CHAPTER ONE

INTRODUCTION

1.1 Background of Study

Reports by the United Nations Food and Agriculture Organization (FAO; Rome, Italy), states that the Earth holds about 1,400 million km³ of water (FAO, 2002). Water is a very essential and important aspect for the growth and development of the human body and other life forms, the human body constitute of 70% of water, (FAO, 2002) which is vital for man's survival and life. Also, the earth is covered with about 70% of water which is important for life forms and nonliving things (FAO, 2002). Water can be contaminated from both natural as well as anthropogenic sources, water provides an environment in which a broad range of microorganisms can survive and function. Water is guaranteed as safe and pure for consumption when it is free from contamination by physical, biological, and chemical means and also, colourless, tasteless, and odourless which is regarded as portable or good quality water (WHO, 2007). It serves various purposes; for instance in the industry, it is used in manufacturing beverages, then for domestic purposes like cooking, cleaning, bathing, washing etc. It is also used in the laboratory as distilled water for carrying out various analysis and procedures. Water is used for recreation purposes and tourism, transportation, hydroelectric power generation and agriculture (especially in irrigation and aquaculture) According to (WHO, 2007); safe drinking water is described as microbial, chemical and physical water that complies with rules of the WHO on domestic drinking water quality.

From the physiochemical point of view, some parameters like hardness, alkalinity, dissolved oxygen, total dissolved solid, chloride and others add to the pleasant appearance, and value of water while, those with adverse health effect are lead, ammonia, nitrate, zinc, etc (WHO, 2002). Water low or high pH, high turbidity is unpleasant to use. In microbiological aspect,

some pathogenic microorganisms found in water are capable of causing diseases; drinking water should therefore be free from pathogenic microorganisms like *salmonella* spp., *shigella* spp, coliforms and others. Coliforms are used as indicator organism when dealing with drinking water quality because contaminated water majorly contains coliform most especially *E.Coli*. These microorganisms cause different diseases like cholera, typhoid, dysentery and hepatitis etc. (WHO, 2002), reports that unsafe water supply is a significant issue and faecal water and treated water contamination sources and treated water is a global problem worldwide, especially in developing countries. The WHO has estimated that inadequate sanitation, pollution or contaminated water causes about 80% of all diseases and illnesses in the world (WHO, 2004).

Reports by (WHO, 2004), drinking water- contacted illnesses kill about 5 million kids annually and create 1/6th of the world's population sick. In Nigeria, mainly the rural residents do not have access to potable water and therefore depend on wells, streams, river etc for domestic use which could be contaminated and therefore pose a health hazard. According to (WHO, 2011), water is the most crucial product to the endurance of life.

Fresh water is a water body majorly with salinity less than that of 0.5% (Yozzo and Diaz, 1999). Fresh water comprises lakes, streams, ponds, rivers etc (Yozzo and Diaz, 1999). The two primary freshwater types are; static (lentic) and flowing (lotic) water. Examples of flowing water are rivers and stream, while examples of static water are ponds and lakes, (Saliu *et al.*, 2018). According to (Dirligen, 2001), the contamination of fresh water with a broad scope of pollutants has become an issue of apprehension over the previous few decades. Organic pollution of fresh water is on the high side and results to high level of eutrophication and low dissolved oxygen content (Schindler and Vallentyne, 2004). The main cause of increased eutrophication is the availability of abundant nutrients available in the water that causes the uncontrollable algal growth (Schindler and Vallentyne, 2004). Turbidity

also decreases the level of light transmission in the water, and high production of algae will lead to increase turbidity and low dissolved oxygen (Saliu *et al.*, 2018).

The Ocean grasp about 97% of surface water, glaciers and polar ice caps 2.4%, and other land surface water such as lakes, rivers and ponds 0.6% (Priya, 2011). A very minute quantity of the Earth's water within biological organs and man-made produce is limited (Priya, 2011). Liquid water can be discovered in water bodies, for Example Sea, ocean, lake, river, canal, stream, and pond. The mainstream of earth's water is sea water. You can also find water in the atmosphere in solid, liquid, and vapour states; it also exists as groundwater in aquifers. Water plays a crucial role in today's global economy, as it functions as a solvent for a large diversity of chemical compounds (Priya, 2011). Farming consumes about 70% of fresh water (Baroni *et al.*, 2007).

A fresh water ecosystem consists of natural vegetation, plants, minerals, micro-organisms, fish, some algae growth, plenty of sunshine and a natural undisturbed, unpolluted environment (Baroni *et al.*, 2007). A fresh water spring flows naturally through the mountains and rolling hills and can be enjoyed by many for drinking purposes. A good sign of any fresh water is abundant life and natural growing vegetation. Fish, insects, algae, and a rich natural growing plant system is all part of the natural fresh water and all thrive and survive as long as the water is not polluted or disturbed. Lakes may have one usual drainage in the system of a river or stream, which retain a lake's normal level by permitting the drainage of superfluous water (Baroni *et al.*, 2007). Specific ones may not have a passage but misplace water merely by evaporation or underground outflow or both. These water bodies are called endorheic lakes, which are virtually confined or closed lakes (Baroni *et al.*, 2007). These lakes are generally lakes with no significant outflow, either through waterways or through subterranean distribution. Any water contained by an endorheic pool leaves the structure simply by evaporation (Baroni *et al.*, 2007). The size, morphometry, accessibility of

water, water chemistry, physics, hydrology and biology of each lake is distinctive. Lakes are great shelter for a huge flora and fauna range. Freshwater lakes are the majority of the lakes on earth and most of them are located in the Northern Hemisphere. The Dead sea lies about 400 meters below mean sea level between Israel and Jordan. Lake Eyre, the largest lake in Australia, is at about 16 metre (Baroni *et al.*, 2007).

Based on the remarkable information stated above, the formation of lakes, their physicchemical conditions and their biotic life (especially the bacteria population) will not be an exemption from what is obtainable at Mountain Top University.

1.2 STATEMENT OF PROBLEM

Water is an important component in any community of which Magada Lake that passes through Mountain Top University, Ibafo is not an exception. In addition to the time dependent ecological variation, that is expected in Magada Lake, heterotrophic bacteria is also expected to be an important component of this lake. Globally, ecological factors (physico-chemical factors) have the possibility to influence (positively or negatively) the bacterial population of a lake. Consequently, the ecological and bacterial population can pose unexpected challenges to the environment.

1.3 RESEARCH QUESTIONS

- 1. Does the ecological parameter influence the bacterial population?
- 2. Does bacteria population at each time varies with ecological parameters?
- 3. Which of the time point favours bacterial population and which one acts otherwise or contrary?

1.4 JUSTIFICATION

Water quality has a great impact on health and well- being of man and animals. The influence of Magada lake's ecological factors on bacteria population have not been studied and documented. Therefore, this study is aimed at assessing the effect of time related ecological factors on bacterial population in Magada Lake that passed through Mountain Top University due to the activities like children playing in the nearby stream in kuro community during raining season. Specifically, to ascertain the effect of anthropogenic and non- anthropogenic activities on ecological and biological activities (i.e. bacterial population) in Magada lake of Mountain Top University.

1.5 OBJECTIVES OF STUDY

1. To determine the physico-chemical factors associated with the water in Magada Lake.

2. To determine the diurnal variation in the physico-chemical parameters of the water in Magada lake.

3. To assess the bacteria population 0f water in Magada Lake.

4. To determine the diurnal variation in bacteria population of the water in Magada Lake.

5. To study the effects of diurnal variation of the physico-chemical parameters of Magada lake on the bacterial population.

CHAPTER TWO

2.0 WATER AS AN ESSENTIAL COMMODITY

Water is a vital commodity for human expenditure and is one of the most important renewable resources, having a major function in determining drinking water portability (MacDonnell *et al.*, 2014). Water is the origin of all lives–environmental resources for the fauna and flora of our earth and a primary need for all lives and nature's most abundant, wonderful and valuable compound (MacDonnell *et al.*, 2014). According to a research carried out by (MacDonnell *et al.*, 2014), he observed that bulk of this water is supplied directly by people who use it or by organizations well-known by such users to supply water. (for example to form irrigation districts, farmers bore their own wells or band together). Most of the remainder is provided by public agencies (including the U.S. Bureau of Reclamation for irrigation).

Roman jurists in the 6th century A.D. poised that there is a very old belief that use of water should be without limit and accessible to those requiring its use (MacDonnell *et al.*, 2014). Water has been considered as a "thing" that belongs to all individuals and is therefore incapable of being confidentially owned as it flows in a river (MacDonnell *et al.*, 2014). (MacDonnell *et al.*, 2014) opined that of all natural income necessary to humans, only the order for water is not satisfied through the marketplace. Water is essential to live, but many individuals are not allowed to drink clean and secure water and many die from waterborne bacterial diseases. (Cabral, 2010).Worldwide wetlands areas global contribution is estimated at up to US\$ 15 trillion per year. Freshwater accounts for only some 6 percent of the world's water supply, but is vital for human uses such as agriculture, drinking, manufacturing, and domestic activities. As discussed above, two-thirds of global freshwater is found underground (Simon, 2013).

2.1 WATER RESOURCES

Earth's water resources, includes:

- 1. Ground water (underground aquifers)
- 2. Salt water resources (oceans, seas)
- 3. Surface water sources (lakes, streams)

Rivers, lakes, and underground aquifers provide fresh water for drinking, irrigation, domestic use, and sanitation etc, while the oceans supply habitation for a huge share of the planet's food supply. Today, on the other hand, increase of agriculture, damming, diversion, over-use, and pollution threatening these distinctive resources in many areas of the world (Fiore, 2018). This research discusses how significant reserves such as oceans, ice caps, and groundwater allocate the world's water supply. It is generally believed that about 97% of the world's water supply by volume is detained in the oceans. The other large reserves are groundwater 4% and icecaps and glaciers 2%, with all other water bodies collectively accounting for a fraction of 1% (Fiore, 2018).

Freshwater accounts for only some 6% of the world's water supply, but is vital for human uses such as agriculture, drinking, manufacturing, general hygiene and domestic uses (Fiore, 2018).

