ALGAL DIVERSITY AND WATER QUALITY OF ASATA RIVER ENUGU, ENUGU STATE, NIGERIA

BY

ASOGWA, EDITH OYINNEM

PG/M.Sc./10/52604.

DEPARTMENT OF PLANT SCIENCE AND BIOTECHNOLOGY

UNIVERSITY OF NIGERIA, NSUKKA

AUGUST, 2016

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A PROJECT REPORT SUBMITTED IN PARTIAL FULFILLMENT OF THE REQUIREMENT OF THE AWARD OF THE DEGREE OF MASTER OF SCIENCE (FRESH WATER ALGOLOGY AND LIMNOLOGY) IN THE DEPARTMENT OF PLANT SCIENCE AND BIOTECHNOLOGY, UNIVERSITY OF NIGERIA, NSUKKA

AUGUST, 2016

APPROVAL PAGE

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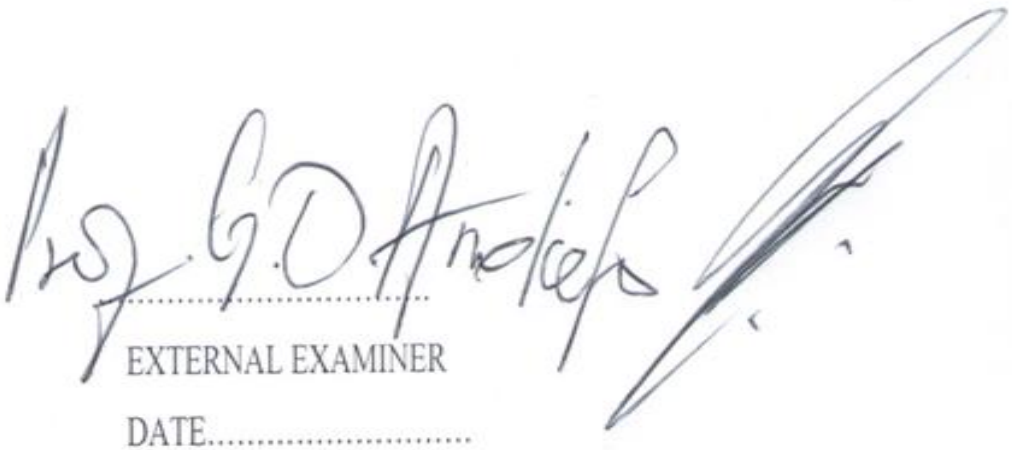

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DEDICATION

Dedicated to my mother Asogwa, Philomena; my husband Asogwa, Peter; my children Anthony Okechukwu, Immaculata Ifunanya, Bonaventure Chukwuma and Vitus Onyedikachi.

ACKNOWLEDGEMENT

I thank the almighty God for His mercies and protection given to me, my family and my Supervisor for the completion of this work. My unreserved gratitude goes to my Supervisor, Dr Nkechi O. Nweze for all the opportunities given to me to carry out this research. May God replenish her and her family abundantly. I also appreciate all my lecturers of Plant Science and Biotechnology, may God grant them good health.

I am immensely grateful to my sister Mrs Osyï Nnamani Irene Ndidiamaka who made this research work to be possible. My thanks also go to my cousin Amos Nwamba for his support in this my research work. May God grant you and your families long life to reap the fruit of this research. .I am also grateful to my colleagues in the Department especially my brothers in Phycology (Ude, Batholomew, Mamoud, Liman and Eze, Ejiofor). May the almighty God pay you back abundantly.

To my destiny helper Ezema, Raymond Anikwe (MY OBSERVER). Thank you for observing the Aï Z of this work and may the almighty God grant your heart desires and give you and your family long life and prosperity. To you Godôs messenger Asogwa, Isaac, thank you for your encouragement and I pray to God that your vision might be accomplished in my life through Jesus Christ our Lord Amen.

For my beloved husband Mr Asogwa, Peter Okechukwu and my children thank you for your prayer, love and understanding of my long absence from home, God will answer our prayer and we will live long to reap the fruit of this research. Finally to my mother, thank you for showing me the way to education and may the good God give you long life and prosperity to reap the fruit of your labour.

ABSTRACT

An assessment of the physicochemical, parameters, algal diversity and bacterial flora of Asata River Enugu, Enugu State, Nigeria was carried out for six months. Water samples and periphyton were collected fortnightly from four locations namely: Okpara Coal Mine, University of Nigeria Teaching Hospital (UNTH) Old Site, Ogbete market and Ogui and analysed using standard procedures. Algal diversity and species richness were determined using Shannnon ï Wiener index. Spread plate method on Macconkey agar was used for bacterial count. Results showed monthly variations (PÒ0.05) in air temperature, velocity, alkalinity, aluminum, dissolved oxygen, biological oxygen demand, chemical oxygen demand, calcium, pH, total suspended solids, iron, lead and zinc, at the various locations. Significant (PÒ0.05) positive (colour and depth, transparency, iron and Bacillariophyta) and negative (pH and algal divisions; lead and Bacillariophyta; mercury and Cyanophyta) correlations were also observed among some parameters investigated. Thirty-four algal taxa belonging to four divisions in the following decreasing order: Bacillariophyta (33 %), Chlorophyta (32 %), Euglenophyta (21 %), and Cyanophyta (14 %) were encountered. Some pollution tolerant algae such as *Gomphonema*, *Nitzschia*, *Navicula*, *Surirella*, *Euglena* and *Oscillatoria* were dominant. Based on Shannonï Wiener index, the water at UNTH and Ogbete were classified as mesoptrophic whereas, Okpara coal mine and Ogui were classified as eutrophic. The diversity index showed no complete evenness in the river. The lowest and highest enteric bacterial counts ($1.029 \times 10^4 \pm 1.174$ and $5.05 \times 10^4 \pm 1.472$) were observed in August and November respectively. Gram ï positive and gram ï negative bacteria such as *Escherichia, Salmonella*, *Micrococcus*, *Proteus* and *Enterobacter* were isolated. *Escherichia* was dominant indicating fecal pollution. pH was within the permissible limit; iron, lead and phosphate beyond the limit; and calcium, magnesium, biological oxygen demand, chemical oxygen demand, dissolved oxygen below WHO standard for drinking water, indicating that the river was eutrophic.

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CHAPTER ONE

INTRODUCTION

Rivers are important pathways for flow of energy, matter and organisms through the landscape. They have always provided a focus of attraction for environmental studies. They offer a number of benefits and services to man. A wide range of human activities at the catchment areas may lead to environmental deterioration of river quality (Kagalou *et al.,* 2002). The increasing environmental pollution is alarming and is on a global scale by anthropogenic activities.

The productivity of the river is determined by the water quality parameters, the aquatic species composition and abundance are affected, hence there is the need to monitor the water quality (Agboola *et al.,* 2011). Rivers are useful in multiple ways such as source of water for man and for domestic animals, in fisheries, agriculture and generation of hydroelectric power. Also tropical rivers play a crucial role in the epidemiology of several tropical diseases (Kadiri, 2007).

The river system presents interesting feature for aquatic organisms, in relation to extended periods of dryness, land use practices and intense human activities, all of which influence water quality (Papastergradou and Babalonas, 1993). Knowledge of specific relationships between particular organisms and environmental factors enable a quick look and fairly reliable evaluation of the essential features of a given biotope (Fytainos *et al.,* 2001).

Pollution of fresh water bodies such as rivers, streams, lakes and ponds is mostly experienced as a result of industrial discharge, municipal waste disposal and surface

runoff. Indiscriminate and uncontrolled discharge of waste into river impact negatively on river ecosystem and human health (Chima *et al.,* 2009).

Water quality is crucial in order that man can benefit from river through series of users (Oliveira *et al.,* 2006). Water quality deals with physical, biological and chemical characteristics in relation to all other hydrological properties. Any characteristics of water that affect survival, reproduction; growth and production of aquaculture species; influence management decisions; cause environmental impact or reduce product quality and safety can be considered a variable quality. The use of physicochemical properties of water to assess water quality gives a good impression of the status, productivities, and suitability of such water body. The change in physical characteristics like depth, temperature, transparency and chemical element of water such as dissolved oxygen, chemical oxygen demand, nitrate, phosphate provides valuable information of the quality of the water, the source (s) of the variation and biodiversity (Mustapha, 2006, 2008; Sumita *et al.,* 2010).

Nutrients play a major role in growth and development of plants in tropical water where there is enough light for photosynthetic activities throughout the year, resulting in light and temperature probably not being limiting. Aquatic autotrophs have nutritional and energetic requirement for their photosynthetic carbon function and growth. Nutrient limitation is important in the regulation of phytoplankton abundance (Hirose *et al.,* 2003). Phytoplankton diversity with the seasonal fluctuation indicates the condition of ecological niches.

Algae are simple plants that vary considerably in size, shape and colour and are found in a range of habitats (Christi *et al.,* 2011). Algae are frequently found in polluted and unpolluted waters and due to this behavior they are considered useful in determining the

quality of water. They are very suitable organisms for the determination of the impact of toxic substances on the aquatic environment because any effect on the lower level of the food chain will also have consequences on higher levels (Joubert, 1980; Jafari and Gunale, 2006). Algae are used for assessing the degree of pollution or as indicators of water pollution of different water bodies (Trivedy, 1986; Sudhaker *et al.,* 1994; Dwivedi and Pandey, 2002). Due to their short life cycle, algae respond quickly to environmental changes and are thus a valuable indicator of water quality (Hotzel and Croome, 1999). Pollution stress reduces the number of algal species but increases the number of individuals hence species diversity of fresh water habitats are very much affected by eutrophication (Christi *et al.,* 2011) As sunlight driven cell factories micro - algae convert carbon dioxide to potential biofuels, food and feeds (Chisti, 2008). These photosynthetic microorganisms are also useful in bioremediation (Navid, 2005; Kadiri 2010).

The anthropogenic inputs of complex mixtures from neighboring communities and agricultural waste such as runoff of manures and fertilizers could lead to alteration of water quality (Mustapha, 2006; Garg et *al.,* 2009). Of primary concern of these anthropogenic activities are their effects on the water quality and aquatic life. Depending on the quality and quantity of waste input, the physical, chemical and biological balance of the receiving water may be significantly modified resulting in pollution and its associated consequences (Akpan *et al.,* 2002; Osondu, 2007).

Water quality deterioration may also be as a result of acidification, heavy metals, organic pollution and obnoxious fishing practices. The effects of these ñimportsò bring loss of structural biodiversity of the water (Mustapha, 2008). Water quality monitoring is of immense importance in the use of water bodies in the management of fisheries (Mustapha, 2006; Nweze, 2009b; Agboola *et al*., 2011). However, the overall condition

of this complex and dynamic system where biotic and abiotic components are constantly interacting with each other brings structural and functional changes in an ecological unit such as aquatic ecosystems (pond, lake, river or ocean) (UNESCO, 2006; Chia *et al.,* 2011).

Water quality assessment may be determined by physical, chemical and biological attributes in relation to standards developed by U.S. Environmental Protection Agency (USEPA, 1976), World Health Organization (WHO, 1993) and American Public Health Association (APHA, 1995), The water required for domestic consumption should possess a high degree of purity and it should be free from suspended and dissolved impurities and bacteria (Alexander, 2008). Whatever the source of water, it is expected to be less contaminated depending upon management and temperature gradient of the water environment (Radojevic and Bashkin, 1999; APHA, 2005).

1.1 Statement of the Problem

Biological assessment is a useful alternative for assessing the ecological quality of aquatic ecosystems since biological communities indicate the environmental effect of water chemistry. The impact of increased environmental pollution in recent times has become a global menace and even much devastating at regional levels. This is often constituted by anthropogenic activities. The diversity of algae in water bodies shows the pollution status of water bodies including that of River Asata. This necessitated this study on physicochemical parameters of Asata River to determine those factors that affect the water quality and algal diversity. Enugu being a metropolitan city where River Asata is located experiences increase in population due to urbanization and industrialization. As the population increases, the introduction of waste into the river also increases, and this leads

to contamination of the river, thereby constituting environmental and health hazards to the inhabitants of the urban area.

