added to the bacterial suspension. A dark brown colour (an indication of positive reaction) was recorded as such. Two drops of indole (Kovacs) reagent were added to the indole test for indole production. Development of a pink colour was an indication of a positive reaction. A drop (each of VP1 and VP2 reagents) was added to well ten (10) and allowed for fifteen (15) minutes and this was observed for reaction. A pink colour was an indication of a positive reaction. One drop each of Nitrate reagent A and Nitrate reagent B were also added to well seven (7) after reading result of hydrolysis of *B*-nitrophenyl-B-*d*- galactopyranoside. This was done to show nitrate reduction.

(e) Interpretation and identification of organisms

Patterns of the reactions were coded into a numerical profile using Oxoid microbact results sheet. On the results sheet, the tests were separated into groups of 3 and a value 4, 2 and 1 as designed by the manufacturer was indicated for each. Adding the values corresponding to reactions within each group, a nine (9) digit profile number (code number) was obtained for all the tests on a strip. Results (code number) were keyed into an octal code in the software and identity of the organism observed was recorded (Plates VI and VII).

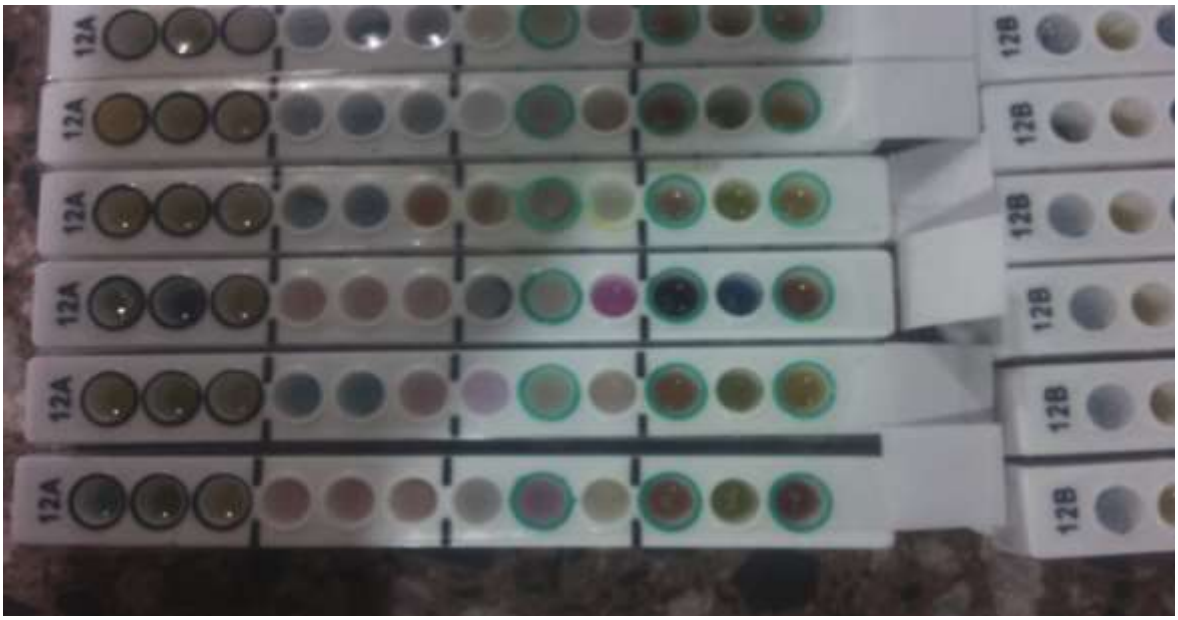

Plate VI: Microbact Strip

Plate VII: Microbact Result Sheet

3.6.2 Molecular characterization of bacterial isolates from water samples from Kainji and Jebba Lakes

Bacterial isolates from water samples were characterized using molecular technique (Luo *et al*., 2017). Although standard biochemical identification methods were been carried out, the use of 16S-rDNA identification has become more acceptable due to its greater degree of accuracy and specificity. Recently, it become apparent that the classical importance attributed to *Aeromonas hydrophila* is actually the result of a bias analysis produced by culture and biochemical identification methods (Beaz – Hidalgo *et al*., 2015) and many *Aeromonas* species were formally masked under *Aeromonas hydrophila* (Figueras *et al*., 2017). The use of 16S-rDNA has re-classified most of these bacterial species. Molecular characterizations of ten bacterial isolates were conducted at Erevna Laboratories, Ibadan, Nigeria. The procedures used for molecular analysis were as follows:

A. DNA Extraction

Genomic DNA extraction was carried out using heat method of extraction (Luo *et al*., 2017). Bacterial cells were harvested from 1000 µL of bacterial broth culture using a microcentrifuge at 10,000 g for 1 minute. Five hundred microlitres (500 µL) of PCR grade water was added to the pellet and centrifuged at 10,000 g for 1 minute to wash off residual broth. Recovered cells were resuspended in 200 µL of PCR grade water. The resultant mixture was heated at 95 ^{o}C for 15 minutes and cooled at room temperature (25 ^{o}C) for 5 minutes. The mixture was then centrifuged at 5000 g (using Druncker diagnostics, Horizon 6 flex, USA) for 5 minutes to separate the DNA from other cell debris. The supernatant containing the DNA was transferred to a clean Eppendorf tube.

B. 16S rDNA PCR Amplification

Each PCR reaction mixture consisted of 5 µL mastermix (Jena Bioscience Red Load Taq Master mix (5x)), 1 µL of 10 pmol each of 27F-AGAGTTTGATCCTGGCTCAG and 1492R- GGTTACCTTGTTACGACTT, 1 µL DNA template and 17 µL sterile nuclease free water to make up a total reaction mixture of 25 µL. PCR was carried out on a GeneAmp 9700 PCR System Thermalcycler (Applied Biosystem Inc., USA) following the protocol described by Luo *et al*. (2017). The mixture was subjected to an initial denaturation at 94 °C for 3 minutes; followed by 35 cycles of 94 °C for 45 seconds, 55 °C for 60 seconds and 72 °C for 60 seconds; and a final extension at 72 °C for 10 minutes (Luo *et al*., 2017).

C. Sequencing and Identification

PCR products were electrophoresed on 1.5 % agarose gel along with Jena Biosciences 100 bp ladder, which was then photographed using a camera in dark room. The amplified fragments were sequenced using a Genetic Analyzer 3130 x l sequencer (Applied Biosystems). The sequencing kit used was BigDye terminator v3.1 cycle sequencing kit. Bio- Edit software and molecular evolutionary genetics analysis (MEGA 6) were used for all genetic analysis. Identities and accession numbers of the isolates were determined using Basic Local Alignment Search Tool (BLAST) from the GENE BANK at www.ncbi.nlm.nih.gov .

3.6.3 Bacterial identification from fish samples

With the help of a sterilized wire loop, swab from the fish samples (gills, intestines and muscles) was collected and inoculated into an appropriate medium, differential and selective media (Nutrient agar, MacConkey agar, Sorbitol MacConkey agar, Eosin Methylene blue agar, Starch Ampicillin agar, Bismuth sulphite, Thiosulfate Citrate Bile

Salt Sucrose agar). These inoculated plates were incubated at 37 ^{o}C for 24 hours using an incubator. After incubation, colonies were picked, re-inoculated into fresh media and incubated at 37 ^{o}C to obtain pure colony. The pure colonies were sub-cultured into agar slants and stored in the refrigerator for further biochemical tests. Isolates were further screened biochemically using Microbact Identification Kits, 12A and 12B (MB1132A/ Australia) (Cheesbrough, 2000).

3.7 Sample Preparation for Determination of Heavy Metals

3.7.1 Water samples

Water samples were digested according to APHA (2005). One hundred millilitres (100 mL) of water sample was transferred into a 125 mL conical flask. Five millilitres (5 mL) of concentrated HNO_3 was added. The solution was allowed to boil slowly and evaporated on a hot plate to 2 mL, to allow precipitation. Heating and addition of concentrated HNO_3 continued until a light coloured clear solution was obtained. The sample was not allowed to dry during the digestion. The wall of the flask was rinsed with distilled water and the solution filtered. The filtrate was transferred into a 100 mL volumetric flask and diluted to the mark and mixed thoroughly. This was stored in a plastic vial prior to heavy metal determination. Portion of this solution was used for heavy metals determination using Atomic Absorption Spectrophotometer (PG instrument model, AA500 Spectrophotometer, UK).

3.7.2 Sediment samples

One gram (1.0 g) of the dry homogenized sediment was weighed into a clean beaker. Five millilitres (5 mL) of 50 % HNO_3 and 15 ml of concentrated HCl (ratio 1:3) were added. This mixture was heated gently on a hot plate until clear and white fumes of $HClO_4$

appeared. This was allowed to cool down and 50 mL of distilled water was added. This was filtered and made up to 100 mL in a volumetric flask and mixed thoroughly. A portion of this solution was used to determine heavy metals according to the procedure of APHA (1992).

3.7.3 Fish samples

The dried fish organs (muscles, gills, intestines) were ground separately in a blender and filtered through a 125 µm sieve (Bio-Elite, 2000). One gram (1.0 g) of homogenate each was digested with 5 mL portion of concentrated HNO_3 acid and after which 50 mL of distilled water was added. This was filtered and the filtrate was made up to 100 mL in a volumetric flask using distilled water (APHA, 1992). A portion of this solution was used for the determination of heavy metals.

3.8 Determination of Heavy Metals

After digestion, the filtrate of each sample was thoroughly mixed and analyzed using atomic absorption spectrophotometer (PG instrument model, AA500 Spectrophotometer, UK). A standard solution of each of the heavy metals was prepared from their salts. Reagent blank was similarly prepared. The atomic absorption spectrophotometer (PG instrument model, AA500 Spectrophotometer, UK) was set at optimum conditions for maximum absorbance signal at the wavelength (283.306 nm, 324.754 nm, 228.802 nm, 357.806 nm and 278 nm for Pb, Cu, Cd, Cr and As respectively) of each element and this was used to determine concentration of each element in the samples, standards and the blanks. The concentration for each of the elements was obtained based on Beer Lambert's law, which states that absorbance is directly proportional to the concentration of metal at a

particular wavelength. Standard calibration curve was drawn using signals (absorbance) of the standard solutions of each element. Concentration of elements in the samples was determined by extrapolation of the absorbance on the calibration curve.

3.9 Analysis of Physico-chemical Parameters of Water

3.9.1 Temperature

This was determined *in situ* using mercury thermometer (T1 62101 Wika USA). The thermometer was placed vertically by immersing the bulb containing the mercury inside the water sample. This was allowed to stand until the rise in mercury level was steady. The temperature reading was then taken in degree centigrade (^{o}C) according to the method of Ademoroti (1996).

3.9.2 Dissolved Oxygen (DO)

Dissolved oxygen was determined using Modified Winkler's method according to the procedure of APHA (1992). Two hundred millilitres (200 mL) sample bottle was dipped into the Lake water and allowed to fill and overflow. With the help of a pipette, 1.0 mL of manganese sulfate ($MnSO_4$) was introduced into the sample followed by the addition of one millilitre (1.0 mL) of alkali – iodide – azide reagent (NaOH + NaI). The stopper was replaced and mixed thoroughly by inverting the bottle five times. One millilitre (1.0 mL) of concentrated tetraoxosulphate VI acid (H_2SO_4) was added to the sample and the stopper replaced and mixed thoroughly. Ten millilitres (10.0 mL) of the sample was transferred into a conical flask and titrated agains sodium thiosulphate ($Na_2S_2O_3.5H_2O$) until an end point (pale straw colour) (APHA, 1992). Dissolved oxygen was calculated using equation 1.

$$\text{DO (mg/L)} = \frac{\text{Volume of sample collected X Titre value}}{\text{Volume of sample used in titration}} \qquad \text{Equation (1)}$$

3.9.3 Biological Oxygen Demand (BOD)

Determination of BOD followed the same procedure as that of dissolved oxygen except that the sample bottles were covered with thick black polythene bag and the sample after collection was allowed to stand for 5 days before titration (APHA, 1992).

3.9.4 Water transparency

It was measured using Secchi disc. The disc (which was coloured in black and white) was attached to a long rope, graduated in centimeter. This disc was lowered into the Lake water. At a point where the disc disappeared and reappeared (while lowering and withdrawing respectively), the reading was taken from the graduation and expressed in centimeter (APHA, 1992). Transparency was calculated using equation 2.

$$\text{Transparency} = \frac{A + B}{2} \qquad \text{Equation (2)}$$

Where, A = Depth at which Secchi disc disappeared.

B = Depth at which Secchi disc reappeared.

3.9.5 Total alkalinity

One hundred millilitres (100 mL) of water sample was measured into a conical flask and one drop of phenolphthalein indicator was added. A drop of methyl orange indicator was added (which changed the colour to yellow). A 0.1 M of hydrochloric acid (HCl) was used to titrate against the mixture in the conical flask up to an end point (reddish coloration). This was marked the end-point of the first titration and the reading was taken and recorded.

The mixture in the conical flask was boiled and allowed to cool and 0.1 M HCl was again used to titrate against the mixture (that is the second titration up to the formation of faint yellow coloration) and the second reading was taken and recorded. The first and the second readings were summed up and multiplied by fifty (50), which is the approved standard conversion factor to obtain the total alkalinity of the water (expressed in mg/l) (Ikeme *et al.*, 2014).

3.9.6 Electrolytic conductivity

Electrolytic conductivity was determined using conductivity meter. The meter was calibrated using potassium chloride solution (KCl_2 {0.7455 g} + D/W {1000 mL}), which gave a conductivity of 1412 μscm^{-1}. One hundred millilitres (100 mL) of water sample was filled into a beaker. The electrode of the conductivity meter was rinsed in distilled water and placed into the water sample. The button on the meter was ON and the reading displayed. This was expressed in microsiemiens/centimeter.

3.9.7 pH

The pH of the water samples was measured using Jenway pH meter. The electrode was rinsed with de-ionised water and dried with soft tissue paper. It was placed in a buffer with pH value of 7.0. Calibration button was pressed and the pH reading of the buffer taken. This was allowed to stabilize and remain for one to two minutes. The electrode was then remove and rinse with distilled water and dried before taking pH reading of the sample. One hundred milliliter (100 ml) of the sample was measured and dispensed in a sample bottle after which the electrode was placed in the water sample, and the meter reading was recorded.

3.10 Screening of the Bacterial Isolates for Biosorption Potentials

Screening of isolates for metal biosorption potential was carried out using well diffusion method. (Abioye *et al*. 2018). Five (5) ppm of each heavy metal (lead, copper, cadmium, chromium and arsenic) was prepared and the metal pH solution was adjusted to 7.0 using NaOH and HCl. Culture medium (Nutrient agar) was sterilized using autoclaved at 121 ^{o}C for 15 minutes and poured into Petri dishes and allowed to solidify. With the help of a sterilized cork borer of 5.0 mm diameter, a well was made on the media. The test organisms were inoculated and spread using sterilized bent glass rod as previously carried out by Abioye *et al*. (2018). One millilitre (1 mL) of 5 ppm heavy metal was inoculated into the well and the plate was incubated at 37 ^{o}C $\pm$ 2 ^{o}C for 24 hours. Development or presence of bacterial colonies around the well was an indication of its ability to tolerate the heavy metal. Absence of bacterial growth was an indication that the test organism could not tolerate the heavy metal, hence, could not grow in presence of the heavy metal.

3.11 Preparation of Metal Stock Solution

Stock solution of ($K_2Cr_2O_7$), $Pb(NO_3)_2$, AS_2O_3, $Cd(NO_3)_24H_2O$, $CuSO_45H_2O$ was prepared by dissolving 1.40 g, 0.39 g, 0.66 g, 0.67 g, 0.98 g respectively, each in 250 mL of distilled water. The solution was agitated for 15 minutes and allowed to stand for 24 hours in order to obtain complete dissolution of the salt (Abioye *et al*., 2017). The pH of the solution was adjusted to 7.0 using NaOH and HCl. Concentrations of 1.0 ppm, 3.0 ppm and 5.0 ppm were serially prepared and determined using atomic absorption spectrophotometer (PG instrument model, AA500 Spectrophotometer, UK).

3.12 Biosorption Analysis

Fifty millilitres (50 mL) of nutrient broth was measured into a two hundred and fifty millilitres (250 ml) flask and this was sterilized at 121 ^{o}C for 15 minutes and allowed to cool. The pH solution was adjusted to 7.0 using NaOH and HCl. Two millilitres (2 mL) of inoculum of a known bacterial isolate was added to the nutrient broth. A known concentration of heavy metal (Pb, Cu, Cr, Cd, As) stock solution was added to the nutrient broth in the flask. This was incubated at 37 ^{o}C and examined at 7 days interval for a period of 28 days, (Abioye *et al*., 2015). The supernatant and the cells were separated by centrifugation at 4000 revolutions per minutes (rpm) for 25 minutes. Concentration of the heavy metals was determined on the 7th, 14th, 21st and 28th day of incubation using atomic absorption spectrophotometer (PG instrument model, AA500 Spectrophotometer, UK). Percentage removal (% R) of each heavy metal was calculated using equation 3.

$$(\%\ R) = \frac{C1 - C2}{C1} \times 100 \qquad \text{Equation (3)}$$

Where C_1 = Initial concentration, C_2 = Final concentration, % R = Percentage removal.

Environmental factors such as temperature, pH, biomass concentration and metal concentration are very vital in this process. The metal ion speciation in solution, surface charge of the biomass, and chemistry of biomass binding sites are greatly affected by the pH (Amirnia *et al*., 2012). In this study, environmental factors such as pH, temperature and biomass concentration were the same; however, heavy metal concentrations (1 ppm, 3 ppm, 5 ppm) and contact time (7 days, 14 days, 21 days, 28 days) varied.

3.13 Data Analysis

Coefficient of correlation between total viable bacterial count, faecal coliform count and the physicochemical parameters were calculated using Pearson correlation test. Statistical significance was set at $P \leq 0.05$. Student T test was used to compare physicochemical parameters of wet season (April, May, June, July, August, September) and dry season (October, November, December, January, February, March). Similarly, total viable bacterial count (TVC) and faecal coliform (FC) in wet season was compared with the count during dry season using T test. T test was employed to determine significance between stations and seasons. Duncan's multiple range tests was used to compare the variables at each sample station.

CHAPTER FOUR

4.0 RESULTS AND DISCUSSION

4.1 Mean Faecal Coliform Counts of Water Samples from Kainji and Jebba Lakes using Most Probable Number (MPN) Technique

Table 4.1 shows the mean faecal coliform counts of water samples from Kainji and Jebba Lakes. It was observed that faecal coliform count in Kainji Lake Station 1 had the highest coliform count (305.17 MPN/100 mL) while Station 4 had the lowest count (171.83 MPN/100 mL). In Jebba Lake, Station 2 had the highest count (700.83 MPN/100 mL) while Station 5 had the lowest count (200.92 MPN/100 mL). Stations 1 and 2 of Jebba Lake, which were at the upper course, had higher faecal coliform counts than Stations 3, 4 and 5, which were at the lower course.

It was also observed that the faecal coliform counts at Stations 1 and 2 of Jebba Lake were above maximum permissible limit of 5.0×10^2 MPN/100 mL according to FEPA (2003). This might be due to increased human activities like washing, bathing and animal husbandry among others. This increase above permissible limit is an indication of potential human and other animal health risks to be caused by pathogens as earlier reported (Byamukama *et al*., 2000), The most reliable tool for assessing health risks posed by pathogens in water is faecal coliform counts and detection of *Escherichia coli*.

Stations 3, 4 and 5 of Jebba Lake had counts that were within acceptable limits. This might be as a result of self purification as the Lake flows downstream as well as the absence of pronounced sources of pollution into the Lake. In general, however, Jebba Lake was observed to have higher coliform counts due to increased human activities than Kainji Lake.

Table 4.1: Mean Faecal Coliform Counts (MPN/100 mL) of Water from Kainji and Jebba Lakes

Sample Station	Kainji Lake Min-Max	Mean ± SD	Jebba Lake Min-Max	Mean ± SD	Max permissible limit (FEPA, 2003)
1	8.00 - 540.00	305.17 ± 184.02	6.00 - 1600.00	580.83 ± 475.50	500
2	6.00 - 350.00	206.33 ± 139.11	20.00 - 1600.00	700.83 ± 531.27	500
3	14.00 -540.00	303.58 ± 190.31	12.00 - 920.00	368.92 ± 317.72	500
4	20.00 - 430.00	171.83 ± 136.85	31.00 - 920.00	368.92 ± 317.72	500
5	10.00 - 430.00	191.83 ± 127.86	14.00 - 540.00	200.92 ± 171.12	500

4.2 Seasonal Faecal Coliform Count of Water from Kainji and Jebba Lakes using Most Probable Number (MPN) technique

In both Kainji and Jebba Lakes, faecal coliform count during the period of wet season (May, June, July, August, September and October, 2017) were higher than those of the period of the dry season (January, February, March, April, November and December, 2017). Minimum faecal coliform count in Kainji Lake during the dry season was 6.0 MPN/100 mL while in the wet season was 63.0 MPN/100 ml. In Jebba Lake, minimum faecal coliform count during the dry season was 6.0 MPN/100 ml while that of wet season was 84.0 MPN/100 mL (Table 4.2). This higher count during the wet season above that of the dry season is an indication that pollution of the Lakes was majorly caused by runoffs from adjacent farms into the Lakes. This agrees with the study of Ampofo and Clerk (2010), who reported that lack of animal waste management could directly affect water quality as a result of surface run offs. The presence of faecal coliform in the Lakes is an indication of contamination since faecal coliforms are not normal flora of surface water. It therefore means that this contamination could have arisen from human and livestock activities. T-test result shows that there was significant difference ($P < 0.05$) between wet and dry seasons in both Lakes (Appendices A and B).

Table 4.2: Mean Seasonal Faecal Coliform Counts (MPN/100 mL) of Water from Kainji and Jebba Lakes

Parameter	Kainji		Jebba	
	Wet	Dry	Wet	Dry
Minimum	63.00[a]	6.00[b]	84.00[a]	6.00[b]
Maximum	540.00[a]	430.00[b]	1600.00[a]	920.00[b]
Mean ± SD	325.90±152.13	145.65±118.43	649.13±428.90	219.30±239.11

Means with dissimilar letter (s) differ significantly according to student t test.**Significant at p ≤ 0.05**

4.3 Mean Viable Bacterial Count (VBC) of Water from Kainji and Jebba Lakes

The mean viable bacterial count at Station 1 in Kainji Lake was higher (5.4×10^5 CFU/100 mL) than other Stations in the same Lake. Station 5 in Kainji Lake had the lowest viable bacterial count (6.3×10^4 CFU/100 mL). In Jebba Lake, Station 3 had the highest viable bacterial count (4.9×10^5 CFU/100 mL), followed by Station 2, which recorded 3.0×10^5 CFU/100 mL and Station 1 (2.7×10^5 CFU/100 mL). Station 4 had the lowest count (2.3×10^5 CFU/100 mL). In Kainji Lake, Sample stations 1, 2 and 3 were observed to be above maximum permissible limit of 1.0×10^5 CFU/100 mL according to Buras *et al*. (1987) while Stations 4 and 5 were within acceptable limit. Unlike Kainji Lake, the viable bacterial counts in all the sample Stations of Jebba Lake were observed to be above the maximum permissible Limit (Table 4.3).

Table 4.3: Mean Viable Bacteria Count (VBC) of Water Samples (cfu/mL) from Kainji and Jebba Lakes

Sample station	Kainji Lake Minimum-Maximum	Mean	Jebba Lake Minimum-Maximum	Mean
Station 1	$3.1x10^3 - 2.6x10^6$	$5.4x10^5$	$4.2x10^2 - 2.6x10^6$	$2.7x10^5$
Station 2	$1.3x10^4 - 2.1x10^6$	$3.5x10^5$	$4.9x10^2 - 2.2x10^6$	$3.0x10^5$
Station 3	$6.0x10^3 - 2.9x10^6$	$5.0x10^5$	$6.2x10^2 - 3.0x10^6$	$4.9x10^5$
Station 4	$1.7x10^3 - 2.6x10^5$	$6.9x10^4$	$1.2x10^3 - 2.1x10^6$	$2.3x10^5$
Station 5	$3.4x10^2 - 2.0x10^5$	$6.3x10^4$	$8.5x10^2 - 1.9x10^6$	$2.7x10^5$
Max permissible limit (CFU/mL) Buras *et al*. (1987)		**$1.0x10^5$**		**$1.0x10^5$**

4.4 Mean Seasonal Viable Bacterial Count (VBC) of Water from Kainji and Jebba Lakes.

Table 4.4 shows the viable bacterial count (VBC) of water samples from Kainji and Jebba Lakes based on season. It was observed that viable bacterial count of Kainji Lake during the wet season was higher (5.5×10^5 CFU/mL) than the dry season (6.2×10^4 CFU/ mL) and that of minimum count for Jebba wet season was higher (1.2×10^3 CFU/ mL) than the dry season (4.2×10^2 CFU/mL). Generally the VBC of water samples from Kainji and Jebba were observed to be higher than maximum permissible limit of 1.0×10^5 CFU/mL (Buras *et al.*, 1987), except the count during dry season in Kainji Lake, which was $6.2x10^4$ CFU/mL. T test result shows that there was significant difference ($P < 0.05$) between the two seasons in both Lakes (Appendices C and D).

Table 4.4: Mean Seasonal Viable Bacterial Count (VBC) of Water from Kainji and Jebba Lakes

Parameter	Kainji		Jebba	
	Wet	**Dry**	**Wet**	**Dry**
Minimum	$3.2x10^{3a}$	$3.4x10^{2b}$	$1.2x10^{3\ a}$	$4.2x10^{2\ b}$
Maximum	$2.9x10^{6\ a}$	$2.6x10^{5\ b}$	$3.0x10^{6\ a}$	$3.0x10^{6\ a}$
Mean	$5.5x10^{5\ a}$	$6.2x10^{4\ b}$	$5.5x10^{5\ a}$	$5.5x10^{5\ a}$
Maximum permissible limit (cfu/mL) (Buras *et al.*, 1987)	**$1.0x10^5$**	**$1.0x10^5$**	**$1.0x10^5$**	**$1.0x10^5$**

Means with dissimilar letter (s) differ significantly according to student t test.**Significant at $p \leq 0.05$**

4.5 Bacterial Species Isolated from Water Samples in Kainji Lake

Biochemical characterization of the bacterial isolates from Kainji Lake water revealed the presence of bacterial species shown in Tables 4.5. These bacterial species could have emanated from both human and other animal sources, including runoffs from animal and municipal wastes. A study carried out by Abdelhamid *et al*. (2006) showed that different kinds of livestock manure were contaminated with pathogenic bacteria such as *Pseudomonas, Vibrio, E. coli, Shigella* and *Salmonella*.

Table 4.5: Bacterial Species Isolated from Water Samples in Kainji Lake

Sample Station	Bacterial isolates
KL.1	*Aeromonas caviae, Aeromonas Veronii bio sobria, Aeromonas aquatilis* strain AE 207*, Pseudomonas fluorescens – 35, Burkholderia pseudomallei, Vibrio alginolyticus, Mannheimia (pasturella) haemolytica, Escherichia coli, Alcaligenes faecalis* strain Sihong_663_1, *Actinobacillus* species
KL.2	*Escherichia hermannii, Aeromonas aquatilis* strain AE 207*, Pseudomonas donghuensis* HYS*, Burkholderia pseudomallei, Bacillus lacus* AK 74*, Vibrio alginolyticus, Escherichia coli*
KL .3	*Aeromonas aquatilis* strain AE 207*, Alcaligenes faecalis* strain Sihong_663_1, *Escherichia hermannii, Pseudomonas donghuensis* HYS*, Escherichia coli, Actinobacillus* species
KL. 4	*Vibrio alginolyticus, Herbaspirillum aquaticum* strain IEH 4430*, Aeromonas caviae, Pseudomonas donghuensis* HYS
KL. 5	*Oceanobacillus oncorhynchi* subsp. *incaldanensis* strain AM-75*, Actinobacillus* species*, Aeromonas aquatilis* strain AE 207*, Pseudomonas fluorescens – 35*

Key: KL = Kainji Lake

4.6 Bacterial Species Isolated from Kainji Lake Sediment Samples

Table 4.6 present different bacterial species isolated from Kainji Lake sediment samples and identified using Microbact kits. This might be from decomposed animal sources from the aquatic environment or it might be from suspended species from the water samples. Drummond *et al*. (2014) stated that "a large proportion of the pathogenic organisms present in water might also become associated with the sediment, which could be subject to resuspension".