2.1.1 Ground water resources

Ground water is the most copious of all fresh water resources (Fiore, 2018). As water percolates, into the ground during layers of soil, rock, and clay, some of it adheres to the uppermost layers to supply water to plant. This water is called the unsaturated or vadose zone. Most of the pores in the vadose zone are filled with air rather than water (Fiore, 2018).

Though air in the vadose zone is at atmospheric pressures, the soil humidity is under stress, with suctions of a magnitude a lot superior to atmospheric pressure. This fluid pressure is formed by strong adhesive forces among the water and the solid grains, and by surface tension at the small interfaces between water and air. Aquifers are areas of porous rock that hold water. Usually, aquifers are made of foundation that has a lot of fractures and linked pores such as, sand stone, limestone and gravel. An aquifer is recharged through the layers of soil and rock (Fiore, 2018). Therefore, there is a major relation between surface water and ground water (Fiore, 2018). Aquifers may be either capped by an impermeable layer (confined) or open to accept water from the surface (unconfined). Confined aquifers are often artesian as the confining layer prevents groundwater from flowing upward, but non-confined aquifers are also artesian in the vicinity of discharge fields. This is why it discharges groundwater into rivers and streams (Fiore, 2018).

A confined aquifer is overlaid by a waterproof layer that keeps rainfall or surface water from recharging (and contamination). Recharge of confined aquifers happens when the permeable rock outcrops on or near the ground, which may be some distance from the surface area. This characteristic may make it harder to regulate quality and pollution. Some aquifers are not restricted to perfection and are termed semi-confined or leaky (Chilton 1996).



Plate 2.1: Confined and unconfined aquifer

2.1.2 Salt water resources

Salt water is rich in the earth's surface. Still, salt water is presently not mostly of use when it comes to potable water provisions, because of its high salt content. It includes oceans and seas.

2.1.3 Surface water resources

Surface water is the water that exists in reservoir streams, lakes and rivers. This water is mainly used for supplying drinking water, leisure, recreation, business, agriculture (including irrigation and aquaculture), transportation, and hydroelectric power generation. The public water supply is removed from surface water by more than 63%. Irrigation receives 58% of its withdrawals from ground water supply (Fiore, 2018).

Lakes and reservoirs: stratification is an significant factor affecting the quality of water in comparatively still profound waters, such as lakes and reservoirs. Stratification happens when water acts as two distinct bodies with distinct densities in a lake or reservoir, one floating on the other (Meybeck, *et al.*, 1996). It is most commonly caused by differences in temperature,

resulting in density differences (water has a maximum density of 4^{0} C), but occasionally by differences in solute concentrations (Meybeck, *et al.*, 1996).

Rivers: An understanding of the discharge regime of a river is extremely important to the interpretation of water quality measurements, especially those including suspended sediment or intended to determine the flux of sediment or contaminants (Meybeck, *et al.*, 1996). A river's discharge is linked to the nature of its catchment, especially its geological, geographical and climatic influences (Meybeck, *et al.*, 1996).

Sources of Water	Surface area	Volume (million km)	Volume (%)	Equivalent	Residence time
				ucptii (iii)	
Oceans and	361	1,370	94	2,500	~4,000 years
Seas					
Lakes and	1.55	0.13	< 0.01	0.25	~10 years
Reservoirs					
Swamps	<0.1	< 0.01	< 0.01	0.007	1-10 years
River channels	<0.1	< 0.01	<0.01	0.003	~2 weeks
Soil moisture	130	0.07	< 0.01	0.13	2 weeks to 50 years
Groundwater	130	60	4	120	2 weeks to 100,000 years
Icecaps and	17.8	30	2	60	10 to 1,000 years
Glaciers					
Atmospheric	504	0.01	< 0.01	0.025	~10 days
Water					
Biospheric	<0.1	< 0.01	<0.01	0.001	~1 week
Water					

 Table 2.1: Estimate of the World water balance

2.2 ECONOMIC IMPORTANCE OF LAKE

Due to their outstanding water quality lakes serves as a drinking water reservoir for large percentage of the world's population, especially in developing countries, and can also serve as form of tourism and recreational activities. Due to the varied relationships between water and human activities, socio-economic growth procedures are strongly linked to the water resource (United Nations, 1987). The most fundamental role of water in socio-economic development is its use for domestic purposes, drinking, personal hygiene, and other domestic purposes constitutes a primary component of welfare that is inadequately provided in much of the developing world (Biswas, 1978). While national water supply is in its position as a direct consumer commodity serves as a fundamental component of welfare, it also acts as an aspect of socio-economic infrastructure (Biswas, 1978). Existence of an adequate domestic water supply is factors in community stability that can affect the success of many of the other components of development (United nations, 1987) fisheries provide an example. Fishing activities contribute to human welfare in several ways, most directly by providing a source of food. Although fish provides only about one percent of the world's total food energy and about 11 percent of all animal protein, the importance is much greater in some nations and regions.(Norse, 1979).Improvement and expansion of irrigation are generally seen as essential to increasing agricultural production (FAO, 1978).

2.3 CLASSIFICATION OF LAKES

The scientific study of in-land waters (both fresh and saline), specifically ponds, lakes and rivers (both manmade and natural), including their physical, biological, chemical, and hydrological aspect is called Limnology.

Lake can be characterized by

a) Total dissolved load of nutrients and sediments

- b) Its maximum depth of water
- c) Quality of water
- d) Biotic species and their density
- e) Rate of Inflow and outflow of water
- f) Its volume of water
- g) Its basin, which is the depression holding the water
- h) Its surface area
- Lakes are classified on the basis of various categories/factors
- a) Mixing of water.
- b) Trophic levels
- c) Nature of Inflow-outflow.
- d) Origin

LAKES ARE GENERALLY CLASSIFIED INTO 8 CATEGORIES

- a) Oligotrophic lakes
- b) Mesotrophic
- c) Eutrophic
- d) Dystrophic lakes
- e) Acidotrophic lakes
- f) Alkalitrophic lakes
- g) Argillotrophic lakes
- h) Siderotrophic lakes

2.4 ECOLOGICAL FACTORS ASSOCIATED WITH LAKE

Statistical assessment of the information in relation to the features of the lakes showed that pH, temperature, and the theoretical hydrological retention time were most closely associated with differences in the distribution of bacterial taxa (Lindstrom, 2005). This indicates that pH and temperature are driving variables in taxa choice and promotes the concept that the input of bacterial cells from the drainage basins influences communities in ponds with brief water turnover times (Lindstrom, 2005). Zooplankton biodiversity was researched in the Tigris River running in Baghdad City, central Iraq. Fourteen physical and chemical parameters have been evaluated, including water and air temperature, pH, EC, turbidity, TDS, DO, BOD⁵, complete hardness, Ca⁺², Mg⁺², chloride, nitrate and phosphate reactive(Abdulwahab, and Rabee, 2015). In the coastal fields of the southern Baltic Sea and in the three lakes, the impacts of environmental variables on the population dynamics of pikeperch have been analysed. The research disclosed that temperature was the most significant environmental factor influencing Pikeperch's population dynamics. The temperature of the water had favourable impacts on annual abundances, development, starts and returns of the year. An apparent increase in significances among different temperature estimates was found from air temperature to water temperature, and to water temperature estimated as degree-days over a certain threshold (Lappalainen, 2001). The comparison results show a variability of the mentioned parameters. At Site II, due to arid climate, the high summer water temperature value (26.95°C) caused a sharp decrease in dissolved oxygen (3 mg/ l^{-1}) and also a decrease in the pH value. During the study period, the salinity of the lagoon ranged from 12.01 to 24.6 mgl⁻¹. A gradient of increasing salinity is observed from site III to site I. Summer recorded high evaporation caused by the highest values (Badsi, 2010). The interaction between these parameters at this station can be explained by the elevated microbial activity in sludge sediment, leading in a reduction in dissolved oxygen concentration and an increase of nitrate and phosphate. (Kagalou *et al.*, 2006)

2.5 MICROORGANISMS ASSOCIATED WITH LAKE

Freshwater lakes are a major contributor of methane to the atmosphere-more so than the world's oceans combined. Some anaerobic microorganisms produce methane in sediments or deep anoxic water. Heterotrophic bacteria break down methylphosphonate as a phosphorus source, releasing methane in the process (Vignieri and Smith 2016). Everywhere on Earth there are microorganisms that will support life. These include habitats with which we are all familiar, soil, water, animals, and plants as well as virtually any manmade structures. Some microbial habitats are those where humans are too hot or too cold, too acidic or too acidic or too caustic, or too salty (Corliss, et al., 1990). About 6000 prokaryote species and 100,000 protists species were officially defined (Corliss, et al., 1990). Even the correct order of magnitude is unknown in the event of microorganism diversity and the problem is extremely contentious (Hedlund and Staley 2004). It was estimated that the complete amount of prokaryotic cells in the oceans was 1029 (Whitman, 1998). All prokaryotic organisms are categorized as bacteria, while eukaryotic organisms include both fungi, protozoa, helminths and higher animals including man. Prokaryotic organisms are split into two main groups: eubacteria, comprising all medically important bacteria, and archaebacteria, a collection of evolutionarily separate organisms (Corliss et al., 1990). Limnetic area areas with adequate oxygen contain Cytophaga, Caulobacter, and Hyphomicrobium pseudomonads and species. There are photosynthetic algae in the limnetic zone. In the deep zone, violet and green sulfur bacteria are found. These bacteria are photosynthetic anaerobic organisms that metabolize H2S into sulfur and sulfate in the benthic zone's lower sediments. (Tortoraand and Funke 2008). Clostridium species are prevalent in low sediments and may include botulism

organisms, especially those that cause botulism outbreaks in waterfowl (Aryal *et al.*, 2008). Fungi from several pathogenic plant genera including *Botryosporium, Cercosporidium, Chaetophoma, Diplodia,* and *Pyrenochaeta* were discovered primarily in soil samples. Very few *actinomycetes* with a few from pond *hydrilla* were retrieved, mostly from pond soils.