1.2 Objectives of the Study

The objectives of the study are to:

- Assess the physicochemical parameters of Asata River, Enugu.
- Identify various algae present and microbial content of Asata River.
- Correlate the physicochemical parameters with algal population of Asata River.
- Determine the trophic status of the various sampling points on Asata River.

CHAPTER TWO

LITERATURE REVIEW

A lot of work has been done on physicochemical and algal bio-diversity of water bodies in Nigeria. They include Akpan and Offem (1993), Cross River; Nweze (2003), on phytoplankton production of Ogelube lake; Kadiri (2006), on the phytoplankton flora and physicochemical attributes of some waters in the Delta Area; Nweze and Chumboh (2006), on physical and chemical characteristic alga flora and coliform content of the oxidation pond at the University of Nigeria, Nsukka; Mustapha (2008), on Oyun Reservoir, Offa; Agboola *et al.,* (2011), on Abesan River, Lagos and Akubuko and Duru (2011), on Ottamiri, Owerri, Imo state.

2.1 Physicochemical or Abiotic factors

The physical parameters define those characteristics of water such as taste or smell, suspended solids, turbidity, colour, temperature, depth / water level. The water level plays an important role in governing the water quality (Garg *et al.,* 2009). Nweze (2006) noted that as the volume of water increases, the pH, dissolved oxygen, silica, phosphate and nitrate decrease. This is due to the dilution of the nutrients by rain. Davis (2009) observed that low depth of water results in low transparency. Furthermore, low input from the catchment area may result in low water volume (Nweze and Chumboh, 2006)

Increase in water temperature and nutrient load, produces annual variation in mean chlorophyll ñaò concentration with the largest changes caused by variation in nutrient load (Elloit and Linday, 2008). Temperature fluctuations, both diurnal and seasonal are more evident in fresh water habitats although flowing waters lack wide fluctuation in temperature (Elloit and Linday, 2008).

Light penetration is an important factor that affects life in the aquatic ecosystem. Rainwater reduces transparency. Onyema (2007) noted that the reason for this is probably an outcome of mixing by rain. Also Mustapha (2006) noted that bloom of algae; more turbid floodwater input and re-suspension of bottom materials considerable reduce transparency. In addition, washing of silt, sediments, debris, organic and inorganic suspended particles contribute to low transparency (Mustapha, 2008). Davis (2009) observed that low depth of water results in low transparency. When transparency increases there is sufficient sunlight penetration causing an increase in photosynthetic activities and increased dissolved oxygen (Adeogun *et al.,* 2011).

Low rate of flow favours the buildup of phytoplankton population in rivers; otherwise they flush out of the system. Phytobenthos can tolerate high rate of flow and as such predominate over phytoplankton in fast flowing rivers and streams (Bellinger and Sigee, 2010).

pH values depend on the carbonate system (free carbon dioxide, carbonic acid, bicarbonate, carbonates), which buffer the water (Onyema, 2007). High pH values are due to utilization of bicarbonates and carbonate buffer system (Kadiri, 2006; Mahima and Pandey, 2007). pH has been observed to be high during the rainy season. Low pH values are associated with the influence of fresh water influx, dilution of lake, low temperature and organic matter decomposition (Zachariya *et al.,* 2011).

Mustapha (2008) noted that dissolved oxygen is an important indicator of water quality, ecological status, productivity and health of a water body, due to its importance as a respiratory gas and its use in biological and chemical reaction High temperature coupled with high rate of decomposition in the dry season results to low oxygen concentration (Kadiri, 2000; Garg *et al.,* 2009; Mustapha, 2008). Algal bloom and explosiveness of the

aquatic macrophyte reduce dissolved oxygen in water (Mustapha, 2006). Low dissolved oxygen during wet season could be attributed to increased current, wave action and relatively shallow bottom of the system, having a mixing effect of the inorganic component of the water column thus increasing the oxygen demand of the system. In contrast the increased photosynthetic activities of the planktonic algae following high photoperiod associated with the dry season increases dissolved oxygen (Egborge and Fagade, 1994: Ogamba *et al.,* 2005: Adeogun *et al.,* 2011). Mohammad and Saminu (2012) reported that dissolved oxygen had significant negative correlation with temperature, hence, when the temperature is high, dissolved oxygen is low and vice versa.

Biological oxygen demand (BOD) is a measure of amount of oxygen that bacteria will consume while decomposing organic matter under aerobic condition (APHA, 1995). If effluents with high BOD levels are discharged into a stream or river, it will accelerate bacterial growth and consume the oxygen in the river. Due to high BOD, oxygen may diminish to levels that are lethal to fish and other aquatic organisms (Adeogun *et al.,* 2011). BOD increases in summer. This is due to the fact that in summer, several microbes present in the water bodies accelerate their metabolic activities with concentrated amount of organic matter in the form of municipal and domestic wastes discharged into the water bodies and hence their demand for oxygen increased (Kuma and Sharma, 2005, Ogamba *et al.,* 2005). Moreover, high BOD values could be attributed to low DO level, since low DO will result to high BOD and this is a strong indication of pollution (Chukwu and Odunzeh, 2006; Adeogun *et al.,* 2011). Discharge of sewage onto the river also increases the BOD of the river (Fikrat *et al.,* 2010). Biochemical oxygen demand (BOD) above 1mg/l is associated with waste water contamination (UNESCO/WHO/UNEP, 1996) Based on BOD, aquatic bodies can be

classified into unpolluted (BOD<1.0 mg/l), moderately polluted (BOD<10.0 mg/l) and heavily polluted (BOD>10.0 mg/l) (Adakole *et al*, 1999).

Chemical oxygen demand (COD) is a measure of the total quantity of oxygen required to oxidize all organic materials into carbon dioxide and water (APHA, 1995; Garg *et al.,* 2009). COD increases with increasing concentration of organic matter (Mustapha, 2008). Low COD is attributed to net dilution and decomposition of organic load (Adeogun *et al.,* 2011). Mustapha (2008) revealed that high COD occurs during the dry season and it is linked with pollution due to high rate of organic decomposition resulting from human activities on the watershed, which produce sewage and agricultural run-off into water bodies and these have negative influence on the water quality.

Decrease in alkalinity and other factors such as calcium, sulphide, phosphate, zinc, water temperature, and colour with time could be attributed to dilution effects of rains (Biswas, 1992; Kadiri, 1993). Evurunobi (1984) noted that coloration of water could be caused by selective transparency of water, suspended living and inanimate matter, the colour of the surrounding country, the varying composition of sky reflection and colour of the bottom (in shallow water). The colour of water and NH_4N concentration possibly have a limiting effect on the abundance of desmids **(**Sumita *et al.,* 2010).

Water hardness is the traditional measure of the capacity of water to react with soap. The concentration of Na^+, Ca^+ and Mg^+ indicates hardness but Na^+ is very low in fresh waters; therefore, hardness is generally measured as the concentrations of calcium and magnesium only, which are far higher in quantities over other hardness producing ions (Mustapha, 2002).

The increased application of fertilizers in agricultural land has resulted in water pollution due to leaching of nutrients like nitrogen and phosphorus. Nitrates may be

found in concentrations of up to 30 mg nitrates as nitrogen /L in some effluent of nitrifying biological treatment plants (Mustapha, 2006). Phosphorus occurs in natural waters and in wastewaters almost solely as phosphates and is essential to the growth of algae as low phosphorus concentration inhibits photosynthesis in algae. As noted by Wetzel (2001) it can be the nutrient that limits primary productivity in water. Where phosphate is a growth limiting nutrient, the discharge of raw or treated wastewater, agricultural drainage, or certain industrial waste to that water may stimulate the growth of photosynthetic aquatic micro and macro organisms in consequential quantities. High concentration of phosphate 0.5 mg/l maximum could also be responsible for the prolific growth of macrophytes. The source of phosphate could also be from fertilizer runoff from agricultural land (Mustapha, 2006).

Heavy metals affect biota adversely. Dissolved metallic ions create turbidity and discoloration; and can precipitate and form bottom sludge. Various heavy metals including those which are essential micronutrients are toxic to organism in their higher concentrations (Furhan *et al*., 2006). Lead can adversely affect plants and invertebrates at the concentration of 500 to 1000 ppm, allowing more lead tolerant populations of the same or different species to take their place and this will change the type of community present (Greene, 1993). In children, acute exposure to very high level of lead may produce encephalopathy and other accompanying sign of coma, ataxic convulsion and hyperirritability (ATSDR, 2010). Although the effects of lead exposure are potential concern for humans, young children less than seven year are most vulnerable (Reagan and Silbergeld, 1989).

Mercury is a highly toxic element that is found both naturally and as an introduced contaminant in the environment (USGS, 2000). The emission of mercury into the atmosphere originates in part from energy generation and from the burning of fossil

fuels (Lacerda and Malm, 2008). Once in the atmosphere, mercury is highly disseminated and can circulate for years accounting for its wide spread distribution (USGS, 2000). The mercury in air may settle into the water bodies and can be methylated to methylmercury through microbial activities (USEPA, 2014). Exposure to methylmercury is usually by eating contaminated fish and wildlife that are at the top of aquatic food chain (USGS, 2000). Effect of methylmercury exposure on wildlife can include mortality (death), reduced fertility, slower growth and development and abnormal behavior that affect survival, depending on the level of exposure (USEPA, 2014). Methylmercury is liposoluble, it is therefore easily absorbed by biological membranes in general and by the digestive tracts of practically all food chain (Lacerda and Malm, 2008). The organification of mercury therefore accelerates its bioaccumulation in the food chain and maximizes its threat to natural ecosystem and to human health (Porcella, 1994).

Some heavy metals are also important for the growth and development of algae and their low concentration can limit the growth of plant biota. There is accumulating evidence that iron limits phytoplankton biomass in the north Pacific gyre, the equatorial Pacific and the Southern Ocean which have been termed high nitrate but low chlorophyll area because of low concentration of iron. Studies have shown that nitrogen and carbon quotas were decreased by 30 % and 25 % respectively in iron limited cultures (Millgan and Harrison, 2000).

Low level of zinc (about 30 mg/l) has been shown to inhibit carbon dioxide fixation in marine phytoplankton. Zinc results from either the donor or acceptor side of PS I1 Chlorosis which occurred in cells grown in high zinc level, suggest the effect of the metal on cellular pigment accumulation. Beta carotene, zeaxatine and some other photosynthetic pigment equally reduce together with chlorophyll a at high zinc level (de

Magalhaes *et al.,* 2004). Besides, cell culture in the presence of 3.30 mg/l zinc level showed that the electron ï transparent space between the cytoplasmic membrane and the peptidoglycan layer was thicker than usual, moreover the cell contains bigger intercellular inclusion than those of the control with reduced number of chlorophyll a and the thykaloid membrane tended to aggregate (de Magalhaes *et al.,* 2004). Thick mucilage produced around cell grown in high zinc concentration may reduce carbon dioxide diffusion into the cell and this could be very critical at low carbon dioxide concentration in the air. The ability of Cyanobacteria to sequester metal ions in the cellular surface in response to high zinc could promote a decline in the uptake of other trace elements like iron (de Magalhaes *et al.,* 2004).

2.2 Biological or Biotic Factors

Algae are simple plants that vary considerably in size, shape and colour and are found in a range of habitats (Christi *et al.,* 2011). Algae are frequently found in polluted and unpolluted waters and due to this behavior they are considered useful in determining the quality of water. They are very suitable organisms for the determination of the impact of toxic substances on the aquatic environment (Joubert, 1980; Jafari and Gunale, 2006). Algae are used for assessing the degree of pollution or as indicators of water pollution of different water bodies (Trivedy, 1986; Sudhaker *et al.,* 1994; Dwivedi and Pandey, 2002).

Studies have shown that there are variations in the algal flora of various water bodies Mohammad and Saminu (2012) reported that algae of Salatan River in Kano State, Nigeria are dominated by Chlorophyta both in terms of number of species and number of individuals. The abundance followed the order Chlorophyta > Bacillariophyta > Cyanophyta > Euglenophyta > Dinophyta. Zakariya *et al.,* (2011) reported the following

order Bacillariophyta > Chlorophyta > Cyanophyta > Chrysophyta > Prryophyta in the lower Niger River in Kogi State and Offem *et a*l., 2011 encountered algal species from four phyla at Ikwori Lake, South ï Easthern Nigeria in the following order of abundance Chlorophyta > Bacillariophyta > Cyanophyta > Dinophyta.