Table 4.6: Bacterial Species Isolated from Kainji Lake Sediment Samples

Sample Station	Bacterial isolates
KL.1	*Bacillus sp., Vibrio alginolyticus, Actinobacillus* species, *Pseudomonas fluorescens - 25,*
KL.2	*Vibrio alginolyticus, Actinobacillus* species, *Pseudomonas aeruginosa*
KL. 3	*Actinobacillus* species, *Pseudomonas aeruginosa*
KL. 4	*Vibrio alginolyticus, Bacillus sp., Pseudomonas fluorescens - 25,*
KL .5	*Vibrio alginolyticus, Pseudomonas aeruginosa*

Key: KL = Kainji Lake

4.7 Bacterial Species Isolated from Fish Samples in Kainji Lake

Following biochemical characterization carried out, the following bacterial species were identified: *Aeromonas hydrophila*, *Escherichia coli* and *Vibrio alginolyticus* (Table 4.7). These might be from animals that drink from and defeacate into the Lake and / or from humans through open defeacation. The presence of coliform bacteria (*Escherichia coli*) and other human pathogens like *Aeromonas hydrophila, Vibrio alginolyticus* and *Pseudomonas* agrees with Emikpe *et al.* (2011), who reported that pathogenic bacteria particularly the coliforms, are harbored by fishes in both fresh and brackish waters.

Table 4.7: Bacterial Species Isolated from Fish Samples in Kainji Lake

Organ	Bacterial isolates
ONFI	*Vibrio alginolyticus, Escherichia coli*
ONFG	*Aeromonas hydrophila, Pseudomonas fluorescens – 25, Burkholderia pseudomallei, Acinetobacter haemolyticus, Moraxella* species, *Cepacia meningocepticum, Vibrio alginolyticus, Bacillus* sp.
ONFM	ND
CGFI	*Aeromonas hydrophila, Escherichia* coli, *Vibrio alginolyticus*
CGFG	*Cepacia meningocepticum, Aeromonas hydrophila, Pseudomonas fluorescens – 25, Moraxella* species, *Bacillus* sp.
CGFM	ND

Key: ONFI = *Oreochromis niloticus* fish intestines, ONFG = *Oreochromis niloticus* fish gills, ONFM = *Oreochromis niloticus* fish muscles, CGFI = *Clarias gareipinus* fish intestines, CGFG = *Clarias gareipinus* fish gills, CGFM = *Clarias gareipinus* fish muscles, ND = Not detected.

4.8 Bacterial Species Isolated from Water Samples in Jebba Lake

Biochemical characterization of bacterial isolates in water samples revealed the presence of different bacterial species (Table 4.8). The sources of these bacterial species might be from animal husbandry operation in which the cattle roam about looking for greener pasture and, as such, defecate any where and into the Lake. This agrees with a study reported by Abdelhamid *et al*. (2006) where different kinds of livestock manure were contaminated with pathogenic bacteria such as *Pseudomonas, Vibrio, E. coli, Shigella* and *Salmonella*, among others.

Table 4.8: Bacterial Species Isolated from Jebba Lake Water Samples

Sample Station	Bacterial isolates
JL.1	*Aeromonas caviae, Aeromonas aquatilis* strain AE 207*, Pseudomonas donghuensis* HYS*, Escherichia coli, Alcaligenes faecalis* strain HPRTAK198, *Bacillus lacus* AK 74*, Mannheimia (pasturella) haemolytica, Pseudomonas aeruginosa, Pseudomonas fluorescens – 35, Escherichia hermannii.*
JL.2	*Aeromonas aquatilis* strain AE 207*, Pseudomonas donghuensis* HYS*, Burkholderia pseudomallei, Mannheimia (pasturella) haemolytica, Bacillus lacus* AK 74, *Pseudomonas aeruginosa, Escherichia hermannii, Escherichia coli, Alcaligenes faecalis* strain HPRTAK198*, Herbaspirillum aquaticum* strain IEH 4430, *Actinobacillus* species.
JL .3	*Pseudomonas aeruginosa, Aeromonas aquatilis* strain AE 207*, Pseudomonas donghuensis* HYS*, Yersinia pestis, Escherichia coli, Actinobacillus* species.
JL. 4	*Aeromonas caviae, Pseudomonas donghuensis* HYS*, Mannheimia (pasturella) haemolytica, Aeromonas aquatilis* strain AE 207*, Burkholderia pseudomallei, Pseudomonas aeruginosa, Escherichia coli, Alcaligenes faecalis* strain HPRTAK198.
JL. 5	*Aeromonas aquatilis* strain AE 207*, Pseudomonas donghuensis* HYS*, Mannheimia (pasturella) haemolytica, Escherichia hermannii, Oceanobacillus oncorhynchi subsp. incaldanensis* strain AM-75*, Actinobacillus* species.

Key: JL = Jebba Lake

4.9 Bacterial Species Isolated from Jebba Lake Sediment Samples

Biochemical characterization of bacterial isolates from sediment in Jebba Lake shows the presence of *Escherichia coli*, *Aeromonas hydrophila*, *Vibrio alginolyticus*, *Pseudomonas*

aeruginosa (Table 4.9). These might be as a result of effluents from domestic wastes, decomposed animal sources from the aquatic environment or from suspended species from the water samples. Previous researches had defined coastal or estuarine sediments as sink of faecally derived bacteria (Perkins *et al*., 2014).

Table 4.9: Bacterial Species Isolated from Jebba Lake Sediment Samples

Sample Station	Bacterial isolates
JL.1	*Aeromonas hydrophila, Vibrio alginolyticus*
JL.2	*Pseudomonas aeruginosa, Aeromonas hydrophila, Pseudomonas fluorescens - 25, Vibrio alginolyticus, Escherichia coli*
JL. 3	*Vibrio alginolyticus, Actinobacillus* species
JL. 4	*Aeromonas hydrophila, Vibrio alginolyticus, Pseudomonas aeruginosa, Pseudomonas fluorescens - 25, Actinobacillus* species
JL. 5	*Vibrio alginolyticus, Pseudomonas aeruginosa, Actinobacillus* species

Key: Jl = Jebba Lake

4.10 Bacterial Species Isolated from Fish Samples in Jebba Lake

Table 4.10 revealed bacterial species isolated from fish samples in Jebba Lake. These microbial species associated with different organs of the fish may not be unconnected with the different human activities being carried out in and around the Lake. The presence of *Escherichia* coli is an indication of recent pollution. Adewoye and Adegunlola (2010)

found similar bacteria in their studies on *Clarias gariepinus*. However in this study, no bacterium was found in the fish muscles.

Table 4.10: Bacterial Species Isolated from Fish Samples in Jebba Lake

Organ	Bacterial isolates
ONFI	*Vibrio alginolyticus, Moraxella* species, *Escherichia coli, Escherichia hermannii, Proteus Vulgaris*.
ONFG	*Cepacia meningocepticum, Moraxella* species, *Pasturella multocida, Pseudomonas fluorescens – 25, Escherichia hermannii, Escherichia coli*.
ONFM	ND
CGFI	*Vibrio alginolyticus, Escherichia* coli, *Aeromonas hydrophila, Vibrio parahaemolyticus, Proteus Vulgaris*.
CGFG	*Vibrio parahaemolyticus, Aeromonas hydrophila, Bacillus sp., Proteus Vulgaris, Pseudomonas fluorescens – 25, Escherichia coli*.
CGFM	ND

Key: ONFI = *Oreochromis niloticus* fish intestines, ONFG = *Oreochromis niloticus* fish gills, ONFM = *Oreochromis niloticus* fish muscles, CGFI = *Clarias gareipinus* fish intestines, CGFG = *Clarias gareipinus* fish gills, CGFM = *Clarias gareipinus* fish muscles, ND = Not detected

4.11 Molecular Characterization of Some Bacterial Isolates from Kainji and Jebba Lakes Water

The amplicon base pair size of the bacterial isolates was 1500 bp as revealed by electrophoresis (Plate VIII and IX). The results of the sequence (Appendices E – N) and the BLAST were compared with the reference strains on the website of the National Centre for Biotechnology Information (NCBI). Details are indicated in Table 4.11.

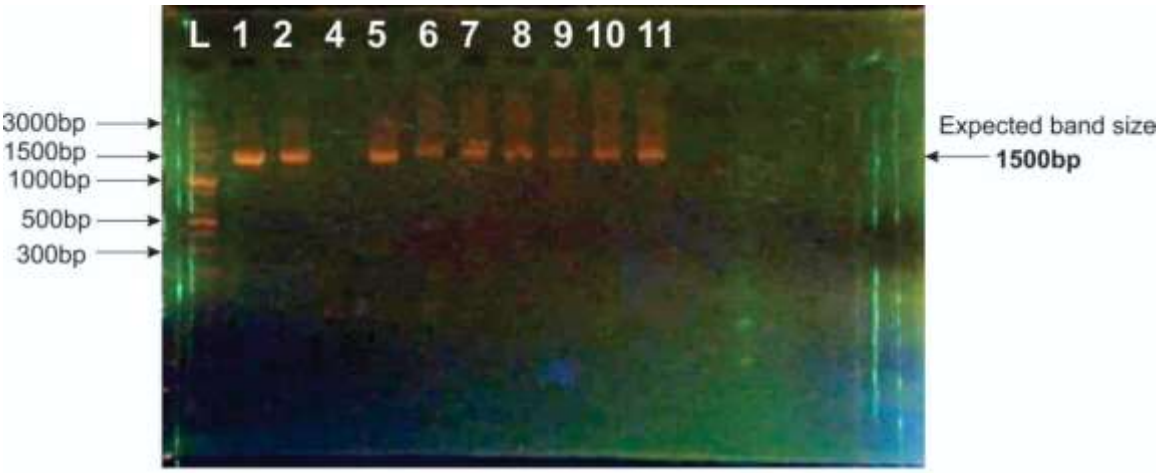

Plate VIII: Agarose gel electrophoresis of 16S rRNA gene of the isolated bacteria

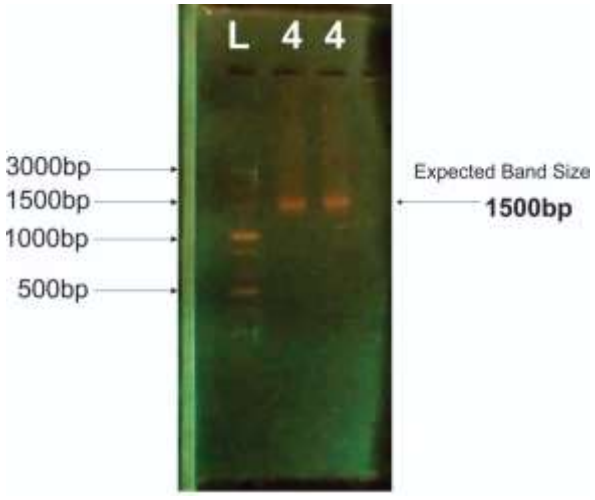

Plate IX: Agarose gel electrophoresis of 16S rRNA gene of isolate No. 4

Table 4.11: BLAST Pairwise Alignment of Ten Amplicon Sequenced

Sample	Genotype	Length	Score	E-Value	Identity (%)	Reference Strains
1	*Aeromonas aquatilis* strain AE207	1629	2021	0	96.29	LT630766.1
2	*Bacillus lacus* strain AK74	1569	1655	0	92.28	NR_159904.1
4	*Aeromonas aquatilis* strain AE207	1334	815	0	93.48	LT630765.1
5	*Bacillus lacus* strain AK74	1547	1413	0	91.43	NR_159904.1
6	*Pseudomonas donghuensis* strain HYS	1196	726	0	91.53	NR_136501.2
7	*Bacillus lacus* strain AK74	1730	1578	0	91.78	NR_159904.1
8	*Herbaspirillum aquaticum* strain *IEH* 4430	1748	1094	0	88.09	NR_116605.1
9	*Alcaligenes faecalis* strain Sihong_663_1	1873	1459	0	94.89	MN309905.1
10	*Alcaligenes faecalis* strain HPRTAK 198	1748	1546	0	96.4	MH392489.1
11	*Oceanobacillus oncorhynchi* subsp. *incaldanensis* strain AM-75	1236	1546	0	91.14	KF817687.1

4.12 Concentrations of Heavy Metals in Kainji Lake Water (January – December, 2017)

Figure 4.1 shows the concentrations of heavy metals in Kainji Lake water (January – December, 2017). The concentration of lead (Pb) was observed to be within the maximum permissible limit of 0.05 mg/l according to FEPA (2003), except in the months of April (0.07 mg/l) and September (0.29 mg/l). Concentration of copper (Cu) was observed to be within permissible limit of 2.0 mg/l according to WHO (1993). Chromium (Cr) was observed to be within permissible limit of 0.05 mg/l except in the months of March, April and June. This might be due to surface runoff from onset of rainfall, thereby washing wastes from adjacent lands into the Lake. The concentration of arsenic (As) was also observed to be within acceptable limit of 0.10 mg/l according to FEPA (2003), except in the months of July, August and September (with the values of 0.15 mg/l, 1.16 mg/l and 0.18 mg/l respectively). The increase in arsenic concentration within the months of July, August and September might be due to increased rainfall within these months and this might have led to increased runoffs. Concentration of cadmium (Cd) in Kainji Lake water was higher than the maximum permissible limit of 0.01 mg/l according to FEPA (2003), except in the months of January and November. However there was no significant difference ($P > 0.05$) in the concentration of Cd except in the months of July, August and September (with the values of 0.29 mg/l, 0.12 mg/l and 0.41 mg/l respectively) (Appendix E). Increased concentration of cadmium above the maximum permissible limit might be due to the application of inorganic fertilizers to the neighboring farm lands, which might easily be washed down into the Lake. This result tends to agree with Agency for Toxic Substance and Disease Registry (2012), which states that natural as well as anthropogenic sources of

cadmium (which include industrial effluents, fertilizers and sewage sludge to farmland) could increase environmental level of cadmium.

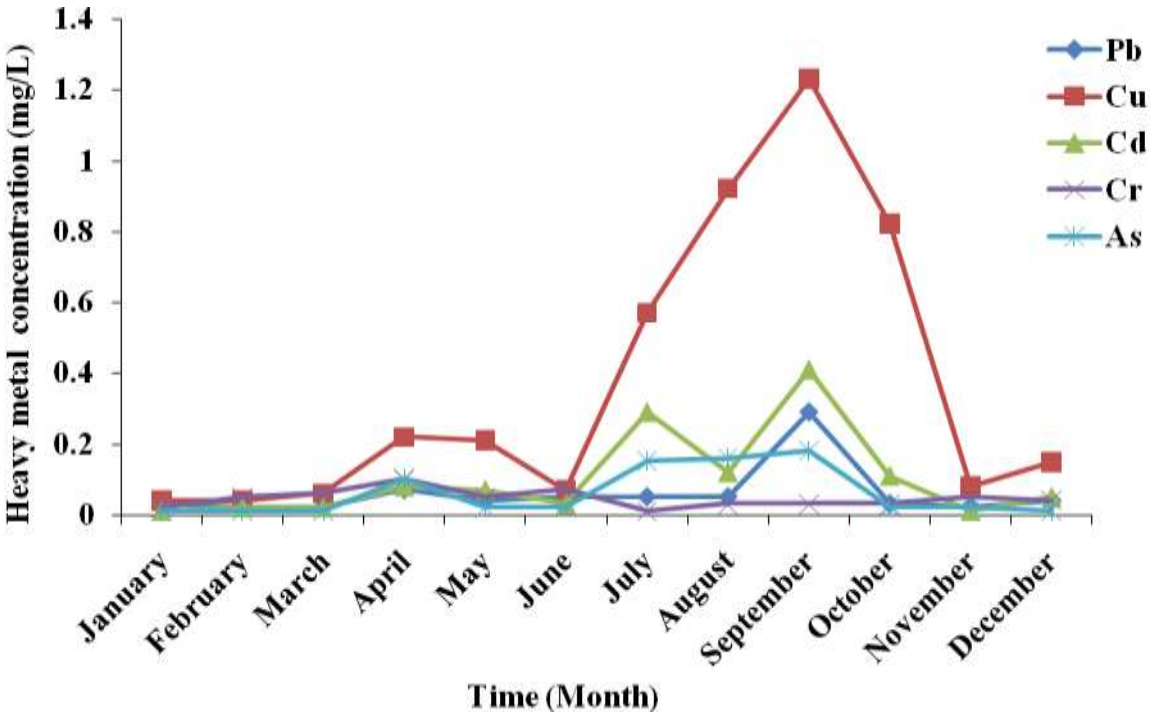

Figure 4.1: Concentrations of heavy metals in Kainji Lake water (January-December, 2017)

4.13 Heavy Metal Concentrations in Kainji Lake Water based on Season

Table 4.12 shows the seasonal heavy metal concentrations in Kainji Lake water. The results showed that the concentrations of heavy metals during the wet season were higher when compared to those of the dry season. This might be as a result of surface runoffs from adjacent mining areas into the Lake during the wet season. This result agrees with the findings of Nsofor and Ekpeze (2014), who reported that during the wet season, concentration of heavy metals in water is always higher than during the dry season. There was significant difference ($P < 0.05$) between values in the dry and wet seasons.

Table 4.12. Heavy Metal Concentrations in Kainji Lake Water based on Season

Season	**Heavy metal concentration (mg/L)**				
	Pb	**Cu**	**Cd**	**Cr**	**As**
Wet	0.09 ± 0.00^{a}	0.54 ± 0.00^{a}	0.17 ± 0.00^{a}	0.05 ± 0.00^{a}	0.10 ± 0.00^{a}
Dry	0.02 ± 0.00^{b}	0.20 ± 0.00^{b}	0.04 ± 0.00^{b}	0.04 ± 0.00^{b}	0.01 ± 0.00^{b}

Values are means of five determinations. Means with dissimilar letter (s) differ significantly according to student t test. **Significant at $P \leq 0.05$**

4.14 Heavy Metal Concentrations of Water from Kainji Sampling Station

The concentration of heavy metals from different sample stations of Kainji Lake shows that sample station 2 (Kl.2) had higher concentration of Pb (0.082 mg/l), Cu (0.464 mg/l), Cd (0.187 mg/l) and As (0.085 mg/l) than other sample stations. Sample station 3 (Kl.3) had higher concentration of Cr (0.061 mg/l) than other stations (Figure 4.2). Higher concentration of heavy metals in station 2 might be due to high mining activities (including excarvation and washings of pulp) in Garafini area (station 2) than other stations. This activity goes along with farming activities where inorganic fertilizers and herbicides are being used in farm lands. This result agrees with Salaudeen *et al*. (2016), who reported that mining areas are the chief areas where heavy metal pollution readily occurs, in which the surrounding environment receives it first.

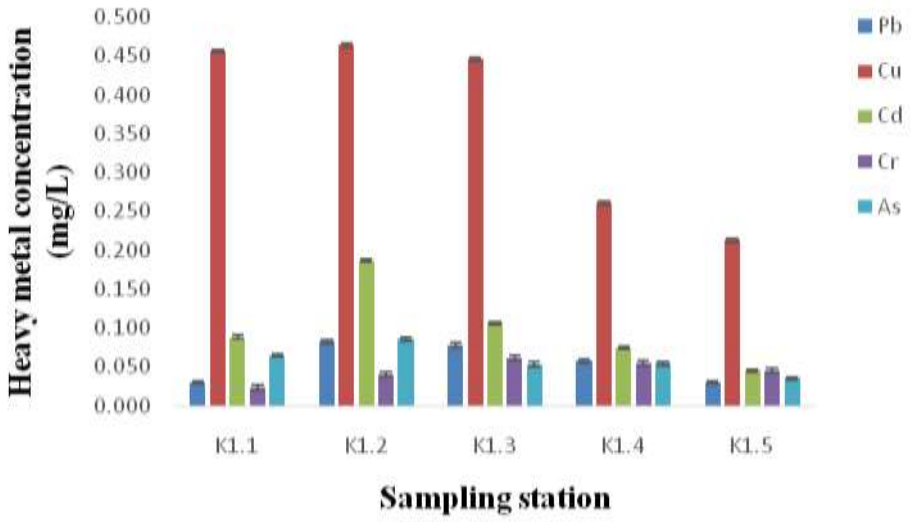

Figure 4.2: Heavy metal concentrations of water from Kainji sampling station

4.15 Heavy Metal Concentrations in Water from Jebba Lake (January – December, 2017)

Results showed that the concentration of lead was within acceptable limit of 0.05 mg/l (FEPA, 2003) except in the months of April and September. Copper was within acceptable limit of 2.0 mg/l except in the month of September, which was 2.350 mg/l. Cadmium was generally higher than acceptable limit of 0.01 mg/l (FEPA, 2003), with its highest concentration in the months of August and September. The concentrations of chromium in the months of January, March and December were above acceptable limit of 0.05 mg/l while arsenic was within acceptable limit of 0.10 mg/l (FEPA, 2003) except in the month of September, which was 0.190 mg/l. The month of April was the month when rainfall started and September was the month with the peak of flooding, hence, these must have caused the washing of effluents from domestic and other wastes from other human activities into the Lake. Therefore the afore-mentioned could have been responsible for the increase in heavy metal concentrations. High concentration of cadmium above maximum permissible limit of 0.01 mg/l might be due to high mining activities, effluents from power generation plant and inorganic fertilizers from farmlands along the course of the Lake. Result is shown in Figure 4.3. Agency for Toxic Substance and Disease Registry (ATSDR, 2012) state that natural as well as anthropogenic sources of cadmium, which include industrial effluents as well as fertilizers from farmlands, could increase environmental levels of cadmium. Tang *et al*. (2006) also stated that the release of cadmium into the environment could be through mining and smelting activities, incineration of plastics, land application of sewage sludge and burning of fossils.

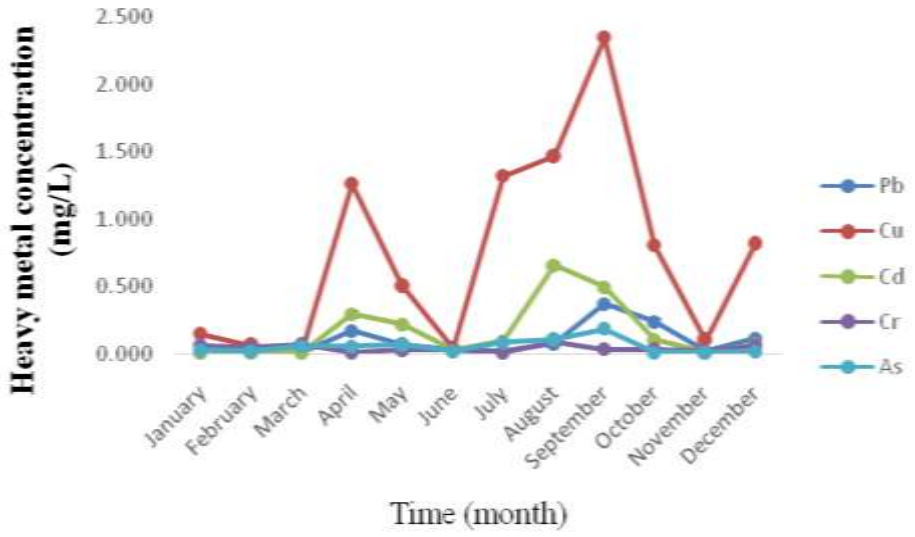

Figure 4.3: Heavy metal concentrations in water from Jebba Lake (January – December, 2017)

4.16 Heavy Metal Concentrations in Jebba Lake Water based on Season

Heavy metal concentrations in Jebba Lake water showed that the wet season had higher concentrations of lead, copper, cadmium and arsenic than the dry season, while reverse was the case for chromium, which had higher concentration during the dry season (0.06 mg/l) than the wet season (0.04 mg/l). There was significant difference ($P < 0.05$) in the concentrations of Pb, Cu, Cd and As between the two seasons (Table 4.13). Higher concentrations of lead, copper, cadmium, and arsenic metals during the wet season might be due to increased introduction of pollutants from adjacent farms through runoffs. The higher concentration of chromium in the Lake water during the dry season compared to the wet season might be that higher concentration of the chromium was being released from oils and lubricants that were used in the turbines and engines of the hydro electric plant. Hence, increased in rainfall during wet season increases the volume of water and would dilute and reduce the concentration while reduction in water volume during the dry season

would concentrate the element. Higher concentration of Pb, Cu, Cd and As in the wet season agrees with the findings of Peter *et al*. (2018), who revealed higher concentration of lead, copper, zinc, iron and manganese during wet season than the dry season. This might be due to flooding.

Table 4.13: Heavy Metal Concentrations in Jebba Lake Water based on Season

Season	**Heavy metal concentration (mg/L)**				
	Pb	**Cu**	**Cd**	**Cr**	**As**
Wet	0.21 ± 0.00^{a}	0.91 ± 0.00^{a}	0.22 ± 0.00^{a}	0.04 ± 0.00^{a}	0.12 ± 0.00^{a}
Dry	0.08 ± 0.00^{b}	0.21 ± 0.00^{b}	0.04 ± 0.00^{b}	0.06 ± 0.00^{a}	0.03 ± 0.00^{b}

Values are means of five determinations. Means with dissimilar letter (s) differ significantly according to student T test. **Significant at $P \leq 0.05$**

4.17 Heavy Metal Concentrations in Water from Jebba Lake Sample Stations

Figure 4.4 shows heavy metal concentrations in water from Jebba Lake sample stations. Sample station Jl.2 had the highest concentration of lead, copper and cadmium, (0.380 mg/l, 0.774 mg/l, 0.227 mg/l, respectively) while station Jl.3 had the highest concentration of chromium and arsenic (0.066 mg/l and 0.143 mg/l, respectively). Higher concentrations of lead, copper and cadmium in sample station Jl.2 might be due to the washings of gold and automobiles (including farming activities) being carried out around the station and these might have been easily washed down into the Lake. This result agrees with Salaudeen *et al*. (2016), who reported that mining areas are the chief areas where heavy metal pollution readily occurs, in which the surrounding environment receives it first.

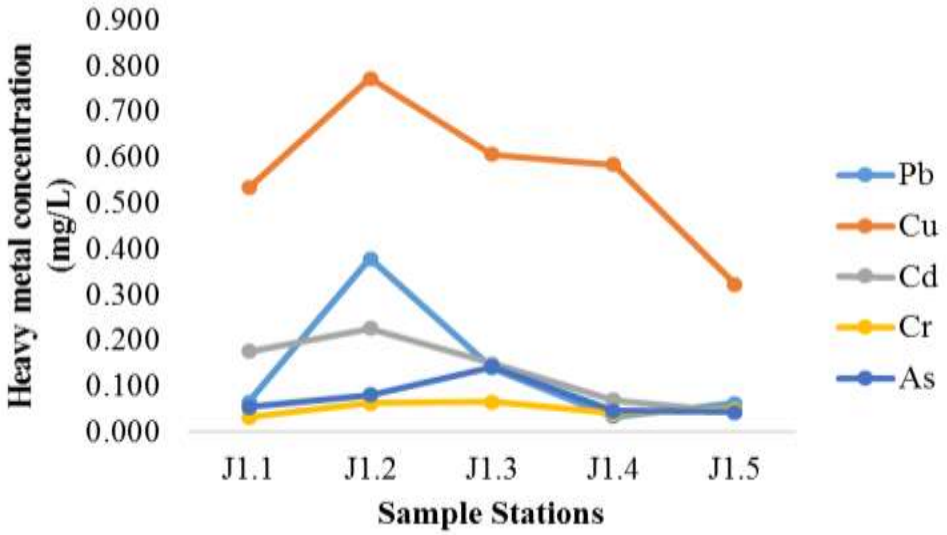

Figure 4.4: Heavy metal concentrations in water from Jebba sample stations

4.18 Heavy Metal Concentrations in Water from Kainji and Jebba Lakes

Table 4.14 shows the mean of heavy metal concentration in water from Kainji and Jebba Lakes. It was observed that the concentration of Pb, Cu and Cd from Jebba Lake was higher than that of Kainji Lake. This may be due to higher human activities on Jebba Lake of recent (including mining, washing of automobiles and domestic effluents). Chromium and Arsenic concentration was higher in Kainji Lake than Jebba Lake. This may be as a result of lubricant and oil as effluent from Kainji hydroelectric plant. These have their source from Kainji Lake and flows down stream to Jebba Lake. This result agrees with Salaudeen *et al*. (2016), who states that mining areas are the chief areas where heavy metal pollution readily occurs, in which the surrounding land receives the pollutants first and latter adjacent surface water bodies through runoff.