2.6 BACTERIAL POPULATION AND THEIR VARIATION IN LAKE

Blelham Tarn's microbial littoral (shallow water) and deep (deep water) ground sediments and Windermere's South Basin were examined (Jones, 1989). Microbial numbers (immediate numbers), biomass (ATP) and activity (system activity of electron transport, CO_2 evolution and glucose mineralization) were consistently higher in the profundal zone of both lakes, the difference being greater and average values higher in the more productive Blelham Tarn (Jones, 1989). The material analysed consisted of 1976 water beetles (24.2 percent of the total collected material) (Pakulnicka, 2008), which accounted for 87 species (Online Appendix), which is 68 percent of the species wealth determined in all the water bodies analyzed. Seventy-eight species were discovered in clay pits, while 37 species lived in gravel pits. The Dohuk River was regarded as the main cause of this pollution. The occurrence of heterotrophic organisms in the summer is 57% higher than the winter values (Khattab and Merkel, 2012). A linear correlation between heterotrophic bacteria and water quality parameters (R2>0.7) could be observed in winter in a layer 12–15 m below the lake's surface at a temperature of 10.9 ° C (Khattab and Merkel, 2012).

CHAPTER THREE

3.0 MATERIALS AND METHODS

3.1 STUDY AREA

The study of Magada Lake was carried out in rainy season in the month of June, 2019 which flows from the Kuro community. The actual part of the water body studied in this research work is located within Mountain Top University Campus which is bounded with Latitudes 743700 and 744100mN and Longitude 545100 and 545900mE. The Mountain Top University Campus is located at Prayer City, Kilometre 12 Lagos- Ibadan Express Way, Ogun State. The water body is typically lentic and thus does not have a defined unidirectional flow pattern, except limited movement as a result of wind action,



Figure 3.1: Map of Mountain Top University Showing the Longitude and Latitude



Plate 3.1: Composite sampling station1 in Magada Lake flowing into MTU.



Plate 3.2: Composite sampling station 2 in Magada Lake flowing into MTU.



Plate 3.3: Composite sampling station 3 in Magada Lake flowing into MTU.



Plate 3.4: Control sampling station 4 in close proximity with Magada Lake flowing into MTU.

3.2 APPARATUS USED

Petri dishes, cover slips, slides, conical flask, inoculating needle, cotton wool, aluminium foil, dropper, Test tubes

3.3 EQUIPMENTS USED

Bunsen burner, water bath, incubator, autoclave, magnetic stirrer, weighing balance, Microscope

3.4 AGAR USED: Nutrient agar

3.5 WATER COLLECTION FOR PHYSICO-CHEMICAL ANALYSIS

The water samples were collected in the month of June, 2019 in a clean sterilized reagent bottles (in three replicates) of 50ml from sampling stations (station 1, 2, 3 and control) at time variations of 12am in the morning, 6am in the morning, 12noon and 6pm in the evening from MTU Magada Lake, Lagos-Ibadan express way, Ogun state. Water samples including control in triplicates were collected from three stations in Mountain Top University, and one control station from the untreated borehole in close proximity with the water body. The sampling points were selected based on anthropogenic activities around such study stations. For the physico-chemical analysis, the water samples were collected using sterilized reagent bottles at all the four stations, they were submerged into the water before opening the cover to avoid air from entering into it. In collecting water samples from the control station the water from the borehole was first filled into a pre-cleaned and sterilized bucket, and samples were immediately collected as with the other stations. The water samples used for dissolved oxygen analysis was collected in a dark amber bottles to avoid light penetration into the bottles while for heavy metal analysis, one drop of nitric acid was added to the water samples. The bottles containing water samples used for ions and nutrients were immediately

placed in a water cooler containing ice blocks (and later transferred into the refrigerator in the laboratory). For microbiological analysis, the water samples were carefully and aseptically collected, stored in ice packs before transporting them to the laboratory to maintain the original state, while some physico-chemical parameters like (pH, temperature, salinity, conductivity, turbidity, and dissolved oxygen) were done in-situ.

3.6 DETERMINATION OF ECOLOGICAL FACTORS

In-situ observations were made for some physico-chemical parameters during this research. These physical features are as follows: Air and water temperature, depth, pH, turbidity, conductivity, salinity and dissolved oxygen.

3.6.1 In-situ determination of physico-chemical parameters

3.6.1.1pH

The pH of the water samples collected at each station was done in-situ using a probe by dipping the probe into a beaker containing the water samples and left for 2-3 minutes before readings were taken. This process was done three times.

3.6.1.2 Temperature

Using a probe, the air and water temperatures were taken at each station where by the probe was left inside each medium (air and water) for 3 - 5 minutes before the readings were taken. This was done in triplicate.

3.6.1.3 Depth

The depth was measured using a straight calibrated wooden pole, which was dipped into the water body until it got to the bottom and the reading was taken directly for each station.

3.6.1.4 Turbidity

Turbidity was done with the aid of a turbidimeter, where the water samples were placed in a sample compartment and the result was read, this sampling was done in triplicate for every station and time variance

3.6.1.5 Conductivity

The samples were measured using a HACH Conductivity metre which was first calibrated before dipping the probe into the water body, and the readings were then taken directly, the readings were taken in triplicate for each study station.

3.6.1.6 Salinity

Salinity was carried out by using a calibrated salinity probe. The probe was dipped into a beaker containing the water samples, which was done in triplicate for each station at different time frame as earlier indicated. The probe was left in the medium for some minutes before readings were taken.

3.6.1.7 Dissolved Oxygen

A reagent bottle was immersed beneath the water surface, to exclude air bubbles, the cover was opened beneath the water and it was stoppered tightly. The samples were then placed in a cell and put in a sample compartment in the equipment and the readings were recorded after 1minutes, this was equally done in all the stations at the appointed time intervals and in triplicates.

3.6.2 Ex- situ determination of physico-chemical parameters

During this research period, ex situ analysis was carried out for the following parameters which include: Heavy metals (Zn, Fe, Pb, Arsenic, Cr, and Cu), nutrients (nitrate, phosphate, and sulphate), hardness, BOD₅, calcium, magnesium, Total alkalinity, acidity and solids (total, dissolved and suspended).

3.6.2.1 Biochemical Oxygen Demand (BOD₅)

Dark reagent bottles were used to collect water samples from the various stations, and were wrapped in black polythene bags. The various samples were taken back to the laboratory and incubated at a temperature of $20 - 25^{\circ}$ C for 5 days. Then after 5 days the oxygen was fixed and determined by the same process as that of dissolved oxygen, and then the results were compared with dissolved oxygen. This was calculated as thus;

 $BOD_5 (mgl^{-1}) = DO_0 - DO_5$

Where DO_0 = the dissolved oxygen at the time the water samples were collected

 DO_5 = the dissolved oxygen after 5 days

3.6.2.2 Total Alkalinity

To test for total alkalinity, two drops of phenolphthalein indicator was added to the water sample in a conical titration beaker, if the solution was colourless, it meant phenolphthalein alkalinity was zero and methyl indicator was therefore added. The volume of HCL needed to reach the first end point was used to calculate phenolphthalein alkalinity. The total alkalinity was then estimated by the addition of 2 drops of methyl orange indicator to the water sample, it turned blue black and was then titrated with 0.01N HCL. A faint pink colour appearance was observed at the end point. The total alkalinity in mgl⁻¹ CaCO₃ was obtained from the formula,

Total Alkalinity =
$$\frac{ml \times Molarity \ of \ titrant}{ml \ of \ sample} \ge 50,000$$

3.6.2.3 Phosphate

The determination of phosphate was carried out by measuring 25ml of the water sample with 1ml of ethanol added to the samples. A ready mixture of reagent A (ammonium molybdate and antimony potassium tartate) and ascorbic acid was made and 1ml was added to the 25ml of the sample ethanol. Absorbence interpretation using a Milton Roy 21D Spectrophotometer at 952nm was noted and this was in turn multiplied by a multiplying factor of 30.6 to estimate the value of phosphate.

3.6.2.4 Nitrate

A beaker containing 10ml of water sample was heated to dry in an oven. After being brought out, to extract nitrate, 1ml of phenyldisulphuric acid was added to the beaker. A paste was formed by using a glass rod and the paste was diluted with 5ml of distilled water, then 3ml of concentrated ammonia solution was added to the sample and a yellow colour was developed. Using a spectrophotometer, absorbance reading was taken at 410nm and the obtained value was multiplied by a multiplying factor of 4.43 to get the value of the nitrate.

3.6.2.5 Sulphate

25ml of the samples were measured in a conical flask and 1.25ml of condition reagent, then 50ml glycerol, 30ml conc. Hydrochloric acid, 300ml distilled water, 175g Nacl solid 100ml 95% alcohol of condition reagent in 1.25mlwere added. Using spatula, few grammes of barium chloride was added and absorbance reading of the sample was taken at 530nm wave length and the value was multiplied by a multiplying factor of 100 to get the value of sulphate.

3.6.2.6 Calcium

This was done titrimetrically using 0.01M EDTA APHA (1975). 50mls of the water sample was put in a 250ml Erlenmeyer flask with the addition of 2ml of 1M sodium hydroxide solution with some grams of (about 0.2g) of the murexide indicator mixture and 1ml of hydroxylamine chloride which will serve as the inhibitor. Thereafter, the 0.01M EDTA was added until the pink colour of the sample turned to purple. To calculate the concentration of calcium in mgl⁻¹ by multiplying the titre value obtained with 0.4008, the concentration of calcium in mgl⁻¹ was therefore estimated.

3.6.2.7 Magnesium

This was also done titrimetrically by using the EDTA method. In this method, the calcium and magnesium concentrations were determined together before the concentration of magnesium was extrapolated. Thereafter, 50ml of sample was put in an Erlenmeyer flask and 3ml of ammonical buffer was added followed by 2 drops of Eriochrome Black T, the indicator/inhibitor solution. The titre 0.01M EDTA solution was added to the sample until it changed from wine to sky blue end point. The value of magnesium (mg) was obtained by the following equation.

$$Mg = (a-b) 0.2432$$

Where k= value of magnesium in mgl⁻¹

a= titre volume for calcium and magnesium titration

b= titre volume for calcium titration only.