Carbon dioxide is converted to potential biofuel, food and feeds by micro ï algae that are natural sun driven cell factories (Chisti, 2008). Phytoplankton are micro algae that are unicellular, colonial, or filamentous and occur as drifters in water bodies (Onyema, 2007; Kadiri, 2010). These organisms serve as chief primary producers in the aquaculture industry (Nweze, 2009a). They are increasingly used to monitor the ecological quality and health of water environment and also to measure the effectiveness of management or restoration programme or regulation action (Brieley *et al*., 2007). They are also useful in bioremediation (Navid, 2005; Kadiri 2010). While phytoplankton are important primary producers and the basis of food chain in open water, some species on the other hand can be harmful to human and other animals by releasing toxic substances into the water (Ariyadeji, *et al.,* 2004). Phytoplankton satisfy condition to qualify as suitable indicators in that they are simple, capable of quantifying changes in water quality, applicable over large geographical area and can also furnish data on background condition and natural variability (Onyema, 2007).

Periphyton is a complex mixture of algae, cyanobacteria, heterotrophic microbes and detritus that attached to submerged surfaces in most aquatic ecosystem *(*Azim *et al.,* 2006). They are an essential component of the aquatic food chain. The algal component are primary producers in fresh water bodies including rivers where different forms are present in various locations as epilithic, epipsamic, epiphythic, epipelic and epizoic forms (Osagie, 2010). Osagie (2010) observed that they serve as an important food source for invertebrates, tadpoles and some fish and if exposed to contaminants in the

water column they can absorb them. Moreover, they are important indicators of water quality.

The biodiversity of planktonic algae in aquatic ecosystems represents the pollution status (Sumita *et al.,* 2010). The presence of desmids indicates that the water is polluted (Kumar and Hosmani, 2006); temperature helps in the growth of Cyanobacteria (Wilkwozaniak, 1998); carbon dioxide shows positive relationship with Chlorophyceae (Hosmani *et al,* 1999) and alkaline environment favours the growth of eugeniods. Harish (2002) reported that phosphates nitrates and nitrites control the growth of euglenoids. High concentrations of calcium favour diatoms (Sumita, *et al*., 2010). Euglenophyceae are common in environment rich in decaying organic matter and large population of Euglena are favoured by the presence of high level of dissolved organic compound and high temperature. High level of nitrates and phosphates lead to excessive blooming of Cyanophyta (Zakariya *et al.,* 2011).

Jafari and Gunale (2006) reported that some species of algae are tolerant to organic pollution, noting that the genera like *Oscillatoria, Euglena, Scenedesmus, Chlamydomonas, Navicula, Nitzschia, Stegeoclonium and Ankistrodesmus* are the species found in organically polluted water. Also that *Oscillatoria, Lyngbya, Anabaena* and *Microcystis (*blue green algae*)* and *Navicula, Nitzschia, Synedra* and *Gomphonema* (diatoms) are pollution tolerant algae. Moreover that the presence of some green algae like *Scenedesmus, Stigeoclonium, Ankistrodesmus, Chlamydomonas, Coelastrum* indicate higher degree of organic pollution in fresh water.

Water is undoubtedly the most precious natural resource that exists on the planet that is very vital for domestic, irrigation, fisheries, and recreational among other services. However, anthropogenic activities adversely affect water bodies via organic and

inorganic fertilizers containing nutrients, such as nitrates, phosphates in a level that over stimulate the growth of aquatic plants and algae. Such dense growths/blooms consequently clog water ways, use up dissolved oxygen and block light. This in turn proves very harmful to aquatic organisms, as it affects the photosynthetic activities of plankton and increase respiratory rates.

Microorganisms often play a major role in determining the extent of pollution (Saha *et al*, 2009)). Municipal waste is a primary contributor of bacteria to the aquatic environment (Linton *et al* 1974). Saha *et al* (2009) noted that fecal coliform is considered presumptive evidence of fecal pollution. Pathogenic bacteria that have been transmitted by water or waste water includes; *Salmonella, Escherichia coli, Campylobacter, Vibrio cholerae; Leptospira* and *Yersinia (*Saha *et al,* 2009). They are the causative organisms of some diseases like *E.coli* diaorrhorea, typhoid fever caused by *Salmonella Typhi,* shigellosis/shigella dysentery caused by *Shigella Salmonella*, Gastroenteritties caused by *Salmonella* sp., cholera caused by *Vibrio Cholerae*, compylobaccteriosis caused by *Compylobacter*, urinary tract infection caused by *Proteuse* sp., and Oppurtunic urinary tract infection caused by *Enterobacter* sp. (Extonet, 1997).

2.3 Economic Importance of Algae

Beneficial Effect of Algae:

The benefits of algae to man are enormous. Directly some are food for man (Zolkowska and Wlodawer, 2006). Sea weeds and certain algae are important sources of food for people especially in coastal areas such as Korea, Japan, and China (Kadiri, 2010), and food for zooplankton and fish (Ezra and Nwankwo, 2000). The most commonly exploited algae are: *Ulva, Chlorella, Nostoc, Monostroma, Caulerpa*, *Undaria*

(Wakame*), Laminaria*, and *Spirulina* (Kadiri, 2010). Algae play a great role in the complex web of life as the main primary producers in the aquatic environment. They are the foundation of the aquatic ecosystem. Algal metabolism and productivity are critical to the natural flora of these habitats. They help to replenish the oxygen balance of the environment. The brown algae*; Ascophyllum nodusum, Laminaria* and *Fucus* are used as feed supplement for animals. These algae supply the animals with a wide range of vitamins, inorganic materials and trace elements (Kadiri, 2010).

Nostoc ellipsoporum is under investigation for potency against serious ailments like HIV and severe acute respiratory syndrome (Zolkwska and Wlodawer, 2006); diatoms have been used as raw material for bioremediation (Kadiri, 2010*)* and *Chlorella* sp has been reported as a bio-cleanser for organic pollution (Zeyaullah *et al.*, 2009). USEPA (2002) reported how algae can be used for environmental monitoring against pollution. Besides, the nitrogen-fixing potentials, Cyanobacteria have been exploited in agriculture as biofertilizers (Mishra and Pubbi, 2004; Kadiri 2010). Boussiba *et al* (2000) stated that some blue-green algae (BGA) *Anabaena* spp. can be used as gene delivery system. In experimental biology, algae have frequently been used as tools for research studies; a notable example is the Calvinôs lollipop experiment that demonstrated the photosynthetic pathways for carbon fixation using *Chlorella* spp.

Deleterious Effects:

Toxic species of *Microscystis, Dinophysis and Gynodinium* cause mortality of fish and other animals. Algae can cause death in the aquatic ecosystem via oxygen depletion; physically, by choking and by release of noxious compounds. They can grow on anything defacing it, for example cars, windows, louvers and metal framework or wall of houses (Nweze, 2009c; Kadiri, 2010). They also lead to unpleasant odour in water

produced by substances such as geosmin, rendering it useless for domestic and recreation purpose (Nweze, 2009b: Kadiri, 2010). Blue green algae (BGA) are often associated with off-flavor problems in fish because they produce geosmin and 2-methylisobomuol (MIB) which impart undesirable flavor in fish. Some species of the blue-green algae also produce toxins that affect farm animals, birds and human negatively thereby reducing food production. *Microcystis* is known to cause death of animals that drink from infested pond. Symptoms in human such as skin irritation, eye irritation, rashes, stomach cramps, vomiting, nausea, diarrhea, asthma, fever and sore throat have been associated with them (Nweze, 2009b: Kadiri, 2010).

CHAPTER THREE

MATERIALS AND METHODS

3.1 STUDY AREA

Enugu is the administrative capital of Enugu state, Nigeria (Fig 1), with a population of about 722,600 (Chima *et al.,* 2009). The city is located between $6^{o}21^{1}$ N and **6^{o}**30^{1} N and $7^{o}\,26^{1}$ E $7^{o}30^{1}$ E, while River Asata lies between Latitude 6^{0} 24` 50``N and $6^{0}$ 27` 53`` N and Longitude 7^{0} 27` 23`` E and $7^{0}$ 31` 26`` E (Fig 2). The study area has a humid tropical climate. Mean annual rainfall ranges between 1600 mm and 2500 mm (with three to four dry months) with the driest month having at least 29 mm of rainfall. Mean monthly temperature ranges between 27^{o}C and 29^{o}C. Vegetation type is rainforest savanna (Chima *et al.,* 2009).

The main river system (Nyaba, Ekulu, Idaw, Aria, Ogbete and Asata) that drains the city originate westward from the base of Udi escarpment and flow eastward into the Cross River. Asata River a third order stream is a tributary of the Ekulu River and has a catchment area of 40 km^{2}. The Asata River basin falls within the larger Abione basin which is geographically made up of a variety of sedimentary rocks (Chima *et al.,* 2009). They noted that these sedimentary rocks which consist of sandstones and shales are generally of cretaceous form.