Table 4.14 Heavy Metal Concentrations in Water from Kainji and Jebba Lakes

	Heavy metal concentration (mg/L)				
Lake water	Pb	Cu	Cd	Cr	As
Kainji	0.08 ± 0.00^{b}	0.56 ± 0.00^{b}	0.09 ± 0.00^{b}	0.05 ± 0.00^{a}	0.07 ± 0.00^{a}
Jebba	0.10 ± 0.00^{a}	0.74 ± 0.00^{a}	0.17 ± 0.00^{a}	0.04 ± 0.00^{b}	0.06 ± 0.00^{b}

Values are means of five determinations. Means with dissimilar letter (s) differ significantly according to student T test. **Significant at $P \leq 0.05$**

4.19 Heavy Metal Concentration of Sediment from Kainji Lake

The results of heavy metal analysis of sediment from Kainji Lake (Table 4.15) showed that Pb, Cu, Cr and As were below threshold effect concentration (TEC) of Sediment Quality Guidelines Values (SQGV) by the National Oceanic and Atmospheric Administration (NOAA, 2009), which has standard TEC values of 35.8 mg/kg, 31.6 mg/kg, 0.99 mg/kg, 43.4 mg/kg respectively and 40.0 mg/kg by European Water Framework Directive for Suspended Matter and Sediment(EWFDSS, 2000). Cadmium recorded a higher concentration (1.062 mg/kg) above TEC value of 0.99 mg/kg (NOAA, 2009) in the month of september. One major source of cadmium in an environment is inorganic fertilizers and since the application of fertilizer to crops in the month of August and September is always very high and also September month being the peak of flooding, these fertilizers must have found their way into the Lake thereby increasing sharply their concentrations at sediment level. There was a significant difference ($P < 0.05$) between the concentrations of cadmium in the month of September and other months.

Table 4.15 Heavy Metal Concentrations of Sediment from Kainji Lake

Time	Heavy metal concentration (mg/kg)				
(Month)	Pb	Cu	Cd	Cr	As
January	0.276^{g}	1.409^{g}	0.100^{h}	0.448^{f}	0.050^{j}
February	0.242^{h}	1.766^{de}	0.244^{g}	0.406^{gf}	0.076^{hi}
March	0.294^{g}	1.718^{ef}	0.324^{f}	0.365^{g}	0.108^{g}
April	0.330^{f}	1.686^{ef}	0.776^{c}	2.068^{a}	0.128^{f}
May	0.722^{d}	2.110^{c}	0.880^{b}	0.864^{e}	0.068^{i}
June	0.634^{e}	2.290^{b}	0.526^{d}	1.379^{d}	0.036^{j}
July	0.740^{d}	2.843^{a}	0.872^{d}	1.690^{b}	0.258^{e}
August	0.943^{c}	2.262^{b}	0.350^{ef}	1.502^{c}	0.086^{h}
September	2.294^{ab}	1.803^{d}	1.062^{a}	0.112^{h}	1.186^{c}
October	2.324^{a}	1.653^{c}	0.576^{d}	0.120^{h}	1.257^{b}
November	2.326^{a}	1.476^{g}	0.408^{e}	1.371^{d}	1.284^{a}
December	2.290^{b}	1.714^{ef}	0.568^{d}	0.136^{h}	1.156^{d}
TEC (NOAA ,2009)	**35.8**	**31.6**	**0.99**	**43.4**	**40.0**

Values are means of five determinations. Means with dissimilar letter (s) differ significantly according to the Duncan's Multiple Range Test (DMRT).
Significant at $P \leq 0.05$

4.20 Heavy Metal Concentrations in Kainji Lake Sediment based on Season

Table 4.16 shows heavy metal concentrations in Kainji Lake sediment based on season. It was observed that the concentrations of Cu, Cd and Cr in Kainji Lake sediment during the wet season were higher (2.17 mg/kg, 0.73 mg/kg and 1.25 mg/kg respectively) than those of dry season (1.63 mg/kg, 0.40 mg/kg and 0.48 mg/kg respectively). This might be due to increase in runoffs from adjacent farms and mining areas. The concentration of Pb and As, however, tends to have higher values in the dry season (1.29 mg/kg and 0.65 mg/kg

respectively) than wet season (0.87 mg/kg and 0.29 mg/kg respectively). There was a significant difference ($P < 0.05$) between the two seasons. This increase in concentration during dry season might be due to direct washings by miners into the Lake. It was generally observed that concentrations of heavy metals analyzed (Pb, Cu, Cd, Cr and As) were found to be below threshold effect concentration according to NOAA (2009). The concentrations of heavy metals in sediment from Kainji Lake was lower when compared to those of Asvar Dam Lake (Ozturk *et al*., 2009), Lake Geneva (Pote *et al*., 2008) and Lake Victoria (Kishe and Machiwa, 2003).

Table 4.16: Heavy Metal Concentrations in Kainji Lake Sediment based on Season

	Heavy metal concentration (mg/kg)				
Season	**Pb**	**Cu**	**Cd**	**Cr**	**As**
Wet	0.87 ± 0.00^{b}	2.17 ± 0.00^{a}	0.73 ± 0.00^{a}	1.25 ± 0.00^{a}	0.29 ± 0.00^{b}
Dry	1.29 ± 0.00^{a}	1.63 ± 0.00^{b}	0.40 ± 0.00^{b}	0.48 ± 0.00^{b}	0.65 ± 0.00^{a}

Values are means of five determinations. Means with dissimilar letter (s) differ significantly according to student T test. **Significant at $P \leq 0.05$**

4.21 Heavy Metal Concentrations in Sediment of Kainji Lake Sample Stations

The heavy metal concentrations in sediment from different sample stations of Kainji Lake (Figure 4.5) revealed that all the heavy metals analyzed were within acceptable limit (threshold effect concentration limit) of sediment quality guidelines value (EWFDSS, 2000; NOAA, 2009). Station 3 (KL.3) had higher concentration of Cu (2.327 mg/kg) than other stations, while in station 4 (Kl.4), Pb, Cr and As had higher concentrations than other stations.

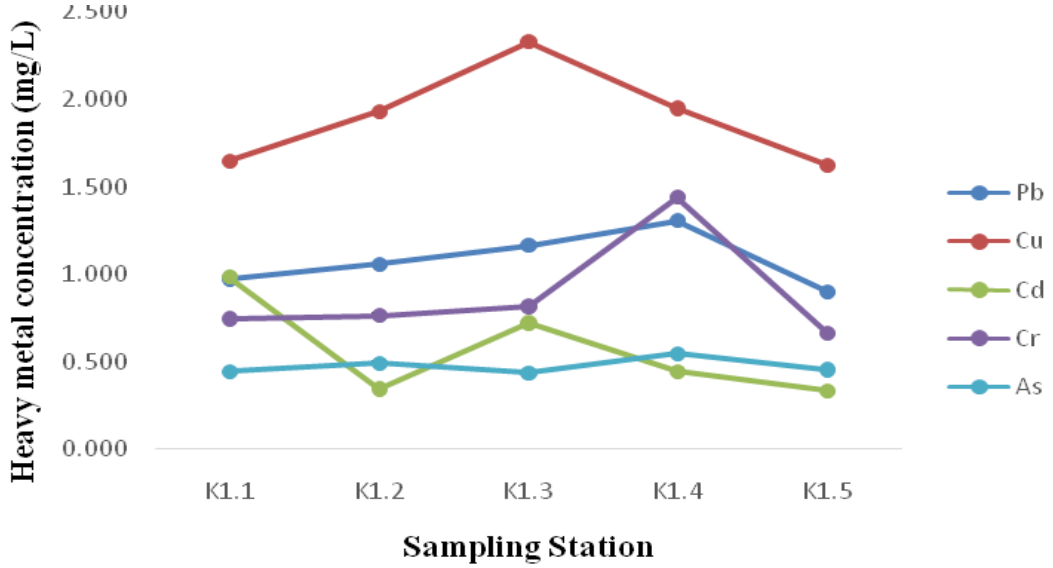

Figure 4.5: Heavy metal concentration in sediments of Kainji Lake sampling station

4.22 Concentrations of Heavy Metals in Jebba Lake Sediments based on Months

Table 4.17 shows the results of concentrations of heavy metals in Jebba Lake sediments based on months. It was observed that the concentration of all heavy metals (Pb, Cu, Cd, Cr and As) analyzed were within acceptable limit of sediment quality guidelines value by NOAA (2009). Concentration of lead and arsenic tended to increase sharply in the months of September to December. However, the increase was not above the threshold effect concentration. This sharp increase of Pb and As in these months (September, October, November and December) might be due to runoffs from domestic wastes and farm lands as a result of increased rainfall in the month of September. Concentration of Pb and Cu in this study was far less compared to the study of Jacob *et al*. (2009) on lead, copper and zinc accumulation in soils of urban farms in Minna.

Table 4.17: Concentrations of Heavy Metals in Jebba Lake Sediments based on Months

	Heavy metal concentration (mg/kg)				
Month	Pb	Cu	Cd	Cr	As
January	0.066^{h}	0.816^{i}	0.414^{d}	0.748^{d}	0.030^{e}
February	0.196^{g}	1.292^{g}	0.458^{d}	0.168^{g}	0.050^{de}
March	0.304^{f}	1.666^{e}	0.580^{c}	0.396^{f}	0.088^{d}
April	0.408^{e}	1.466^{f}	0.816^{a}	0.970^{b}	0.092^{d}
May	0.460^{de}	1.836^{d}	0.668^{bc}	0.582^{e}	0.056^{de}
June	0.268^{fg}	2.092^{c}	0.784^{ab}	0.914^{bc}	0.052^{de}
July	0.278^{fg}	2.870^{a}	0.422^{d}	0.876^{c}	0.048^{de}
August	0.506^{d}	2.456^{b}	0.664^{bc}	0.872^{c}	0.056^{de}
September	1.780^{c}	1.072^{h}	0.700^{bc}	0.152^{g}	1.128^{b}
October	2.268^{b}	1.376^{g}	0.650^{c}	0.172^{g}	0.948^{c}
November	2.542^{a}	1.058^{h}	0.380^{d}	1.198^{a}	1.270^{a}
December	2.274^{b}	1.068^{h}	0.470^{e}	0.176^{g}	1.174^{b}
TEC(NOAA, 2009)	**35.8**	**31.6**	**0.99**	**43.4**	**40.0**

Values are means of five determinations. Means with dissimilar letter (s) differ significantly according to the Duncan's Multiple Range Test (DMRT).
Significant at $P \leq 0.05$

4.23 Mean Seasonal Heavy Metal Concentrations in Jebba Lake Sediment

In Jebba Lake, the mean seasonal concentrations of Cu, Cd and Cr (Table 4.18) during the wet season were higher (1.97 mg/kg, 0.68 mg/kg and 0.73 mg/kg respectively) than the concentrations during the dry season (1.21 mg/kg, 0.49 mg/kg and 0.48 mg/kg respectively). However, the concentrations of Pb and As were higher during the dry season than those of the wet season. This might be due to increase in direct washings of the ore pulp into the Lake by miners. Significant difference ($P < 0.05$) was observed between the wet and dry seasons.

Table 4.18: Mean Seasonal Heavy Metal Concentrations in Jebba Lake Sediment

	Heavy metal concentration (mg/kg)				
Season	**Pb**	**Cu**	**Cd**	**Cr**	**As**
Wet	0.62 ± 0.00^{b}	1.97 ± 0.00^{a}	0.68 ± 0.00^{a}	0.73 ± 0.00^{a}	0.24 ± 0.00^{b}
Dry	1.28 ± 0.00^{a}	1.21 ± 0.00^{b}	0.49 ± 0.00^{b}	0.48 ± 0.00^{b}	0.59 ± 0.00^{a}

Values are means of five determinations. Means with dissimilar letter (s) differ significantly according to student T test. **Significant at $P \leq 0.05$**

4.24 Concentrations of Heavy Metals in Sediments of Jebba Lake Sample Stations

Figure 4.6 shows the concentrations of heavy metals in sediments of Jebba Lake at different sample stations. It was observed that the heavy metal concentrations in sediments from Jebba Lake sample station were below threshold effect concentration (TEC) of NOAA (2009). Sample station 2 (Jl.2) had higher concentrations of Pb, Cu and Cr than other sample stations (but values were still below TEC). The higher concentrations of Pb and Cu in sediment sample from station 2 were also reflected in their concentrations in water samples from the same station. This higher concentration might be due to increased washings of the ore pulp by miners in the station, washing of automobiles as well as runoffs from farm lands around the station.

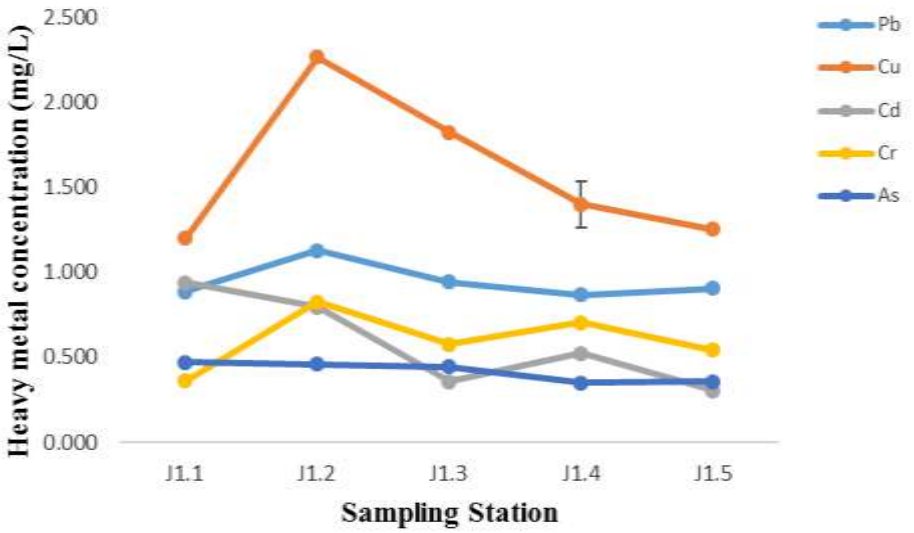

Figure 4.6: Heavy metals in sediments of Jebba Lake sampling station

4.25 Heavy Metal Concentrations in Sediments from Kainji and Jebba Lakes.

The concentrations of heavy metals in sediments from Kainji and Jebba Lakes revealed that Pb, Cu, Cd, Cr and As had higher concentrations in sediment from Kainji compared to sediment from Jebba Lake (Table 4.19). This might be as a result of remote and long standing mining activities and hydroelectric power generation plant in Kainji Lake, thereby causing heavy metals to settle and associate more with sediment from the Kainji Lake.

Table 4.19: Heavy Metal Concentrations in Sediments from Kainji and Jebba Lakes

	Heavy metal concentration (mg/kg)				
Lake Sediment	Pb	Cu	Cd	Cr	As
Kainji	1.12 ± 0.00^{a}	1.89 ± 0.00^{a}	0.56 ± 0.00^{a}	0.87 ± 0.00^{a}	0.47 ± 0.00^{a}
Jebba	0.09 ± 0.00^{b}	1.59 ± 0.00^{b}	0.55 ± 0.00^{b}	0.61 ± 0.00^{b}	0.42 ± 0.00^{b}

Values are means of five determinations. Means with dissimilar letter (s) differ significantly according to student T test. **Significant at $P \leq 0.05$**

4.26 Concentrations of Heavy Metals in *Oreochromis niloticus* Fish Intestines (ONFI) from Kainji Lake Sample Stations

The concentrations of Cu, Cd and Cr (Figure 4.7) in *Oreochromis niloticus* fish intestines (ONFI) from Kainji Lake water in all the sampled stations were below maximum permissible limit of 3.0 mg/kg, 0.5mg/kg and 0.15 mg/kg respectively according to FEPA (2003) and WHO (2003). However, the concentrations of Pb in all the sampled stations were above maximum permissible limit of 0.3 mg/kg according to FAO/WHO (2011). The concentrations of As in all the sampled stations were also above maximum permissible limit of 0.02 mg/kg. This poses potential danger to the fish and the consumer if not properly handled. Presence of Pb and As in an aquatic environment could be associated with mining activities, combusion of leaded oil, among others. The long-standing activities of illegal miners in Kainji Lake could have been responsible for the high concentrations of Pb and As in the intestines of the fish above tolerable limit. This result agrees with the findings of Ekwu (2010), who reported high concentrations of Pb (2.84 µg/g and 5.1 µg/g) in the intestines of *Tilapia* and *Clarias gariepinus* respectively in a fish farm in south-eastern Nigeria. Ozturk *et al*. (2009) also reported higher concentrations of lead, copper and chromium above tolerable values in the intestines of *Cyprinus carpio* when compared with the gills and muscles of the same fish. These, they attributed to geological mining history of the localities and urban as well as domestic activities.

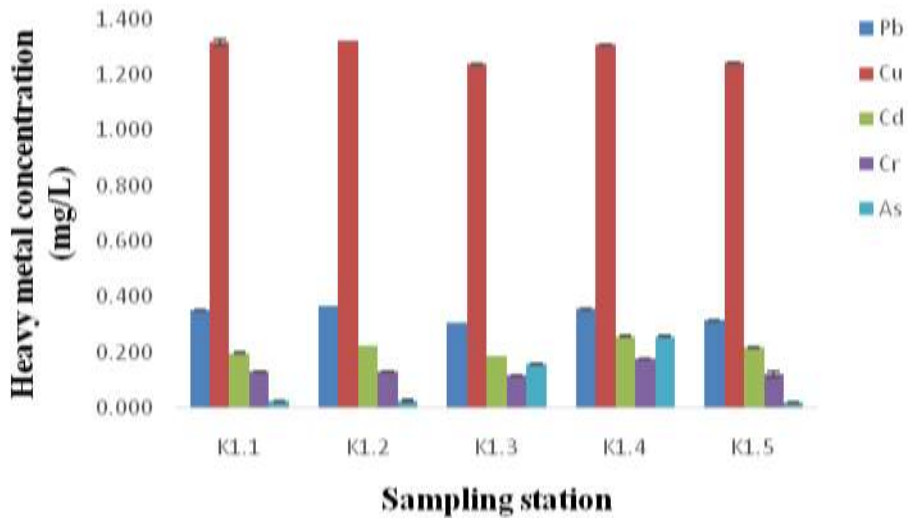

Figure 4.7: Heavy metal concentrations in ONFI of Kainji sampling station

4.27 Heavy Metal Concentrations in *Oreochromis niloticus* Fish Intestines (ONFI) of Kainji Lake based on Season

The concentrations of Pb and Cr were higher (0.35 mg/kg, 0.14 mg/kg respectively) during the wet season compared to the dry season (0.33 mg/kg, 0.13 mg/kg respectively). There was higher concentration of copper (Cu) and cadmium (Cd) during the dry season than the wet season. High concentration of Pb and Cr during the wet season might be attributed to flood while high concentration of Cu and Cd during the dry season might be due to direct washings or point source of pollution. There was significant difference ($P < 0.05$) between wet and dry season (Table 4.20).

Table 4.20: Mean Seasonal Heavy Metal Concentrations in *Oreochromis niloticus* Fish Intestines (ONFI) from Kainji Lake

	Heavy metal concentration (mg/kg)				
Season	**Pb**	**Cu**	**Cd**	**Cr**	**As**
Wet	0.35 ± 0.00^{a}	1.26 ± 0.00^{b}	0.19 ± 0.00^{b}	0.14 ± 0.00^{a}	0.02 ± 0.00^{a}
Dry	0.33 ± 0.00^{b}	1.31 ± 0.00^{a}	0.24 ± 0.00^{a}	0.13 ± 0.00^{b}	0.02 ± 0.00^{a}

Values are means of five determinations. Means with dissimilar letter (s) differ significantly according to student T test. **Significant at $P \leq 0.05$**

4.28 Heavy Metal Concentrations in *Oreochromis niloticus* Fish Gills (ONFG) of Kainji Sampling Station

The concentrations of Cu and Cd in the gills (Figure 4.8) were found to be lower than maximum permissible limit of 3.0 mg/kg and 0.5 mg/kg respectively (WHO 2003; FEPA 2003). Chromium concentration was lower than its maximum permissible limit except in sample station Kl.4. Concentration of As in all sample stations were higher than the maximum permissible limit of 0.02 mg/kg (Appendix T). Lead in sample station Kl.2 was above maximum permissible limit of 0.30 mg/kg (FAO/WHO, 2011). These high concentrations of arsenic and lead might be due to long-time status of mining activities around the Kainji Lake region and its proximity to sample station Kl.2 which is characterized by high mining activities and agricultural farms.

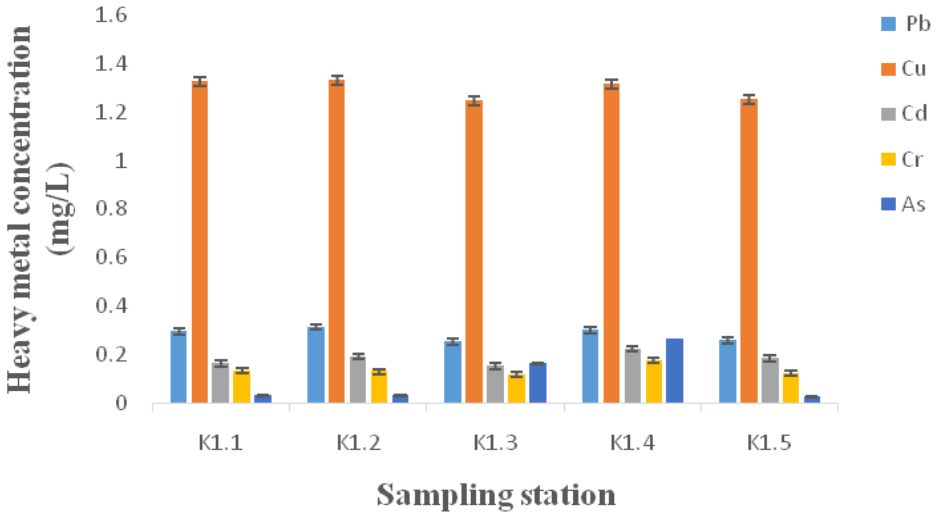

Figure 4.8: Heavy metal concentrations in *Oreochromis niloticus* fish gills (ONFG) of Kainji sampling station

4.29 Heavy Metal Concentrations in *Oreochromis niloticus* Fish Gills (ONFG) from Kainji Lake based on Season

Seasonal concentrations of heavy metals in ONFG showed that lead and copper (Table 4.21) were higher (0.18 mg/kg, 0.61 mg/kg respectively) during the wet season, compared to their concentrations during the dry season (0.12 mg/kg, 0.52 mg/kg respectively). The higher concentrations of lead and copper during the wet season compared to the dry season might be due to runoffs and flooding during the wet season. This agrees with the findings of Nsofor and Ikpeze (2014), who had higher concentrations of Zn, Fe, Cu and Pb in *Clarias gariepinus* during the wet season compared to the dry season. Cadmium and chromium, however, had higher concentrations during the dry season than the wet season. This might be as a result of direct and point sources of pollution. Significant difference ($P < 0.05$) was observed between the two seasons.

Table 4.21: Mean Heavy Metal Concentrations in ONFG Kainji Lake based on Season

	Heavy metal concentration (mg/kg)				
Season	Pb	Cu	Cd	Cr	As
Wet	0.18 ± 0.00^{a}	0.61 ± 0.00^{a}	0.06 ± 0.00^{b}	0.06 ± 0.00^{b}	0.01 ± 0.00^{a}
Dry	0.12 ± 0.00^{b}	0.52 ± 0.00^{b}	0.13 ± 0.00^{a}	0.08 ± 0.00^{a}	0.01 ± 0.00^{a}

Values are means of five determinations. Means with dissimilar letter (s) differ significantly according to student T test. **Significant at $P \leq 0.05$**

4.30 Heavy Metal Concentrations in *Oreochromis niloticus* Fish Muscles (ONFM) of Kainji Sample Station

The concentrations of Pb, Cu, Cd, Cr and As in ONFM in all the sample stations (Table 4.22) were below maximum permissible limit (FAO/WHO, 2011; WHO, 2001; FEPA, 2003; WHO 2003), however there was a significant difference ($P < 0.05$) between the sample stations. Concentrations of the heavy metals in the muscles were in this order: Cu > Pb > Cd > Cr > As. This result agrees with Karadede and Unlu (2000), Ozturk *et al.* (2009) and Ekwu (2010), whose reports from a number of fish species studied showed that muscles were not an active tissue in the accumulation of heavy metals when compared with other organs of the fishes studied.

Table 4.22: Heavy Metal Concentrations in ONFM of Kainji Sample Station

Sample station	Heavy metal concentration (mg/kg)				
	Pb	**Cu**	**Cd**	**Cr**	**As**
K1.1	0.048^{a}	0.122^{b}	0.027^{c}	0.018^{b}	0.003^{c}
K1.2	0.046^{a}	0.123^{b}	0.031^{b}	0.012^{d}	0.006^{b}
K1.3	0.015^{d}	0.103^{d}	0.036^{a}	0.016^{c}	0.001^{d}
K1.4	0.024^{c}	0.195^{a}	0.026^{c}	0.024^{a}	0.008^{a}
K1.5	0.034^{b}	0.106^{c}	0.024^{d}	0.013^{d}	0.010^{a}

Values are means of five determinations. Means with dissimilar letter (s) differ significantly according to the Duncan's Multiple Range Test (DMRT).
Significant at $P \leq 0.05$

4.31 Heavy Metal Concentrations in *Oreochromis niloticus* Fish Muscles (ONFM) of Kainji Lake based on Season

Table 4.23 shows **h**eavy metal concentrations in ONFM of Kainji Lake based on season. It was observed that the concentrations of Pb, Cu, and Cd during the wet season were higher (0.04 mg/kg, 0.17 mg/kg and 0.14 mg/kg respectively) than the dry season (0.02 mg/kg, 0.12 mg/kg and 0.04 mg/kg respectively). This result agrees with the findings of Nsofor and Ekpeze (2014), who reported higher concentrations of heavy metals in fish during the wet season compared to the dry season. Concentration of Cr, however, was observed to be higher during the dry season (0.02 mg/kg) than the wet season (0.01 mg/kg). Significant difference ($P < 0.05$) was observed between the concentrations during the wet and dry seasons.

Table 4.23: Heavy Metal Concentrations in ONFM of Kainji Lake based on Season

	Heavy metal concentration (mg/kg)				
Season	**Pb**	**Cu**	**Cd**	**Cr**	**As**
Wet	0.04 ± 0.00^{a}	0.17 ± 0.10^{a}	0.14 ± 0.00^{a}	0.01 ± 0.00^{b}	0.01 ± 0.00^{a}
Dry	0.02 ± 0.00^{b}	0.12 ± 0.00^{b}	0.04 ± 0.00^{b}	0.02 ± 0.00^{a}	0.01 ± 0.00^{a}

Values are means of five determinations. Means with dissimilar letter (s) differ significantly according to student t test. **Significant at $P \leq 0.05$**

4.32 Heavy Metal Concentrations in *Clarias gariepinus* Fish Intestines (CGFI), Kainji Lake Sampling Station

Figure 4.9 shows heavy metal concentrations in *Clarias gariepinus* fish intestines (CGFI) from Kainji sample stations. It was observed that the concentrations of Pb in the intestines of *Clarias gariepinus* in all the sample stations were higher than the maximum permissible limit of 0.3 mg/kg according to FAO/WHO (2011). This might be as a result of high concentration of lead in the Kainji water samples especially in the months of April and September. This tends to agree with previous studies of Yilmaz (2003) and Ozturk *et al.* (2009) that reported different concentations of heavy metals in fish species, which were attributed to chemical characteristics of the water from which the fishes were sampled. Copper (Cu) and Cadmium (Cd) were observed to be below maximum permissible limit of 3.0 mg/kg and 0.5 mg/kg respectively according to WHO (2003). Chromium (Cr) was seen to be within acceptable range, that is below 0.15 mg/kg according to FEPA (2003), except in stations 3 (Kl.3) and 4 (Kl.4). Arsenic was also observed to be slightly higher than maximum permissible limit of 0.02 mg/kg according to WHO (2001), except in sample stations Kl.3 and Kl.5, which were observed to be within acceptable limit of 0.02 mg/kg (Appendix U). This arsenic level could be associated with agricultural effluents. Oti (2001)

reported high level of arsenic (0.082 mg/l) in *Clarias gariepinus* from Aba River in Anambara State, Nigeria.