3.6.2.8 Heavy Metals

Using the Aluminium Block Digester BD 110, ten millilitre of water sample was digested having added 4ml perchloric acid; 20ml concentrated nitric acid and 2ml concentrated sulphuric acid. The mixture was then heated until white fume evolved and a clear solution was obtained. Iron was determined colourimetrically using Milton Roy Spectrophotometer by Orthophenamthroline method. Lead, chromium and cadmium were determined colourimetrically using Milton Roy 21D Spectrophotometer.

3.7 COLLECTION OF WATER SAMPLES FOR MICROBIAL ANALYSIS

Water sample was collected for the microbial analysis of this project. The water samples were collected in a clean and sterilized 50mlMcCartney bottles from the various sampling stations including the control, at time variations of 6:00am in the morning, 12pm in the afternoon, 6:00pm in the evening and 12am in the morning from MTU Magada Lake, Lagos-Ibadan express way, Ogun state and analytical study of the water sample was carried out in the Microbiology laboratory of Mountain Top University for identification of possible micro-organism in the water body.

3.8 PREPARATION OF MEDIA

The media used for isolation was Nutrient Agar. The Petri dishes and Durham bottles to be used for isolation were sterilized using the dry heat sterilization method (Oven) at 160°C for 1hr. It was prepared by weighing 28g of Nutrient Agar in 1000ml in a conical flask using a weighing balance. They were sterilized respectively and kept in the water bath for homogenization. After which they were transferred to the autoclave and sterilized at 120mmHg for 1hr. The media were then allowed to cool down to and maintain at temperature of 45° C using a water bath. The media were then poured into the Petri dishes aseptically and allowed to set.

3.9 SERIAL DILUTION AND ISOLATION OF BACTERIA

Serial dilution of the water samples were made from stock, by putting 1ml of the water sample into 9ml of distilled water in a test tube. Using a five-fold dilution, 10^{-1} dilution factor was made by taking 1ml of water from the stock and then 10^{-2} dilution factor was made by taking 1ml of water from 10^{-1} dilution factor until 10^{-5} dilution factor was made. Isolation was carried out using the pour plate method whereby 0.1ml of the water samples were siphoned from the dilution factors 10^{-1} and dispensed in a petri dish using a micropipette, and the agar was allowed to cool but not solidify and 20ml was aseptically poured into the sterilized glass petri dishes containing dilution factors 10^{-1} and allowed to set and the process was done for 10^{-3} and 10^{-5} and also for the control which was taken from the untreated borehole water. This process was done aseptically to avoid contamination. The petri dishes were left for 20mins before inverting them and transferring into the incubator at 37° C for 24-48hrs. Colonies grown on media were enumerated and calculated as colony forming units per cubic meter (CFU ml⁻⁵). The results were interpreted and documented

3.9.1 SUBCULTURING OF ISOLATES

Distinct colonies from each culture plates were sub-cultured by streaking them on fresh agar plates in order to obtain pure culture to carry out biochemical tests to identify bacteria. The purpose of sub-culturing colonies is to isolate a colony from various colonies inside a medium and plate inside a fresh nutrient medium. The pure isolates were transferred onto agar slant in McCartney, inoculated for 14-18hrs before transferring into a refrigerator at 4°C to serve as stock culture for subsequent test during identification. The sub-culturing process was carried out aseptically to prevent contamination. The petri dishes were inverted and transferred into the incubator at 37°C for 24hrs



Plate 3.5: Bacteria Culture plate

3.9.2 BIOCHEMICAL TESTS FOR IDENTIFYING THE ISOLATES

The tests carried out in identifying the isolates are: Gram staining, catalase test, oxidase test, sugar fermentation test, and indole test

3.9.3 GRAMS STAINING: Reagents used were: Crystal violet, Gram's iodine, safranin and 70% alcohol. The inoculating loop was sterilized in a flame of a bunsen burner, then a pure culture was smeared on a sterile slide and heat fixed by passing it across the flame quickly with the smear facing up. The slides were placed on the staining rack for staining. The smear was covered with crystal violet stain and allowed to stand for 1 minute, then washed off carefully under running tap water. Gram's iodine was then used to flood the smear and allowed to stand for 1 minute, and then drained off under gentle running tap. Afterwards, the slide was flooded with decolorizing agent (70% alcohol) and then allowed to stand for 10 seconds. The slide was then washed under running tap water, drained completely and counterstained with safranin for 30 minutes. Then, slide was then washed under gently running tap water until no colour appeared in the effluent and then blot dried with filter paper and then viewed under the microscope







Plate 3.6: Microscopic view of Bacteria Isolate

3.9.4 CATALASE TEST:

Reagent used: Hydrogen peroxide

The pure culture was smeared on a sterile slide using a sterilized inoculating loop. Then, a drop of hydrogen peroxide was dropped on the smear. The result was then observed. The presence of oxygen bubbles showed the presence of catalase and the absence of the oxygen bubbles showed the absence of catalase.

3.9.5 OXIDASE TEST:

Reagent used: Oxidase reagent

The pure culture was smeared on the filter paper and few drops of the oxidase reagent was added and the results were observed. A purple colouration was produced within 10 seconds by oxidase positive cultures. Absence of purple colouration was produced by oxidase negative cultures

3.9.6 INDOLE TEST:

Reagent used: Kovac's Reagent

Peptone water was poured in to 19 test tubes and inoculated with a loopful of the 19 isolates respectively. Then, it was incubated for 5-7 days at 37^oC. Afterwards, 0.5ml of Kovac's reagent was added to the test tubes and gently shook and allowed to stand. The colour was observed

CHAPTER FOUR

4.1 RESULTS

In-situ physico-chemical parameters with respect to time (as shown in table 4.1); the air temperature was relatively high at 6am which is nearly similar to that of the control (27.30 °c). However, the air temperature at 12am (23.62°c) and 12pm (26.86°c) are similar, while that of 6pm (23.61°c) was the least of time in terms of value. The water temperature of the control (28.26°c) was outstanding in comparison to 12am (27.03°c), 6am (27.93°c), 12pm (26.82°c) and 6pm (27.05°c), though 12pm (28.26) had the lowest value. At 12am (7.63) and 12pm (7.16), the pH value is neutral which has the highest pH value. In comparison to 6pm (6.34), the pH is slightly acidic; however, the control was completely acidic. At 6am, the salinity is remarkably high with a value of (130.4µcm) which is closely followed by 12pm (113.7µcm). The salinity at 6pm (80.61µcm) is nearly comparable with the control (86.00µcm), while that of 12am is the smallest with value of (12.63µcm). The turbidity at 12am (17.94NTU) and 6pm (18.67NTU) are relatively close, though, 6pm is having the highest value of (18.60NTU). Remarkably, control has the lowest value of (0.07NTU). Dissolved oxygen (DO) is outstanding at 12pm (15.32mgl⁻¹) and is closely tailed by 6pm (13.57mgl⁻¹). However, DO at 6am (11.53 mgl⁻¹) is also significantly higher than that of 12am (9.68mgl⁻¹) and with that of control having the least value (0.066mgl⁻¹). Conductivity at 12pm (0.14µscm¹) is similar to that of 6pm (0.14µscm¹) having the least values. The conductivity at 6am (0.26µscm¹) increases above that of 12pm and 6pm. The conductivity of the control is the highest value $(0.846 \mu \text{scm}^1)$ which is then followed by the conductivity at $12am (0.654 \mu s cm^{1}).$

The ex-situ physico-chemical values are as stated in Table 4.2 The phosphate level at 12am (0.009mgl^{-1}) and $12 \text{pm} (0.009 \text{mgl}^{-1})$ are similar, while the phosphate level at 6am (0.008mgl^{-1}) and 6pm (0.008mgl^{-1}) are also similar with the control having the highest value (0.014mgl^{-1})

¹) .The sulphate levels at 6pm (6.24mgl⁻¹) is greater than the rest of the other time variation, with the control (2.54mgl⁻¹) being the least. The 12am (3.48mgl⁻¹) is slightly above the control with 6am (4.91mgl⁻¹) and 12pm (4.88mgl⁻¹) similar in comparison with each other. The control of nitrate has a remarkably low value (0.078mgl⁻¹). The 12pm (8.62mgl⁻¹) value is incomparable to the control but is the highest among the other time. The 12am (4.04mgl⁻¹) is followed by 6am (7.16mgl⁻¹) and 6pm (7.53mgl⁻¹). The control (10.00mgl⁻¹) of calcium is the least in value and 12am (17.34mgl⁻¹) having the highest value. At 12am, the level of calcium is remarkable and is incomparable with the control. The content of calcium at 12pm (16.37mgl⁻¹) is lower than the value of 12am (17.34mgl⁻¹). At 6pm (15.74mgl⁻¹) the value is incomparable with 6am (14.09 mgl⁻¹). Hence, 6pm (15.74mgl⁻¹) has higher calcium content than that of 6am (14.09mgl⁻¹).

The heavy metal values are as represented in table 4.3. The 12pm (0.37mgl⁻¹) and 6pm (0.37mgl⁻¹) have similar cadmium content which is the highest content. Similarly, the 12am (0.003mgl⁻¹) and 6am (0.007mgl⁻¹) also had low content of cadmium; hence they are significantly lower in value than 6pm and 12pm, while the control value was not detected. There was reduction in the cadmium values from 6am followed by 12am and 12pm and with the value at 6pm being visibly lower than the other values while, the control station cadmium content was however not detectable. The content of lead as at 6pm (0.036mgl⁻¹) showed a visible variation when compared with the other times of 12pm (0.004mgl⁻¹), 6am (0.001mgl⁻¹), and 12am (0.0003mgl⁻¹) which were on the decrease while the control lead value was still not detectable. The results of chromium show that the 6pm (0.71mgl⁻¹) was outstandingly higher than the rest of the three other sampling periods. The values decreased from 12pm (0.024mgl⁻¹), to 12am (0.036mgl⁻¹) and then to 6am (0.075mgl⁻¹) and 6pm (0.710mgl⁻¹). The control value was however undetectable. The hardness value at 6pm (14.98mgl⁻¹) and 6am (14.82mgl⁻¹) were corresponding while the values at 12pm (12.26mgl⁻¹) and 12am (12.67mgl⁻¹)

¹) were however lower, with the control having a value of (10.13 mg^{-1}) . The highest alkalinity value was recorded at 12pm (1.528mgl⁻¹) while lowest values were recorded at 6pm $(0.562 \text{ mg}l^{-1})$ and the control value is $(1.13 \text{ mg}l^{-1})$.