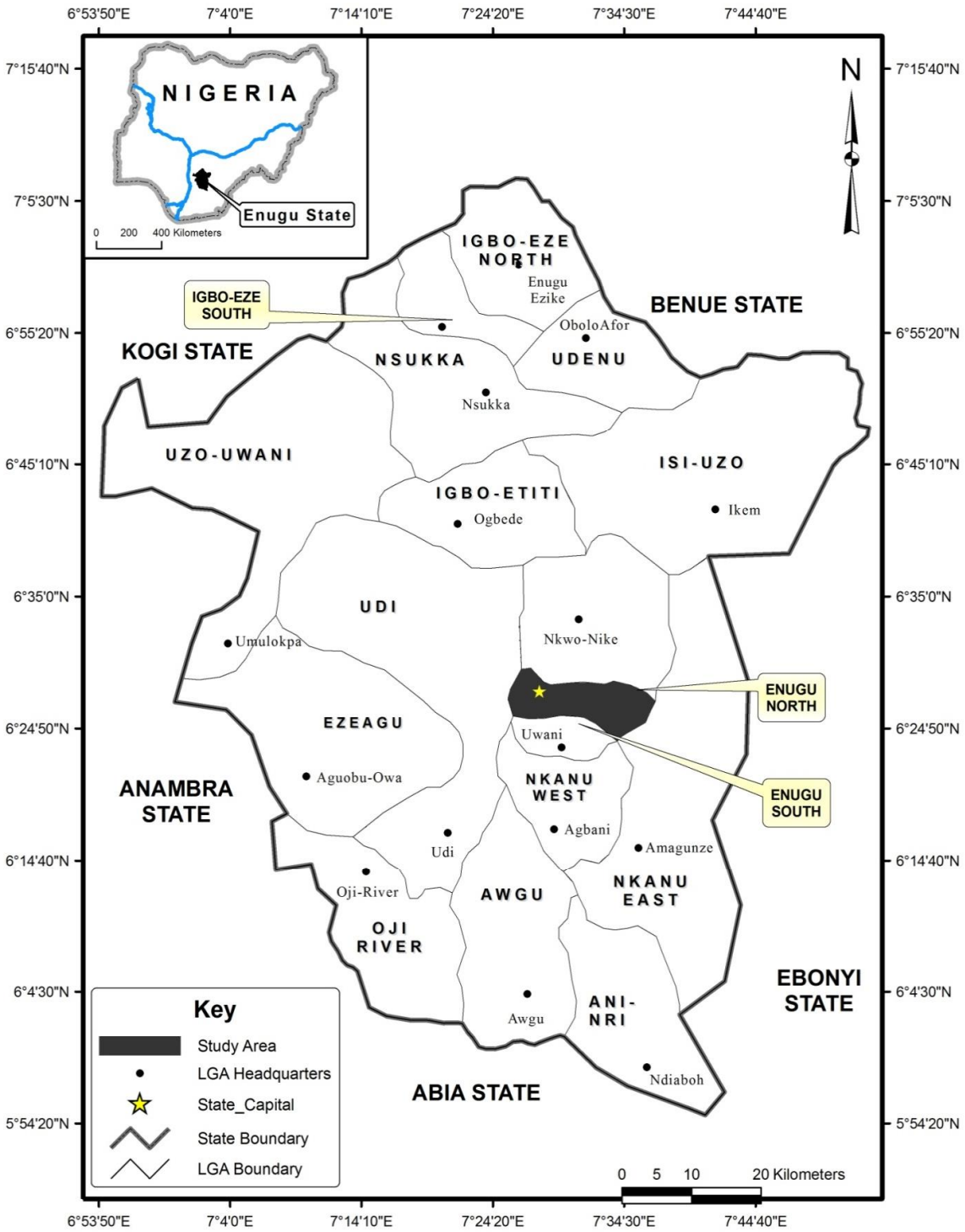

Fig. 1: Enugu State showing the Studv Area

Source: Department of Geography, University of Nigeria, Nsukka

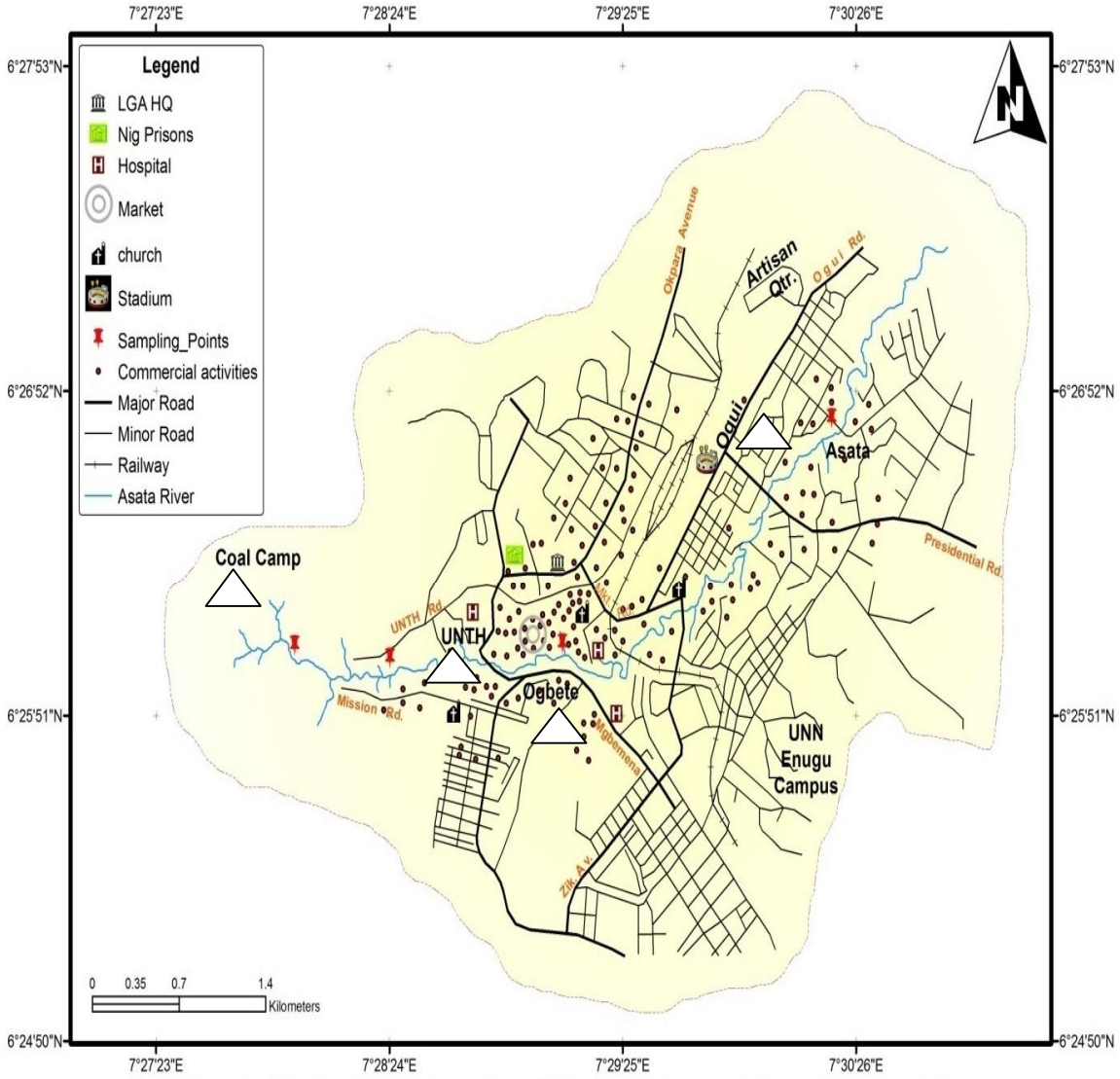

Fig. 2: Study Area showing River Asata and Sampling points

Source: Department of Geography, ***University of Nigeria, Nsukka***

Key △ = Sampling points

3.2 COLLECTION OF SAMPLES

Samples were collected fortnightly from four locations for analyses of the physicochemical, algal and microbial content. These stations were Okpara mine (abandoned coal mine), University of Nigerian Teaching Hospital Enugu (UNTH) old site, Ogbete market and Ogui, designated S_1, S_2, S_3 and S_4 respectively along the course of the river, from upstream to downstream. S_1 is near an abandoned coal mine where there is surface runoff from farm land, also bathing and laundering of clothes take place at this point; S_2 is beside a newly constructed bridge where the river passed behind the old site of University of Nigerian Teaching Hospital Enugu; S_3 is located at the Ogbete main market characterized by discharge from market waste products and waste from the abattoir and S_4 is surrounded by thick dense grass vegetation (Fig. 2).

Samples for chemical analyses were collected with a one - litre glass bottle for laboratory analysis. Samples for algal studies were collected with about 100 ml wide mouth glass bottles and preserved immediately with Lugols iodine solution (Bellinger, 1992).

3.3 Meteorological Data

Temperature, rainfall, wind speed and sunlight data for the study period were accessed from Akanu Ibiam Air-port Enugu, Enugu State.

3.4 Physicochemical Methods

The methods described by American Public Health Association (APHA, 1995), were adopted unless otherwise stated.

3.4.1 **Physical Methods**

The following physical parameters were assessed Temperature, depth, transparency, colour and rate of flow.

3.4.1.1 Temperature

Air and water temperatures were measured using mercury in glass centigrade thermometer with a range of 0 - 100^0C. At every location, the thermometer was raised up into the air to determine the air temperature of the location and thereafter the thermometer was dipped into the water sample in a beaker to measure the water temperature. The reading was recorded in degree celcius (^{0}C).

3.4.1.2 Depth Depth of the water was measured using a calibrated pole. Values were recorded in centimetre (cm).

3.4.1.3 Transparency

The Secchi disc transparency was measured with a Secchi disc following the method of Wetzel (2001). This is a disc of 20 cm diameter suspended by a rope graduated at 10 cm intervals The reading was obtained by lowering the Secchi disc into in the water and noting the depth of disappearance from view, and the disc was then pulled up and the depth of reappearance on raising was also noted. The mean of the values gave the Secchi disc transparency and it was expressed in centimetres.

3.4.1.4 Colour

The colour of the water sample was determined using a Lovibond comparator following the technique stated by British Drug House Limited (BDH). Values were recorded in Hazen units (a measure of the colour produced by 1 mg of platinum present as chloroplatinate in association with cobaltous chloride per litre of water and this is equivalent to 1 mg/l of platinum).

Procedure Nessleriser discs NSA and NSB were used. Fifty millilitre of distilled water was placed in a Nessleriser glass and placed in the left hand compartment of the Lovibond Nessleriser to serves as a blank. A matching Nessleriser glass was filled to the 50 ml mark with the sample and placed in the right hand compartment. The colour of the sample was compared with the colour on the disc until a colour match was obtained. Values were expressed in Hazen units. Samples indicating colour values beyond the available scales on the disc were diluted and the appropriate correction made.

3.4.1.5 Rate of flow

To determine the rate of flow, a known distance was measured along the course of the river .A cork was dropped in the water and the time taken for the cork to move from one point to the end of the measured distance was noted. The distance moved by the object over the time gave the rate of flow. Values were recorded in m/sec.

3.4.2 Chemical methods:

The chemical parameters investigated were as follows: pH .dissolved oxygen content, biochemical oxygen demand (BOD), chemical oxygen demand (COD), alkalinity, calcium, nitrates, ammonia, silicate, phosphate, sulphide, magnesium, total dissolved solids (TDS), total suspended solids (TSS), lead, mercury, iron and zinc, pH and dissolved oxygen were measured immediately after collection of samples in the field to prevent deterioration.

3.4.2.1 pH

This was determined using a battery operated field pH meter, Hanna model, with combined electrode and digital meter.

3.4.2.2 Dissolved oxygen content

The dissolved oxygen was determined titrimetrically using the Azide modification of Winklerôs method as described by APHA, (1995). This method is used for water containing more than 0.05 mg/l but less than 1.0 mg/l ferrous ion, or more than 0.1 mg/l nitrite nitrogen and certain organic compounds.

Reagents for Azide Modification

1. Manganous sulphate solution; Manganous sulphate (480 g) was dissolved in some quantity of distilled water and made up to 1 litre.

2. Alkaline-Iodide ï Azide solution: Potassium hydroxide (700 g) and 150 g of potassium iodide were dissolved in some quantity of distilled water and made up to 960 ml. Sodium azide (10 g) was dissolved in 40 ml of distilled water and poured into the KOH-KI solution.

3. Concentrated H_2SO_4 (sulphuric acid) Sp Gr 1.84g.

4. Sodium thiosulphate solution (0.025N): $Na_2S_2O_3.5H_2O$, 6.205 g was dissolved in freshly boiled and cooled distilled water, made up to 1 litre and standardized with (0.025N) Potassium dichromate ($K_2Cr_2O_7$).

5. Starch solution: Potato starch (1 g) was dissolved in a small quantity of distilled water and 200 ml of boiling water added, cooled and preserved with 0.25 g salicylic acid.

6. **Standardization of sodium thiosulphate solution**: Some quantity of potassium dichromate ($K_2Cr_2O_7$) was oven dried at 130 ^{0}C for 30 minutes and dissolved in 200 ml of distilled water. $K_2Cr_2O_7$ solution (10 ml) was placed in a beaker in an ice bath and 1 ml KOH-KI solution added drop by drop rotating the beaker at the same time. Then 1 ml of conc. H_2SO_4 was added and iodine released in the solution was titrated with 0.025 N $Na_2S_2O_3.5H_2O$ solutions until iodine colour reduced to pale yellow. Starch solution (2

ml) was added as indicator turning the solution blue black. Titration was continued until the blue black colour disappeared leaving a clear solution. Exactly 10 ml of 0.025 N sodium thiosulphate neutralized 10 ml of potassium dichromate. When there was variation, the appropriate correction factor was be calculated.

Procedure for Fixing Oxygen in the Field: Glass stoppered oxygen/reagent bottle (300 ml) was filled with sample to be analyzed. To this, 2 ml of $MnSO_4.4H_2O$ solution was added below the surface, followed by 2 ml of KOH /KI solution. The stopper was replaced and the content mixed by inverting the bottle several times and a precipitate of manganous sulphate (Mn (OH_2) was formed. To this, 2 ml of concentrated H_2SO_4 was added down the neck of the bottle, mixed by inverting the bottle several times; the precipitate dissolves and iodine was released from the sample. This was allowed to stand for 5 - 10 minutes and titrated with sodium thiosulphate solution.

Titration Procedure: Two hundred millilitres of sample was placed in a conical flask and titrated rapidly with 0.025 N sodium thiosulphate until the iodine colour is reduced to pale yellow colour. Then, 2 ml of starch solution was added and titration continued until blue colour completely disappeared (any blue colour formed when the solution has stayed for some time was ignored), The volume of sodium thiosulphate used was numerically equal to the dissolved oxygen content in milligrams per litre.