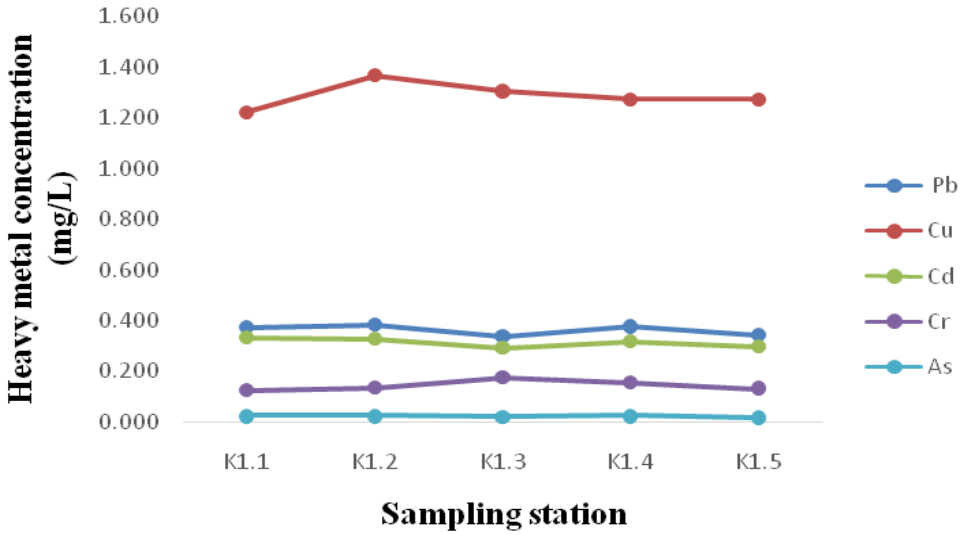

Figure 4.9: Heavy metal concentrations in CGFI of Kainji sampling station

4.33 Heavy Metal Concentrations in *Clarias gariepinus* Fish Intestines (CGFI) of Kainji Lake based on Season

The concentrations of Cu, Cd and As in *Clarias gariepinus* fish intestines (CGFI) for both seasons (Table 4.24) were not above maximum permissible limit of 3.0 mg/kg, 0.5 mg/kg and 0.02 mg/kg respectively according to WHO (2003) and WHO (2001) respectively. The concentration of Pb was above maximum permissible limit of 0.3 mg/kg according to FAO/WHO (2011). This also might be as a result of high concentrations of lead in the Kainji water samples especially at the onset of rainfall (month of April) and the peak of rainfall (September), which were accompanied with flooding. This result agrees with previous studies of Yilmaz *et al*, (2003) and Ozturk *et al.* (2009) that reported different

concentations of heavy metals in fish species, which were attributed to chemical characteristics of the water from which fishes were sampled.

Table 4.24: Heavy metal concentrations in CGFI of Kainji Lake based on Season

	Heavy metal concentration (mg/kg)				
Season	**Pb**	**Cu**	**Cd**	**Cr**	**As**
Wet	0.39 ± 0.00^{a}	1.29 ± 0.00^{a}	0.29 ± 0.00^{b}	0.13 ± 0.00^{b}	0.02 ± 0.00^{a}
Dry	0.34 ± 0.00^{b}	1.28 ± 0.00^{b}	0.34 ± 0.00^{a}	0.16 ± 0.00^{a}	0.02 ± 0.00^{a}

Values are means of five determinations. Means with dissimilar letter (s) differ significantly according to student T test. **Significant at $P \leq 0.05$**

4.34 Heavy Metal Concentrations in *Clarias gariepinus* Fish Gill (CGFG) of Kainji Lake Sample Station

The concentrations of heavy metals (Pb, Cu, Cd, Cr, and As) in CGFG in all the sample stations (Table 4.25) were observed to be below maximum permissible limit of 0.3 mg/kg, 3.0 mg/kg, 0.5 mg/kg, 0.15 mg/kg and 0.02 mg/kg respectively. This shows that this organ (the gills) is safe for human consumption. The concentrations of Pb, Cd and Cr in CGFG of Kainji Lake were reportedly lower than the concentrations in *Clarias gariepinus* from Itu River, Akwa Ibom State (Etim *et al*., 2013).

Table 4.25 Heavy Metal Concentrations in CGFG of Kainji Lake Sample Stations

	Heavy metal concentration (mg/kg)				
Sample Stations	**Pb**	**Cu**	**Cd**	**Cr**	**As**
K1.1	0.157^{b}	0.637^{a}	0.167^{a}	0.062^{d}	0.017^{a}
K1.2	0.147^{c}	0.486^{d}	0.153^{b}	0.066^{c}	0.010^{b}
K1.3	0.123^{d}	0.548^{b}	0.155^{b}	0.104^{a}	0.00b
K1.4	0.176^{a}	0.537^{c}	0.130^{c}	0.096^{b}	0.018^{a}
K1.5	0.095^{e}	0.474^{e}	0.113^{d}	0.058^{e}	0.010^{b}

Values are means of five determinations. Means with dissimilar letter (s) differ significantly according to the Duncan's Multiple Range Test (DMRT).
Significant at P ≤ 0.05

4.35 Heavy Metal Concentrations in *Clarias gariepinus* Fish Gills (CGFG) of Kainji Lake based on Season

Table 4.26 shows the concentrations of heavy metals in *Clarias gariepinus* fish gills (CGFG) of Kainji Lake based on season. It was observed that the concentrations of Pb, Cu, Cd, Cr and Arsenic in CGFG in both seasons were below maximum permissible limit. Concentration of Pb during wet season was higher (0.16 mg/kg) than the concentration during the dry season (0.12 mg/kg). This might be due to the effect of runoffs during the wet season whereas the concentration of Cu, Cd and Cr was higher during the dry season compared to that of the wet season. This might be due to direct washings by miners. Significant difference ($p < 0.05$) existed between the concentration of metals during the wet and dry seasons.

Table 4. 26: Seasonal Heavy Metal Concentrations in CGFG of Kainji Lake

	Heavy metal concentration (mg/kg)				
Season	**Pb**	**Cu**	**Cd**	**Cr**	**As**
Wet	0.16 ± 0.00^{a}	0.52 ± 0.00^{b}	0.10 ± 0.00^{b}	0.06 ± 0.00^{b}	0.01 ± 0.00^{a}
Dry	0.12 ± 0.00^{b}	0.55 ± 0.00^{a}	0.19 ± 0.00^{a}	0.10 ± 0.00^{a}	0.01 ± 0.00^{a}

Values are means of five determinations. Means with dissimilar letter (s) differ significantly according to student t test. **Significant at $P \leq 0.05$**

4.36 Heavy Metal Concentrations in *Clarias gariepinus* Fish Muscles (CGFM) of Kainji Sample Station

Table 4.27 shows heavy metal concentrations in *Clarias gariepinus* fish muscles (CGFM) of Kainji sample station. It was observed that CGFM in sample station Kl.4 recorded higher concentrations of Pb, Cd, Cr (0.126 mg/kg, 0.133 mg/kg, 0.020 mg/kg respectively) than all other stations. This might be due to increased human activities (washing by miners, agricultural farms) around the sample station Kl.4 compared to the other stations. Sample station Kl.1 recorded the highest concentration of Cu (0.156 mg/kg) while sample station Kl.2 had the highest concentration of As (0.004 mg/kg) compared to all other sample stations. The concentration of cadmium from this study was generally lower compared to the result of cadmium concentration in *Clarias gariepinus* from Niger River reported by Effiong *et al*. (2019).

It was generally observed that the concentrations of all the heavy metals analyzed in CGFM were below maximum permissible limit according to WHO (2001), FEPA (2003), WHO (2003) and FAO/WHO (2011). This result is in consistence with the findings of Ozturk *et al*. (2009), whose report from a number of fish species examined showed that fish muscles

were not active tissues in accumulating heavy metals. Hence, it could be said that the muscles (flesh) of *Clarias gariepinus* from Kainji Lake are safe for human consumption.

Table 4.27: Heavy Metal Concentrations in CGFM of Kainji Sample Station

	Heavy metal concentration (mg/kg)				
Sample Station	**Pb**	**Cu**	**Cd**	**Cr**	**As**
K1.1	0.021^{d}	0.156^{a}	0.019^{d}	0.013^{c}	0.003^{b}
K1.2	0.023^{c}	0.110^{c}	0.022^{c}	0.008^{e}	0.004^{a}
K1.3	0.028^{b}	0.126^{b}	0.094^{b}	0.011^{d}	0.000^{c}
K1.4	0.126^{a}	0.073^{e}	0.133^{a}	0.020^{a}	0.000^{c}
K1.5	0.020^{d}	0.097^{d}	0.017^{e}	0.017^{b}	0.000^{c}

Values are means of five determinations. Means with dissimilar letter (s) differ significantly according to the Duncan's Multiple Range Test (DMRT).
Significant at $P \leq 0.05$

4.37 Heavy Metal Concentrations in *Clarias gariepinus* Fish Muscles (CGFM) of Kainji Lake based on Season

Concentrations of Pb, Cu and Cr during the wet season (Table 4.28) were observed to be higher (0.03 mg/kg, 0.12 mg/kg and 0.02 mg/kg respectively) than the concentrations during the dry season (0.02 mg/kg, 0.11 mg/kg and 0.01 mg/kg respectively). This higher concentration during the wet season might be as a result of runoffs and flooding during the wet season. This result agrees with the findings of Nsofor and Ekpeze (2014), who reported higher concentrations of heavy metals in fish during the wet season compared to the dry season. The concentration of Cd during the dry season was, however, higher (0.05 mg/kg) than that during the wet season (0.02 mg/kg).

Table 4.28: Seasonal Heavy Metal Concentrations in CGFM of Kainji Lake

	Heavy metal concentration (mg/kg)				
Season	**Pb**	**Cu**	**Cd**	**Cr**	**As**
Wet	0.03 ± 0.00^{a}	0.12 ± 0.00^{a}	0.02 ± 0.00^{b}	0.02 ± 0.00^{a}	0.00 ± 0.00^{a}
Dry	0.02 ± 0.00^{b}	0.11 ± 0.00^{b}	0.05 ± 0.00^{a}	0.01 ± 0.00^{b}	0.00 ± 0.00^{a}

Values are means of five determinations. Means with dissimilar letter (s) differ significantly according to student t test. **Significant at $P \leq 0.05$**

4.38 Heavy Metal Concentrations in *Oreochromis niloticus* Fish Intestines (ONFI) of Jebba Lake Sampling Station

Figure 4.10 shows heavy metal concentrations in *Oreochromis niloticus* fish intestines (ONFI) from different sample stations of Jebba Lake. It was observed that the concentrations of Pb in the intestines of *Oreochromis niloticus* in all the sample stations were above the maximum permissible limit of 0.3 mg/kg according to FAO/WHO (2011). Copper and cadmium were, however, below maximum permissible limit. The concentration of chromium was above maximum permissible limit of 0.15 mg/kg except in sample stations Jl.3 and Jl.5. Concentration of arsenic was also observed to be above maximum permissible limit of 0.02 mg/kg according to WHO (2001), except in sample station Jl.5.

In all the samples analyzed, sample station Jl.2 had the highest concentration of heavy metals. This might be due to the washing of precious metals by miners and effluents from car wash, which was being carried out in and around the station. These high cncentrations were also aided by much agricultural farms in which inorganic fertilizers were being applied to adjacent farms, which might have been easily washed down into the Lake through surface runoffs. Hence it could be deduced that the quality of heavy metals in an

aquatic ecosystem, is to a great extent, dependent on the amount of human activities around the water body.

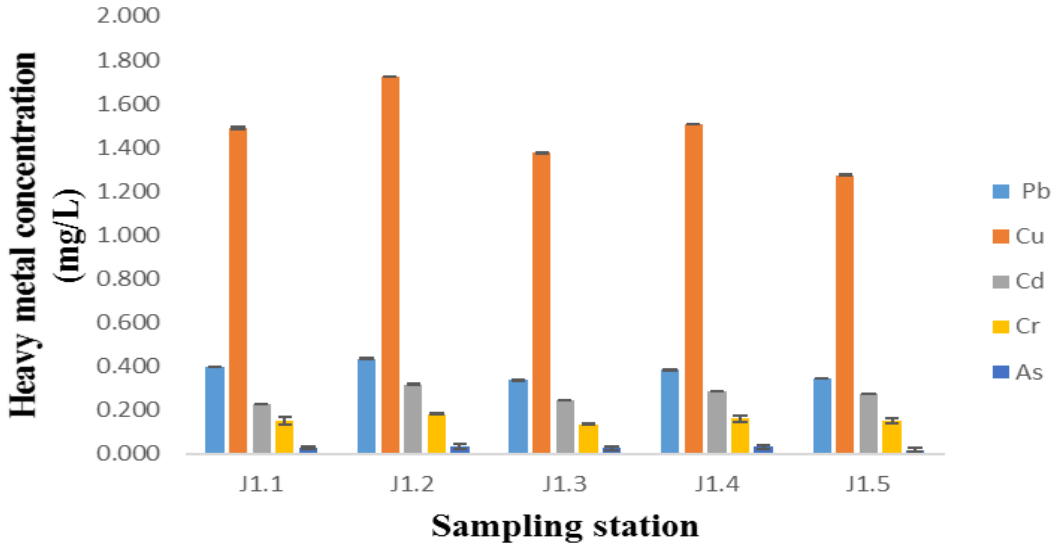

Figure 4.10: Heavy metal concentrations in ONFI of Jebba Lake sampling station

4.39 Heavy Metal Concentrations in *Oreochromis niloticus* Fish Intestines (ONFI) of Jebba Lake based on Season.

Table 4.29 shows heavy metal concentrations in *Oreochromis niloticus* fish intestines (ONFI) of Jebba Lake based on season. It was observed that the concentrations of Cu, Cd and Cr in ONFI were higher during the dry season compared to the wet season, whereas Pb had a higher concentration (0.04 mg/kg) during the wet season than the dry season (0.36 mg/kg). Significant difference ($P < 0.05$) existed in concentrations of metals between the two seasons, whereas there was no significant difference ($P > 0.05$) in concentration of As in the two seasons. Higher concentrations of Cu, Cd and Cr during the dry season might have arisen from direct washing of ore pulp by miners into the water body and also increased oil and lubricant from hydroelectric power plant. This tends to agree with

Jonathan and Maina (2009), who reported that fish species mostly absorbed heavy metals from its surrounding waters thereby resulting to the accumulation of some in reasonable amount.

Table 4.29: Heavy Metal Concentrations in ONFI of Jebba Lake based on Season

	Heavy metal concentration (mg/kg)				
Season	**Pb**	**Cu**	**Cd**	**Cr**	**As**
Wet	0.40 ± 0.00^{a}	1.41 ± 0.00^{b}	0.25 ± 0.00^{b}	0.15 ± 0.00^{b}	0.03 ± 0.01^{a}
Dry	0.36 ± 0.00^{b}	1.54 ± 0.00^{a}	0.29 ± 0.00^{a}	0.16 ± 0.00^{a}	0.03 ± 0.01^{a}

Values are means of five determinations. Means with dissimilar letter (s) differ significantly according to student T test. **Significant at $P \leq 0.05$**

4.40 Heavy Metal Concentrations in *Oreochromis niloticus* Fish Gills (ONFG) of Jebba Lake Sampling Station

Heavy metal concentrations in *Oreochromis niloticus* fish gills **(**ONFG) of Jebba Lake sample stations (Figure 4.11) showed that the concentrations of all the heavy metals analysed were below maximum permissible limit except the concentration of As from sample station Jl.4, which was 0.021 mg/kg. The concentrations of Cu and Cd were highest in Jl. 2 while Pb and Cr were highest in JL.1. Sample stations J1. 1 and Jl. 2 were at the upper course of the Jebba Lake. Unlike other heavy metals, As had its highest concentration at sample station Jl.4. Sample station Jl.1 was directly below the hydroelectric power plant, hence, effluents, from there might be responsible for the increase of Pb and Cr. Station Jl.2 was characterized by high mining activities, washing of automobiles and other domestic effluents. These might be responsible for the increase in Cd and Cu concentrations. The slight increase of As concentration (0.21 mg/kg) in Jl.4 might be due to effluents from old Awuru market (as it lies adjacent and often time received wastes from the market). Thus, it

could be said that the quality of fish depends on the quality of its aquatic environment, which in turn depends on the anthropogenic activities in and around the water body.

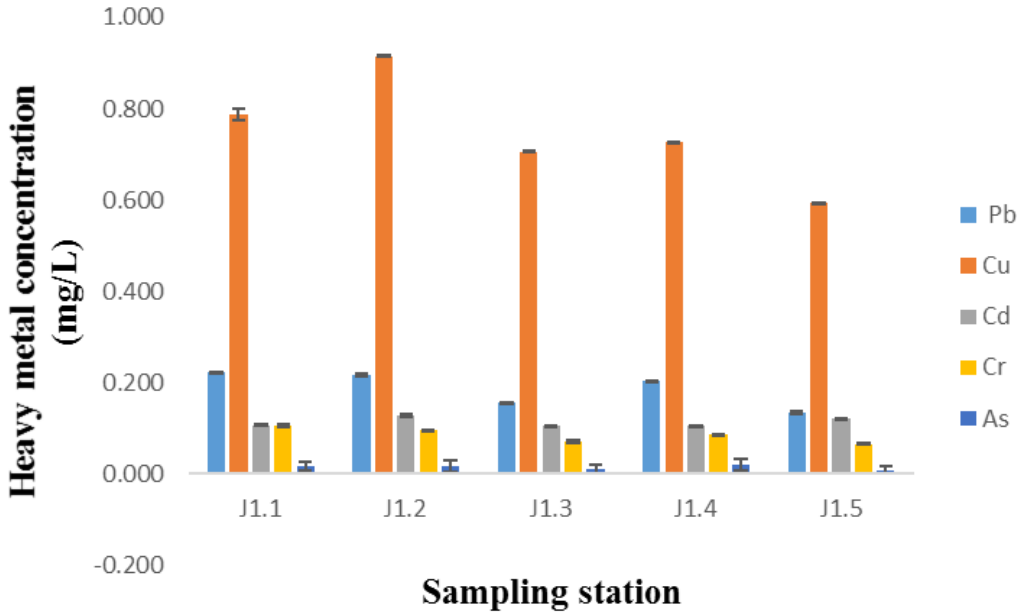

Figure 4.11: Heavy metal concentrations in ONFG of Jebba Lake sampling station

4.41 Heavy Metal Concentration in *Oreochromis niloticus* Fish Gills (ONFG) from Jebba Lake based on Season.

Table 4.30 shows heavy metal concentrations in *Oreochromis niloticus* fish gills (ONFG) from Jebba Lake based on season. It was observed that all the concentrations of heavy metals (Pb, Cu, Cd, Cr and As) in ONFG were below maximum permissible limits. This shows that this organ of fish (ONFG) from Jebba Lake is safe for human consumption. Significant difference ($P < 0.05$) existed in concentrations of heavy metals between the two seasons. It was also observed that the metals had higher concentrations during the dry season compared to the wet season except the concentration of Pb (0.19 mg/kg) which was the same in both seasons. The observed variations in concentration of heavy metals in the

gills alongside seasonal variation might be as a result of direct washings of ore into the Lake by miners and surface runoffs from the neighboring farms and adjacent mining zones.

Table 4.30: Seasonal heavy Metal Concentrations in *Oreochromis niloticus* Fish Gills (ONFG) from Jebba Lake.

	Heavy metal concentration (mg/kg)				
Season	**Pb**	**Cu**	**Cd**	**Cr**	**As**
Wet	0.19 ± 0.00^{a}	0.73 ± 0.01^{b}	0.09 ± 0.00^{b}	0.07 ± 0.00^{b}	0.01 ± 0.01^{b}
Dry	0.19 ± 0.00^{a}	0.76 ± 0.00^{a}	0.14 ± 0.00^{a}	0.10 ± 0.00^{a}	0.02 ± 0.01^{a}

Values are means of five determinations. Means with dissimilar letter (s) differ significantly according to student T test. **Significant at $P \leq 0.05$**

4.42 Heavy Metal Concentrations in *Oreochromis niloticus* Fish Muscles (ONFM) of Jebba Lake Sample Station

Table 4.31 shows heavy metal concentrations in *Oreochromis niloticus* fish muscles (ONFM) of Jebba Lake sample station. It was observed that the concentrations of Pb, Cu, Cd, Cr and As in ONFM were below maximum permissible limit of 0.3 mg/kg, 3.0 mg/kg, 0.5 mg/kg, 0.15 mg/kg and 0.02 mg/kg respectively. This result agrees with that of Karadede and Unlu (2000), who reported that fish muscles was not an active tissue in accumulation of heavy metals when compared with other organs. Samples of ONFM from station Jl.2 had the highest concentrations of Pb, Cu, Cd and Cr compared to other sample stations. This might be because of increased anthropogenic activities such as illegal mining, washings by miners, high concentrations of agricultural farms among others, around the sample station Jl.2. This agrees with the findings of Salaudeen *et al*. (2016) who reported that mining areas were the chief areas where heavy-metal pollution readily occured, in

which the surrounding land receives the pollutants first and later adjacent water bodies through runoffs. With the concentration of these metals in ONFM below maximum permissible limit, it could be said that the muscles of *Oreochromis niloticus* from Jebba Lake are safe for human consumption.

Table 4.31 Heavy Metal Concentrations in *Oreochromis niloticus* Fish muscles (ONFM) of Jebba Lake Sample Station

	Heavy metal concentration (mg/kg)				
Sample Station	**Pb**	**Cu**	**Cd**	**Cr**	**As**
J1.1	0.045^{b}	0.115^{c}	0.033^{c}	0.024^{b}	0.005^{c}
J1.2	0.061^{a}	0.217^{a}	0.275^{a}	0.025^{a}	0.006^{b}
J1.3	0.033^{d}	0.195^{b}	0.022^{d}	0.013^{e}	0.000^{d}
J1.4	0.046^{b}	0.217^{a}	0.044^{b}	0.021^{c}	0.007^{a}
J1.5	0.036^{c}	0.104^{d}	0.033^{c}	0.018^{d}	0.000^{d}

Values are means of five determinations. Means with dissimilar letter (s) differ significantly according to the Duncan's Multiple Range Test (DMRT).
Significant at $P \leq 0.05$

4.43 Heavy Metal Concentrations in *Oreochromis niloticus* Fish Muscles (ONFM) of Jebba Lake based on Season

Table 4.32 shows heavy metal concentrations in *Oreochromis niloticus* fish muscles (ONFM) of Jebba Lake based on season. It was observed that the concentrations of Pb and Cu were higher during the wet season than the dry season. This might be due to runoffs and flooding from adjacent farms and mining sites. This result agrees with the findings of Peter *et al*. (2018), who reported higher concentrations of lead, copper, zinc, iron and manganese

during wet season than the dry season. There was a significant difference ($P < 0.05$) in the concentrations of Pb, Cu and Cr for wet and dry seasons. However, Cd and As showed no significant difference.

Table 4.32: Heavy metal Concentrations in *Oreochromis niloticus* Fish Muscles (ONFM) of Jebba Lake based on Season.

	Heavy metal concentration (mg/kg)				
Season	**Pb**	**Cu**	**Cd**	**Cr**	**As**
Wet	0.046 ± 0.00^{a}	0.174 ± 0.00^{a}	0.031 ± 0.00^{a}	0.018 ± 0.00^{b}	0.004 ± 0.00^{a}
Dry	0.041 ± 0.00^{b}	0.165 ± 0.00^{b}	0.031 ± 0.00^{a}	0.022 ± 0.00^{a}	0.004 ± 0.00^{a}

Values are means of five determinations. Means with dissimilar letter (s) differ significantly according to student T test. **Significant at $P \leq 0.05$**

4.44 Heavy Metal Concentrations in *Clarias gariepinus* Fish Intestines (CGFI) of Jebba Lake Sample Station

Table 4.33 shows heavy metal concentrations in *Clarias gariepinus* fish intestines (CGFI) of Jebba Lake sample stations. The concentrations of Pb, Cd, Cr and As in sample station Jl.2 were higher than that in other sample stations. Sample station Jl.4 had higher concentration of Cu than other sample stations. It was observed that the concentrations of Pb in CGFI of Jebba Lake in all the sample stations were above maximum permissible limit of 0.3 mg/kg (FAO/WHO, 2011). The concentrations of Cu in all the sample stations were below maximum permissible limit, so also was the concentration of Cd in all the sample stations except in station Jl.2, which was 1.695 mg/kg and, this was above the maximum permissible limit of 0.5 mg/kg according to WHO (2003). The concentrations of Cr in sample stations Jl.1, Jl.2 and Jl.4 were also observed to be above the maximum permissible

limit of 0.15 mg/kg (FEPA, 2003). This might be due to wastes (lubricants and oil) from hydro-electric power generation plant. Concentration of As was also above its permissible limit of 0.02 mg/kg (WHO, 2001). However, there was no significant difference ($P > 0.05$) between the sample stations. Higher concentrations of Pb, Cd, Cr and As in station Jl.2 than other stations might be due to increased human activities such as illegal mining, agricultural farms and washings of automobiles around the station. This agrees with the work of Salaudeen *et al*. (2016), who reported that "mining areas are the chief areas where heavy metal pollution readily occurs, in which the surrounding land receives it first and later adjacent water body through surface runoffs" The high concentrations of Pb, Cd, Cr and As was also reflected in concentration of heavy metals in the water and sediment samples analysedfrom same station. Effiong *et al*. (2019), however, reported lower concentrations of cadmium in CGFI from River Niger at Onitsha axis.

Table 4.33: Heavy Metal Concentrations in *Clarias gariepinus* Fish Intestines (CGFI) of Jebba Lake Sample Station

	Heavy metal concentration (mg/kg)				
Sample Station	**Pb**	**Cu**	**Cd**	**Cr**	**As**
J1.1	0.413^{b}	1.446^{b}	0.376^{b}	0.156^{b}	0.025^{a}
J1.2	0.447^{a}	1.691^{b}	1.695^{a}	0.173^{a}	0.032^{a}
J1.3	0.375^{c}	1.293^{c}	0.317^{d}	0.146^{c}	0.028^{a}
J1.4	0.358^{d}	1.604^{a}	0.345^{c}	0.157^{b}	0.028^{a}
J1.5	0.352^{d}	1.386^{bc}	0.303^{e}	0.132^{d}	0.023^{a}

Values are means of five determinations. Means with dissimilar letter (s) differ significantly according to the Duncan's Multiple Range Test (DMRT).
Significant at $P \leq 0.05$

4.45 Heavy Metal Concentrations in *Clarias gariepinus* Fish Intestines (CGFI) of Jebba Lake based on Season

The concentrations of Cu, Cd and Cr were observed to be higher during the dry season than the concentration during the wet season (Table 4.34). This might be due to direct washings and increase in waste released from hydroelectric plant as the water volume decreased during the dry season. Lead (Pb) was observed to be higher during the wet season compared to the dry season. This might be due to surface runoffs from mining sites and other domestic wastes. There was a significant difference ($P < 0.05$) in concentrations of Pb, Cu, Cd and Cr between wet and dry seasons while there was no significant difference ($P > 0.05$) in concentrations of As between the two seasons.

Table 4.34: Heavy Metal Concentrations in *Clarias gariepinus* Fish Intestines (CGFI) of Jebba Lake based on Season

	Heavy metal concentration (mg/kg)				
Season	**Pb**	**Cu**	**Cd**	**Cr**	**As**
Wet	0.43 ± 0.00^{a}	1.45 ± 0.01^{b}	0.34 ± 0.00^{b}	0.14 ± 0.00^{b}	0.03 ± 0.01^{a}
Dry	0.36 ± 0.00^{b}	1.52 ± 0.00^{a}	0.35 ± 0.00^{a}	0.17 ± 0.00^{a}	0.03 ± 0.01^{a}

Values are means of five determinations. Means with dissimilar letter (s) differ significantly according to student T test. **Significant at $P \leq 0.05$**

4.46 Heavy Metal Concentration in *Clarias gariepinus* Fish Gills (CGFG) in Jebba Lake Sample Station

The concentrations of heavy metals (Pb, Cu, Cd, Cr and As) in *Clarias gariepinus* fish gills **(**CGFG) of Jebba Lake were observed to be below maximum permissible limit in all the sample stations (Figure 4.12). Sample station Jl. 2 was observed to have higher

concentrations of Pb, Cu, Cr and As than other sample stations while sample station Jl.1 had higher concentration of Cd than other stations. This might be due to the presence of oil and lubricant used in powergeneration plant, which flows directly to station Jl. 1 at its lower course before other stations. Farombi *et al*. (2007) reported higher concentrations of lead, copper, cadmium and arsenic in fish gills of *Clarias gariepinus* obtained from Ogun River.