The Biochemical oxygen demand (BOD₅) values for the various time periods are as stated in table 4.4. At 12pm (0.16), the biochemical oxygen demand was relatively high and however closely followed by the value 6pm (0.15), which were still correspondingly higher when compared with the values at 6am (0.07) and 12am (0.05) decrease respectively. Total dissolved solid values were relatively higher at 12am (33.26mgl⁻¹) and 6pm (34.36mgl⁻¹), followed by the 6am (16.04mgl⁻¹) values, by the 12pm (13.69mgl⁻¹) value, and with the control (0.866mgl⁻¹) having the lowest value. The values of the total suspended solids and total solids also followed the same pattern as that of total dissolved solid. In table 1, comparatively; apart from pH that shows a significant difference (p<0.05) other parameters such as air temperature, water temperature, salinity, turbidity, dissolved oxygen; conductivity and depth were not significantly different.

The bacterial population with time is as showed in figure 4.23The result of the study showed that at 6am (81) and 12am (80), the bacterial population was very high in comparison to 12pm (6), and 6pm (55), and with the bacterial population load being lowest at 12pm.

Figure 4.24 represents the bacterial population across the time variation. The result of the study showed that at 12am (45) the bacterial population was very high in comparison to 12pm (20), 6am (18), and 6pm (41). Likewise, the bacterial population load was very low at 12pm.

Figure 4.25 represents the bacterial population cross the time variation. The result shows that at 6am (26), the bacterial load was low and in comparison to 12am (37), and 12pm (50), and, the bacterial load was relatively high for 6pm (66).

Figure 4.26 represents the bacterial population loads in control. At 6pm (18), the bacterial load was on the high side in comparison to 6am (15), 12pm (9), and 12am (7). The time at which the bacterial population is on the low side is at 12am.

Figure 4.27: Shows the overall graph of the bacterial population with respect to time, the bacteria population was at the peak at 12am (80).

Physico-Chemical Parameters				p-Value
	Time	Mean	SD	
Air Temperature(°c)	12AM	25.6222	±0.78975	P<0.05
	6AM	27.8667	± 0.88882	
	12PM	26.3889	±1.03297	
	6PM	23.6111	±3.77629	
	CONTROL	27.3000	±0.000	
Water Temperature(°c)	12AM	27.0333	±0.43333	
	6AM	27.9333	±0.53645	
	12PM	26.8222	±0.60123	
	6PM	27.0556	± 1.02974	
	CONTROL	28.2667	±0.000	
рН	12AM	7.6333	±0.31798	
	6AM	6.9889	±0.15396	
	12PM	7.1667	±0.65574	
	6PM	6.3444	±0.45010	
	CONTROL	5.888		
Salinity(mgl ⁻¹)	12AM	7.3333	±0.25166	

Table 4.1: Summary of Insitu physico-chemical characteristics of Magada Lake.

	6AM	130.4444	±48.55962	
	12PM	113.7778	±54.75839	
	6PM	80.6144	±78.44701	
	CONTROL	86.0000	±0.000	
Turbidity(NTU)	12AM	17.9478	±8.07649	
	6AM	7.9178	±3.74110	
	12PM	6.5844	±4.68186	
	6PM	18.6700	±15.26461	
	CONTROL	.0700	±0.000	
Dissolved oxygen(mgl ⁻¹)	12AM	9.6800	±1.13210	
	6AM	11.5367	±3.77971	
	12PM	15.3222	±3.01300	
	6PM	13.5778	±4.99793	
	CONTROL	0.0667	±0.000	
Conductivity(µscm ¹)	12AM	0.6544	±0.68406	
	6AM	0.2656	±0.10178	
	12PM	0.1467	±0.08083	
	6PM	0.1456	±0.07042	
	CONTROL	0.8467	±0.000	
Depth(cm)	12AM	3.6000	±1.47309	
	6AM	3.6000	±1.47309	
	12PM	3.4667	±1.30512	
	6PM	3.4667	±1.30512	
	CONTROL	4.5000	±0.000	

PHYSICO-		STD	P -VALUE/ SIG
CHEMICAL			
PARAME I ERS		MEAN DEVIATIO	
Phosphate(mgl ⁻)	12AM	0.0090 ±0.00624	p>0.05
	6AM	0.0087 ±0.01016	
	12PM	0.0092 ±0.00366	
	6PM	0.0085 ± 0.00585	
	CONTROL	0.0140 ±0.00000	
Sulphate(mgl ⁻¹)	12AM	3.4810 ±3.00319	
	6AM	4.9139 ±2.14602	
	12PM	4.8880 ±4.12575	
	6PM	6.2476 ±0.57927	
	CONTROL	2.5447 ±0.00000	
Nitrate(mgl ⁻¹)	12AM	4.0456 ±3.60322	
	6AM	7.1667 ±2.43333	
	12PM	8.6278 ±01.1219	
	6PM	7.5356 ±0.12760	
	CONTROL	0.0788 ±0.00000	
Magnesium(mgl ⁻¹)	12AM	1.5033 ±1.29950	
	6AM	2.6300 ±0.59851	
	12PM	4.5956 ±03.4622	
	6PM	2.2989 ±0.29410	
	CONTROL	2.3800 ±0.00000	
Calcium(mgl ⁻¹)	12AM	17.344 ±1.52036	
	6AM	14.094 ±2.12723	
	12PM	16.377 ±2.93359	
	6PM	15.744 ±3.73651	
	CONTROL	10.000 ±0.00000	

 Table 4.2: Summary of Nutrients characteristics in Magada Lake

 Table 4.3: Summary of ex situ physico-chemical characteristics of Magada Lake

PHYSICO- CHEMICAI			STD		
PARAMETERS	TIME	MEAN	DEVIATION		P- VALUE
Cadmium(mgl^{-1})	12AM	0.0032	+0.00291	p>0.005	I VILLE
Cuulinum(ingi)	6AM	0.0077	+0.01002		
	12PM	0.3747	+0.64894		
	6PM	0.3760	± 0.01091 ± 0.63945		
	CONTROL	0.0000	+0.00000		
	CONTROL	0.0000	0.00000		
Iron(mgl ⁻¹)	12AM	0.7310	±0.63950		
	6AM	1.0457	±0.97769		
	12PM	0.8386	±0.62883		
	6PM	0.3401	±0.42383		
	CONTROL	0.0000	± 0.00000		
$Lead(mgl^{-1})$	12AM	0.0003	± 0.00058		
	6AM	0.0010	±0.00173		
	12PM	0.0043	±0.00751		
	6PM	0.0363	± 0.05950		
	CONTROL	0.0000	± 0.00000		
		0.0267	0.06251		
Chromium(mgl ⁻¹)	12AM	0.0367	± 0.06351		
	6AM	0.0750	±0.07744		
	12PM	0.0241	± 0.03835		
	6PM	0.7102	±0.61796		
	CONTROL	0.000.00			
		0			
Hardness(mgl ⁻¹)	12AM	12.6778	±4.47367		
	6AM	14.8200	± 2.25868		
	12PM	12.2611	±5.88837		
	6PM	14.9844	± 5.70780		
	CONTROL	10.3167	± 0.00000		
Alkalinity(mgl ⁻¹)	12AM	1.3656	±0.37144		
	6AM	1.0200	±0.19757		
	12PM	1.5289	±0.01388		
	6PM	0.5622	±0.45928		
	CONTROL	1 1 3 0 0	+0.0000		
Table 4.4: Summary of other ex situ physico-chemical characteristics of Magada Lake

PHYSICO-				P VALUE
CHEMICAL			STD.	
PARAMETERS		MEAN	DEVIATION	P< 0.05
BOD ⁵	12AM	0.0556	± 0.09623	1 < 0.05
	6AM	0.0722	0.048110	
	12PM	0.1667	±0.10000	
	6PM	0.1556	±0.03849	
	CONTROL	0.0000	± 0.00000	
TDS (mg^{-1})	12AM	33.2611	±14.20043	
	6AM	16.0400	±8.20978	
	12PM	13.6989	±8.67599	
	6PM	34.3611	±27.08495	
	CONTROL	0.866±.0		
TSS (mg^{-1})	12AM	20.3289	±9.82747	
	6AM	8.6356	±4.15949	
	12PM	7.3856	±5.84371	
	6PM	20.7033	±18.00330	
	CONTROL	0.0500	± 0.000000	
Total solid (mg ⁻¹)	12AM	53.5900	±24.01837	
	6AM	24.6756	±12.36826	
	12PM	21.0511	±14.53360	
	6PM	55.0611	±45.07567	
	CONTROL	0.93670	$\pm .0.00000$	