3.4.2.3 Biochemical Oxygen Demand

This was determined titrimetrically following the methods of APHA (1995). A five day inoculation period was used for BOD determination. Dissolved oxygen was determined in the oxygen bottle in the first day. Another portion of the sample in another oxygen bottle was incubated for five days at 25^0 C in an oven. Then; dissolved oxygen was determined at the end of the incubation period. There was a depletion of oxygen in the

incubated sample. The difference in the oxygen level gives the BOD of the sample as D_1-D_2.Where D_1 is dissolved oxygen in the first day and D_2 is dissolved oxygen after the five days of incubation.

3.4.2.4 Chemical Oxygen Demand:

This was determined titrimetrically using the methods of APHA (1995).

Procedure: Sulphuric acid (H_2SO_4) 0.4 g and 20 ml of the sample were put in a reflux flask. To this, 10 ml of 0.25 N $K_2Cr_2O_7$ was added. Pumice stone was dropped followed by 30 ml of concentrated H_2SO_4. $AgSO_4$ which was added slowly. The content of the flask was mixed thoroughly and connected to a condenser for 2 hours after which the condenser was cooled and washed down. The mixture was diluted to 150 ml with distilled water and titrated against N/10 ferrous ammonium sulphate solution, using 3 drops of ferroin as indicator (the solution changed from green colour to wine red). A blank was set up using distilled water instead of the sample.

3.4.2.5 Total Alkalinity

Total alkalinity ($CaCO_3$) of unfiltered samples was determined by titrating 100 ml of the sample with 0.02 N sulphuric acid using 2 drops of methyl orange indicator. Colour change from the yellow to faint orange indicates the end point. The volume of acid used multiplied by 10 gives the methyl orange alkalinity in part per million (mg/L). Total alkalinity (mg/l) = $\frac{ml \times N \times 50{,}000}{V}$

N = Normality of titrant, V = volume of sample (100 ml), ml = volume of titrant used**.**

3.4.2.6 Total Iron

This was determined colorimetrically by thioglycollic acid (mecarptoacetic acid) method using BDH Lovibond disc as described by British Drug Houses (BDH) with slight modifications.

Principle of the method: The method is based on the colour produced by the action of thioglycollic acid on iron (both ferric and ferrous), in the presence of ammonia.

The following reagents were used:

1. Citric Acid Solution: Citric acid (CH_8O_7) in water containing 20% W/V of the chemical.

2. Thioglycollic acid ($HSCH_2COOH$)

3. Aqueous Ammonia containing 10% W/W NH_3

Two Nessleriser Discs NEA and NEB were used depending on the range of value of iron in the sample. Disc NEA covered 0.002-0.0018 mg iron/ 50 ml of sample, while NEB covers 0.002 to 0.006 mg iron/50 ml of sample. Values were converted to mg/l by multiplying by 20.

Technique: One Nessleriser glass was filled to 50 ml mark with sample to be analyzed. To this 2 ml of citric acid was added followed by 0.1 ml of thiogycollic acid and 2 ml of 10% of aqueous ammonia. The solution was mixed and allowed to stand for 5 minutes and placed in the right hand compartment of the Nessleriser. To eliminate interference by the color of the sample, untreated water sample was placed in the left hand compartment as the blank instead of distilled water with reagent added.

The colour produced by the test solution was compared with that on the standard disc rotating the disc until a colour match was obtained. The amount of iron was expressed in mg /l. Samples with very high values were diluted with distilled water before determination and the appropriate correction was made.

3.4.2.7 Ammonia

This was determined colorimetrically using Nesslerôs reagent and BDH Lovibond Nessleriser following the instruction of the manufacturers. Disc NAA and NAB were used. Disc NAA covered 0.02 - 0.2 mg ammonia/50 ml of sample, while NAB covered 0.02 - 0.53 mg ammonia / 50 ml of sample.

Reagent Used: Nesslerôs Reagent (BDH)

Procedure: A Nessleriser glass was filled to the 50 ml mark with the sample to be tested and 2 ml of Nesslerôs reagent added. The solution was mixed and allowed to stand for 5 minutes. A second (matching) Nessleriser glass with 50 ml sample was placed in the left hand compartment to serve as blank instead of distilled water with reagent added. This was to eliminate interference by colour. The test solution was placed in the right compartment. The colour produced was compared with the colour of the standard disc, rotating the disc until a color match was obtained. Values obtained were expressed in mg/L. Samples with small quantities of ammonia from 0.02 - 0.10 mg per litre were allowed to stand for 15 minutes before matching.

2.4.2.8 Silica

This was determined colorimetrically using Lovibond disc and Nessleriser following the method of BDH Limited. Disc NN was used in all cases. It covered 1-20 mg/ L of silica.

Reagents Used

1. Aqueous solution of ammonium molybdate ($NH_4 MO_7O_{24}.4H_2O$). 10 % W/V

2. Sulphuric acid (H_2SO_4) 2 N.

3. Mixture of 1 volume of 10% ammonium molybdate solution and two volumes of 2N sulphuric acid.

Procedure: One Nessleriser was filled to 50 ml mark with the sample under test and placed in the left hand compartment to serve as blank and eliminate interference by colour. Another Nessleriser (matching) glass was filled with the solution under test (between 25 - 35^0C), 6 ml of the mixture of ammonium molybdate and sulphuric acid was added, mixed and allowed to stand for 10 minutes.

The test solution was placed in the right hand compartment of the Nessleriser and the colour produced compared with the colour in the standard disc rotating the disc until a colour match was obtained. Values were expressed in mg/l.

3.4.2.9 Phosphate

This was determined colorimetrically using ammonium molybdate and stannous chloride (Denigeôs method) with BDH Lovibond disc and Nessleriser following the procedure given by British Drug House Limited (BDH).

Reagent used

1. Ammonium Molybdate Reagent: Ten grams of ammonium molybdate was dissolved in 100 ml of distilled water. One hundred and fifty millilitres of concentrated H_2SO_4 was diluted in 150 ml of distilled water stirring continuously. The sulphuric acid solution was allowed to cool and ammonium molybdate added to it. This solution was stored in a polyethene bottle and protected from the action of light by masking the bottle with sheets of carbon paper and storing away from light.

2. Stannous Chloride Solution: In 100 ml of glycerol, 2.5 g stannous chloride ($SnCl_2.2H_2O$) was dissolved.

Procedure: Nessleriser disc NMB was used. It covered 0.02 - 2 mg/l phosphate present as phosphate oxide (P_2O_3). For samples found to contain very low levels of phosphate, 45 ml was used instead of 10 ml as directed by BDH.

Forty five millilitres of sample was put in a beaker and immersed in iced water so that the sample will attain the temperature (25^0C) specified in the method. One millilitre of ammonium molybdate reagent was added and mixed followed by 0.15 ml of stannous chloride solution. The test solution was mixed properly, put in a Nessleriser glass and made up to the 50 ml mark with distilled water. It was allowed to stand for 5 minutes and placed in the right compartment of the Nessleriser.

A blank was set up in a second Nessleriser glass by diluting 45 ml of the sample to be tested to 50 ml. This was placed in the left hand compartment and the colour produced in the test solution was compared with that on the standard disc until a colour match was obtained.

Values obtained in mg/l of phosphoric oxide (P_2O_3) were converted to mg/l phosphate by multiplying the value by 1.34 as directed by BDH.

3.4.2.10 Nitrate

This was determined colorimetrically with phenol disulphoric acid using BDH Nessleriser following standard methods (APHA, 1995). Disc covering the range of 0.5 - 6 mg nitrate nitrogen was used in all determinations.

Principles of the Method: This was based on the colour produced by the nitrate on phenol-2- 4 - disulphonic acid.

Reagents used

1. Phenol-Disulphonic acid Solution: Approximately 25% W/V in concentrated sulphuric acid.
2. Ammonia Solution (NH_4OH) 25%W/V Ammonia

Procedure: Thirty millilitres of unfiltered samples was evaporated to dryness in a beaker over a water bath and 1 ml of phenol disulphonic acid was added. The acid was smeared on all the dried matter to dissolve it and allowed to stand for 10 minutes. Ten millilitres of distilled water was added and the solution allowed to cool. Ten millilitres of 25% aqueous ammonia was added and the solution made up to 50 ml in a volumetric flask. A blank was set up in a second Nessleriser glass by pouring 50 ml of the untreated sample. This was placed in the left hand compartment and the colour produced in the test solution was compared with that on the standard disc until a colour match was obtained. Values were recorded in mg/L.

3.4.2.11 Hydrogen Sulphide

This was determined colorimetrically using Lovibond disc and Nessleriser using a method described by BDH.

Reagents Used

Lead acetate solution: Fifty percent solution of lead acetate was prepared by dissolving 0.5 g in 100 ml of distilled water. This quantity of lead was used instead of 1 g of lead acetate basic stated in the standard method due to unavailability of lead acetate basic. The suitability of lead acetate was confirmed by trying out the test on standard solutions of sulphide using the 50% lead acetate solution prepared. This was possible because 1 g of lead acetate dissolved in 100 ml of distilled water contains 50% lead acetate in solution while the lead hydroxide component remains insoluble.

Procedure: To 50 ml of sample, 0.1 ml of lead acetate solution was added, mixed, put in a Nessleriser glass and placed in the right compartment of the Nessleriser. Untreated sample of equal volume was put in another Nessleriser glass and placed in the left hand compartment to serve as blank. The colour produced in the treated sample was compared with the colour on the BDH disc. Values were recorded in mg/L.

3.4.2.12 Calcium

This was determined by the EDTA complexiometric method.

Procedure: A known volume of the sample was titrated with 0.01M EDTA solution using mureoxide as indicator. The solution changed from pink to purple. From the result of the titration, the calcium content was calculated using the formula below.

mg/l Ca = A × B × 400.5/ml Sample

Where A = ml titration for sample and

B = ml $CaCO_3$ equivalent to 100 ml EDTA titrant.

Values were recorded in mg/L.

3.4.2.13 Magnesium

Magnesium was determined by EDTA complexiometric method using Erichrome black-T as indicator.

Procedure: Twenty one millilitres of sample was diluted with 100 ml of distilled water and 0.25 ml of buffer solution was added (same as buffer for testing for hardness), followed by 3 - 4 drops of Erichrome black indicator. The sample was titrated slowly with 0.1M EDTA until the colour changed from red to blue. The concentration of magnesium was calculated using the formula

mg/l Mg = A× 2.4328×100//ml Sample

Where A = EDTA

Note: 1 ml 0.1 M EDTA = 2.432 ml of magnesium.

3.4.2.14 Lead

Lead was determined by sodium sulphide method.

Principle: When a sulphide is added to a solution containing lead ions, a brown colour due to formation of colloidal lead sulphide is produced.

Procedure: To 10 ml of the sample, 6 drops of 10% potassium cyanide solution and 25 ml of 1.2 ml ammonia solutions were added. Finally, 0.5 ml of 10% sodium solution was added and mixed. The absorbance of the resulting brown solution was measured at a wave length of 430 nm with a VIS (S23A model) spectrophotometer. A calibration curve obtained from standard lead solution was drawn with the above method and from the calibration curve the lead concentration in the sample was determined.

The values were calculated as follow;

$$\text{mg Pb/l} = \frac{\text{ug Pb (in 10 ml, from calibrated curve)}}{\text{ml sample}}$$

3.4.2.15 Mercury

This was determined by spectrophotometeric method as described by Ramachandra and Solaki (2007). VIS (S23A model) spectrophotometer was used.

Procedure: In two test tubes, 2.5 ml aliquots were put (standard and bland) and 2.5 ml EDTA solution, 2.5 ml of potassium iodide buffer, 0.5 ml of rhodamine 6G and 1 ml of gelatin solution were added. Then, it was made up to 25 ml with distilled water. After thorough mixing, the intensity of colour was read at 565 mm in a VIS (S23A model) spectrophotmetre and the concentration was determined from the calibration curve.

3.5 Biological Methods

3.5.1 Algal Studies

3.5.1.1 Labeling of Samples

The samples collected were labeled with information such as date, time of sampling, study area, station number and volume collected. These were written with a permanent maker on a masking tape and pasted on the container.