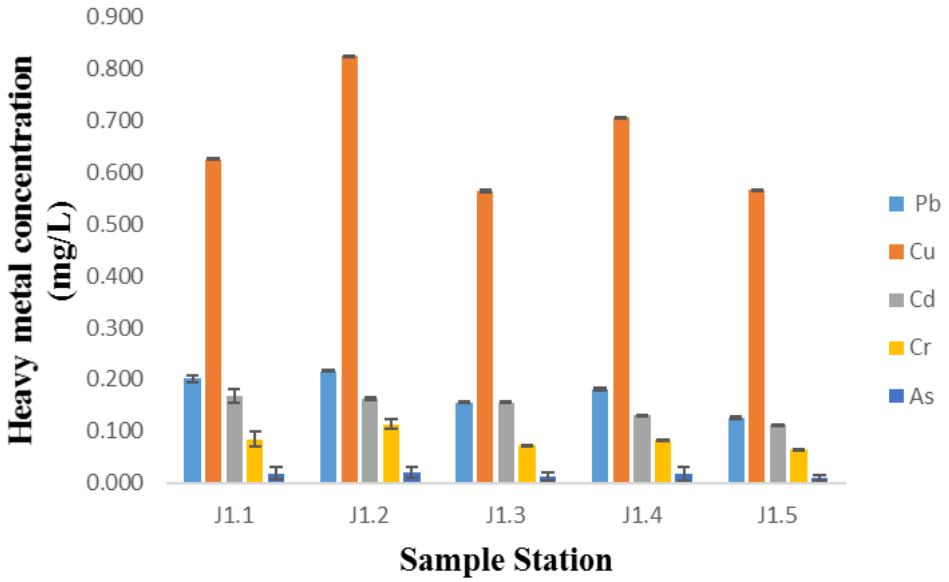

Figure 4.12: Heavy metal concentration in CGFG in Jebba Lake sample station

4.47 Heavy Metal Concentrations in *Clarias gariepinus* Fish Gills (CGFG) in Jebba Lake based on Season

It was observed that the concentrations of Cu, Cd and Cr were higher during the dry season compared to the wet season (Table 4.35). This might be due to direct washings of ore pulp into the Lake as well as release of effluents from hydroelectric power plant, thereby increasing the concentrations of heavy metal as water volume decreased. Increase heavy metal concentration in water may led to increased sorption. Lead was higher during the wet

season compared to the dry season, but there was no significant difference between wet and the dry seasons. These results showed that the concentrations of these heavy metals were below maximum permissible limit (that is within acceptable limit).

Table 4.35: Heavy Metal Concentrations in *Clarias gariepinus* Fish Gills (CGFG) of Jebba Lake based on Season

	Heavy metal concentration (mg/kg)				
Season	**Pb**	**Cu**	**Cd**	**Cr**	**As**
Wet	0.18 ± 0.00^{a}	0.61 ± 0.00^{b}	0.11 ± 0.00^{b}	0.07 ± 0.00^{b}	0.02 ± 0.01^{a}
Dry	0.17 ± 0.00^{a}	0.70 ± 0.00^{a}	0.19 ± 0.00^{a}	0.09 ± 0.00^{a}	0.02 ± 0.01^{a}

Values are means of five determinations. Means with dissimilar letter (s) differ significantly according to student T test.
Significant at $P \leq 0.05$

4.48 Heavy Metal Concentrations in *Clarias gariepinus* Fish Muscles (CGFM) from Jebba Lake Sampling Station

Concentrations of heavy metals in *Clarias gariepinus* fish muscles (CGFM) from Jebba Lake sampling stations showed that Cu, Cd, Cr and As were higher in sampling station Jl. 2, with high mining activities and washing of automobiles, compared to their concentrations in other sampling stations while the concentration of Pb was highest in Jl. 4, which lies adjacent Old Awuru market and receives efflunts from the market (Figure 4.13). It was also observed that the concentrations of all the metals (Pb, Cu, Cd, Cr, As) were found to be within acceptable limit. Thus, it could be said that the muscles of *Clarias gariepinus* in Jebba Lake are safe for human consumption.

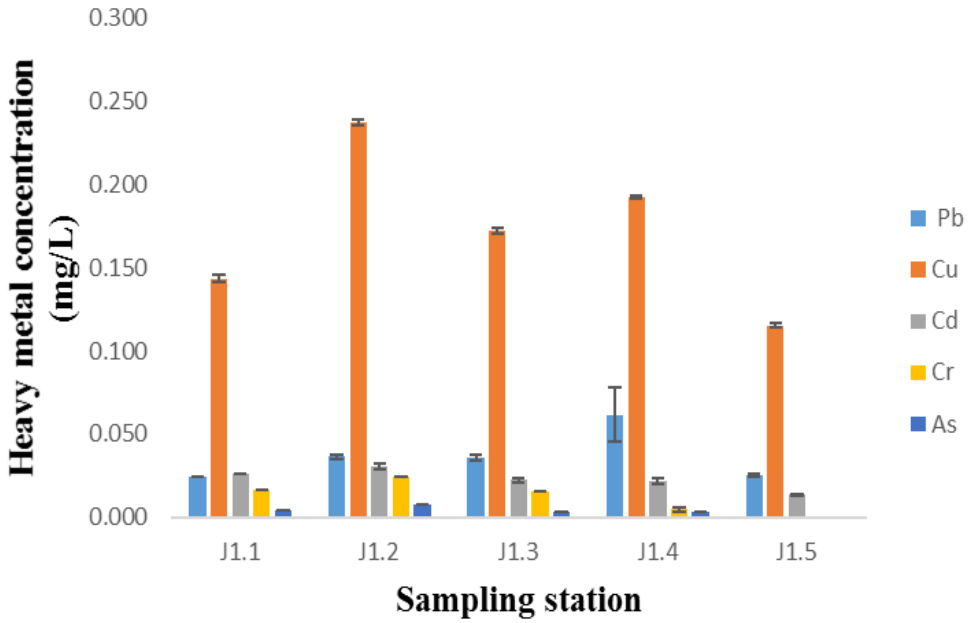

Figure 4.13: Heavy metal concentrations in CGFM, Jebba Lake sampling station

4.49 Heavy Metal Concentrations in *Clarias gariepinus* Fish Muscles (CGFM) of Jebba Lake based on Season

The concentrations of heavy metals in *Clarias gariepinus* fish muscles (CGFM) of Jebba Lake (Table 4.36) (which was based on seasonal variations showed that Pb was higher (0.05 mg/kg) during the wet season compared to the dry season (0.04 mg/kg). This might be due to runoffs from adjacent mining sites, thereby increasing its concentration in the water body and, by extension, the fish organs. Copper (Cu) and Arsenic (As), on the other hand, were higher during the dry season (0.19 mg/kg, 0.01 mg/kg respectively) compared to the wet season (0.16 mg/kg, 0.00 mg/kg respectively). However, there were no significant differences ($P > 0.05$) in the concentrations of cadmium (Cd) and chromium (Cr) between the two seasons. Effiong *et al*. (2019) reported higher concentrations of cadmium in CGFM from Niger river , Onitsha. All the heavy metals examined (Pb, Cu, Cd, Cr, As) were observed to be within acceptable limit.

Table 4.36: Heavy Metal Concentration in *Clarias gariepinus* Fish Muscles (CGFM) of Jebba Lake based on Season

	Heavy metal concentration (mg/kg)				
Season	**Pb**	**Cu**	**Cd**	**Cr**	**As**
Wet	0.05 ± 0.00^{a}	0.16 ± 0.00^{b}	0.02 ± 0.00^{a}	0.01 ± 0.00^{a}	0.00 ± 0.00^{b}
Dry	0.04 ± 0.00^{b}	0.19 ± 0.00^{a}	0.02 ± 0.00^{a}	0.01 ± 0.00^{a}	0.01 ± 0.00^{a}

Values are means of five determinations. Means with dissimilar letter (s) differed significantly according to student T test.
Significant at $P \leq 0.05$

4.50 Concentrations of Heavy Metals in Organs of *Oreochromis niloticus* (ON) and *Clarias gariepinus* (CG) in Kainji Lake

Concentrations of heavy metals in the intestines, gills and muscles of *Oreochromis niloticus* (ON) and *Clarias gariepinus* (CG) from Kainji Lake were compared (Figure 4.14). It was observed that the intestines of *Oreochromis niloticus* (ONFI) accumulated more heavy metals (Pb, Cu, Cd, Cr, As) than the intestines of *Clarias gariepinus* (CGFI). On the other hand, it was observed that the gills of *Clarias gariepinus* (CGFG) accumulated more heavy metals than the gills of *Oreochromis niloticus* (ONFG). This might be due to differences in organs affinity, ability to accumulate and excrete these heavy metals. The muscles of *Oreochromis niloticus* (ONFM) also accumulated more concentrations of heavy metals than the muscles of *Clarias gariepinus* (CGFM). Previous study by Lawal – Are *et al*. (2018) showed higher concentrations of cadmium, chomium and lead in *Oreochromis niloticus* than in *Clarias gariepinus.* It was also observed that the concentrations of Cu, Cd and Cr in the intestines of both fishes were below maximum permissible limit. Concentrations of Pb and As in the intestines of *Oreochromis niloticus*

were above maximum permissible limit. This might be due to the *Oreochromis niloticus* high tendency to bioaccumulate Pb and As.

Concentrations of Cu, Cd and Cr in the gills of both fishes were below maximum permissible limit. Concentrations of Pb and As in *Clarias gariepinus* fish gills were higher than the maximum permissible limit of 0.3 mg/kg and 0.02 mg/kg respectively. This might be due to *Clarias gariepinus* high tendency to bioaccumulate Pb and As. In gills of *Oreochromis niloticus*, the concentrations of Pb and As were within acceptable range. This might be due to the gills poor tendency to bioaccumulate Pb and As or the gills good ability to excrete it. In fish muscles, though the concentrations of the metals in *Oreochromis niloticus* were higher than that of *Clarias gariepinus*, they were not, however, above maximum permissible limit. All were observed to be within acceptable range.

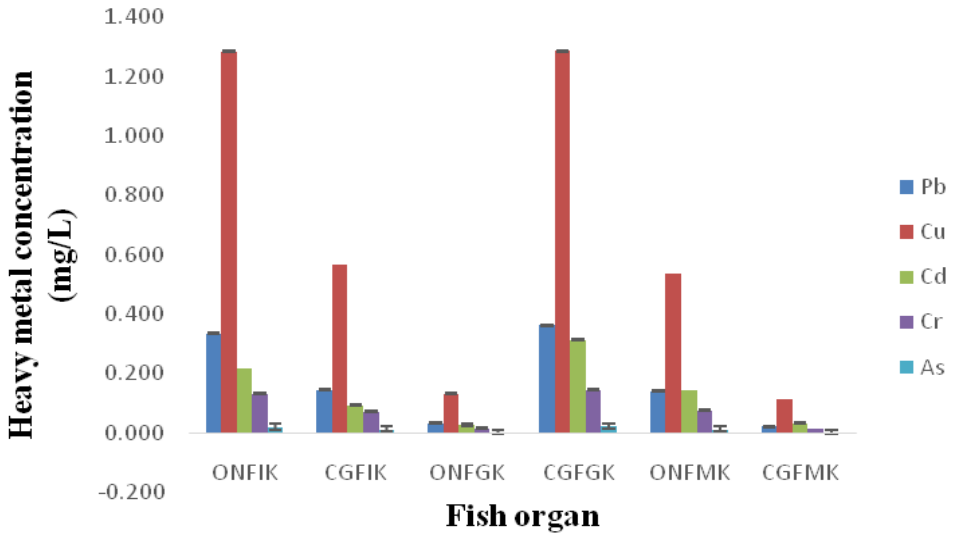

Figure 4.14: Concentrations of heavy metals in organs of *Oreochromis niloticus* (ON) and *Clarias gariepinus* (CG) in Kainji Lake

Key: ONFIK = *Oreochromis niloticus* Fish intestines from Kainji, CGFIK = *Clarias gariepinus* Fish intestines from Kainji, ONFGK = *Oreochromis niloticus* Fish gills from kainji, CGFGK = *Clarias gariepinus* fish gills from Kainji, ONFMK = *Oreochromis niloticus* Fish muscles from Kainji, CGFMK = *Clarias gariepinus* Fish muscles from kainji.

4.51 Heavy Metal Concentrations in Organs of *Oreochromis niloticus* in Kainji and Jebba Lakes.

Heavy metal concentrations in the organs of *Oreochromis niloticus* in Kainji and Jebba Lakes (Table 4.37) showed that Pb, Cu, Cd, Cr and As in *Oreochromis niloticus* fish intestines from Jebba Lake (ONFIJ) were higher than the concentrations in *Oreochromis niloticus* fish intestines from Kainji Lake (ONFIK).

It was also observed that the concentrations of these heavy metals (Pb, Cu, Cd and Cr) found in *Oreochromis niloticus* fish gills from Jebba (ONFGJ) were higher than the concentrations in *Oreochromis niloticus* fish gills from Kainji Lake (ONFGK). However the concentrations of As in ONFG from both Lakes had no significant difference ($P > 0.05$).

Similarly, the concentrations of Pb, Cu, Cd and Cr in *Oreochromis niloticus* fish muscles from Jebba (ONFMJ) were higher than the concentrations in *Oreochromis niloticus* fish muscles from Kainji Lake (ONFMK). However, arsenic was not found in *Oreochromis niloticus* fish muscles (ONFM) of both Lakes. The concentration of heavy metals from the organs of *Oreochromis niloticus* was in this order: muscles < gills < intestines. This order agrees with the findings (concentrations of heavy metals in fish organs) of Ozturk *et al.* (2009). The variation in concentrations of heavy metals in same organ from different water bodies (Kainji and Jebba Lakes), which revealed organ from Jebba Lake as having higher concentrations is an indication that Jebba Lake water was more polluted in terms of heavy metals than Kainji Lake water (as at the time of this study). The higher pollution status of Jebba Lake water might be due to higher mining activities (excarvation and washing of ore pulp), washings from automobile centres, effluents from hydroelectric plant and domestic

wastes as well as runoffs from farm lands. This agrees with the findings of Zhang and Wong (2007), who stated that concentrations of heavy metal levels in fish depended on different factors such as physicochemical parameters of the water and sediments as well as the season of capture.

Table 4.37 Heavy Metal Concentrations in Organs of *Oreochromis niloticus* in Kainji and Jebba Lakes.

		Heavy metal concentration (mg/kg)				
		Pb	**Cu**	**Cd**	**Cr**	**As**
Intestine	ONFIK	0.34±0.00^{b}	1.28±0.00^{b}	0.22±0.00^{b}	0.13±0.00^{b}	0.02±0.00^{b}
	ONFIJ	0.38±0.00^{a}	1.47±0.00^{a}	0.27±0.00^{a}	0.15±0.00^{a}	0.03±0.00^{a}
Gills	ONFGK	0.15±0.00^{b}	0.56±0.00^{b}	0.09±0.00^{b}	0.07±0.00^{b}	0.01±0.00^{a}
	ONFGJ	0.19±0.00^{a}	0.75±0.00^{a}	0.11±0.00^{a}	0.09±0.00^{a}	0.01±0.00^{a}
Muscles	ONFMK	0.03±0.00^{b}	0.13±0.00^{b}	0.03±0.00^{b}	0.02±0.00^{b}	0.00±0.00^{a}
	ONFMJ	0.05±0.00^{a}	0.17±0.00^{a}	0.04±0.00^{a}	0.03±0.00^{a}	0.00±0.00^{a}

Values are means of five determinations. Means with dissimilar letter (s) differ significantly according to the Duncan's Multiple Range Test (DMRT).
Significant at P ≤ 0.05

Key: ONFIK = *Oreochromis niloticus* Fish intestines from Kainji, ONFIJ = *Oreochromis niloticus* Fish intestines from Jebba, ONFGK = *Oreochromis niloticus* Fish gills from Kainji, ONFGJ = *Oreochromis niloticus* Fish gills from Jebba, ONFMK = *Oreochromis niloticus* Fish muscles from Kainji, ONFMJ = *Oreochromis niloticus* Fish muscles from Jebba

4.52 Heavy Metal Concentrations in Organs of *Clarias gariepinus* in Kainji and Jebba Lakes.

Concentrations of heavy metals in the organs of *Clarias gariepinus* in Kainji and Jebba Lakes (Table 4.38) showed that all the heavy metals analyzed in *Clarias gariepinus* fish intestines from Jebba Lake (CGFIJ) had higher concentrations than the *Clarias gariepinus* fish intestines from Kainji Lake (CGFIK). In addition, the gills of *Clarias gariepinus* from Jebba Lake had higher concentrations of the heavy metals compared to that from Kainji Lake. Jebba Lake water generally had higher concentrations of heavy metals compared to Kainji Lake water. This might be responsible for the higher concentrations of these heavy metals in the organs of fishes from Jebba Lake. Heavy metal analysis of *Clarias gariepinus* fish muscles from Jebba Lake (CGFMJ) revealed higher concentrations of Pb and Cu compared to that of Kainji Lake (CGFMK). The concentration of Cd, was however, higher in fish muscles from Kainji Lake compared to that of Jebba Lake. *Clarias gariepinus* is a bottom feeder and from this study, the heavy metal concentrations in the sediment from Kainji were higher than those from Jebba Lake. This could be because of the long-standing mining activities on Kainji Lake. Hence, *Clarias gariepinus* might have acquired this cadmium from the sediment into its muscles and might have thus poor ability to excrete it. This result agrees with the report of Jonathan and Maina (2009), who stated that fish species mostly absorbed heavy metals from its feeding diets, sediment and surrounding water, resulting to their accumulation of heavy metals in reasonable amounts. However, there was no significant difference ($P > 0.05$) in the concentrations of Cr from the two Lakes. On the other hand, arsenic was not detected in the muscles of *Clarias gariepinus* from the two Lakes.

Table 4.38: Heavy Metal Concentrations in Organs of *Clarias gariepinus* in Kainji and Jebba Lakes.

		Heavy metal concentration (mg/kg)				
		Pb	**Cu**	**Cd**	**Cr**	**As**
Intestine	CGFIK	0.35 ± 0.04^{b}	1.29 ± 0.00^{b}	0.32 ± 0.00^{b}	0.14 ± 0.00^{b}	0.02 ± 0.00^{b}
	CGFIJ	0.40 ± 0.00^{a}	1.49 ± 0.00^{a}	0.35 ± 0.00^{a}	0.15 ± 0.00^{a}	0.03 ± 0.01^{a}
Gills	CGFGK	0.14 ± 0.00^{b}	0.53 ± 0.00^{b}	0.14 ± 0.00^{b}	0.08 ± 0.00^{b}	0.01 ± 0.01^{b}
	CGFGJ	0.18 ± 0.00^{a}	0.66 ± 0.00^{a}	0.15 ± 0.00^{a}	0.09 ± 0.00^{a}	0.02 ± 0.01^{a}
Muscles	CGFMK	0.02 ± 0.00^{b}	0.11 ± 0.00^{b}	0.03 ± 0.00^{a}	0.01 ± 0.00^{a}	0.00 ± 0.00^{a}
	CGFMJ	0.04 ± 0.00^{a}	0.17 ± 0.00^{a}	0.02 ± 0.00^{b}	0.01 ± 0.00^{a}	0.00 ± 0.00^{a}

Values are means of five determinations. Means with dissimilar letter (s) differ significantly according to the Duncan's Multiple Range Test (DMRT).
Significant at $P \leq 0.05$

Key: CGFIK =*Clarias gariepinus* fish intestines from Kainji Lake, CGFIJ = *Clarias gariepinus* fish intestines from Jebba Lake, CGFGK = *Clarias gariepinus* fish gills from Kainji Lake, CGFGJ = *Clarias gariepinus* fish gills from Jebba Lake, CGFMK= *Clarias gariepinus* fish muscles from Kainji, CGFMJ = *Clarias gariepinus* fish muscles from Jebba Lake.

4.53 Physicochemical Qualities of Kainji and Jebba Lake Water

Table 4.39 shows the minimum and maximum readings and means of the physico-chemical parameters of Kainji and Jebba Lake water samples. Mean temperature value of Kainji Lake was 29.80 ^{o}C while that of Jebba Lake was 28.87 ^{o}C. Mean transparency of water in Jebba Lake was 0.38 m while that of Kainji was 0.56 m. This shows that Jebba Lake water was more turbid than that of Kainji Lake. This might be due to increased human activities (including farming, washings by miners) around the Jebba Lake compared to Kainji Lake. It might also be that more run offs from farms got into Jebba Lake than Kainji Lake. This agrees with the findings of Abubakar *et al*. (2006), who reported that'reduced activity in

Geriyo Lake and complete absence of rainfall accounted for high transparency'. In addition, the size of Jebba Lake which was smaller than that of Kainji Lake, causing fast flowing of the water and, hence, might erode the banks and contribute to its turbid nature. The pH level of the two Lakes ranged from 7.0 and above. This implies that the two Lakes could support more growth of bacteria than fungi. Total alkalinity, biochemical oxygen demand, total dissolved solids and nitrate contents in Jebba Lake were higher than those of Kainji Lake (Table 4.39). This might be attributed to higher agricultural and domestic activities on Jebba Lake, including the use of fertilizers and herbicides. Kainji Lake had higher phosphate level (0.30 mg/l) than Jebba Lake (0.28 mg/l). This might be due to detergents and soaps from domestic effluents. All the parameters analysed in this study were found to be within acceptable range for aquatic life according to FMARD (2010) and WHO (2006). Correlation analysis showed that temperature, pH, conductivity, DO, total alkalinity, BOD, TDS and Nitrates are correlated with faecal coliform (FC) in Kainji Lake (Appendix Z), while transparency, pH, conductivity, DO, total alkalinity, BOD, TDS and Nitrates are correlated with faecal coliform (FC) in Jebba Lake (Appendix Z(i)).

Table 4.39: Physicochemical Qualities of Kainji and Jebba Lakes Water

Parameter	Kainji Lake		Jebba Lake		Standard
	Min. – Max.	Mean ± SD	Min. – Max.	Mean ± SD	
Temperature (^{o}C)	25.00 - 32.50	29.80±1.96	25.00-31.50	28.87±1.92	25-32[a]
Transparency (m)	0.10 - 1.40	0.56±0.34	0.04-0.80	0.38±0.22	0.3-0.6[a]
pH	6.60 - 8.00	7.30±0.27	6.80-7.80	7.20±0.23	6.5-9.0[a]
Conductivity (µS/cm)	32.00 - 78.00	49.10±11.57	16.00-74.00	48.60±11.90	<500[a]
Dissolved Oxygen (mg/l)	4.00 - 10.00	7.26±1.12	4.00-10.40	7.25±1.80	5.0-9.0[a]
Total Alkalinity (mg/l)	4.40 - 14.00	10.03±2.95	4.20-14.50	10.06±2.93	5.0-500[a]
Biological Oxygen Demand (mg/l)	0.50 - 6.00	2.10±1.44	0.20-6.60	3.30±1.64	<15.0[a]
Total Dissolved Solids (mg/l)	0.002 - 0.05	0.02±0.01	0.01-0.055	0.03±0.01	<1200[a]
Nitrate (mg/l)	6.80 - 18.77	12.70±4.12	10.20-18.80	14.30±2.82	≤50.0[b]
Phosphate (mg/l)	0.098 - 1.16	0.30±0.18	0.00-0.75	0.28±0.14	<0.5[b]

Key: a = FMARD 2010; b = WHO 2006

4.54 Physicochemical Qualities of Kainji and Jebba Lake Water based on Season

Table 4.40 shows the physicochemical qualities of Kainji and Jebba Lake water based on season. Seasonal variation in the physicochemical parameters of Kainji Lake water showed that temperature, conductivity, total alkalinity, total dissolved solids, nitrate and phosphate were higher during the wet season compared to the dry season. This might be due to run offs from adjacent farms during the wet season. Unlike Kainji Lake, Jebba Lake had higher conductivity, total alkalinity, total dissolved solids, nitrate and phosphate during the dry season than the wet season. This might be due to the fact that during the dry season, all the spill ways of Kainji Dam were open, which spilled water into the Jebba Lake, hence, increasing the volume of water in Jebba Lake and often time overflowing its bank.This agrees with the findings of Peter *et al*. (2018) who showed higher concentration of conductivity, total alkalinity, total dissolved solids and nitrate in the dry season compared to the wet season. Changes or variations observed in some parameters in the two Lakes could be attributed to the flow variability and changes in watershed conditions. This result also agrees with Ali *et al.* (2007) and Abubakar (2013), who reported that high variability of water quality might be due to the impact of many factors such as rainfall, surface runoffs and catchment activities, which prevailed during wet and dry season periods. Water volume from Futa Jallon gets into Kainji Lake after 3 months, hence, increasing the volume of its water by November, December and January, thereby leading to the opening of its spill ways. There was significant difference ($P < 0.05$) between the two seasons in Kainji Lake (with regards to temperature, nitrate, conductivity, total dissolved solids and phosphate), while pH, transparency, BOD and DO did not show any significant difference ($P > 0.05$) (Appendix V). Result of statistical analysis for temperature, transparency, pH, conductivity,

Nitrate, phosphate, DO, BOD and TDS showed that there was no significant difference ($P > 0.05$) between the two seasons in Jebba Lake (Appendix W).

Table 4.40: Physicochemical Qualities of Kainji and Jebba Lake Water based on Season

Parameter	Kainji		Jebba		Standard
	Wet Mean ± SD	Dry Mean ± SD	Wet Mean ± SD	Dry Mean ± SD	
Temperature (^{o}C)	31.10±0.88	28.60±1.96	29.95±1.27	27.80±1.86	25-32[a]
Transparency (m)	0.67±0.35	0.45±0.30	0.34±0.21	0.42±0.23	0.3-0.6[a]
pH	7.29±0.24	7.30±0.31	7.30±0.19	7.10±0.21	6.5-9.0[a]
Conductivity (µS/cm)	52.93±13.23	45.30±8.20	53.7±10.71	43.50±10.95	<500[a]
Dissolved Oxygen (mg/l)	7.21±1.00	7.31±1.25	7.69±1.65	6.80±1.86	5.0-9.0[a]
Total Alkalinity (mg/l)	11.10±2.80	9.00±2.80	11.01±2.29	9.12±3.22	5.0-500[a]
Biochemical Oxygen Demand (mg/l)	1.10±0.65	3.06±1.34	3.34±1.69	3.19±1.62	<15.0[a]
Total Dissolved Solids(mg/l)	0.03±0.01	0.02±0.01	0.03±0.008	0.02±0.01	<1200[a]
Nitrate (mg/l)	15.70±3.40	9.70±2.10	16.87±0.96	11.64±1.10	≤50.0[b]
Phosphate (mg/l)	0.34±0.15	0.25±0.19	0.34±0.14	0.20±0.08	<0.5[b]

Key: a = FMARD 2010; b = WHO 2006

4.55 Mean Physicochemical Qualities of Water from Sample Stations in Kainji Lake

It was observed in Table 4.41 that station 1 (Kl.1) of Kainji Lake had the highest conductivity (51.25 µS/cm) and total dissolved solids content (0.03 mg/l). Sample station 1 (Kl.1) lies adjacent Warra market and might receive wastes of dissolved solids and salts from the market. Station 3 (Kl.3) had the highest biological oxygen demand (2.39 mg/l) and high phosphate (0.37 mg/l). Sample station 3, which is at Yunawa, had high animal husbandry operation (thereby releasing organic manure into the Lake). It was also characterized by high domestic washings, which might have caused increased phosphate content. This result agrees with Chukwu and Nwanko (2003), who reported that high variability of water quality might be due to the impact of factors such as rainfall, surface runoffs and catchment activities. Station 4 (Kl.4) had the highest temperature and nitrate. However, there was no significance difference ($P > 0.05$) in the physicochemical parameters determined in all the sample stations.

Table 4.41: Mean Physicochemical Qualities of Water from Sample Stations in Kainji Lake

Sample station	Temperature ^{o}C	Transp. (m)	pH	Cond. (µS/cm)	DO (mg/l)	TA (mg/l)	BOD (mg/l)	TDS (mg/l)	Nitrate (mg/l)	Phosphate (mg/l)
Kl. 1	29.50±2.05[a]	0.46±0.30[a]	7.31±0.25[a]	51.25±12.3[a]	7.00±0.8[a]	10.13±2.93[a]	1.78±1.4[a]	0.03±0.0[a]	12.11±4.19[a]	0.30±0.1[a]
Kl. 2	29.67±2.05[a]	0.58±0.28[a]	7.35±0.24[a]	46.00±10.6[a]	6.91±1.4[a]	9.34±2.69 [a]	2.05±1.3[a]	0.02±0.0[a]	12.25±4.45[a]	0.30±0.1[a]
Kl. 3	29.90±2.18[a]	0.60±0.35[a]	7.32±0.21[a]	49.83±13.5[a]	7.82±1.1[a]	9.78±2.90 [a]	2.39±1.7[a]	0.02±0.0[a]	12.91±4.32[a]	0.37±0.2[a]
Kl. 4	29.96±1.93[a]	0.57±0.34[a]	7.26±0.29[a]	48.75±10.6[a]	6.99±1.1[a]	10.20±3.14 [a]	2.09±1.4[a]	0.02±0.0[a]	13.03±4.39[a]	0.27±0.1[a]
Kl. 5	30.04±1.96 [a]	0.63±0.42[a]	7.25±0.38[a]	48.67±12.0[a]	7.58±0.8[a]	10.43±3.45 [a]	2.08±1.5[a]	0.02±0.0[a]	13.00±4.10[a]	0.24±0.1[a]

Values on the same column with different superscript are significantly different ($p \leq 0.05$).