Figure 4.1: water temperature



Figure 4.2: Air Temperature







Figure 4.4: Salinity



Figure 4.5: Turbidity



Figure 4.6: Dissolved Oxygen



Figure 4.7: Conductivity



Figure 4.8: Depth



Figure 4.9: Phosphate



Figure 4.10: Nitrate



Figure 4.11: Magnesium



Figure 4.12: Calcium



Figure 4.13: Cadmium



Figure 4.14: Iron



Figure 4.15: Lead

Figure 4.16: Chromium



Figure 4.17: Hardness



Figure 4.18: Alkalinity



Figure 4.19: BOD



Figure 4.20: TDS



Figure 4.21: TSS



Figure 4.22: TS



Figure 4.23: Bacterial population across the time variation



Figure 4.24: Bacterial population across the time variation



Figure 4.25: Bacterial population across the time variation



Figure 4.26: Bacterial population in control across the time variation



Figure 4.27: bacterial populations across time variation

Table: 4.5PRESUMPTIVE IDENTITIES OF BACTERIA ASSOCIATED WITHWATER

S/N	LAB CODE	ISOLATE IDENTITY
1	6awm1a	Bacillus anthracis, Staphylococcus aureus
2	6awm1b	Bacillus anthracis, Staphylococcus aureus
3	6awm2a	Bacillus cereus, Staphylococcus aureus
4	6awm2b	Bacillus cereus, Staphylococcus aureus
5	6awm3a	Bacillus thuringiensis Staphylococcus epidermidis
6	6awm3b	Bacillus thuringiensis, Staphylococcus epidermidis
7	12pwm1a	Bacillus subtilis, Staphylococcus aureus
8	12pwm1b	Bacillus subtilis, Staphylococcus aureus
9	12pwm2a	Bacillus anthracis, Staphylococcus aureus
10	12pwm2b	Bacillus anthracis, Staphylococcus aureus
11	12pwm3a	Bacillus thuringiensis, Staphylococcus epidermidis
12	12pwm3b	Bacillus thuringiensis, Staphylococcus epidermidis
13	6pwm1a	Bacillus cereus, Staphylococcus epidermidis
14	6pwm1b	Bacillus cereus, Staphylococcus epidermidis
15	6pwm2a	Staphylococcus epidermidis, Bacillus thuringiensis

16	6pwm2b	Staphylococcus epidermidis, Bacillus cereus,
17	6pwm3a	Staphylococcus aureus, Bacillus cereus,
18	6pwm3b	Staphylococcus aureus, Bacillus cereus,
19	12awm1a	Staphylococcus saprophyticus, Bacillus cereus,
20	12awm1b	Staphylococcus saprophyticus, Bacillus thuringiensis
21	12awm2a	Staphylococcus epidermidis, Bacillus subtilis
22	12awm2b	Staphylococcus epidermidis, Bacillus subtilis
23	12awm3a	Staphylococcus aureus, Bacillus thuringiensis
24	12awm3b	Staphylococcus aureus, Bacillus subtilis

TABLE 4.6 MICROBIOLOGICAL CHARACTERISTICS

S/N	ISOLATES	GRAM STAINING +/-	GRAM STAINING SHAPE	COLOUR	SHAPE OF COLONY	APPEARANCE	CATALASE +/-	OXIDASE +/-	INDOLE +/-
1.	6awm1a	+	Long rods	White, cream,	Circular, punctiform	dull,	_	_	_
2.	6awm1b	+	Short rods	White, cream	Circular, punctiform	Dull	_	_	_
3.	6awm2a	+	Long rods	White, cream	Round, lobate, punctiform	Translucent	_	+	+
4.	6awm2b	+	Cocci, in form of diplococci, staphylococci	White, cream, yellow	Circular, punctiform	Dull	+	_	+
5.	6awm3a	_	Cocci	Yellow, white, cream	Circular, punctiform, lobate	Dull	+	_	_
6.	6awm3b	_	Cocci	White, cream	Round, punctiform	Translucent, dull	+		
7.	12pwm1a	+	Cocci	Cream,	Circular,	Shiny, dull,			

					punctiform,	translucent	_	_	_
8.	12pwm1 b	+	Cocci	Yellow, white, cream	Circular, punctiform, lobate,	Dull, shiny, translucent	+	+	_
9.	12pwm2a	+	Cocci	Cream, yellow, white	Circular, undulate, punctiform	Dull, transparent, translucent	+	_	+
10.	12pwm2 b	+	Cocci	Cream, yellow, white	Circular, undulate, punctiform	Dull, transparent, translucent	+	+	+
11.	12pwm3a	+	Long and short rods	Cream, yellow, white	Circular, undulate, punctiform	Dull, transparent, translucent	+	+	_
12.	12pwm3 b	+	Short rods in chain	Yellow, white, cream, pink,	Circular, punctiform, lobate	Translucent, shiny, dull	+	+	_
13.	6pwm1a	+	Clustered Cocci	Cream, white,	Punctiform, circular	Dull, shiny, translucent	+		+
14.	6pwm1b	+	Clustered Cocci	Cream, white,	Punctiform, circular	Dull, shiny, translucent	_	+	+
15.	6pwm2a	_	Clustered short rods	White, cream,	Punctiform, circular	Shiny, dull, translucent	+	+	
16.	6pwm2b	+	Clustered short rods	White, cream,	Circular, punctiform,	Translucent, shiny, dull	+	+	_
17.	6pwm3a	+	Cocci	White,	Circular,	Translucent,	+	_	_

				cream,	punctiform,	shiny, dull			
18.	6pwm3b	_	Cocci	White, cream,	Circular, punctiform,	Translucent, shiny, dull	+	_	-
19.	12awm1a	_	Cocci	White,	Circular, punctiform, lobate	Dull	+	_	_
20.	12awm1b	+	Long and short rods	White,	Circular, punctiform, lobate	Dull	+	+	Ι
21.	12awm2a	+	Cocci	White, yellow, cream	Circular, punctiform	Translucent	+	_	
22.	12awm2b	+	Cocci	White, yellow, cream	Circular, punctiform	translucent	+	_	_
23.	12awm3a	+	Cocci	White,	Circular, punctiform, irregular	Dull, shiny	+	+	+
24.	12awm3b	_	Long and short rods	Pink, white,	Irregular	Shiny, dull,	+	_	_

CHAPTER FIVE

5.0 DISCUSSION

This research study established the diurnal variation in the physico-chemical parameters and bacterial population in Magada lake. It is observed that some ecological influences physicochemical factors subsequently contributed to the high bacterial population at 6am and 12pm especially in station 1, where there was anthropogenic influence. This suggests that the high presence of human activities in this station affected the microbial load thereby leading to an increase. The microbial load was higher in this station probably due to point charges of sewage from toilets, residential hostels, kitchen substances from construction sites which is near the lake and flood / runoff water during the rains. This was also observed by (Stewart et al., 2008), when they worked on lakes and observed that microbial load can be on the increase due to run-off from point discharge and sewage. According to (Mamora, 2012) dissolved oxygen is one of the most important parameters in aquatic systems. The mean dissolved oxygen values recorded in Magada lake (9.68 mgl⁻¹) was lowest at 12 am and highest (15.32 mgl⁻¹) at 12 noon. This is however at variance with the report of (AWQA, 2012) which states that cold water hold more dissolved oxygen than warmers waters, as the water temperature was colder at 12am than at 12pm. An acceptable reason for this disparity could be as a result of the production of more oxygen in the water due to photosynthesis which is directly proportional to the amount of sunlight, which is highly evident at 12pm. However, all the values recorded exceeded the minimum limit of FEPA and WHO, which is therefore an indication of lesser amount of organic pollution. (Mahre et al., 2007) has stated that BOD₅ of a system is usually increased by the addition of organic and inorganic substances to the system. The BOD₅ values ranged from (0.056 mgl^{-1}) recorded at 12am to (0.167 mgl^{-1}) recorded at 12pm. According to them BOD₅ is used as an indication of organic

pollution, and the results therefore shows that the BOD_5 levels were far lower than the FEPA and WHO permissible limits for drinking water and aquatic life (Chapman,1996). Thus it can therefore be interpreted that Magada lake was low in organic pollution. Depth did not show any form of variation, and ranged from 2.0m (station 1) to 4.9m (station3).

A statement by (Turner Designs, 2012) indicates that phosphate can be found as free ions in water systems and as a salt in terrestrial environment used in detergents as water softeners Phosphate values ranged from (0.0085 mgl⁻¹) 6 pm to (0.0092 mgl⁻¹) 12pm. These values are extremely low compared with the values recorded by Mahre et al, 2007 in River Kaduna (5.5 mgl⁻¹ 44.7mgl⁻¹). However all the values were lower than the value of the control which was (0.0140mgl⁻¹). The reason for the control value being higher could be as a result of plants and bacteria using up the nutrients for growth, while there were no plants, and negligible bacteria population in the control water (bore hole). The values were also lower than the FEPA and WHO permissible limits for drinking water.

Sulphate levels ranged from $(3.48 \text{ mgl}^{-1}) 12$ am to (6.25 mgl^{-1}) 6pm. These values are however lower than that on the control (2.54 mgl^{-1}) . The slightly higher sulphate levels could be due to the usage of fertilizers for horticultural practices around the water body. Another reason for this could also be as a result of burning of fossil fuel for power generation and from transportation around the water body which ends up as runoff water which flow into the water body. The values were however lower that that recorded by (Akan *et al.*, 2007) which ranged from (22.60 mgl⁻¹) to (45.45 mgl⁻¹). However the levels were below FEPA and WHO maximum permissible level for drinking water (20 mgl⁻¹) and (200 mgl⁻¹) respectively. The values were however lower than that of the control (2.54 mgl⁻¹)

The nitrate values recorded in this study were between (4.05 mgl^{-1}) 12am and (8.62 mgl^{-1}) (12pm). These values were higher than that the control (0.0788 mgl⁻¹). According to Bush

and Meyer (1982) nitrate toxicity is capable of causing anaemia in infants and pregnant women, and formation of carcinogenic nitrosamines. According to them a nitrate content of more than (100 mgl⁻¹) impact bitter taste to water and may cause physiological problem. According to (Akan et al., 2007) this can lead to high growth of algae and other organisms which can result to eutrophication. However, the nitrate levels found in this study were lower than permissible limits of FEPA and WHO (45mgl⁻¹). Magnesium and Calcium are main indicators of water hardness. The levels of magnesium recorded in the study ranged from (1.50 mgl⁻¹) 12 am to (4.60 mgl⁻¹), while the level of calcium recorded ranged from (14.09 mgl⁻¹) 6 am to (17.34 mgl⁻¹) 12 am. The hardness of drinking water can be determined by its calcium and magnesium content, which can be expressed as the equivalent amount of calcium carbonates that, is usually formed from the calcium and magnesium in solution. One major source of calcium in freshwater is the bedrock and weathering of calcium bearing rocks (Waite, 1984). Calcium is often the most common metallic ion in fresh surface water and among the most common in ground water. However the levels of calcium recorded in this study is relatively high compared to values in other water bodies in Nigeria; (Okogwu and Ugwumba, 2006) for Ologe Lagoon; (Omoigberale and Ogbeibu, 2007) for River Osse, who recorded calcium values less than 10 mgl⁻¹. According to (Omoigberale and Ogbeibu, 2007) the level of magnesium concentration recorded in this study is however high when compared with some Nigerian water bodies.