3.5.1.2 Preservation of Samples

Between the time of sampling and its analysis in the laboratory, physical, chemical and biochemical changes may have taken place altering the intrinsic quality of the samples. Therefore it was necessary to preserve the samples before taking them to the laboratory to prevent or minimize change. This was done by keeping the samples in the dark, adding preservatives, lowering the temperature to retard reaction by freezing or by a combination of both methods. For a phytoplankton sample to be analyzed for extended period, Lugolôs iodine which stained cell brownish yellow and maintain cell morphology was used.

Lugolôs iodine was prepared by dissolving 20 g of potassium iodide in 200 ml of distilled water. Then 10 g of pure iodide was dissolved in this solution. This was preserved by adding 20 g of glacial acetic acid few days before use (Hotzel and Chroome, 1999).

3.5.1.3 Evaluation of algal biodiversity

The analysis of the population was carried out by the following methods of Radojevic and Bashkin (1999). The phytoplankton consisting of individual cells, filaments and colonies were counted as individual cells. When colonies were counted, the average

number of cells per colony was counted, and in filamentous algae, the average length of the filament was determined if possible.

3.5.1.3.1 Preparation and mounting of Slides

Fresh preserved samples in the bottles were mixed uniformly. A wet mount was prepared by collecting the sample with a calibrated pipette and one drop of the sample was placed on a glass slide. A cover glass was carefully placed to avoid air bubbles. The cover glass was held in position with transparent nail vanish to prevent evaporation during the identification and counting processes. A binocular compound microscope Leica Gallen model with objective lenses X10 X40 and X100 was used for algal studies). A digital camera Cannon camera 10.0 mega pixels, 3.3X optical zoom was used for photomicrography.

3.5.1.3.2 Microscopy

Dichotomous keys were used to identify algae following the methods of Prescott (1962 and 1970), Belcher and Swale (1976), Bellinger (1992) and materials from the internet (http:/www.lifescience.napier.ac/uk/algalweb/tribo.a.jpi).

3.5.1.3.3 Counting Method

Algal enumeration was done using ôDrop count methodô as described by Ramachandra and Solanki, (2007). In this method the plankton in a drop of the sample on the glass slide was counted in transects. The total area under the cover slip represented the number of organisms present in a drop of the sample. The appropriate factor was calculated to give the number of organisms per mililitre of water sample.

Some plankton are unicellular while others are multicellular (colonial), posing problems for enumeration (Ramachandra and Solanki, 2007). For analysis, a colony of plankton

was counted as single count. According to Ramachandra and Solanki (2007), formula for calculating organism per litre is shown below.

Total plankton count per liter = A x (1/L) x (n/v)

Where;

A = number of organism per drop

L = volume of original sample (l)

n = total volume of concentrated sample (ml)

V = volume of one drop (ml)

3.5.1.8 Diversity Index

Diversity indices are used to characterize species abundance relationship in natural communities. Relative abundance of species was determined using Shannon-Wiener index (H) as described by Ogbeibu (2005).

$$H^1 = \tilde{e}_{i-I}^{S} (PilnPI)$$

Where: H^1= Shannon Diversity index

Pi = relative abundance of speciesòIò

ln = natural logarithm

ni = number of individual of species ñIò

N= total number of individual of all species

S = total number of species

Evenness index:: This is the ratio of the observed diversity (H^1) to the maximum diversity (H^1_{max})

$$\text{Evenness (E)} = \frac{H^1}{H_{max}} = \frac{H^1}{\ln S}$$

Where: H^1 = observed Shannon Diversity index, lnS = natural logarithm, H_{max} = maximum index

3.5.2 Microbial Analyses of Water

Manual for Systematic Bacteriology (Ezeonu *et al.*, 2011) was used to identify the microorganisms.

3.5.2.1 Media Preparation

Macconkey agar (52 g) was suspended in 1000 ml of distilled water. The medium was homogenized and autoclaved with auto ï control portable type stainless pressure at 121^0C for 15 minutes at 103 Kilonewton per metre squared (KN / m^2).

3.5.2.2 Microbial Count

Spread-plating method was used for the bacterial count. Separate sterile pipette was used to transfer 0.1 ml from each dilution of water sample onto the corresponding Macconkey agar. Then 0.1 ml of the water sample was spread evenly over the plate count agar surface with sterile glass spreader. The plate was labeled appropriately based on the water sample (S_1, S_2, S_3 and S_4) and incubated in Gallenhamp Hot Box for 24 hours at 37^0 C.

3.5.2.3 Microbial Characterization

Discrete bacteria colonies were isolated immediately for counting. The isolates were characterized by gram staining, sugar fermentation test, methyl red test, indole

production test, citrate utilization test, oxidized test and catalase test (Cheesbrough, 2004).

3.5.2.4 Catalase Test

This test was used to differentiate those bacteria that produce the enzyme catalase, such as staphylococci, from non-catalase producing bacteria such as streptococci.

Method: Hydrogen peroxide solution (2 - 3 ml) was poured into a test tube. Using a sterile wooden stick or glass rod, several colonies of the test organism were removed and immersed in the hydrogen peroxide. Active bubbling indicated that the test was positive but no bubbling showed that the test was negative.

3.5.2.5 Citrate utilization test

This test was used occasionally to assist in the identification of enterobacteria. The test is based on the ability of an organism to use citrate as its only source of carbon.

Method: A dense bacterial suspension of the test organism was prepared in 0.25 ml sterile physiological saline in a small tube. To this a citrate tablet was added and the tube was stoppered and incubated overnight at 35 - 37^0C. Red colouration showed that the test was positive while yellow-orange colour showed a negative test.

3.5.2.6 Oxidase test

The oxidase test is used to identify *Pseudomonas, Nesseria, Vibro, Brucelis* a*nd Pasteurella* species, all of which produce the enzyme cytochrome oxidase.

Method Freshly prepared oxidase reagent (2-3 drops) was placed on a piece of filter paper in a clean petri dish. A colony of the test organism was removed using a piece of stick or glass rod and smeared on the filter paper. Deep blue or purple colouration after 10 seconds showed a positive test**.**

3.5.2.7 Urease test

Testing for urease enzymes activity is actively used in differentiating enterobacteria. *Proteus* strains are strong urease producers. *Y-enterococolitrica* also shows activity (at 35 - 37^{0}C). *Salmonellae* and *Shigellae* do not produce urease**.**

Method: A bijou bottle containing 2-3 ml of the sterile test organism was inoculated heavily and incubated at 35-37^{0}C for 3-12 hours. A pink colour showed positive test, while no pink colour showed negative test.

3.5.2.8 Indole test

This test is important in the identification of enterobacteria. Most strains of *E. coli, P. vulgaris, P. rettgeri, M. morgani, and Providenca* species break down the amino acid tryptophan with the release of indole.

Method: The test organism was inoculated in a bijou bottle containing 3 ml of sterile tryptone water, incubated at 35-37^{0}C for up to 48 hours. Kovacôs reagent (0. 5 ml) was added to the test solution, shaken gently and examined for a red colour at the surface layer within 10 minutes. A red surface layer indicated positive test while no red colour indicated negative test.

3.6 DATA ANALYSIS

Genstat package (Discovery edition version 10) was used for the Analysis of Variance (ANOVA) and correlation analysis of physicochemical parameters with algal population and the Spearmanôs correlation coefficients noted. Paleontology Statistics (PAST) was used to determine the diversity of the algal species in each sampling point.

CHAPTER FOUR

RESULTS

4.1 CLIMATE

4.1.1 **Rainfall**

The monthly mean rainfall ranged from 21.6 - 393.2 mm, observed in the months of September and October respectively.

4.1.2 Relative Humidity

The monthly mean relative humidity was highest in the month of July (87 %) and least in the month of November (79%).

4.1.3 Solar Radiation

The maximum mean value was recorded in the month of November 2012 (13.5 watts/m^2/sec) while the minimum was recorded in August 2012 (8.8 watts/m^2/sec).

4.1.4 **Atmospheric temperature**

The maximum ranged from 28.9 ï 32.7^{o}C recorded in the months of August and November respectively. While the minimum temperature ranged from 21.5 ï 22.2^{o}C recorded in the months of July and November respectively.

4.1.5 Wind Speed

The maximum was recorded in the month of October, 2012 (7 m/s), while the minimum values were recorded in the months of July and August 2012 (5 m/s). The monthly fluctuations in the meteorological data are shown in Fig. 3.

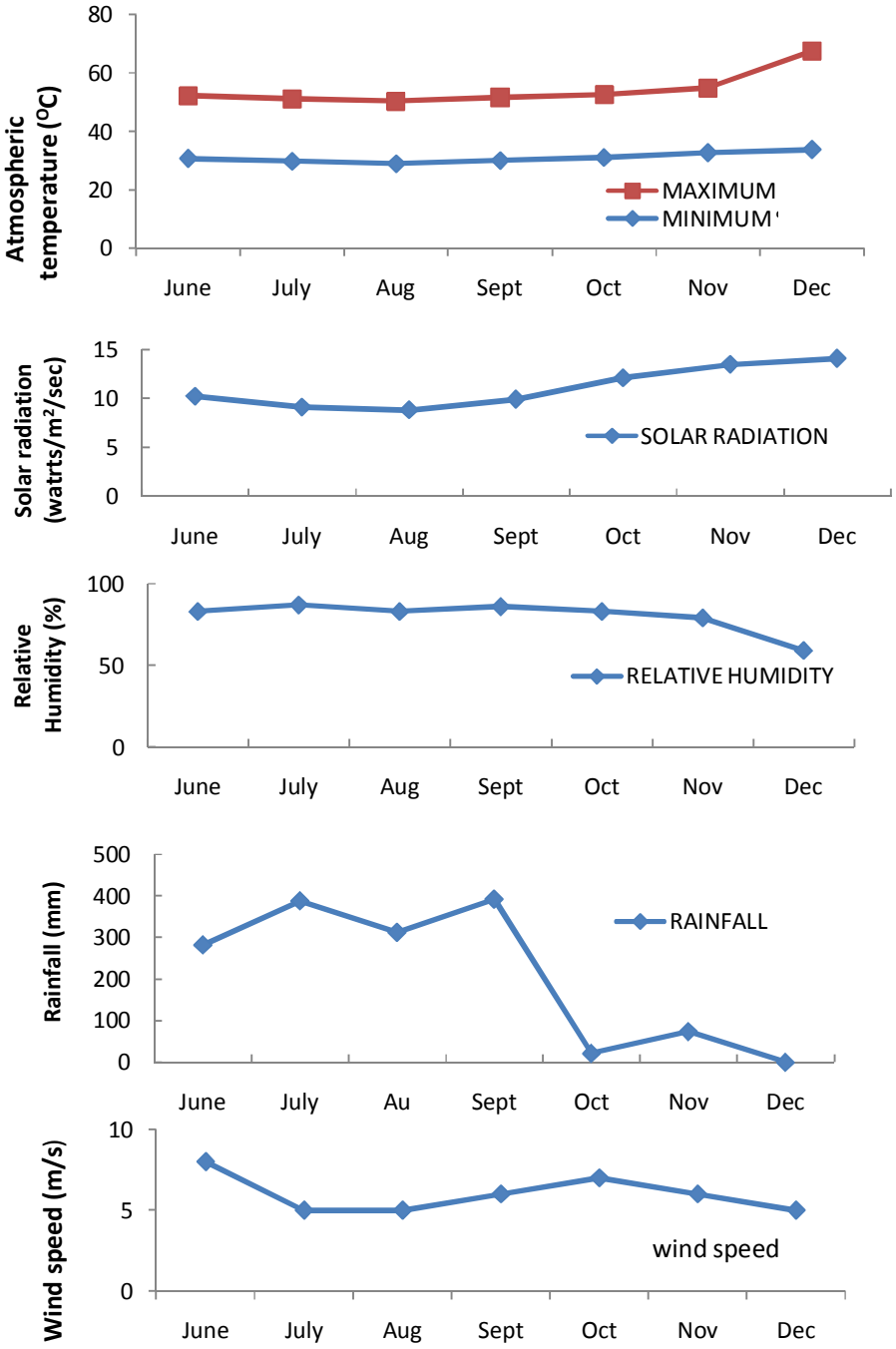

Figure 3: Meteorological data for atmospheric temperature, solar radiation, relative humidity, rainfall and wind speed

Source: Akanu Ibiam Airport Enugu, Enugu State, Nigeria.