Key: DO: Dissolved Oxygen, TA: Total Alkalinity, BOD: Biological Oxygen Demand, TDS: Total Dissolved Solids, Transp: Transparency, Cond: Conductivity

4.56 Mean Physicochemical Qualities of Water from Sample Stations in Jebba Lake Water

The physico-chemical parameters of water from Jebba sample stations showed that temperature, conductivity, total alkalinity, biological oxygen demand and phosphate were higher in sample station 2 (Jl.2) of the Lake than all other stations (Table 4.42). This might be due to high human activities in station 2 above that of other stations, such as domestic washings, washing of ore pulp by miners, washing of automobiles and even its proximity to agricultural farms. This might have caused increase of these parameters above that of other stations. Station 4 (Jl.4) had higher nitrate content (14.88mg/l) and pH level (7.22) than other sample stations of Jebba Lake. Station 4 (Jl.4) at Old Awuru lies adjacent Old Awuru market, hence, might have received more organic pollutants as wastes from the market, thus, increasing nitrate and pH of the water. Statistical analysis showed that there was no significant difference ($P > 0.05$) between sample stations.

Table 4.42: Mean Physicochemical Qualities of Water from Sampling Stations in Jebba Lake Water

Sample station	Temperature ^{o}C	Transp. (m)	pH	Cond. (µS/cm)	DO (mg/l)	TA (mg/l)	BOD (mg/l)	TDS (mg/l)	Nitrate (mg/l)	Phosphate (mg/l)
Jl. 1	28.88±2.00[a]	0.42±0.2[a]	7.20±0.1[a]	45.50±10.01[a]	7.14±1.9[a]	9.75±3.01[a]	3.38±1.76[a]	0.03±0.01[a]	13.78±2.82[a]	0.31±0.1[a]
Jl. 2	28.92±1.9[a]	0.32±0.1[a]	7.20±0.3[a]	53.17±12.83[a]	7.05±1.85[a]	10.56±3.27[a]	3.61±1.64[a]	0.03±0.01[a]	14.61±2.82[a]	0.34±0.1[a]
Jl. 3	28.88±1.96[a]	0.38±0.2[a]	7.16±0.2[a]	44.33±11.81[a]	7.20±1.56[a]	10.26±3.08[a]	2.55±1.79[a]	0.03±0.01[a]	14.09±2.80[a]	0.29±0.1[a]
Jl. 4	28.73±2.0[a]	0.36±0.2[a]	7.22±0.2[a]	49.91±8.23[a]	7.24±2.11[a]	10.07±2.54[a]	3.60±1.55[a]	0.03±0.008[a]	14.88±2.87[a]	0.27±0.1[a]
Jl. 5	28.68±2.05[a]	0.37±0.2[a]	7.19±0.2[a]	51.45±15.36[a]	7.58±1.89[a]	9.99±2.97[a]	3.47±1.57[a]	0.03±0.01[a]	14.00±3.15[a]	0.24±0.1[a]

Values on the same column with different superscript are significantly different ($p \leq 0.05$).

KEY: DO: Dissolved Oxygen, TA: Total Alkalinity, BOD: Biochemical Oxygen Demand, TDS: Total Dissolved Solids, Transp: Transparency, Cond: Conductivity

4.57 Bacteria Screened for Biosorption Potentials

Table 4.43 shows the results of some non-pathogenic bacterial species, which were selected and screened for biosorption analysis of lead, copper, cadmium, chromium and arsenic. It was observed that *Pseudomonas donghuensis* strain HYS and *Aeromonas aquatilis* AE 207 were sensitive to lead and, hence, could not tolerate it. In addition, *Alcaligenes faecalis* strain HPRTAK 198 was highly sensitive to cadmium and thus could not tolerate and remove cadmium. *Bacillus lacus* strain AK 74, *Herbaspirillum aquaticum* strain IEH 4433, *Alcaligenes faecalis* strain sihong 663 and *Oceanobacillus oncorhynchi* subsp. *incaldanensis* strain AM 75 were not sensitive to all the heavy metals tested. Hence these bacteria could be used in biosorption of all the selected heavy metals (Pb, Cu, Cd, Cr, As).

Table 4.43: Bacteria Screened for Biosorption Potentials

S/N	Bacteria	Pb	Cu	Cd	Cr	As
1	*Pseudomonas donghuensis* strain *HYS*	+	-	-	-	-
2	*Aeromonas aquatilis AE* 207	+	-	-	-	-
3	*Alcaligenes faecalis* strain *HPRTAK* 198	-	-	+	-	-
4	*Bacillus lacus* strain *AK* 74	-	-	-	-	-
5	*Herbaspirillum aquaticum* strain *IEH* 4433	-	-	-	-	-
6	*Alcaligenes faecalis* strain sihong 663	-	-	-	-	-
7	*Oceanobacillus oncorhynchi* subsp. *Incaldanensis* strain AM 75	-	-	-	-	-

Key: - = Not sensitive/ Ability to tolerate heavy metal hence development of bacterial colony

+ = Sensitive/ Inability to tolerate heavy metal hence no development of bacterial colony

4.58 Biosorption of Heavy Metals by *Pseudomonas donghunsis* strain HYS

Biosorption of copper, cadmium, chromium and arsenic by *Pseudomonas donghunsis* strain HYS was studied at different concentrations of the tested heavy metals (1.0ppm, 3.0 ppm and 5.0 ppm) and different contact time (7 days, 14 days, 21 days, 28 days) (Table 4.44). The highest percentage removal of Cu, Cd, Cr, As was observed at 1.0 ppm as 81.5%, 62.0%, 86.5% and 68.5% respectively while the lowest metal removal was observed at a concentration of 5.0 ppm as 25.5%, 13.68%, 56.3% and 18.5% respectively. Changes in percentage removal varied in the order of 1.0 ppm > 3.0 ppm > 5.0 ppm. In percentage removal of heavy metals with respect to contact time, it was observed that day 7 generally recorded the highest percentage removal for all the metals studied. The rate of biosorption in this order: 1.0 ppm > 3.0 ppm > 5.0 ppm shows decrease in sorption rate as metal concentration increases. This result agrees with the findings of Puranik and Paknikar (2009) as well as Abioye *et al*. (2015), who reported that an increase in metal concentration beyond the optimum level in a biosorption experiment resulted in a retardation of the sorption process due to the fact that the metal binding site on the sorbent becomes saturated, leaving no space for more molecules to occupy. Percentage removal of heavy metals, which was also higher in day 7 than in day 14, 21 and 28, might be due to the interplay of more binding sites from day 1 – 7 and its gradual saturation with increase in contact time from day 7 – 28.

Pseudomonas donghunsis strain HYS was found to remove heavy metals in this order: Chromium > Copper > Arsenic > Cadmium, which means it (*Pseudomonas donghunsis* strain HYS) has greater affinity for chromium.

Table 4.44: Biosorption of Heavy Metals by *Pseudomonas donghunsis* strain HYS

Time (Day)	Cu			Cd			Cr			As		
1	1.00 ppm	3.00 ppm	5.00 ppm	1.00 ppm	3.00 ppm	5.00 ppm	1.00 ppm	3.00 ppm	5.00 ppm	1.00 ppm	3.00 ppm	5.00 ppm
7	0.410	2.485	4.525	0.620	2.815	4.668	0.300	2.012	3.506	0.682	2.528	4.235
14	0.350	2.162	4.300	0.445	2.776	4.546	0.210	1.650	2.850	0.456	2.342	4.156
21	0.220	2.096	4.146	0.415	2.676	4.358	0.160	1.400	2.416	0.380	2.285	4.088
28	0.185	2.046	3.725	0.380	2.500	4.316	0.135	1.090	2.185	0.315	2.225	4.075
% R	81.5	31.8	25.5	62.0	16.7	13.68	86.5	63.7	56.3	68.5	25.8	18.5

Key: ppm = part per million, % R = Percentage removal of heavy metal

4.59 Biosorption of Heavy Metals by *Aeromonas aquatilis* strain AE 207

Table 4.45 shows biosorption potentials of *Aeromonas aquatilis* strain AE 207 on copper, cadmium, chromium and arsenic. Highest percentage removal of copper, cadmium, chromium and arsenic by *Aeromonas aquatilis* strain AE 207 was observed at 1.0 ppm concentration and these were 93.5%, 92.5%, 84.0% and 72.3% respectively. The percentage removal followed this order: Cu > Cd > Cr > As. The lowest percentage removal was observed at 5.0 ppm. For percentage removal of heavy metals with respect to contact time, it was observed that higher percentage was removed within the first 7 days compared to day 14, 21 and 28. The higher percentage removal of heavy metals at lower concentration (1.0 ppm) than at higher concentration (5.0 ppm) agrees with the report of Meenambigail *et al*. (2016), who stated that microorganisms play an important role in bioremediation of heavy metals from contaminated soil and wastewater, but when microorganisms are exposed to higher concentrations of heavy metals, this might have a deleterious effects on their growth and activities.

Table 4.45: Biosorption of Heavy Metals by *Aeromonas aquatilis* strain AE 207

Time (Day)	Cu			Cd			Cr			As		
1	1.00 ppm	3.00 ppm	5.00 ppm	1.00 ppm	3.00 ppm	5.00 ppm	1.00 ppm	3.00 ppm	5.00 ppm	1.00 ppm	3.00 ppm	5.00 ppm
7	0.450	1.251	2.241	0.402	2.010	3.014	0.420	1.950	2.895	0.602	2.050	3.612
14	0.250	0.812	2.132	0.210	1.602	2.565	0.320	1.752	2.401	0.410	1.715	3.085
21	0.125	0.716	1.902	0.110	1.305	2.270	0.200	1.421	2.325	0.340	1.540	2.740
28	0.065	0.625	1.852	0.075	1.185	2.055	0.160	1.305	2.300	0.277	1.460	2.618
% R	93.5	79.2	63.0	92.5	60.5	58.9	84.0	56.5	54.0	72.3	51.3	47.6

Key: ppm = part per million, % R = Percentage removal of heavy metal

4.60 Biosorption of Heavy Metals by *Alcaligenes faecalis* HPRTAK 198

Biosorption of heavy metals by *Alcaligenes faecalis* HPRTAK 198 (Table 4.46) showed that high percentage removal of lead, copper, chromium and arsenic were observed at low concentration (1.0 ppm) as 72.5 %, 91.0 %, 95.0 % and 99.4 % respectively compared to higher concentration (5.0 ppm), which were 61.9 %, 66.0 %, 70.4 % and 79.5 % respectively. Decrease in percentage removal of heavy metals as its concentration increased might be due to decrease in binding site. This result agrees with the findings of Puranik and Paknikar (2009), who reported that in a biosorption setup, increase in metal concentration above optimum level brings about retardation of sorption rate. This is due to the fact that the metal binding sites on the sorbent become saturated, leaving no space for more molecules of the metal to bind. Percentage removal was in this order: As > Cr > Cu > Pb. *Alcaligenes faecalis* HPRTAK 198 showed highest percentage removal (99.4 %) of arsenic compared to other metals. This is an indication that *Alcaligenes faecalis* HPRTAK 198 had greater affinity and ability to utilize arsenic than the other metals. *Alcaligenes faecalis* HPRTAK 198 is a Gram negative bacterium with potentials to biodegrade organic and inorganic compounds. They are considered to be non – pathogenic.

Table 4.46 Biosorption of Heavy Metals by *Alcaligenes faecalis* HPRTAK 198

Time(Day)	Pb			Cu			Cr			As		
1	1.00 ppm	3.00 ppm	5.00 ppm	1.00 ppm	3.00 ppm	5.00 ppm	1.00 ppm	3.00 ppm	5.00 ppm	1.00 ppm	3.00 ppm	5.00 ppm
7	0.310	1.800	2.600	0.350	1.020	2.340	0.400	1.095	2.420	0.250	2.005	2.718
14	0.290	1.512	2.305	0.280	1.015	2.130	0.210	0.775	2.010	0.130	1.060	2.511
21	0.280	1.221	2.098	0.160	0.816	1.910	0.116	0.420	1.650	0.102	0.606	1.206
28	0.275	1.010	1.905	0.090	0.612	1.700	0.050	0.375	1.480	0.006	0.300	1.026
% R	72.5	66.3	61.9	91.0	79.6	66.0	95.0	87.5	70.4	99.4	90.4	79.5

Key: ppm = part per million, % R = Percentage removal of heavy metal

4.61 Biosorption of Heavy Metals by *Bacillus lacus* strain AK 74

Biosorption of heavy metals by *Bacillus lacus* strain AK 74 shows that there were significant removal of lead, copper, cadmium, chromium and arsenic (Table 4.47). Percentage removal followed this order: Pb > Cu > Cr >As > Cd. It was also observed that at lower concentration (1.0 ppm) of heavy metals, percentage removal was higher (99.0 %, 98.0 %, 96.5 %, 96.0 % and 94.5 % respectively) than at higher concentration (5.0 ppm) which was observed to be 74.4 %, 74.2 %, 58.2 %, 48.2 % and 42.0 % respectively. *Bacillus lacus* strain AK 74 recorded higher percentage removal of lead (99.0 %) when compared to chromium (96.5 %) at 1.0 ppm. This result agrees with the findings of Abioye *et al*. (2018), where *Bacillus* species had higher uptake level of lead in tannery effluent compared to uptake of chromium. It was generally observed that *Bacillus lacus* AK 74 was a strong biosorbent for lead, copper, cadmium, chromium and arsenic with over 90.0 % removal within 28 days. Hydrolytic enzymes could be secreted by *Bacillus* species, which are capable of tolerating heavy metals (Abioye *et al*., 2018).

Bacillus comprises more than 300 species for those that have been vividly published (Harjodh *et al.,* 2018). *Bacillus lacus* strain AK 74 is a Gram positive, motile, spore-forming aerobic bacterium or facultatively anaerobic bacterium. It could be found in water, soil and in the air. They are alkaliphilic or alkalitolerant species.

Table 4.47 Biosorption of Heavy Metals by *Bacillus lacus* strain AK 74

Time (Day)	Pb			Cu			Cd			Cr			As		
1	1.00 ppm	3.00 ppm	5.00 ppm	1.00 ppm	3.00 ppm	5.00 ppm	1.00 ppm	3.00 ppm	5.00 ppm	1.00 ppm	3.00 ppm	5.00 ppm	1.00 ppm	3.00 ppm	5.00 ppm
7	0.310	1.125	1.508	0.300	1.012	1.826	0.260	2.000	3.525	0.340	1.136	3.615	0.328	2.000	3.015
14	0.185	0.730	1.307	0.150	0.901	1.415	0.145	1.520	3.180	0.106	0.767	2.600	0.200	1.532	2.680
21	0.050	1.106	2.218	0.060	0.702	1.320	0.095	1.240	2.900	0.040	0.750	0.450	0.065	1.110	2.625
28	0.010	0.316	1.082	0.020	0.446	1.290	0.055	1.110	2.900	0.035	0.506	2.090	0.040	0.725	2.590
% R	99.0	89.5	74.4	98.0	85.1	74.2	94.5	63.0	42.0	96.5	83.1	58.2	96.0	75.8	48.2

Key: ppm = part per million, % R = Percentage removal of heavy metal

4.62 Biosorption of Heavy Metals by *Herbaspirillum aquaticum* strain IEH 4430

Table 4.48 shows the biosorption potential of *Herbaspirillum aquaticum* strain IEH 4430 on heavy metals; Pb, Cu, Cd, Cr and As. Percentage removal of the metal follows this order: As > Cu > Cr > Pb > Cd and these were 75.5 %, 65.5 %, 60.0 %, 48.4 %, and 39.8 % at 1.0 ppm respectively. Highest percentage removal was observed at 1.0 ppm concentration compared to 5.0 ppm concentration in all the heavy metals analyzed. The decrease in percentage removal of the heavy metals as concentration increased might be due to decrease in binding site. This agrees with the findings of Puranik and Paknikar (2009), who reported that in a biosorption setup, increase in metal concentration beyond optimum level results in a retardation of sorption process due to increasing saturation of binding site, thereby leaving no space for more molecules of the metal to bind. *Herbaspirillum aquaticum* IEH 4430 has greater affinity for arsenic, hence, removal rate for arsenic was highest (75.5 %).

Table 4.48 Biosorption of Heavy Metals by *Herbaspirillum aquaticum* strain IEH 4430

Time (Day)	Pb			Cu			Cd			Cr			As		
1	1.00 ppm	3.00 ppm	5.00 ppm	1.00 ppm	3.00 ppm	5.00 ppm	1.00 ppm	3.00 ppm	5.00 ppm	1.00 ppm	3.00 ppm	5.00 ppm	1.00 ppm	3.00 ppm	5.00 ppm
7	0.815	2.685	4.525	0.700	2.185	3.852	0.812	2.815	4.668	0.740	2.348	4.342	0.650	2.008	3.000
14	0.698	2.400	4.300	0.525	2.062	3.450	0.704	2.776	4.546	0.568	2.180	4.236	0.410	1.615	2.452
21	0.584	2.252	4.146	0.418	1.826	3.280	0.618	2.676	4.358	0.430	1.976	4.182	0.295	1.305	2.260
28	0.516	2.105	4.125	0.345	1.576	3.095	0.602	2.500	4.316	0.400	1.846	3.995	0.245	1.180	2.180
% R	48.4	29.8	17.5	65.5	47.5	38.1	39.8	16.7	13.8	60.0	38.5	20.1	75.5	60.7	56.4

Key: ppm = part per million, % R = Percentage removal of heavy metal

4.63 Biosorption of Heavy Metals by *Alcaligenes faecalis* strain sihong 663

Percentage biosorption of heavy metals by *Alcaligenes faecalis* strain sihong 663 was in the order: Arsenic > Copper > chromium > lead > cadmium (Table 4.49). The highest percentage removal was observed at 1.0 ppm concentration in all the heavy metals analyzed. Highest percentage removal was observed in arsenic, that was from 1.0 ppm to 0.005 ppm after 28 days. This indicated that *Alcaligenes faecalis* strain sihong 663 had greater affinity for arsenic than other metals. Other heavy metal that was tolerated and reduced to a permissible value of 0.025 ppm after 28 days was copper.

Batt (2014) reported that *Alcaligenes faecalis* have potentials for biodegradation of organic and inorganic compounds. Aly *et al*. (2015) shows that strains of *Alcaligenes faecalis* manifest high resistant and multiple tolerances to heavy metals (Pb, Cd, Al, Cu, Ag, Sn).

Alcaligenes faecalis strain Sihong 663, being non pathogenic bacteria, which is commonly found in water and soil, could be a useful biosorbent for arsenic and copper polluted environment.

Table 4.49: Biosorption of Heavy Metals by *Alcaligenes faecalis* strain sihong 663

Time (Day)	Pb			Cu			Cd			Cr			As		
1	1.00 ppm	3.00 ppm	5.00 ppm	1.00 ppm	3.00 ppm	5.00 ppm	1.00 ppm	3.00 ppm	5.00 ppm	1.00 ppm	3.00 ppm	5.00 ppm	1.00 ppm	3.00 ppm	5.00 ppm
7	0.300	2.206	3.896	0.110	1.125	2.508	0.605	2.581	3.802	0.450	2.035	3.906	0.311	0.502	2.315
14	0.255	2.008	3.612	0.095	0.601	2.106	0.501	2.310	3.608	0.350	1.605	3.308	0.155	0.420	1.208
21	0.226	1.862	3.508	0.058	0.390	2.056	0.400	2.185	3.185	0.214	1.310	3.006	0.018	0.280	0.935
28	0.198	1.712	3.416	0.025	0.275	1.285	0.325	2.002	3.480	0.112	1.116	2.820	0.005	0.245	0.850
% R	80.2	42.9	31.7	97.5	90.8	74.3	67.5	33.3	30.4	88.8	62.8	43.6	99.5	91.8	83.0

Key: ppm = part per million, % R = Percentage removal of heavy metal

4.64 Biosorption of Heavy Metals by *Oceanobacillus oncorhynchi* subsp. *incaldanensis* strain AM -75

Table 4.50 shows results of the biosorption potential of *Oceanobacillus oncorhynchi* subsp. *incaldanensis* strain AM -75. The highest percentage removal of lead, copper, cadmium, chromium and arsenic were observed at 1.0 ppm concentration as 95.8 %, 98.8 %, 71.0 %, 91.5 % and 96.6 % respectively. Percentage removal of the heavy metals follows this order: Cu > As > Pb > Cr > Cd. Percentage removal/sorption rate of heavy metals was higher at 1.0 ppm concentration compared to the concentration at 5.0 ppm. This reveals that the higher the heavy metal concentrations, the lower the percentage removal of the heavy metal in an environment. This result agrees with the findings of Vijayakumar *et al*. (2011), who reported that percentage cadmium uptake decreased with increase in initial concentration of cadmium. Decrease in percentage sorption as concentration increases might be attributed to reduction in the number of available functional groups (amino, carboxyl, hydroxyl and carbonyl) on the absorbent as the initial concentration was increased (Vijayakumar *et al*., 2011).

Table 4.50 Biosorption of Heavy Metals by *Oceanobacillus oncorhynchi* subsp. *incaldanensis* strain AM -75

Time (Day)	Pb			Cu			Cd			Cr			As		
1	1.00 ppm	3.00 ppm	5.00 ppm	1.00 ppm	3.00 ppm	5.00 ppm	1.00 ppm	3.00 ppm	5.00 ppm	1.00 ppm	3.00 ppm	5.00 ppm	1.00 ppm	3.00 ppm	5.00 ppm
7	0.250	1.300	3.988	0.110	0.310	2.224	0.422	2.085	3.308	0.320	1.525	2.900	0.210	1.225	3.950
14	0.125	1.085	2.625	0.052	0.177	1.188	0.308	1.680	3.105	0.280	1.208	2.625	0.120	1.106	3.625
21	0.080	0.950	2.320	0.030	0.072	0.870	0.295	1.400	3.005	0.150	0.992	2.330	0.060	0.800	2.400
28	0.042	0.680	2.098	0.012	0.041	0.152	0.290	1.260	2.995	0.085	0.702	2.150	0.034	0.615	2.300
% R	95.8	77.3	58.0	98.8	98.6	97.0	71.0	58.0	40.1	91.5	76.6	57.0	96.6	79.5	54.0

Key: ppm = part per million, % R = Percentage removal of heavy metal

CHAPTER FIVE

5.0 CONCLUSION AND RECOMMENDATIONS

5.1 Conclusion

This study revealed the presence of faecal coliforms in both Kainji and Jebba Lakes and bacterial pathogens (*Vibrio alginolyticus*, *Mannheimia haemolytica*, *Aeromonas hydrophila*, *Pseudomonas fluorescens* – 35, *Aeromonas veronii bio sobria*, *Aeromonas caviae*, *Pseudomonas aeruginosa*, *Pasturella multocida*, *Vibrio parahaemolyticus* and *Yersinia pestis*) were isolated from water, sediment and fish samples from both Lakes; however, no pathogen was isolated from the fish muscles.

Heavy metals (Pb, Cu, Cr, As) concentrations in the water were generally observed to be below maximum permissible limit in the water samples. However, the concentration of cadmium (Cd) was higher than the maximum permissible limit. It was generally observed that the concentration of heavy metal in water samples were higher during the wet season compared to the dry season. Heavy metal concentrations in sediments from Kainji Lake had higher concentrations than Jebba Lake. However, the concentrations in sediments from both Lakes were generally found to be below threshold effect concentration (TEC). Concentration of heavy metals in fish samples revealed that the intestines of both *Oreochromis niloticus* and *Clarias gariepinus* had lead (Pd) content, which is above maximum permissible limit. Arsenic content was observed to be above maximum permissible limit in some sample stations. The gills of *Oreochromis niloticus* from Kainji Lake had high arsenic (As) content and lead (Pb) content above maximum permissible limit in some stations. Copper, cadmium, chromium were found to be within acceptable limit. Fish muscles of both *Oreochromis niloticus* and *Clarias gariepinus* had low concentration

(concentration below maximum permissible limit) of all the heavy metals analysed. It therefore means that edible part (muscle) of the fishes is safe for human consumption.

Physicochemical qualities of all the parameters examined in the water bodies were found to be within acceptable range that is favourable for aquatic lives.

Biosorption potentials of bacterial isolates revealed that *Bacillus lacus* strain AK 74, had high potential to biosorp Pb, Cu, Cd, Cr and As, while *Oceanobacillus oncorhynchi* subsp. *incaldanensis* strain AM -75 had high potential to biosorp Pb, Cu, Cr and As. *Alcaligenes faecalis* HPRTAK 198 had high potential to biosorp Cu, Cr and As while *Alcaligenes faecalis* strain sihong 663 had high potential to biosorp Cu and As. *Aeromonas aquatilis* strain AE 207 had high potential to biosorp Cu and Cd.

5.2 Recommendations

1. Although no bacterial pathogen was isolated from the fish muscles (edible part of the fish), but from other organs, it is advisable that these fish species from the Lakes be subjected to proper handling (De-gutting, washing, smoking, boiling) before consumption. This is to prevent cross contamination.

2. Mining activities should follow international best practices so as to reduce heavy metal pollution of surface water bodies.

3. Public health awareness should be carried out to enlighten the community on the danger of indiscriminate dumping of wastes into surface water bodies and illegal mining activities.

4. Bacterial species such as *Bacillus lacus* strain AK 74, *Oceanobacillus oncorhynchi* subsp. *incaldanensis* strain AM -75, *Alcaligens faecalis* HPRTAK 198, *Alcaligenes faecalis* strain sihong 663 and *Aeromonas aquatilis* strain AE 207 be used in clean up/sequesteration process of the respective heavy metals polluted environment.

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APPENDICES

APPENDIX A: t-Test for Seasonal Faecal coliform count of water from Kainji Lake

FC – KL
t-Test: Paired Two Sample for Means

	Wet	***Dry***
Mean	342.5333333	125.6333333
Variance	20805.98161	9624.033333
Observations	30	30
Pearson Correlation	0.713844092	
Hypothesized Mean Difference	0	
Df	29	
t Stat	11.74724485	
P(T<=t) one-tail	7.58498E-13	
t Critical one-tail	1.699127027	
P(T<=t) two-tail	1.517E-12	
t Critical two-tail	2.045229642	

APPENDIX B: t-Test for Seasonal Faecal coliform count of water from Jebba Lake

FC – JL
t-Test: Paired Two Sample for Means

	Wet	***Dry***
Mean	670.4666667	197.9666667
Variance	181289.292	39899.55057
Observations	30	30
Pearson Correlation	0.666899602	
Hypothesized Mean Difference	0	
Df	29	
t Stat	7.884118453	
P(T<=t) one-tail	5.37587E-09	
t Critical one-tail	1.699127027	
P(T<=t) two-tail	1.07517E-08	
t Critical two-tail	2.045229642	

APPENDIX C: t-Test for Seasonal viable bacterial count (VBC) of water from Kainji Lake.

TVC – KL

t-Test: Paired Two Sample for Means

	Wet	***Dry***
Mean	548846.6667	62133.33333
Variance	8.13298E+11	6018796202
Observations	30	30
Pearson Correlation	0.175372118	
Hypothesized Mean Difference	0	
Df	29	
t Stat	2.990275031	
P(T<=t) one-tail	0.002817092	
t Critical one-tail	1.699127027	
P(T<=t) two-tail	0.005634185	
t Critical two-tail	2.045229642	

APPENDIX D: t-Test for Seasonal viable bacterial count (VBC) of water from Jebba Lake.

TVC- JL

t-Test: Paired Two Sample for Means

	Wet	***Dry***
Mean	554340	66126
Variance	8.2336E+11	11512610590
Observations	30	30
Pearson Correlation	-0.141713823	
Hypothesized Mean Difference	0	
Df	29	
t Stat	2.879383304	
P(T<=t) one-tail	0.003706396	
t Critical one-tail	1.699127027	
P(T<=t) two-tail	0.007412792	
t Critical two-tail	2.045229642	

APPENDIX E: Sequence of *Aeromonas aquatilis* strain AE207 partial 16S rRNA gene.