The lead levels in the study ranged from (0.003mgl⁻¹) 12am to (0.036mgl⁻¹) 6pm; chromium ranged from (0.024mgl⁻¹) 12pm to (0.075mgl⁻¹) 6am; Iron ranged from (0.340mgl⁻¹) 6pm to (1.045mgl⁻¹) 6am and Cadmium ranged from (0.003mgl⁻¹) 12am to (0.376mgl⁻¹) 6pm. The heavy metal values recorded in this project did not have any statistical significant difference. Aquatic systems receive large amounts of heavy metals from unregulated sewage, industrial effluents and through runoffs (Tariq*et al.*, 1991). Heavy metals are transported dissolved

form in water or as an integral part of suspended sediments. Toxic heavy metals dissolved in water have the greatest potential of causing the most deleterious effects. Lead, a cumulative poison is widely distributed in the environment. The source of lead to the aquatic environment has been traced to automobile exhaust and rusting of lead pipes used in pipe borne water supply (Mombeshora*et al.*, 1993).

Iron is a major element in various primary minerals. It reaches natural water mainly from leaching and flaking of rust from iron pipes. The presence of chromium in freshwater bodies could be attributed to the fact that it can be carried as runoff water from industrial sites. Also chromium is transported and deposited in natural debris, which precipitates to the bottom of the water body. Cadmium is a cumulative poison that is emitted into the air from burning of fossil fuels and coal or the use of cadmium containing pesticides. The fact that cadmium gets into aquatic habitat from anthropogenic or aerial emissions have been reported by many authors including (Lacerda, 1983). (Lacerda, 1983) also reported cadmium dispersion in the environment and that it enters the aquatic ecosystem from terrestrial ecosystems at a slow rate. The total hardness of a freshwater body can be defined as the total amount soluble magnesium and calcium salts present, and is expressed as CaCO₃. In most natural water the predominant ions present are those of bicarbonates which are associated mainly with calcium mainly, and to a lesser degree with magnesium and still to a lesser degree with sodium and potassium. The values ranged from (12.261mgl⁻¹) 12am to (14.984mgl⁻¹) 6pm. The alkalinity values ranged from (0.562mgl⁻¹) 6pm to (1.528mgl⁻¹) 12pm. According to Wilson (2010) water alkalinity and hardness are primarily a function of the geology of the area where the surface water is located and the dissolution of carbon dioxide (CO₂) from the atmosphere. Wilson (2010) stated that the pH, alkalinity and hardness are capable of affecting the toxicity of many substances in water. In line with his observations, the values recorded in this study can be attributed to the influx of runoff water containing large amount of dissolved and suspended materials. The values for suspended and dissolved solids ranged from (7.385mgl⁻¹) 12pm to (20.073mgl⁻¹) 6pm, and (13.698mgl⁻¹) 12pm to (34.361mgl⁻¹) 6pm respectively. The presence of high values could be attributed to a high influx of runoff water carrying a lot of dissolved and suspended materials (Patra *et al.*, 2001). These high values could also be as a result of the study being done during the rainy season. Suspended solids may settle out on the bottom of the water body thus smothering the benthic organisms and silting up the water body. This may lead to destruction of plant and animal life and therefore the natural food supply of fish. The effects of suspended solids on the delicate respiratory systems of fish have been reported to be lethal at up to (30mgl⁻¹) (Jackson *et al.*, 1989). The high level of dissolved solids observed could also be attributed to high influx of rainy season runoff water. The influx of rain is however capable of diluting dissolved solids in water. However, high level of dissolved solids in water may also be harmful; its effect on water density is minimal and can therefore be ignored.

5.1 CONCLUSION

The work presented in this special profile illustrates some of the current and likely near future's major topics of ecological and microbial factors associated with Magada lake. The study of ecological and microbial factors associated with Magada lake has measured the different aspects of physico chemical parameters and microbial population in respect to time (diurnal variation). According to (WHO, 2009), the temperatures of all stations are still within the permissible limit for aquaculture and also for domestic use. This is in agreement with the work of (Alabaster and Llyod, 1980), who stated that the temperature of natural inland waters in the tropics generally varies between 25-35°C. The pH for all the three stations is within the permissible range for both aquaculture and for domestic use (WHO, 2009). The salinity value shows that the water was fresh in all the stations. Turbidity, a measure of cloudiness is caused by suspended solid particles and also a measure of the ability of the water to receive light. (McClurky, 1991) opined that the turbidity maximal is created by the mixture of suspended solids from the stream and the sea. Relatively, higher turbidity values were observed at around12am and 6pm. MANOVA indicates that there was no significant difference between the time variation (p>0.05). The conductivity of all the time variations is within permissible limit for aquaculture and for domestic utilization (WHO, 2009).

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APPENDIX

Table: 1 IN- SITU PHYSICO-CHEMICAL PARAMETERS

Station	Time	Replicate	Air	Water	pН	Salinity	Turb	Do	Conduct	Depth
			temp	temp		(ppm)	(NTU)	(mg/l)	(ms/cm)	(m)
			(°C)	(°C)						
1	12am	1	26.5	26.3	7.4	7.4	18.93	10.2	1.36	2.0
		2	26.8	26.8	7.8	7.6	18.2	10.5	1.51	2.0
		3	26.3	26.5	7.6	7.8	18.5	10.4	1.4	2.0
	6am	1	27.5	27.8	6.8	182	11.08	19.9	0.25	2.0
		2	27.9	28	7	180	11.51	10	0.28	2.0
		3	27.3	27.5	6.9	179	11.1	17.8	0.25	2.0
	12pm	1	24.2	26.5	7.5	177	14.29	18.5	0.24	2.0
		2	26.7	26	7.8	179	10.33	19.2	0.24	2.0
		3	24.9	26.1	8	175	11.28	18.7	0.24	2.0
	6pm	1	26.3	26.3	5.9	155	36.47	19.5	0.21	2.0
		2	26.5	25	6	160	36.41	19	0.25	2.0
		3	26.2	26.3	5.8	158	36	19.5	0.22	2.0
2	12am	1	25.2	27.2	7.2	7.2	25.71	8.1	0.36	3.9
		2	25.	27.8	7.5	7.5	25.72	8.12	0.42	6.9
		3	25.2	26.8	7.3	7.2	25.7	8.9	0.5	3.9
	6am	1	27.5	28	7.1	130	8.52	9.1	0.18	3.9
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		2	26.5	27	7.5	125	8 44	9.2	0.15	3.9
		- 3	27.5	27.5	6.9	128	8.67	9.51	0.17	3.9
		5	21.5	21.5	0.7	120	0.07	7.51	0.17	5.7
	12pm	1	26.5	27	7.3	80	3.63	12.6	0.1	3.9
		2	26.7	26.5	7.3	82	3.79	14.3	0.1	3.9
		3	26.6	27.1	7.2	82	2.94	13.6	0.1	3.9
	6pm	1	25.1	27.5	6.3	83	10.11	11	0.1	3.9
		2	25.5	28	6.5	85	10.15	11.3	0.1	3.9
		3	25	27.5	6.2	82	10.12	10.9	0.1	3.9
3	12am	1	25.1	27.1	7.9	7.1	9.59	10.2	0.11	4.5
		2	25.5	27.5	8.2	7.2	9.6	10.5	0.12	4.5
		3	25	27.3	7.8	7	9.58	10.2	0.11	4.5
	6am	1	28.7	28.5	6.8	86	3.81	9.72	0.11	4.5
		2	29	28.9	7	81	4	9.7	0.9	4.5
		3	28.9	28.2	6.9	83	3.9	8.9	0.1	4.5
	12pm	1	27.2	27.2	6.9	83	4.7	12.6	0.1	4.5
		2	26.9	27.5	6.2	83	3.9	14.6	0.1	4.5
		3	27.8	27.5	6.3	83	4.4	13.8	0.1	4.5

	брт	1	27.6	27.4	6.7	0.84	9.58	10.3	0.11	4.5
		2	2.8	27.5	6.9	0.85	9.6	10.5	0.11	4.5
		3	27.5	28.00	6.8	0.84	9.59	10.2	0.11	4.5
control	12pm	1	27.10	28.00	5.90	83	0.10	0.1	0.81	0.00
		2	27.50	28.50	5.85	89	0.11	0.1	0.85	0.00
		3	27.30	28.30	5.90	86	0.00	0.0	0.88	0.00