4.2 **Okpara Coal Mine**

4.2.1 **Monthly Variations in physicochemical parameters of Asata River at Okpara Coal Mine**

Air temperature was lowest in the month of August and highest in September with a mean value of 30.33 + 0.93^0C. Water temperature was lowest and highest in the months of September and November with a mean value of 27.9 ± 0.615^0C. Maximum colour value was obtained in the month of June and lowest in July and the mean value was 60 ± 6.742 Hazen; depth was lowest in the month of July and highest in June with a mean value of 25.65 ± 4.797 cm; maximum transparency was obtained in the month of July and the lowest in the month of June the mean value was 24.77 ± 4.214 cm. The lowest velocity value was recorded in the months of June and November and the highest was obtained in July; the mean value was 0.554 ± 0.0934 m/s. Total alkalinity was highest in the month of November and least in June; the mean value was 52.75 ± 6.162 mg/l. Total Solids and total dissolved solids were highest in the month of June and lowest in October and their mean values were 233.3 ± 18.80 mg/l and 211.1 ± 16.68 mg/l respectively. Total suspended solids was lowest in the months of June and November and highest in the month of August; with a mean value of 0.605 ± 0.0249 mg/l. Maximum dissolved oxygen value was obtained in the month of October and least value in the month of June, the mean value was 3.2 ± 0.0521. Chemical oxygen demand was highest in the month of June and least in the months of July and October respectively with a mean value of 1.657 ± 0.0242 mg/l. Maximum biological oxygen demand value was recorded in the month of September and least in the months of June and July. The mean value was 0.609 ± 0.00128 mg/l. Ammonia was slightly higher in the month of July and the mean value was 28.0 ± 0 mg/l; pH was highest in the month of September and

lowest in the month of November with a mean value of 6.6 ± 0.149. Maximum silica was obtained in November and the minimum in September with a mean value of 0.05 mg/l.

Calcium, aluminium and hydrogen sulphide did not vary much with the months and the mean values were of 13.14 ± 0.170 mg//l, 2 ± 0 and 0.035 ± 0.00208 mg/l respectively. Least magnesium value was obtained in the months of June and the highest in the month of July with a mean value of 32.51 ± 0.658 mg/l. The highest value of phosphate was recorded in the month of August and the least in June and September; with a mean value of 2.681 ± 0.000831 mg/l. Nitrate was highest in September and October and least in the month of November, with a mean value of 0.265 ± 0.00707 mg/l. Iron was highest in the month of July and lowest in the month of June, with a mean value of 1.084 ± 0.0891 mg/l. Lead was highest in the month of June and lowest in the month of October with a mean value of 0.0387 ± 0.00400 mg/l. Mercury was highest in the month of July and least in the month of October with a mean value of 0.000306 ± 0.0000489 mg/l. The maximum value of zinc was recorded in the month of October and the least in the month of June. The mean value was 0.386 ± 0.0217 mg/l.

Mean values of pH was within the permissible limit of World Health Organization (2006) drinking water standards, while the values of dissolved oxygen, total alkalinity, BOD, calcium, nitrate, phosphate, TDS and mercury were below the permissible limit of World Health Organization (2006) drinking water standards. The values of iron, lead and phosphate were beyond the permissible limit of World Health Organization (2006) drinking water standards.

Table 1: Variations of physicochemical parameters of Asata River at Okpara Coal Mine

Parameter	June	July	Aug	Sept	Oct	Nov	Mean ± se	Who standard
Air temperature (^{o}C)	33.93	27.38	25.93	34.6	29.95	30	30. ± 0.79	NA
Water temperature (^{0}C)	28	25	27	26	27	29	27.8 ± 0.615	NA
Colour (Hazen)	70.1	70	70.1	40.1	40.2	70	60.27 ± 6.742	NA
Depth (cm)	58.1	11.5	15	23	23	19	25.65 ± 4.797	NA
Transparency (cm)	525	11.5	15	23	24	19	24.77 ± 4.214	NA
Velocity (m/s)	0.3	0.9	0.5	0.4	0.6	0.3	0.554 ± 0.0934	NA
T.S.S. (mg/L)	0.63	0.74	0.84	0.73	0.64	0.63	0.605 ± 0.0249	NA
T.S. (mg/L)	248.75	174.50	237.5	245	145	220	233.3 ± 18.80	NA
TDS (mg/L)**	248.12	173.76	236.66	244.27	144.36	219.37	211.1 ± 16.68	1000
Total Alkalinity. (mg/L)**	90.1	84.5	25	45.1	37	27	52.75 ± 6.162	100
DO (mg/L)**	1.13	2.1	3.46	4.51	5.13	2.59	3.2 ± 0.521	5 ï 8.5
COD (mg/L)	1.73	1.58	1.63	1.65	1.58	1.67	1.657 ± 0.0242	NA
BOD (mg/L)**	0.3	0.3	0.31	0.33	0.32	0.31	0.607 ± 0.00128	2 ï 4
Ammonia (mg/L)	28	28.2	28	28	28	28	28 ± 0	NA
pH (mg/L)	6.7	6.8	6.8	6.9	6.1	5.7	6.6 ± 0.149	6.5 ï 8.5
Silica (mg/L)	0.05	0.04	0.05	0.03	0.04	0.06	0.05 ± 0	NA
Calcium (mg/L)**	13	13	13	13.	13	12	13.14 ± 0.170	100
Aluminum (mg/L)	0.03	0.02	0.03	0.03	0.03	0.03	0.035 ± 0.00208	NA
Magnesium (mg/L)**	30.36	33.72	31.87	32.1	30.77	31.8	32.51 ± 0.658	NA
Nitrate (mg/L)**	0.25	0.25	0.25	0.3	0.3	0.2	0.265 ± 0.00707	50
Hydrogen sulphide (mg/L)	2.0	2.1	2.1	2.0	2.0	2.1	2.0 ± 0	NA
Phosphate (mg/L)*	2.61	2.65	2.67	2.61	2.65	2.66	2.68 ±.0.00833	0.5
Iron (mg/L)*	0.71	1.35	1.28	0.99	0.97	1.23	1.084 ± 0.0891	0.3
Lead (mg/L)*	0.25	0.05	0.24	0.14	0.02	0.03	0.038 ± 0.00271	0.01
Mercury (mg/L)**	0.375×10^{-5}	0.85×10^{-5}	0.2×10^{-4}	0.275×10^{-6}	0.15×10^{-7}	0.518×10^{-6}	$3.06 \times 10^{-4} \pm 4.59 \times 10^{-5}$	0.006
Zinc (mg/L)	0.3	0.4	0.4	0.4	0.5	0.4	0.0217	NA

*** = Beyond WHO 2006 standard NA = Not available**

**** = Below permissible value**

4.2.2 Algal biodiversity of Asata River at Okpara coal mine

The composition of algae observed at Okpara Coal Mine is presented in the Table 2. Four divisions of algae comprising Bacillariophyta (73%), Chlorophyta (20%), Cyanophyta (4%) and Euglenophyta (3%) were encountered. Bacillariophyta was represented by *Achnantes* sp., *Cyclotella* sp., *Gomphonema* sp., *Navicula* sp. *Surirella* sp., and *Tabellaria* sp., Chlorophyta was represented by *Ankistrodesmus* sp., *Closterium* sp., *Cosmarium* sp., *and Spirogyra* sp.. Cyanophyta was represented by *Gloeocapsa* Kutzing and *Spirulina laxa* G.M.Smith. Euglenophyta was represented by *Euglena acus* Erhrenberg, *Euglena mutabilis* F. Schmitz *and Phacus curvicauda* Huebner. *Navicula* spp had the highest population.

Table 2: Composition of algae observed at Okpara Coal Mine

Class	Total number of individuals/ml	% Composition
Bacillariophyta		
Achnantes sp.	150	0.4
Cyclotella sp.	150	0.4
Gomphonema sp.	2250	2
Nitzschia dissipata Kutzing	1950	2
Navicula sp. 1	2800	20
Navicula sp. 2	26000	24
Navicula sp. 3	27000	25
Surirella sp	18000	16
Tabellaria sp.	5850	5
Total	**109350**	**100 (73.17)**
Chlorophyta		
*Ankistrodesmus falcatu*s Ralfs	750	6
Closterium parvulum Nageli	1050	8
Cosmarium panamense Presc.	6000	44
Mougeotia sp.	1500	11
Spirogyra crassa kuetzing	1500	11
Spirogyra ellipsopora Traseau	750	6
Spirogyra weberi Kuetzing	500	4
Ulothrix aequalis Kuetzing	1500	11
Total	**13550**	**100 (9.07)**
Cyanophyta		
Gloeocapsa Kutzing	4500	20
Oscillatoria rubescens DeCand	1500	7
Spirulina laxa G.M. Smith	1500	7
Tolypothrix distorta Kutzing	15000	67
Total	**22500**	**100 (15.06)**
Euglenophyta		
Euglena acus Ehrenberg	1500	40
Euglena mutailis F. Schmitz	1500	40
Phacus curvicauda Huebner	750	20
Total	**3750**	**100 (2.51)**

Note: The percentage in bracket indicates the percentage composition of each class of algae.

4.2.3 Monthly variations in algal densities for Okpara Coal Mine

The values for the monthly algal densities are presented in Table 3. No algae were recorded in the months of August and September. Bacillariophyta recorded ranged from 6000 ï 62300 individuals/ml observed in the months of July and November. They constituted 73.17% of the total population of algae encountered during the study period. Chlorophyta recorded ranged from 150 ï 7450 individuals/ml observed in the month of October and November They constituted 9.07% of the total population of algae encountered. Cyanophyta recorded ranged from 1500 observed in the months of June, July and October to18000 individuals/ml recorded in November. They constituted 15.06% of the total algae encountered during the study period. Euglenophyta was not recorded in the months of June, August and September. The value recorded ranged from 750 -1500 individuals/ml recorded in the months of July, October and November. They constituted 2.51% of the total population of algae encountered during the study period.

Table 3: Monthly variations in algal densities of Asata River (individuals/ml) at Okpara Coal Mine

Division	June	July	August	September	October	November	Total	%
Bacillariophyta	17300	6600	0	0	23150	62300	109350	73.17
Chlorophyta	1800	4150	0	0	150	7450	13550	9.07
Cyanophyta	1500	1500	0	0	1500	18000	22500	15.06
Euglenophyta	0	750	0	0	1500	1500	3750	2.51
Total	20600	13000	0	0	26300	89250	149450	100
%	13.78	8.70	0	0	17.60	59.72	100	

4.3 University of Nigeria Teaching Hospital Enugu (UNTH) Old Site

43.1 Monthly variations in physicochemical parameters of Asata River at UNTH Old Site

The monthly variations in physicochemical parameters of UNTH old site are presented in Table 4. Air temperature was highest in the month of November and lowest in the month of August. The mean value was 30.83 ± 1.110^{o}C. Maximum value of water temperature was obtained in the month of November and lowest in the month of July. The mean value was 28.45 ± 41.4^{o}C. The value of colour was highest in the month of June and the lowest in the month of September. The mean value was 64.17 ± 6.566 Hazen. The maximum value for depth was obtained in September and the least in July. The mean value was 14.37 ± 1.034 cm. Transparency was highest in September and lowest in June. The mean value was 14.37 ± 1.024 cm. The maximum velocity value was obtained in July and least in the months of August and September. The mean value was 0.666 ± 0.128. Total alkalinity was highest in the month of June and lowest in the month of October. The mean value was 52.42 ± 7.342 mg/l. TS and TDS values were obtained in August and least in July. The mean values were 225.0 ± 13.06 mg and 222.4 ± 12.57 mg/l respectively. TSS was highest in the month of July and lowest in August. The mean value was 0.713 ± 0.144 mg/l.