CTCAGCGGATTATACTCATTGACATGCAGTCGAGCGGCAGCGGGAAAGTA
GCTTGCTACTTTTGCCGGCGAGCGGCGGACGGGTGAGTAATGCCTGGGGA
TCTGCCCAGTCGAGGGGGATAACTACTGGAAACGGTAGCTAATACCGCAT
ACGCCCTACGGGGGAAAGCAGGGGACCTTCGGGCCTTGCGCGATTGGATG
AACCCAGGTGGGATTAGCTAGTTGGTGAGGTAATGGCTCACCAAGGCGAC
GATCCCTAGCTGGTCTGAGAGGATGATCAGCCACACTGGAACTGAGACAC
GGTCCAGACTCCTACGGGAGGCAGCAGTGGGGAATATTGCACAATGGGGG
AAACCCTGATGCAGCCATGCCGCGTGTGTGAAGAAGGCCTTCGGGTTGTA
AAGCACTTTCAGCGAGGAGGAAAGGTTGGTAGCTAATAACTGCCAGCTGT
GACGTTACTCGCAGAAGAAGCACCGGCTAACTCCGTGCCAGCAGCCGCGG
TAATACGGAGGGTGCAAGCGTTAATCGGAATTACTGGGCGTAAAGCGCAC
GCAGGCGGTTGGATAAGTTAGATGTGAAAGCCCCGGGCTCAACCTGGGAA
TTGCATTTAAAACTGTCCAGCTAGAGTCTTGTAGAGGGGGGTAGAATTCC
AGGTGTAGCGGTGAAATGCGTAGAGATCTGGAGGAATACCGGTGGCGAAG
GCGGCCCCCTGGACAAAGACTGACGCTCAGGTGCGAAAGCGTGGGGAGCA
AACAGGATTAGATACCCTGGTAGTCCACGCCGTAAACGATGTCGATTTGG
AGGCTGTGTCCTTGAGACGTGGCTTCCGGAGCTAACGCGTTAAATCGACC
GCCTGGGGAGTACGGCCGCAAGGTTAAGACTCATATGAATTGACGGGGGC
CCGCACAAGCGGTGGAGCATGTGGTTTAATTCGATGCAACGCGAAGAACC
TTACCTGGCCTTGACATGTCTGGAATCCTGTAGAGATACGGGAGTGCCTT
CGGGAATCAGAACACAGGTGCTGCATGGCTGTCGTCACCTCCTGTCCTGA
GATGTTGGGTTAGTCCCGCAACGAGCGCAACCCCTGTCCTTGTTGCAGCA
CGTAATGGGGGAACTCAGGGAGACTGCCGTGATAACCGGAGAAAGTGGGG
ATGACGTCAAGTCATCATGTCCTTACGGCAGGCTACCACGTGCTACATGG
CGGTTCGAAGGCTGCAGCTACGAACTGACAATCCGAAAGCCGTCGAATCC
GACGGGGCGGGCCCCTCTCCCCCCACCCCCAGGGGGAAATTAAAATTTTT
TTTATATTTTTTAGGCCCGGAAAGGCCCTCCCCCGGGTTATCCCCCCCCC
CCCCGAGGGGGGGTTGCAAACTTTCCCCCTTTGGGGGGGGGGGTCGCGGC
GAAAAAAAAAGAAGAATGAAAAATAAAAAAAAATAAAAATAAAATATAAA
AAAAAAAAAAGAGGGGGTTATAGAATGAAAAACAAAAAAAATAGAAGAAA
AAAGACAAAAAAAAAAAAAAAAAAGCACGCCGGGCGTTTTCCTTTTCTGC
CCGGCCCCTTTCCGCCCCTTCCTCGCCCCGCCACAAAAACACAAAAAAAA
ATAAAAATAAAAAAAAAAAACAACAAGGC

APPENDIX F: Partial sequence of *Bacillus lacus* strain AK74 16S rRNA

GTCCGAAGAAAGTAAACACTGCTACATGCAGTCGAGCGGACAGAAGGGAG
CTTGCTCCCGGATGTTAGCGGCGGACGGGTGAGTAACACGTGGGTAACCT
GCCTGTAAGACTGGGATAACTCCGGGAAACCGGAGCTAATACCGGATAGT
TCCTTGAACCGCATGGTTCAAGGATGAAAGACGGTTTCGGCTGTCACTTA
CAGATGGACCCGCGGCGCATTAGCTAGTTGGTGGGGTAATGGCTCACCAA
GGCGACGATGCGTAGCCGACCTGAGAGGGTGATCGGCCACACTGGGACTG
AGACACGGCCCAGACTCCTACGGGAGGCAGCAGTAGGGAATCTTCCGCAA
TGGACGAAAGTCTGACGGAGCAACGCCGCGTGAGTGATGAAGGTTTTCGG
ATCGTAAAGCTCTGTTGTTAGGGAAGAACAAGTGCGAGAGTAACTGCTCG
CACCTTGACGGTACCTAACCAGAAAGCCACGGCTAACTACGTGCCAGCAG
CCGCGGTAATACGTAGGTGGCAAGCGTTGTCCGGAATTATTGGGCGTAAA
GGGCTCGCAGGCGGTTTCTTAAGTCTGATGTGAAAGCCCCCGGCTCAACC
GGGGAGGGTCATTGGAAACTGGGAAACTTGAGTGCAGAAGAGGAGAGTGG
AATTCCACGTGTAGCGGTGAAATGCGTAGAGATGTGGAGGAACACCAGTG
GCGAAGGCGACTCTCTGGTCTGTAACTGACGCTGAGGAGCGAAAGCGTGG
GGAGCGAACAGGATTAGATACCCTGGTAGTCCACGCCGTAAACGATGAGT
GCTAAGTGTTAGGGGGTTTCCGCCCCTTAGTGCTGCAGCTAACGCATTAA
GCACTCCGCCTGGCGAGTACGGTCGCAAGACTGAGACTCACAGGAATTGA
CGGGAGCCCGCACAAGCGGTGGAGCATGTGGTTTAATTCGAAGCAACGCG
AAGAACCTTACCAGGTCTTGACATCCTCTGACAACACTAGAGATAGGGCT
TTCCTTCGCGAACAGAGTGACAGGTGATGCATGGTTGTCGTCTGCTCCTG
TCCTGAGATGTTGAGTTAAGTCCCGCAACCCGCGCACCCCTGATCTAGTT
GCATCATTCAATTGGTCCTCTAGGAGACTGCCGCGACCAGCCGCAGAAAG
GGGGGGGGCGTCGGTCTCATCCCCTTTGACCTGGCTACACGGGGTACATG
ACGGACTAGGGTGAAAAACCCCACCTAAACAATGGGTGGTCCGTCTTTTT
TTTTTTTATCCCAACCAACCCTTTCCCGGCGAGGGGGGAAAAAAATTTTG
GGGGAACCTGGCGGCGTGTTACCCCCCTCCCGCCGCGCCCCCCCCCCCCC
TCCCCGCCCTAACGGGACTTCTTTTTTTTTGGGGGGGGGGTGCATAGAAT
AAAAAAAAAAAATAAAAAAAAAAAAAAATGGCCCGCGTAAAAAAAAAAAA
AAAAAAAAAAAAAAAAAAAAAAAAAAAAAATCTTTTTTTTTCTTTTCTCC
CGCCCTTTCTCCTTTCTTTCTTTATATAAAAAAACAAATAAAAAATAATA
AACAAAAAAAATA

APPENDIX G: *Aeromonas aquatilis* strain AE207 partial sequence 16S rRNA gene

ACGGCGAGAAGGACCGGAATGCAGTTCGAGCGGCAGCGGGAAGTATTTTG
CTACTTTTGCCGGCGAGCGGCGGATGGGTGAGTAATGCCTGAGGATCTGC
CCAGTCGAGGGGGATAACTACAGGAAACGGTAGCTAATACCGCACACGCC
CTACGGGGGAAAGCGGGGGACCTTCGGGCCTTGCGCGATTGGATGAACCC
GGGTGGGATTAGCTAGTTGGTGAGGTAATGGCTCACCAAGGCCACGATCC
CTATCTGGTCTGAAAGGATGATCAGCCACACTGGAACTGAGACACGGTCC
AGACTCCTACGGGAGGCAGCAGTGGGGAATATTGCACAATGGGGGAAACC
CTGATGCAGCCATGCCGCGTGTGTGAAGAAGGCCTTCGGGTTGTAAAGCA
CTTTCACCGAGGAGGAAAGGTTGGTAGCTAATAACTGCCAGCTTTGACGT
TACTCGCAGAATAACCACCGGCTAACTCCGTGCCAGCAGCCGCGGGAATA
CGGAGGGTGCAAGCGTTTATCGGAATTATAGGGCGTAGAGCGCACGCAGG
CGGTGGGACAAGTTATATATGTGAGATTTTGGCTCTCGCGGCGAAATTCC
CTTGGAACTGTATTAAAGGTTTTTTTTCCAGAGGGCAGGAATATTAGACG
CCCCCCGTAAATCCGTCTCATTTTAGGGGGGGGAAAAAACGGGGGATTAT
GACCCTCGTTTGCACCCAAAGCTTTTCCACGAGAAACGATAATTTTGAGG
ACCGGGGGGGCCCCCTTCCCCCCGGGTTTTCCCCCCCCAAAAATCTAATC
CATTTAAGGGGGCGGGGGGGATTTCGCACCCCCCCCTCTAAAAAAACTCG
GGTAAACTGTTTAGAAGGGGAGTTCCCAGGGGTGAAGGGCGGAGTCTTTT
AAAATTATTATGATGAGAGAGACAGGGGCGCGGGGGTTTTACCAGGTAAA
TTATATTTTCAATAGAAAAGGGGGACACCTCTTTATAATACGCCGGGGAG
GTGGGAGAGAAAATAATAGACACAGTGTGCTTTTTTTGGGCAGAGAGCAC
TCAGAGACGAGAGATTATTGAGGGTACATCTTTTCTTTCTTGAGGAAAAA
AGAGTGTTATAAGTACAAAGAGCAGGGTTCTCTAACACCACTCGGGAGGT
GTGGGTCTCTCCACGGAGTGTTCATCCCTGCGTAGTGAGTAAGATAAACT
CCACCCGCGGGCCGTCACAAAGAAATAGAAAGCGTGGAGGGGGTCTTCTC
TACTCTCGCGGTGAGGCTAGATACTCTAACGAGCACAAGCGCAACGGAAG
TACGCTGCTCG

APPENDIX H: Partial sequence of *Bacillus lacus* strain AK74 16S rRNA

CTACCGGTGTAGTAACCTGCATTATACATGCAGTCGAGCGGATGCTTTGG
GAGCTTGCTCCCAAAGGTTAGCGGCGGACGGGTGAGTAACACGTGGGCAA
CCTGCCTATGAGACTGGGATAACTCCGGGAAACCGGGGCTAATACCGGAT
AATTCTTTTCTACTCATGTAGAAAAGCTGAAAGATGGTTTCGGCTATCAC
TCATAGATGGGCCCGCGGCGCAGTAGCTAGTTGGTGAGGTAACGGCTCAC
CAAGGCGACGATGCGTAGCCGACCTGAGAGGGTGATCGGCCACACTGGGA
CTGAGACACGGCCCAGACTCCTACGGGAGGCAGCAGTAGGGAATCTTCCG
CAATGGACGAAAGTCTGACGGAGCAACGCCGCGTGAGTGATGAAGGTTTT
CGGATCGTAAAACTCTGTTGTTAGGGAAGAACAAGTACCGGAGTAACTGC
CGGTACCTTGACGGTACCTAACCAGAAAGCCACGGCTAACTACGTGCCAG
CAGCCGCGGTAATACGTAGGTGGCAAGCGTTGTCCGGAATTATTGGGCGT
AAAGCGCGCGCAGGTGGTCTCTTAAGTCTGATGTGAAAGCCCCCGGCTCA
ACCGGGGAGGGTCATTGGAAACTGGGAGACTTGAGTGCAGGAAAGAAGAG
TGGAATTCCACGTGTAGCGGTGAAATGCGTAGAGATGTGGAGGAACACCA
GTGGCGAAGGCGACTCTTTGGCCTGTAACTGACACTGAGGCGCGAAAGCG
TGGGGAGCAAACAGGATTAGATACCCTGGTAGTCCACGCCGTAAACGATG
AGTGCTAAGTGTTAGAGGGTTTCCGCCCTTTAGTGCTGCAGCAAACGCAT
TAAGCACTCCGCCTGGGGAGTACGGCCGCAAGGCTGAAACTCAAAGGAAT
TGACGGGGGCCCGCACAAGCGGTGGAGCATGTGGTTTAATTCGAAGCAAC
GCGAAGAACCTTACCAGGTCTTGACATCCTCTGACACTCCTAAAAATAGG
ACTTTCCCTTCGGGGACAAAATGACAGGTGGGGCGTGGGTGTCTTCATCT
CTGTCCCGAGATATGTGTTTTGTCCCCCACCCGCCCCCCCCTGATCTTTT
TTCCCCATTTATTTTGCACTCTAGGGGACCCCCTGACACCCCCAAAAAGT
GGGGTGAGGTGTATCTCTTCCCCTTTAGCCCGGCCCCCCCCGGGCTCAAC
GGCGGTCCAAGGGGGGAAAACCGGGGCCCAACCCCCCCTAAACCCCCCCC
CTTTTTATTTTTTGGGACCCCCCCCTTTCCCCCCCACCCAAAAAGAAAAA
AAATATTCCGGGGAGAAACCCCGGCGGTTAACCCCCGCGGGCGGGGGGCC
CACCCCCCGGGGCAAAGGGGGGGCCAGAGGGGTGGGGTGGGGGCAGGGGA
GGAGGAAGAAAAAAAAAAAAAAAAAAAAAAAATAAATAATCGGCGCTGCT
CCCGCCTCTATTTTATTTATTTTTTTTACTTTCTCGCCCGCTTTGTCCTT
TTTTGTGGGGGGCCGCCGCCCCCGTTTTCACTCTTGCGCCCGCCCCT

APPENDIX I: Complete sequence of *Pseudomonas donghuensis* strain HYS 16S ribosomal RNA

CCTATCACAGCGGGGCCTACAATGCAGTCGTAACAAGGTAACCGTACATT
GCTCTTGATTCGGCGGCGGACGGGTGAGTAATGCCTAGGAATCTGCCTGG
TAGTGGGGGACAACGTTTCGAAAGGAACGCTAATACCGCATACGTCCTAC
GGGAGAAAGCAGGGGACCTTCGGGCCTTGCGCTAACAAATGAGCCTACGT
CGGATTACCTAGATGGTGGGGTAATGGCTCACCAAGGCGACGATCCAACG
CTGGTCTGAGGGATGATCAGTCACCCAGGAACTGAGACACGGTCCAAACT
CCTACCGGAGGCAGCAGTGGGGAATATTGGACAATGGGCGAAAGCCTGAT
CCACCCATGCCGCGTGTGTGAAGAAGGTCTTCGGATTGTAAAGCACTTTA
GGTTGGGAGGAAGGGCAGTAAGTTAATACGTTGCTGTTTTGACGTTACCG
AATGAATAAACCCCGGCTAACTCTGTGCCAAAAGCCGCGGTAATACAGAG
GGTGCAAGCCGTTAATCATAAATTACTGGGCGTAAAGCGCGCGGTATGTG
TGTCGTTAAGTTGCATGTGAAAGCCCCCTTATCTCTGATGTAACTGCCTC
CGGCTCTGCCGAGCTAGACTATTGTAAACTGTGGTGCTTTTTCCTGTGAG
AGAAGAGTGTGATTCCACATGTGACGGTGAAATGCTGACAGATGTGGACG
AACACCCTTGTCCTGAGCCACTCGTTCGACTACAACTGGAGCTAACGCGA
TTAAACATCCTGATCATACACGACTAGATCCACCGGCTACTCCACGCTGT
AATCCATGAGTGCTTAGTGTTGCAGCTCTATGCACTCATTCTAAGCCGCC
AGCTTAGTAATGACGCCATCGTACATACTGCACAGAGTTGACAGAGTCAA
ACTCAAACGAATTGAGCAGGGAGCTGTATCTCGCAGCGACACTATGAACT
TTATTCTGCACCTCACGTAACAAACGTCTACTCCGCGTAGGAATGCGCGG
CGATCGGGTTATACTCAGAGGCGACATTCCTCATAGGGAGTAGTATGACG
CGTTGCTGCGTAGTCTTGTCATCAGTCGATGTACCGAACAGACCGCTGTC
TTCACTACAGCGGAGTCGTACGTTAATTTTCGCACTTGGGTGAACAACTC
AGACAACGTCGATGAATGTCAGCTAGAACTGCATCTACCCGTAAGA

APPENDIX J: Partial sequence of *Bacillus lacus* strain AK74 16S rRNA

TCAGAGAAAAGAGGAAGGAATTGCAAGTTCGAGCGGAGCAAGGGAGCTTG
CGCCCGGATGTTAGCGACGGACGGGTGAGTAACACGTGGGTAACATGCCT
GTAAGACTGGGATAACTCCGGGAAACCGGAGCTAATACCGGATAGTTCCT
TGAACCGCAGGGTTCAGGGATGAAAGACGGTTTCGGCTGTCACTTACAGA
TGGACCCGCGGCGCATTATCTAGTTGGTGGGGTAATGGCTCACCAAGGCG
ACGATGCGTAGCCGACCTGAGAGGGTGATCGGCCACACTGGGACTGAGAC
ACGGCCCAGACTCCTACGGGAGGCAGCAGTAGGGAATCTTCCGCAATGGA
CGAAAGTCTGACGGAGCAACGCCGCGTGAGTGATGAAGGTTTTCGGATCG
TAAAGCTCTGTTGTTAGGGAAGAACAAGTGCGAGAGTAACTGCTCGCACC
TTGACGGTACCTAACCAGAAAGCCACGGCTAACTACGTGCCAGCAGCCGC
GGTAATACGTAGGTGGCAAGCGTTGTCCGGAATTATTGGGCGTAAAGGGC
TCGCAGGCGGTTTCTTAAGTCTGATGTGAAAGCCCCCGGCTCAACCGGGG
AGGGTCATTGGAAACTGGGAAACTTGAGTGCAGAAGAGGAGAGTGGAATT
CCACGTGTAGCGGTGAAATGCGTAGAGATGTGGAGGAACACCAGTGGCGA
AGGCGACTCTCTGGTCTGTAACTGACGCTGAGGAGCGAAAGCGTGGGGAG
CGAACAGGATTAGATACCCTGGTAGTCCACGCCGTAAACGATGAGTGCTA
AGTGTTAGGGGGTTTCCGCCCCTTAGTGCTGCAGCTAACGCATTAAGCAC
TCCGCCTGGGGAGTACGGTCGCAAGACTGAGACTCAAAGGAATTGACGGG
GGCCCGCACAAGCGGTGGATCATGTGTTTTATTCGAAGCAACGCGAAGAA
CCTTACCAGGTCTTGACATCCTCTGACAACCCTAGAGATAGGGCTTTCCC
TTCGGGACCGAATGACAGGTGGTGCATGGTTGTCGTCTTCTCTTCTCCTG
AGATGTTGGGTTAGTCCTGCAACGACGCCACCCCTGGAGCTGGTTGTCAC
TATTCCATTGGTCCTCTAAGGTAACATCCATGTGACAGACCGCAAGAGGG
TGGGGAGTGGGGGCGGTTTTTTCTTCCCTCTTTTTGCCCTCCCCTTCACT
TGTGGTTCCCAACCGGTTTTGAAAAGGGAGCGAAAACCCCCCCCCCCACA
AACACGCCGTGGGCCTTCCTTTTATCTTTATAGCGCTTTTCTCCTCTCTT
TCCCTCTCGCTCCCGCGGCGAGGAGGGGATTGAGGATAATATTTTTTTGT
GTTCCCCTGTTTTTCCGCCGCATTTGTTTCCTCCTCCCGCCGCCCCCGCC
CGCTCCCCCCTCCCTCACTCACCACGCCGGTGCGCGCGTATAACACCCCG
TCTAGTCCTTGTCTTATGTTTACTGATGGCGCGGGGCGGGCGTGGAGCGG
GTACGCGCGGAGGTGCTATAAAACAGACTAAAACAAACAATAATAAAATT
AATAACAGATTCGGCCGCCGAGCGCGTCTCCTGCCCTCCGCCCCCTCCCC
CTTCTTTTATTGCTTTTTATCTAGCCGCCGCCTCCTATTACTACTGCCTT
CTTCTTTATTTTTTTTGTTTATCGGTGCTACCGCGCCTTTATATTACCCC
TACTCTCTTCGCCCCGTCTCTCCTCCCTCT

APPENDIX K: Partial sequence of *Herbaspirillum aquaticum* strain IEH 4430 16S rRNA

CTCAGGAGATAAGCGACCCGGACTTGCAGTTCGTACGAGCAGTCAGCGCA
GTAGAAGCTTGCTCTCTTGGCGGCGAGTGGCGGACGGGTGAGTAATATAT
CGGAACGTGCCCAGTAGCGGGGGATAACTACTCGAAAGAGTGGCTAATAC
CGCATACGCCCTACGGGGGAAAGGGGGGGATCGCAAGACCTCTCACTATT
GGAGCGGCCGATATCGGATTAGCTAGTTGGTGGGGTAAAGGCTCACCAAG
GCAACGATCCGTAGCTGGTTTGAGAGGACGACCAGCCACACTGGGACTGA
GACACGGCCCAGACTCCTACGGGAGGCAGCAGTGGGGAATTTTGGACAAT
GGGGGAAACCCTGATCCAGCCATCCCGCGTGTATGATGAAGGCCTTCGGG
TTGTAAAGTACTTTTGGCAGAGAAGAAAAGGTATCCCCTAATACGGGATA
CTGCTGACGGTATCTGCAGAATAAGCACCGGCTAACTACGTGCCAGCAGC
CGCGGTAATACGTAGGGTGCAAGCGTTAATCGGAATTACTGGGCGTAAAG
CGTGTGTAGGCGGTTCGGAAAGAAAGATGTGAAATCCCAGGGCTCAACCT
TGGAACTGCATTTTTAACTGCCGAGCTAGAGTATGTCAGAGGGGGGTAGA
ATTCCACGTGTAGCAGTGAAATGCGTAGATATGTGGAGGAATACCGATGG
CGAAGGCAGCCCCCTGGGATAATACTGACGCTCAGACACGAAAGCGTGGG
GAGCAAACAGGATTAGATACCCTGGTAGTCCACGCCCTATACGATGTCAA
CTAGCTGTTGGGGGCCGTTAAGCCCTTAGTATCGCAACTAACGCGTGAAG
TTGACAGACAGGGGAGTACGGTCGCAAGATCAAAACTCATCGCAATTGAC
TGGGACCCGCCACCAGCCGATGCATGGTGTGGATTATTTCTATGCAACGC
GGATAACCCTTACTATACTTGACATAGCGGGAAAGACAAAACAACATGCA
GCATTTGTTCCCTACGAAAAACAAAACCCAAAAGCCAGTAGGGAGGGGGT
CCCCTCCCCCCCCCCAAAAACAATGAATTTATTGCACGAACCCCCCGACCA
CCAAAACCCATTTTGGGATTGTTGTGGGCGCCCCCTTTCCCCCCTTATTC
TCCACACCCCCCCCCCGCCCCCCTACCGGAGGGGTGGTGGTGTTTTTTCT
TCTCTCGTTGGGTGCTTATGTGCCTGAACTACCAAAAAAATTAGATAGGG
GGGTTGTATTAGGGGTGCCCCCCCCCCCCTAAAACGGGATATTCTCTTTC
TAACACCCGCTGATGTGTGCGGGGGGCTTAACCTTTTTCCCCCCCCCGCC
CCCGCTAGGATTTTTTTCTATTAGGCTCCCGATCCTCCGAGTCGCGTTCC
CGCTTACCTTTTTTTTGCGTTTTACCCGCTTCTAACACTCCCTCCTACCC
CCACGGGCGGCCCGGCCTCACGGCAAAACTTTGGTATCTTTCCGACCCTT
CGCTTTTCCCGCCCCGCTTCACCCTGGCTTTTTTTACCTAACGCCCCGCA
ACCAAAACATAAATAAACCCGGCCCCTATTTTCCCGCCTCGTTCCCACCG
CTTAAACTATTATAATATTCATAATACTCAACATATTTGTTTGCGGCTGC
CGCTACTAATTTTCCCTTTACCCTCCCCCGCCCCTTTCTTCCCTTCTTTC
TTACCTCTACCTACACCCCCCTTATTTCCCTCTTTTCCGCGGTCCTGA

APPENDIX L: Partial sequence of *Alcaligenes faecalis* strain Sihong 663-1 16S ribosomal RNA gene

TCCGGCTAAAGAGAGAAGGGATGGCAAGCCGAAAGGCAGCAACGAGACGA
GCTTGCTCTCTTGGCGGCGAGTGGCGGACGGGTGAGTAATATATCGGAAC
GTGCCCAGTAGCGGGGGATAACTACTCGAAAGAGTGGCTAATACCGCATA
CGCCCTACGGGGGAAAGGGGGGGATCGCAAGACCTCTCACTATTGGAGCG
GCCGATATCGGATTAGCTAGTTGGTGGGGTAAAGGCTCACCAAGGCAACG
ATCCGTAGCTGGTTTGAGAGGACGACCAGCCACACTGGGACTGAGACACG
GCCCAGACTCCTACGGGAGGCAGCAGTGGGGAATTTTGGACAATGGGGGA
AACCCTGATCCAGCCATCCCGCGTGTATGATGAAGGCCTTCGGGTTGTAA
AGTACTTTTGGCAGAGAAGAAAAGGTATCCCCTAATACGGGATACTGCTG
ACGGTATCTGCAGAATAAGCACCGGCTAACTACGTGCCAGCAGCCGCGGT
AATACGTAGGGTGCAAGCGTTAATCGGAATTACTGGGCGTAAAGCGTGTG
TAGGCGGTTCGGAAAGAAAGATGTGAAATCCCAGGGCTCAACCTTGGAAC
TGCATTTTTAACTGCCGAGCTAGAGTATGTCAGAGGGGGGTAGAATTCCA
CGTGTAGCAGTGAAATGCGTAGATATGTGGAGGAATACCGATGGCGAAGG
CAGCCCCCTGGGATAATACTGACGCTCAGACACGAAAGCGTGTGGAGCAG
ACAGGATTAGATACCCTGGTAGTCCACGCCCCTATACGATGTCAACTAGC
TGTTGTAGGCGTTAGGACATCAGTAGCGCAACTAACGCATGAAGTTTACC
TCCAGAGGAATACGGTCGGTAGAGTAGGACTCAATGGAATTGATATGGAC
ACACCCCAACAAAGGATGATGTCGGATAATTACGATGTCACGCGAGTATC
CTTACGTATTTTGACCTGCTGACGAGCCTGAAATAAAAAAGTCTCTGTTC
TCAAGAACAATACGAACTACGGATGCTATAGGAGGAAAGAGATCTCTTCC
CCCCGACGCCGTAATTATTTTCATTAAAGGACTTACACCCCATAACTATT
TACATAATTCGGAGCTGCCACCCCCCCTTTAAACCCTACCTCCGCACCCC
CCCCCACGGGGCGGGTTGGGTTTTTTTTTTCTTTTTTTACTTTGTTGTTT
TCCTTTATTTTACATAGTGATATAGGTGGGGTGAGTGGGGTTTTTATTCC
CCACCCGCCAAAAATAATTATCTTTATACTTTAGTTGGTCGTGTGAACTC
CCCCCCCCCTCCCCTTTTCTCCTCGCCCGCGCGGTACATTTTTATTTTCG
TTTACGCTACCACCTCCGTCTCATTCCTCCGCGCCGTTTCGCTATTTTTT
TACACCTATACCCGGTATATACCTCGGCTTGTTTTTGTAGTGGCGGAGCG
GACCCCTCCCCCGTCTTACTGTTGTTTTTTCTTTTACCCCTACGGCCGTA
CGCGCATTATTCCTCGCGGCGTCCTTGCACCAGCATAACTTTATATTAAT
AATATATACACCACCCACGCACCCCCTATCGCCCACCATACCGCCCGCCG
CGCCCGCCTTATTATTGCGCGCACGCCGACCCGGCGGCTGGTTATGCTGA
TTATCATAATTATACGCTGCTATACTGCGTACCTCTTCTACGTTACACCG
TCCCGCCCGGCTTACATTATTGTTGCCCTTTATATGCGTTCGCCCGCCCC
ACACGCCTTATACGCTGCGCCCTGATGTCCTACTGCTCTTACTATGTCTT
ATTGCCCATGTCGACCCGCCGCCTGTTTATATATACGCCGTATCTTTATT
GCGCGCGCGGCTTACTCTCTGAG