NUTRIENTS

s/n	station	time	Replicate	phosphate	Sulphate	Nitrate	Magnesium	Calcium
1	1	12am	1	0.014	3.120	4.50	0.43	16.3
			2	0.016	5.350	7.80	2.43	19.6
			3	0.018	7.320	9.90	4.60	21.4
		6am	1	0.002	2.435	3.500	2.24	10.5
			2	0.004	2.437	4.500	3.23	12.3
			3	0.006	2.439	5.500	4.24	13.5
		12pm	1	0.008	5.334	5.98	0.0	14.4
			2	0.011	7.238	7.98	2.34	16.4
			3	0.015	9.238	9.98	5.50	18.2
		6pm	1	0.0014	8.712	4.50	4.88	10.5
			2	0.0017	6.517	7.80	2.44	12.3
			3	0.0020	4.517	9.90	0.42	13.5
2	2	12am	1	0.002	5.156	3.500	3.50	16.2
			2	0.004	5.166	4.500	1.98	16.5
			3	0.006	5.176	5.500	0.50	16.7
		6am	1	0.0014	3.999	9.80	0.0	14.4
			2	0.0017	5.664	7.78	2.34	16.4
			3	0.0020	8.600	5.62	5.50	18.2
		12pm	1	0.008	5.334	5.98	4.88	21.4
			2	0.011	7.238	7.98	2.44	19.4
			3	0.015	9.238	9.98	0.42	17.2
		6pm	1	0.010	8.712	9.72	3.50	13.5
			2	0.012	6.517	7.52	1.98	15.7
			3	0.014	4.517	5.72	0.50	17.5
3	3	12am	1	0.005	0.013	0.22	0.02	16.2
			2	0.007	0.013	0.24	0.03	16.5

	station	Time	Replicate	BOD	TDS	TSS	TS
s/n							
1	1	12am	1	0.0	30.00	18.95	48.95
			2	0.0	35.87	20.94	56.81
			3	0.0	39.99	27.23	67.22
		6am	1	0.1	19.55	10.00	29.55
			2	0.1	22.40	12.20	34.60
			3	0.1	29.50	15.30	44.8
		12pm	1	0.0	20.33	10.50	30.83
			2	0.0	22.31	13.28	35.59
			3	0.2	28.51	18.50	47.01
		6pm	1	0.1	61.23	39.65	100.88
			2	0.3	65.68	41.25	106.90
			3	0.2	69.99	43.56	113.55
2	2	12am	1	0.1	43.00	23.20	66.2
			2	0.2	46.82	29.17	75.99
			3	0.2	49.20	34.55	83.75
		6am	1	0.0	13.10	8.99	22.09
			2	0.02	17.22	9.18	26.40
			3	0.03	20.22	9.35	29.57
		12pm	1	0.3	8.33	3.30	11.33
			2	0.3	8.54	3.30	11.84
			3	0.2	8.97	3.59	12.56
		брт	1	0.1	22.30	8.70	31.00
			2	0.1	19.43	11.17	30.6
			3	0.2	15.53	9.87	25.40
3	3	12am	1	0.0	20.58	11.00	31.58
			2	0.0	18.59	10.59	29.18

BIOCHEMICAL OXYGEN DEMAND

			3	0.0	15.30	7.33	22.63
		6am	1	0.1	4.43	3.20	7.63
			2	0.2	7.97	4.20	12.17
			3	0.0	9.97	5.30	15.27
		12pm	1	0.1	8.50	4.30	12.80
			2	0.2	8.79	4.75	13.54
			3	0.2	9.01	4.95	13.96
		6pm	1	0.0	17.11	9.99	27.1
			2	0.2	18.39	10.79	29.18
			3	0.2	19.59	11.35	30.94
4	control		1	0.0	0.68	0.01	0.69
			2	0.0	0.98	0.05	1.03
			3	0.0	1.00	0.09	1.09

EX-SITU PHYSICOCHEMICAL PARAMETERS FOR THE LAKE

S/N	STATIO N	TIME	REPLIC ATE	CADMI UM	IRON (Mg/L)	LEAD (Mg/L)	CHRO MIUM	HARDN ESS	sALKA LINITY
1	1	12am	1	0.002	0.000	0.000	0.000	10.50	+1.78
			2	0.006	0.000	0.000	0.000	13.20	+1.70
			3	0.009	0.000	0.000	0.000	15.20	+1.67
		6am	1	0.001	0.003	0.001	0.000	14.50	+0.90
			2	0.004	0.005	0.003	0.000	14.80	+1.00
			3	0.007	0.007	0.005	0.000	13.20	+1.05
		12pm	1	0.000	1.146	0.000	0.000	16.50	+1.60
			2	0.000	1.187	0.000	0.100	19.60	+1.40
			3	0.000	1.197	0.000	0.105	20.00	+1.60

		брт	1	1.112	0.005	0.001	1.012	12.66	+0.70
			2	1.114	0.009	0.004	1.003	16.50	+0.75
			3	1.117	0.012	0.007	1.002	18.60	+0.78
2	2	12am	1	0.000	1.184	0.000	0.000	15.60	+1.00
			2	0.000	1.187	0.000	0.000	16.50	+0.98
			3	0.000	1.190	0.000	0.000	18.90	+0.95
		6am	1	0.000	1.185	0.000	0.008	14.90	+1.20
			2	0.000	1.187	0.000	0.100	17.50	+1.50
			3	0.000	1.189	0.000	0.103	19.60	+1.00
		12pm	1	0.000	1.222	0.000	0.000	4.95	+1.50
			2	0.000	1.226	0.000	0.000	7.50	+1.80
			3	0.000	1.230	0.000	0.000	9.00	+1.32
		6pm	1	0.000	0.815	0.000	0.000	8.50	+0.06.
			2	0.000	0.818	0.000	0.000	8.60	+0.03
			3	0.000	0.820	0.000	0.000	9.50	+0.03
3	3	12am	1	0.002	1.003	0.000	0.108	7.20	+1.51
			2	0.004	1.006	0.001	0.110	7.50	+1.20
			3	0.006	1.009	0.002	0.112	9.50	+1.50
		6am	1	0.016	1.940	0.000	0.152	10.00	+0.80
			2	0.019	1.945	0.000	0.155	13.30	+0.85
			3	0.022	1.950	0.000	0.157	15.58	+0.88

		12pm	1	1.120	0.111	0.009	0.001	9.50	+1.15
			2	1.125	0.113	0.013	0.004	10.5	+1.62
			3	1.127	0.115	0.017	0.007	12.8	+1.77
		брт	1	0.011	0.192	0.103	1.123	17.5	+0.89
			2	0.014	0.194	0.105	1.125	20.5	+0.90
			3	0.016	0.196	0.107	1.127	22.5	+0.92
4	Contro 1		1	0.000	0.000	0.000	0.000	8.95	+1.12
			2	0.000	0.000	0.000	0.000	9.50	+1.13
			3	0.000	0.000	0.000	0.000	12.5	+1.14

MORPHOLOGICAL PARAMETERS

	TIME	DILUTION	REPLICATE	NO OF
		FACTOR		COLONIES
Station 1	12am	10-1	Rep 1	290
			Rep 2	271
			Rep 3	269
		10-3	Rep 1	180
			Rep 2	176
			Rep 3	161
		10-5	Rep 1	90
			Rep 2	83
			Rep 3	60

6am	10-1	Rep 1	280
		Rep 2	279
		Rep 3	250
	10-3	Rep 1	110
		Rep 2	103
		Rep 3	109
	10-5	Rep 1	25
		Rep 2	26
		Rep 3	28
12pm	10-1	Rep 1	130
		Rep 2	127
		Rep 3	120
	10-3	Rep 1	116
		Rep 2	109
		Rep 3	107
	10-5	Rep 1	53
		Rep 2	47
		· r	
		Rep 3	41
6pm	10-1	Rep 1	287
		Rep 2	276
		Rep 3	254
	10-3	Rep 1	159
		P -	

			Rep 2	130
			Rep 3	125
		10-5	Rep 1	80
			Rep 2	60
			Rep 3	55
2	12am	10-1	Rep 1	281
			Rep 2	305
			Rep 3	299
		10-3	Rep 1	180
			Rep 2	176
			Rep 3	149
		10-5	Rep 1	80
			Rep 2	30
			Rep 3	25
	бат	10-1	Rep 1	150
			Rep 2	142
			Rep 3	120
		10-3	Rep 1	78
			Rep 2	50
			Rep 3	45
		10-5	Rep 1	18

		Rep 2	15
		Rep 3	20
12pm	10-1	Rep 1	130
		Rep 2	127
		Rep 3	120
	10-3	Rep 1	118
		Rep 2	116
		Rep 3	109
	10-5	Rep 1	23
		Rep 2	20
		Rep 3	16
6pm	10-1	Rep 1	150
		Rep 2	141
		Rep 3	125
	10-3	Rep 1	69
		Rep 2	65
		Rep 3	54
	10-5	Rep 1	44
		Rep 2	47

		Rep 3	33
12am	10 ⁻¹	Rep 1	140
		Rep 2	136
		Rep 3	129
	10 ⁻³	Rep 1	110
		Rep 2	101
		Rep 3	105
	10-5	Rep 1	50
		Rep 2	35
		Rep 3	28
6am	10 ⁻¹	Rep 1	200
		Rep 2	230
		Rep 3	207
	10-3	Rep 1	90
		Rep 2	92
		Rep 3	94
	10 ⁻⁵	Rep 1	80
	12am 6am	12am 10^{-1} 10^{-3} 6am 10^{-5} 10^{-5} 10^{-5} 10^{-3} 10^{-3} 10^{-3}	Rep 3 Rep 1 12am 10^{-1} Rep 1 Rep 3 Rep 3 10^{-3} Rep 1 Rep 3 Rep 3 10^{-3} Rep 1 Rep 3 Rep 3 10^{-3} Rep 1 Rep 3 Rep 3 $6am$ 10^{-1} Rep 1 Rep 3 Rep 3 $6am$ 10^{-1} Rep 1 Rep 3 Rep 3 $6am$ 10^{-3} Rep 1 Rep 3 Rep 3 10^{-3} Rep 1 Rep 3 Rep 3 10^{-3} Rep 1 Rep 3 Rep 3

			Rep 2	75
			Rep 3	89
	12pm	10-1	Rep 1	141
			Rep 2	136
			Rep 3	139
		10-3	Rep 1	31
			Rep 2	25
			Rep 3	20
		10-5	Rep 1	10
			Rep 2	5
			Rep 3	3
	брт	10-1	Rep 1	150
			Rep 2	164
			Rep 3	168
		10 ⁻³	Rep 1	81
			Rep 2	79
			Rep 2	75
		10-5	Rep 5	15
		10 5	Rep 1	65
			Rep 2	57
			Rep 3	42
control		10-1	Rep 1	50
			Rep 2	47

1				
			Rep 3	55
	10 ⁻³	Rep 1	32	
		Rep 2	22	
			Rep 3	38
		10-5	Rep 1	15
			Rep 2	9
				18
			Rep 3	