APPENDIX M: Partial sequence *Alcaligenes faecalis* strain HPRTAK198 16S ribosomal RNA gene

ACGAGAGAGCGGACGAATTGCAAGTTCGACGGCAGCACGAGAACCTGGCC
CTCTTGGACGCGAGTGGGAGAGGTGAGAAAGTATGGGAGACCGTGCCCTG
GAGGGGGGGATCACTACTCAAAGGAGTGGCTAACACCGCAAACTCCCTAC
AGGGGAGAGGGGGAGATTCCGGCACCTCGCACTCTTGTATCCGCCGATGT
CAGAATCTCTTGTGGGGGGGGTCAGCGCTCCCCAGCAACGATCCCTATCT
GTGGTTTGAGAAGAACACCCCCCACTGTGGGACTGACACACTCCCCACAC
TCCTACGGGAGGCAACTGTGGAAAATTTTGGACAATGGGCGAACCCCTGA
TCCCCCCGTCCCGTGTGTGAAAAAAGCGCCTTCGTTTTGAAATACTTTTT
TGGGGGAGAAAAGTGTGTATCCCCTAATACGGGATACTGCTGACTGTATC
TGCAAAAAACGCACCGGCTAACTATGTCCCCACCCCCGCGGTAATACATG
GGGTGCACGCGTTACTCAAAATTACTGGGCGTAAAGCGTGTGTGCGGGGT
TCGGAAACAAAGATGTGAAATCCCGGGGCTCCCCCTTAAAACTGTTTTTT
AAACTGCCGATCTAAGTAAGACAGAGGGGGGTAGTATTCCACGTGTCGCA
GTGAAGTGCGTAGATATGTGGAAGAATACCGATGGCGAAGGCACCCCCCT
GGCATCACACTGACGCTCACACACAAAAGCGTGGGGAGCAAACAGGGATT
ATACTCCCCTGGTACTCCCCCCCCCCAAAACCATGTTAATTAAGTTGCTG
GGGGGGAAGATGGGGACTTCCTCCCCCCAGCTAAAACCGTGATTTTCCAC
CCTTAGGTAGGGATATTTTCTTGCATGATTAAAAATCAAATTAATTATGC
CCTGTTGAACAGAACCACCCCGTGGAGGTTTTTTATCAATTACATTTTTA
TAGGCGAAAAACCTTTCCCCCCTTTTGTGTGGGGGGGGGGTGAGAGGAAG
AGATAGTTGGTCTTCTCCCCACAAGAAGACA

APPENDIX N: Sequence of *Oceonobacillus oncorhynchi* supsp. *incaldanensis* strain AM - 75

CTCGGGGGGCTGGCGCAAATAATGCAGTCGAGCGCGGCAGCGGAGGAAGT
CTTCGGAGGGAAGTTCGTGGAAGGAGGAGCGGAGGGTGGAATACCGCCTG
AGCAACCTGCCTGTAAAACGGGAATAACTCGCGAAAACGCGAGCTAATAC
CGGACCACACTTTCTATCCTTTGATGGAAAGTTGAAAGGCGGCTTTTGCT
GTCACTTACAGATGGGCCTGCGGCGCAGTACCTAGTTGGTGAGGTAACGG
CTCACCCAGGCAACAATACGTAGCCCACCTGAGAGGGTGATCGGCCACAC
TGGGACTGAGACGCGGCCCAGACTCCTACGGGAGGCAGCACTAGGGAATC
TTCCGCACTGGACGAAAGTCTGACGGAGCAACGCCGCGTGAGTGATGAAG
GTTTTCGGATCGTAAAACTCTGTTGTCAGGGAAGAACAAGTACGATTTTA
ACTGATCGTACCTTGACGGTACCTGACCACAAAGCCACGGCTAACTACGT
GCCAACAGCCGCGGTAATACGTATGTGGCAAGCGTTGTCCGGAATTATTG
GGCGTAAAGCGCTCGCAGGCGGTTCTTTAAATCTGATGTGAAATCCTGCG
GCTCAACCGCGGACGTGCATTGGAAACTGGAGGACTTGAGTGCAGAAGAG
GAGAGTGGAATTCCACGTGTAGCGGTGAAATGCGTAGAGATGTGGAGGAA
CACCAGTGGCGAAGGCGACTCTCTGGTCTGTAACTGACACTGAGGAGCGA
AAGCGTGGGGAGCGAACAGGATTAGATACCCTGGTAGTCCACGCCGTAAC
GATGAGTGCTAGGTGTTAGGGGGTTTCCGCCCCTTATGCTGAAGTTAACG
CATTAAGCACTCCGCCGGGGGAGTACAGCTCGCAAGGCTGAAACTCAAAA
GAATTGACGGGGGACTCGCACAAGCGGTGGAGCATGTGGGTTTAATTCAA
GCAACACTAGCACCTTACCACGTCTTGACATCCTTTGGACTGCTCTACAG
ATAGAGCTTTTCCCATTCAGGGACGATAGTGACATGTGGCTGCATTGTCT
GTCGATCAGCATCGTTGTCATGAAAATGATAGAGATTCAGTTCCCAGCAA
CGAGCGCGAGTCCGTTCATCTTAATGTCAGCCAACTGAGTTAGGTCACCT
CTCAAGTGACTGGCGATGACAGAGCCGAGACGTGGCATAGCGTCGATCGT
ACATGACCCCTACGACGTCGGCTAACACACCGACTC

APPENDIX O: Concentration of heavy metals in Kainji Lake water (Jan-Dec. 2017)

	Heavy metal concentration (mg/L)				
Month/SEM	Pb	Cu	Cd	Cr	As
January	0.03^{b}	0.04^{d}	0.01^{c}	0.02^{de}	0.01^{c}
February	0.02^{b}	0.04^{d}	0.02^{c}	0.05^{bcd}	0.01^{c}
March	0.02^{b}	0.06^{d}	0.02^{c}	0.06^{bc}	0.01^{c}
April	0.07^{b}	0.22^{d}	0.08^{c}	0.10^{a}	0.10^{b}
May	0.04^{b}	0.21^{d}	0.07^{c}	0.05^{bcd}	0.02^{c}
June	0.05^{b}	0.07^{d}	0.03^{c}	0.07^{ab}	0.02^{c}
July	0.05^{b}	0.57^{c}	0.29^{ab}	0.01^{e}	0.15^{ab}
August	0.05^{b}	0.92^{b}	0.12^{bc}	0.03^{cde}	0.16^{a}
September	0.29^{a}	1.23^{a}	0.41^{a}	0.03^{cde}	0.18^{a}
October	0.03^{b}	0.82^{bc}	0.11^{c}	0.03^{cde}	0.02^{c}
November	0.02^{b}	0.08^{d}	0.01^{c}	0.05^{bcd}	0.02^{c}
December	0.04^{b}	0.15^{d}	0.05^{c}	0.04^{cde}	0.01^{c}

Values are means of five determinations. Means with dissimilar alphabet letter along the column differ significantly ($P \leq 0.05$) according to the Duncan Multiple Range Test.

APPENDIX P: Concentration of heavy metals in Jebba Lake water (Jan-Dec. 2017)

	Heavy metal concentration (mg/L)				
Month	**Pb**	**Cu**	**Cd**	**Cr**	**As**
January	0.014^{g}	0.147^{h}	0.013^{h}	0.068^{c}	0.026^{g}
February	0.023^{fg}	0.066^{j}	0.016^{gh}	0.050^{e}	0.016^{i}
March	0.018^{g}	0.026^{l}	0.016^{gh}	0.074^{b}	0.060^{e}
April	0.176^{c}	1.263^{d}	0.296^{c}	0.015^{i}	0.058^{f}
May	0.072^{e}	0.507^{g}	0.220^{d}	0.033^{g}	0.067^{d}
June	0.031^{f}	0.042^{k}	0.028^{g}	0.027^{h}	0.019^{h}
July	0.019^{g}	1.323^{c}	0.094^{e}	0.011^{j}	0.087^{c}
August	0.079^{e}	1.470^{b}	0.658^{a}	0.095^{a}	0.110^{b}
September	0.372^{a}	2.350^{a}	0.495^{b}	0.036^{f}	0.190^{a}
October	0.242^{b}	0.804^{f}	0.104^{e}	0.035^{f}	0.011^{j}
November	0.021^{gf}	0.106^{i}	0.015^{gh}	0.017^{i}	0.012^{j}
December	0.118^{d}	0.822^{e}	0.073^{f}	0.063^{d}	0.020^{h}

Values are means of five determinations. Means with dissimilar letter (s) differ significantly according to the Duncan's Multiple Range Test (DMRT).
Significant at $p \leq 0.05$

APPENDIX Q: Heavy metal concentrations in water from Kainji sample stations

	Heavy metal concentration (mg/L)				
Sampling Stations	**Pb**	**Cu**	**Cd**	**Cr**	**As**
K1.1	0.031^{d}	0.456^{b}	0.088^{c}	0.023^{e}	0.064^{b}
K1.2	0.082^{a}	0.464^{a}	0.187^{a}	0.040^{d}	0.085^{a}
K1.3	0.078^{b}	0.445^{c}	0.107^{b}	0.061^{a}	0.054^{c}
K1.4	0.057^{c}	0.261^{d}	0.075^{d}	0.055^{b}	0.054^{c}
K1.5	0.031^{d}	0.213^{e}	0.046^{e}	0.045^{c}	0.036^{d}

Values are means of five determinations. Means with dissimilar letter (s) differ significantly according to the Duncan's Multiple Range Test (DMRT).
Significant at $p \leq 0.05$

APPENDIX R: Heavy metal concentrations in water from Jebba sample stations

	Heavy metal concentration (mg/L)				
Sampling Stations	**Pb**	**Cu**	**Cd**	**Cr**	**As**
J1.1	0.066^{a}	0.534^{d}	0.176^{b}	0.033^{e}	0.055^{c}
J1.2	0.380^{c}	0.774^{a}	0.227^{a}	0.064^{b}	0.082^{b}
J1.3	0.141^{d}	0.607^{b}	0.150^{c}	0.066^{a}	0.143^{a}
J1.4	0.034^{c}	0.585^{c}	0.070^{d}	0.041^{d}	0.047^{d}
J1.5	0.064^{b}	0.323^{e}	0.044^{e}	0.049^{c}	0.044^{e}

Values are means of five determinations. Means with dissimilar letter (s) differ significantly according to the Duncan's Multiple Range Test (DMRT).
Significant at $p \leq 0.05$

APPENDIX S: Heavy metal concentration in ONFI of Kainji sample station

	Heavy metal concentration (mg/kg)				
Sample Stations	**Pb**	**Cu**	**Cd**	**Cr**	**As**
K1.1	0.351^{c}	1.317^{b}	0.195^{d}	0.130^{b}	0.023^{c}
K1.2	0.367^{a}	1.323^{a}	0.224^{b}	0.128^{b}	0.023^{c}
K1.3	0.306^{e}	1.237^{e}	0.186^{e}	0.116^{c}	0.156^{b}
K1.4	0.355^{b}	1.306^{c}	0.256^{a}	0.174^{a}	0.256^{a}
K1.5	0.313^{d}	1.244^{d}	0.216^{c}	0.118^{c}	0.018^{d}

Values are means of five determinations. Means with dissimilar letter (s) differ significantly according to the Duncan's Multiple Range Test (DMRT).
Significant at P ≤ 0.05

APPENDIX T: Heavy metal concentration in ONFG of Kainji sample station

	Heavy metal concentration (mg/kg)				
Sample Stations	**Pb**	**Cu**	**Cd**	**Cr**	**As**
K1.1	0.296^{c}	1.326^{b}	0.162^{c}	0.132^{b}	0.031^{c}
K1.2	0.312^{a}	1.332^{a}	0.191^{b}	0.130^{c}	0.031^{c}
K1.3	0.251^{d}	1.246^{d}	0.153^{d}	0.118^{e}	0.164^{b}
K1.4	0.300^{b}	1.315^{c}	0.223^{a}	0.176^{a}	0.264^{a}
K1.5	0.258^{d}	1.253^{d}	0.183^{c}	0.120^{d}	0.026^{d}

Values are means of five determinations. Means with dissimilar letter (s) differ significantly according to the Duncan's Multiple Range Test (DMRT).
Significant at P ≤ 0.05

APPENDIX U: Heavy metal concentration in CGFI Kainji sample station

	Heavy metal concentration (mg/kg)				
Sample Stations	**Pb**	**Cu**	**Cd**	**Cr**	**As**
K1.1	0.373^{b}	1.223^{d}	0.333^{a}	0.126^{e}	0.023^{a}
K1.2	0.382^{a}	1.367^{a}	0.329^{a}	0.136^{c}	0.024^{a}
K1.3	0.338^{c}	1.304^{b}	0.292^{c}	0.176^{a}	0.020^{ab}
K1.4	0.375^{ab}	1.272^{c}	0.319^{b}	0.156^{b}	0.024^{a}
K1.5	0.343^{c}	1.273^{d}	0.297^{c}	0.133^{d}	0.018^{b}

Values are means of five determinations. Means with dissimilar letter (s) differ significantly according to the Duncan's Multiple Range Test (DMRT).
Significant at P ≤ 0.05

APPENDIX V: t-Test for seasonal physicochemical qualities -Kainji Lake

Temperature
t-Test: Paired Two Sample for Means

	Wet	***Dry***
Mean	31.05	28.56333
Variance	0.781896552	3.82723
Observations	30	30
Pearson Correlation	-0.144418712	
Hypothesized Mean Difference	0	
Df	29	
t Stat	6.025866852	
P(T<=t) one-tail	7.41855E-07	
t Critical one-tail	1.699127027	
P(T<=t) two-tail	1.48371E-06	
t Critical two-tail	2.045229642	

Transparency
t-Test: Paired Two Sample for Means

	Wet	*Dry*
Mean	0.449333333	28.56333
Variance	0.087723678	3.82723
Observations	30	30
Pearson Correlation	0.007038233	
Hypothesized Mean Difference	0	
Df	29	
t Stat	-77.90634534	
P(T<=t) one-tail	1.53489E-35	
t Critical one-tail	1.699127027	
P(T<=t) two-tail	3.06978E-35	
t Critical two-tail	2.045229642	

Total Alkalinity
t-Test: Paired Two Sample for Means

	Wet	*Dry*
Mean	11.07	8.986667
Variance	7.838034483	7.599816
Observations	30	30
Pearson Correlation	-0.03517078	
Hypothesized Mean Difference	0	
Df	29	
t Stat	2.854439551	
P(T<=t) one-tail	0.003940092	
t Critical one-tail	1.699127027	
P(T<=t) two-tail	0.007880184	
t Critical two-tail	2.045229642	

BOD

t-Test: Paired Two Sample for Means

	Wet	***Dry***
Mean	1.093333333	3.06
Variance	0.420988506	1.804552
Observations	30	30
Pearson Correlation	0.393329006	
Hypothesized Mean Difference	0	
Df	29	
t Stat	-8.68054978	
P(T<=t) one-tail	7.37802E-10	
t Critical one-tail	1.699127027	
P(T<=t) two-tail	1.4756E-09	
t Critical two-tail	2.045229642	

pH

t-Test: Paired Two Sample for Means

	Wet	*Dry*
Mean	7.29	7.3
Variance	0.056103448	0.097241
Observations	30	30
Pearson Correlation	-0.158730574	
Hypothesized Mean Difference	0	
Df	29	
t Stat	-0.130265067	
P(T<=t) one-tail	0.448628061	
t Critical one-tail	1.699127027	
P(T<=t) two-tail	0.897256123	
t Critical two-tail	2.045229642	

Conductivity

t-Test: Paired Two Sample for Means

	Wet	*Dry*
Mean	52.93333333	45.26667
Variance	174.9609195	67.02989
Observations	30	30
Pearson Correlation	-0.145347079	
Hypothesized Mean Difference	0	
Df	29	
t Stat	2.539284486	
P(T<=t) one-tail	0.008364884	
t Critical one-tail	1.699127027	
P(T<=t) two-tail	0.016729768	
t Critical two-tail	2.045229642	

DO

t-Test: Paired Two Sample for Means

	Wet	*Dry*
Mean	7.206666667	7.313333
Variance	1.00754023	1.550161
Observations	30	30
Pearson Correlation	0.055662163	
Hypothesized Mean Difference	0	
Df	29	
t Stat	-0.375672666	
P(T<=t) one-tail	0.354947155	
t Critical one-tail	1.699127027	
P(T<=t) two-tail	0.70989431	
t Critical two-tail	2.045229642	

TDS

t-Test: Paired Two Sample for Means

	Wet	***Dry***
Mean	0.028	0.020593
Variance	0.000105931	0.000102
Observations	30	30
Pearson Correlation	-0.41753483	
Hypothesized Mean Difference	0	
Df	29	
t Stat	2.361120665	
P(T<=t) one-tail	0.012576903	
t Critical one-tail	1.699127027	
P(T<=t) two-tail	0.025153806	
t Critical two-tail	2.045229642	

NO_3

t-Test: Paired Two Sample for Means

	Wet	***Dry***
Mean	15.66	9.655333
Variance	11.53757931	4.396315
Observations	30	30
Pearson Correlation	0.056600264	
Hypothesized Mean Difference	0	
Df	29	
t Stat	8.455968437	
P(T<=t) one-tail	1.28093E-09	
t Critical one-tail	1.699127027	
P(T<=t) two-tail	2.56186E-09	
t Critical two-tail	2.045229642	

PO_4

t-Test: Paired Two Sample for Means

	Wet	*Dry*
Mean	0.343	0.252
Variance	0.022732069	0.035707
Observations	30	30
Pearson Correlation	0.197574529	
Hypothesized Mean Difference	0	
Df	29	
t Stat	2.294651324	
P(T<=t) one-tail	0.014590886	
t Critical one-tail	1.699127027	
P(T<=t) two-tail	0.029181772	
t Critical two-tail	2.045229642	

APPENDIX W: t-Test for seasonal physicochemical qualities - Jebba Lake

Temperature

t-Test: Paired Two Sample for Means

	Dry	*Wet*
Mean	27.78333333	29.95
Variance	3.442816092	1.609482759
Observations	30	30
Pearson Correlation	0.116091707	
Hypothesized Mean Difference	0	
Df	29	
t Stat	-5.59074034	
P(T<=t) one-tail	2.46055E-06	
t Critical one-tail	1.699127027	
P(T<=t) two-tail	4.92109E-06	
t Critical two-tail	2.045229642	

Transparency

t-Test: Paired Two Sample for Means

	Dry	*Wet*
Mean	0.415333333	0.342333333
Variance	0.054474023	0.044542644
Observations	30	30
Pearson Correlation	-0.215941895	
Hypothesized Mean Difference	0	
Df	29	
t Stat	1.152835868	
P(T<=t) one-tail	0.129196027	
t Critical one-tail	1.699127027	
P(T<=t) two-tail	0.258392054	
t Critical two-tail	2.045229642	

pH

t-Test: Paired Two Sample for Means

	Dry	*Wet*
Mean	7.063333333	7.3
Variance	0.044471264	0.037931034
Observations	30	30
Pearson Correlation	-0.025187564	
Hypothesized Mean Difference	0	
Df	29	
t Stat	-4.460085908	
P(T<=t) one-tail	5.66848E-05	
t Critical one-tail	1.699127027	
P(T<=t) two-tail	0.00011337	
t Critical two-tail	2.045229642	

Conductivity

t-Test: Paired Two Sample for Means

	Dry	*Wet*
Mean	43.46666667	53.66666667
Variance	119.9126437	114.7126437
Observations	30	30
Pearson Correlation	0.019600746	
Hypothesized Mean Difference	0	
Df	29	
t Stat	-3.683586206	
P(T<=t) one-tail	0.000468816	
t Critical one-tail	1.699127027	
P(T<=t) two-tail	0.000937632	
t Critical two-tail	2.045229642	

NO_3

t-Test: Paired Two Sample for Means

	Dry	*Wet*
Mean	11.643	16.87133333
Variance	1.179035517	0.914198161
Observations	30	30
Pearson Correlation	0.22526852	
Hypothesized Mean Difference	0	
Df	29	
t Stat	-22.46118347	
P(T<=t) one-tail	3.45596E-20	
t Critical one-tail	1.699127027	
P(T<=t) two-tail	6.91191E-20	
t Critical two-tail	2.045229642	

DO

t-Test: Paired Two Sample for Means

	Dry	*Wet*
Mean	6.796666667	7.693333333
Variance	3.467229885	2.72754023
Observations	30	30
Pearson Correlation	0.302424369	
Hypothesized Mean Difference	0	
Df	29	
t Stat	1.730469058	
P(T<=t) one-tail	0.047088182	
t Critical one-tail	1.699127027	
P(T<=t) two-tail	0.094176364	
t Critical two-tail	2.045229642	

TA

t-Test: Paired Two Sample for Means

	Dry	*Wet*
Mean	9.116	11.005
Variance	10.38202483	5.227991379
Observations	30	30
Pearson Correlation	0.338600545	
Hypothesized Mean Difference	0	
Df	29	
t Stat	3.174771775	
P(T<=t) one-tail	0.001769513	
t Critical one-tail	1.699127027	
P(T<=t) two-tail	0.003539026	
t Critical two-tail	2.045229642	

BOD

t-Test: Paired Two Sample for Means

	Dry	*Wet*
Mean	3.193333333	3.338333333
Variance	2.633747126	2.843048851
Observations	30	30
Pearson Correlation	-0.40416003	
Hypothesized Mean Difference	0	
Df	29	
t Stat	-0.286419582	
P(T<=t) one-tail	0.388296535	
t Critical one-tail	1.699127027	
P(T<=t) two-tail	0.776593071	
t Critical two-tail	2.045229642	

TDS

t-Test: Paired Two Sample for Means

	Dry	*Wet*
Mean	0.02248	0.033
Variance	0.000103751	6.88276E-05
Observations	30	30
Pearson Correlation	0.167386734	
Hypothesized Mean Difference	0	
Df	29	
t Stat	-4.06556715	
P(T<=t) one-tail	0.000167361	
t Critical one-tail	1.699127027	
P(T<=t) two-tail	0.000334722	
t Critical two-tail	2.045229642	

APPENDIX X: Correlation of physicochemical parameters vs VBC of KL

Parameter	Tempt. (oC)	Transp. (m)	pH	Cond.(µS/cm)	DO. (mg/l)	Total Alk. (mg/l)	BOD (mg/l)	TDS.(mg/l)	NO_3 (mg/l)	PO_4 (mg/l)	VBC
Tempt. (oC)	1										
Transp. (m)	0.09498068	1									
pH	-0.360088	0.093968108	1								
Cond.(µS/cm)	0.25046155	-0.35328412	-0.0265	1							
DO. (mg/l)	0.05854734	0.00231867	-0.1629	-0.0867242	1						
Total Alk. (mg/l)	0.27330889	-0.47612524	0.16621	0.690044743	-0.2011339	1					
BOD (mg/l)	-0.5883677	-0.28087443	-0.0318	-0.23208834	0.32140201	-0.247633619	1				
TDS.(mg/l)	0.25209569	-0.19419824	-0.1287	0.613156692	0.14127873	0.445983236	-0.362172	1			
NO3 (mg/l)	0.56458124	-0.04131988	0.15172	0.585436663	-0.2672922	0.795198713	-0.612734	0.357186	1		
PO4 (mg/l)	0.26527165	0.046249343	0.20529	0.09744779	-0.2300844	0.289453486	-0.351396	-0.042269	0.4264601	1	
VBC	-0.1585954	0.189058902	0.26548	-0.29590916	-0.1088608	-0.214160818	-0.160675	-0.157687	0.0018377	0.09644789	1

APPENDIX Y: Correlation of physicochemical parameters vs VBC of JL

Parameter	Tempt. (oC)	Transp (m)	pH	Cond(µS/cm)	DO (mg/l)	Total Alk (mg/l)	BOD (mg/l)	TDS(mg/l)	NO_3 (mg/l)	PO_4 (mg/l)	VBC
Tempt. (oC)	1										
Transp (m)	-0.109049992	1									
pH	0.29668858	-0.100173091	1								
Cond(µS/cm)	0.152949582	0.438764966	0.2641843	1							
DO (mg/l)	0.180256807	-0.16550564	0.0863827	0.282130355	1						
Total Alk (mg/l)	0.03423225	-0.525474713	0.266812	0.659156685	0.514926078	1					
BOD (mg/l)	-0.16642943	0.056818242	0.2709294	0.206593823	0.35365492	0.220497075	1				
TDS(mg/l)	0.315629204	-0.420173044	0.3505629	0.57076373	-0.069693123	-0.507166587	0.331740019	1			
NO3 (mg/l)	0.61579242	-0.235374218	0.5316919	0.468629046	0.269427104	0.345501049	0.098289247	0.493296575	1		
PO4 (mg/l)	0.368545671	-0.085093569	0.3988203	0.432326287	0.385290867	0.521133844	0.146459923	0.319802187	0.61787847	1	
VBC	0.19020712	0.288100539	0.1209778	-0.198828056	-0.055385186	-0.041964415	-0.082679363	-0.173721306	-0.104043864	-0.36182357	1

APPENDIX Z: Correlation of physicochemical parameters vs FC of KL

Parameter	Tempt. (oC)	Transp. (m)	pH	Cond.(µS/cm)	DO. (mg/l)	Total Alk. (mg/l)	BOD (mg/l)	TDS.(mg/l)	NO_3 (mg/l)	PO_4 (mg/l)	FC
Tempt. (oC)	1										
Transp. (m)	-0.094980681	1									
pH	0.360087959	-0.093968108	1								
Cond.(µS/cm)	0.250461547	0.353284117	-0.0265126	1							
DO. (mg/l)	0.058547339	0.00231867	-0.1628753	-0.086724204	1						
Total Alk. (mg/l)	-0.273308894	-0.476125238	0.1662099	0.690044743	-0.201133899	1					
BOD (mg/l)	0.588367745	-0.280874429	-0.0317781	-0.232088344	0.321402008	-0.247633619	1				
TDS.(mg/l)	0.252095688	-0.194198239	-0.1286666	0.613156692	-0.141278734	0.445983236	-0.362171753	1			
NO3 (mg/l)	0.564581239	-0.041319877	0.1517236	0.585436663	-0.267292219	0.795198713	-0.612733669	-0.357186013	1		
PO4 (mg/l)	-0.265271649	-0.046249343	0.2052853	0.09744779	-0.230084439	-0.289453486	-0.351396415	-0.042268607	0.426460079	1	
FC	-0.024713369	0.436876596	0.0226205	-0.540285344	0.052098331	-0.486818181	0.107314633	0.286753309	-0.270986353	0.175897373	1

APPENDIX Z (i): Correlation of physicochemical parameters vs FC of JL

Parameter	Tempt. (oC)	Transp. (m)	pH	Cond.(µS/cm)	DO (mg/l)	Total Alk. (mg/l)	BOD (mg/l)	TDS.(mg/l)	NO_3 (mg/l)	PO_4 (mg/l)	FC
Tempt. (oC)	1										
Transp. (m)	-0.109049992	1									
pH	0.29668858	-0.100173091	1								
Cond.(µS/cm)	0.152949582	-0.438764966	0.2641843	1							
DO (mg/l)	0.180256807	-0.16550564	0.0863827	0.282130355	1						
Total Alk. (mg/l)	0.03423225	-0.525474713	0.266812	0.659156685	0.514926078	1					
BOD (mg/l)	-0.16642943	-0.056818242	0.2709294	0.206593823	0.35365492	0.220497075	1				
TDS.(mg/l)	0.315629204	-0.420173044	0.3505629	0.57076373	-0.069693123	0.507166587	-0.331740019	1			
NO3 (mg/l)	0.61579242	-0.235374218	0.5316919	0.468629046	0.269427104	0.345501049	0.098289247	0.493296575	1		
PO4 (mg/l)	0.383518221	-0.084204708	0.3905387	0.429013103	0.379396667	0.51415889	0.159998244	0.30952905	0.628511259	1	
FC	0.299483874	0.004671213	0.0429237	-0.247618832	-0.196646783	-0.433952475	-0.079653356	-0.113306055	0.074259547	0.051183874	1