DO values was highest in November and lowest in October and the mean value was 5.728 ± 0.529. COD value was obtained in June and least in the months of July and August. The mean value was 1.642 ± 0.0238. BOD value was highest in the months of August and September and lowest in July. The mean value was 0.622 ± 0.00115 mg / l. Ammonia did not vary with time and

location, the mean value of 28.0 mg/l. Maximum pH value was obtained in August and the least in November. The mean value was 7.05 ± 0.143 mg/l. Silica value was constant with a mean value of 0.05 mg/l.

Calcium was highest in the month of June and lowest in November, the mean value was 13.13 ± 0.182 mg/l. Magnesium was highest in November and lowest in August, the mean value was 32.28 ± 0.424 mg/l. Nitrate was high in the months of June, September, October and November and low in the months of July and August. The mean value was 0.0246 ± 0.00753 mg/l. Phosphate was almost constant with the highest value obtained in September. The mean value was 2.681 ± 0.00833 mg/l. Hydrogen sulphide was constant with a mean value of 2.0 mg/l.

Minimum iron value was obtained in June and the least in August. The mean value was 0.998 ± 0.0800 mg/l. Lead was highest in the month of August and lowest in September. The mean value was 0.0376 ± 0.00503 mg/l. Maximum mercury value was highest in the months of September and November and the least in October, the mean value was 0.000292 ± 0.0000398 mg // l. Zinc was almost constant with the highest value obtained in September. The mean value was 0.393 ± 0.0172 mg/l.

Mean values of pH and DO was within the permissible limit of World Health Organization (2006) drinking water standard, while the mean values of total alkalinity, BOD, calcium, nitrate, TDS and mercury were below the permissible limit of World Health Organization (2006) drinking water standard. The mean values of iron, lead and phosphate were beyond WHO standard.

Table 4: Variations of physicochemical parameters of Asata River at UNTH Old Site

Parameter	June	July	Aug	Sept	Oct	Nov	Mean ± SE	WHO Standard
Air temp (^{0}C)	31.55	32.97	27.08	29.98	31.13	33.98	30.83 ± 0.140	NA
Water temp (^{0}C)	29	26	28	29	29	31	28.48 ± 0.414	NA
Colour (Hazen)	85	70	70	40.1	40.5	70	64.17 ± 6.566	NA
Depth (cm)	11.2	10.6	12	19	16	16	14.37 ± 1.034	NA
Transparency (cm)	11.2	10.6	12	19	16	16	14.37 ± 1.024	NA
Velocity (m/s)	0.6	0.9	0.4	0.4	0.6	0.6	0.666 ± 0.128	NA
T.S.S. (mg/L)	0.49	0.74	0.47	0.70	0.68	0.65	0.713 ± 0.144	NA
T.S. (mg/L)	198.25	248	248.5	198.5	247.5	202.5	225.0 ±13.06	NA
TDS. (mg/L)**	197.75	247.26	248.03	197.8	246.82	201.85	222.4 ± 12.57	1000
Total Alkalinity. (mg/L)**	61.5	64.5	54.1	41.3	47.5	32.1	52.42 ± 7.342	100
DO. (mg/L)	2.88	3.4	3.56	4.67	5.87	2.21	5.758 ± 0.529	5 ï 8.5
COD. (mg/L)	1.67	1.61	1.61	1.62	1.64	1.62	1.642 ± 0.0238	NA
BOD. (mg/L)**	0.58	0.6	0.66	0.66	0.64	0.62	0.622 ± 0.0115	2 ï 4
Ammonia. (mg/L)	28	28	28	28	28	28	28.0 ± 0	NA
pH. (mg/L)	6.9	7.0	7.3	7.0	6.8	6.4	7.05 ± 0.143	6.5 ï 8.5
Silica. (mg/L)	0.05	0.05	0.05	0.05	0.05	0.05	0.05 ± 0	NA
Calcium. (mg/L)**	13.5	13	13	13	13	12.5	13.13 ± 0.182	100
Aluminium. (mg/L)	0.03	0.04	0.03	0.03	0.03	0.02	0.0319 ± 0.00122	NA
Magnesium. (mg/L)	31.63	32.71	31.6	32.82	32.57	32.76	32.28 ± 0.424	NA
Hydrogen sulphide. (mg/L)	2.0	2.0	2.0	2.0	2.0	2.0	2.0 ± 0	NA
Phosphate. (mg/L)*	2.68	2.68	2.68	2.69	2.68	2.68	2.681 ± 0.0833	0.5
Nitrate. (mg/L)**	0.25	0.2	0.2	0.25	0.25	0.25	0.246 ± 0.753	50
Iron. (mg/L)*	0.61	1.08	1.27	0.87	0.87	1.25	0.998 ± 0.08	0.3
Lead. (mg/L)*	0.04	0.05	0.26	0.16	0.02	0.03	0.036 ± 0.003	0.01
Mercury. (mg/L)**	0.25 x 10^{-5}	0.25 x 10^{-5}	0.25 x 10^{-5}	0.27 x 10^{-6}	0.15 x 10^{-5}	0.27 x 10^{-6}	0.000292 ± 3.98 × 10^{-5}	0.006
Zinc (mg/L)	0.4	0.4	0.4	0.5	0.4	0.4	0.393 ± 0.017	NA

*** = Beyond WHO 2006 Standard NA = Not available ** = Below permissible value**

4.3.2 Algal biodiversity of Asata River at UNTH Old Site

The composition of algae observed at UNTH Old Site is presented in Table 5. Four divisions comprising Chlorophyta (40.59 %), Bacillariophyta (23.47 %), Cyanophyta (19.30 %) and Euglenophyta (16.64 %) was recorded. Bacillariophyta was represented by *Fragillaria* sp, *Navicula* 1, *Navicula* 2, *Navicula* 3, *Nitzschia dissipata* Kutzing, *Surirella* sp and *Tabellaria* sp.. Chlorophyta was represented by *Ankistrodesmus* sp., *Chlorella* sp., *Closterium* sp., *Cosmarium* sp., *Oedogonium* sp., *Phormidium* sp. and *Ulothrix* sp. Cyanophyta was represented by *Gloeocapsa* Kutzing., *Microcystis* sp. and *Spirulina laxa* G.M.Smith. Euglenophyta was represented by *Phacus curvicauda* Huebner.

Table 5; Composition of algae observed at UNTH Old Site.

Class	Total Number Of Individual/Ml	% Composition
Bacillariophya		
Fragillaria sp	15000	71
Navicula sp. 1	17000	8
Navicula sp. 2	1500	7
Navicula sp. 3	1300	6
Nitzschia dissipata Kutzing	150	1
Surirella sp.	150	1
Tabellaria sp.	1360	6
Total	**21160**	**100 (23.47)**
Chlorophyta		
Ankistrodesmus falcatus Ralfs	2550	7
Chlorella sp.	750	2
Closterium parvalum Nageli.	300	1
Cosmarium panamense Presc	1500	4
Microspora sp.	750	2
Oedogonium sp	750	2
Ulothrix aequalis Kuetz	30000	82
Total	**36600**	**100 (40.59)**
Cyanophyta		
Gloeocapsa Kutzing	900	5
Spirulina laxa G.M.Smith	1500	9
Phormidium sp	15000	86
Total	**17400**	**100 (19.30)**
Euglenophyta		
Phacus *curvicauda* Huebner	15000	100
Total	**15000**	**100 (16.64)**
GRAND TOTAL	**76660**	**100**

Note: The percentage in bracket indicates the percentage composition of each class of algae.

4.3.3 Monthly variation in algal densities at UNTH old site

The values for the monthly algal densities measured as individuals/ml are presented in Table 6. Bacillariophyta ranged from 0 ï 16300 individuals/ml observed in the months of August and July respectively. They constituted 23.47% of the total population. Chlorophyta ranged from 0 ï 16200 individuals/ml. No Chlorophyta was observed in the months of June and August, the highest value was observed in the month of July. They constituted 40.59% of the entire population. Cyanophyta was not observed in the months of June, August and September, the values observed ranged from 150 ï 2250 individuals/ml observed in the months of July and November respectively. They constituted 19.30% of the entire population. Euglenophyta was observed only in the month of November with a value of 1500 individuals/ml. They constituted 16.64% of the entire population.

Table 6: Monthly variations in algal densities (individuals/ml) at UNTH Old Site

Division	June	July	August	September	October	November	Total	%
Chlorophyta	0	16200	0	15000	2250	3150	36600	40.39
Bacillariopyta	1400	16300	0	310	800	2350	21160	23.47
Cyanophyta	0	0	0	15000	0	2250	17400	19.30
Euglenophyta	0	0	0	0	0	15000	15000	16.64
Total	1400	32650	0	30310	3050	22750	90160	100
%	1.55	36.21	0	33.62	3.38	25.23	100	

4.4 Ogbete Market

4.4.1 Physicochemical parameters of Asata River at Ogbete market.

The mean monthly variations in physicochemical parameters of Asata River at Ogbete market are shown in Table 7. Maximum air temperature value was recorded in September and the least in July; the mean value was 29.55 ± 1.054^{o}C. Water temperature was highest in August and lowest in June; the mean value was 28.22 ± 0.374^{o}C. Minimum colour value was obtained in October and the maximum in September; the mean value was 72.8 ± 0.116 Hazen. Depth was highest in October and least in September; the mean value was 13.06 ± 1.647 cm. The least transparency value was obtained in August and the highest in October; the mean value was 13.06 ± 1.647 cm. Velocity was highest in the month of July and lowest in November; the mean value was 0.431 ± 0.0789 m/sec

Total alkalinity was highest in July and lowest in September; the mean value was 67.58 ± 8.516 mg/l. DO was highest in October and lowest in November; the mean value was 2.454 ± 0.582 mg/l. BOD was almost constant; the mean value was 0.626 ± 0.00713 mg/l. COD was highest in the month of November and lowest in June; the mean value was 1.612 ± 0.0159 gm/l. The lowest pH value was recorded in November and highest in June; the mean value was 6.9 ± 0.056. TDS and TS values were highest in September and lowest in November; the mean value were 249.0 ± 15.15mg/l and 246.7 ± 17.29 mg/l.. The least TSS value was recorded in September and the highest in July; the mean value was 0.68 ± 0.990 mg/l.

Aluminium value was almost constant with the mean value of 0.0339 ± 0.00168 mg/l. Ammonia was constant with a mean value of 28.0 mg/l.

Calcium value was almost constant and the mean value was 13.13 ± 0.114 mg / l. Minimum magnesium value was recorded in November and the highest in October; the mean value was 33.46 ± 0.763 mg/l. Nitrate was highest in September and lowest in the months of June, August and November; the mean value was 0.247 ± 0.00553 mg/l. Phosphate was almost constant with a mean value of 2.68 ± 0.00108 mg/l. Silica value was constant with a mean value of 0.05 mg/l. Hydrogen sulphide was constant with a mean value of 2.0 mg/l. Iron was highest in October and lowest in September; the mean value was 1.07 ± 0.854 mg/ l. Lead was highest in the month of June and lowest in the month of September; the mean value was 0.0379 ± 0.00421 mg/l. The least values for mercury were recorded in the months of July and September and the highest in November; the mean value was 0.00035 ± 0.0000289 mg/l. Zinc values were low in the months of June, July and September, and high in the months of August, October and November; the mean value was 0.0399 ± 0.0174 mg/l.

The mean value of pH was within the permissible limit of World Health Organization (2006) drinking water standard, while the values of total alkalinity, BOD, DO, calcium, dissolved oxygen, nitrate, TDS and mercury were below the permissible limit of World Health Organization (2006) drinking water standard, The mean value of iron, lead and phosphate were beyond WHO standard,

Table 7: Variations of physicochemical parameters of Asata River at Ogbete Market.

Parameter	June	July	Aug	Sept	Oct	Nov	Mean ± SE	WHO Standard
Air temp (^{0}C)	30.5	25.4	29	32.15	27.98	28.90	29.55 ± 1.054	NA
Water temp (^{0}C)	27	27.5	30	29	28	29	28.22 ± 0.371	NA
Colour (Hazen)	70	70	70	100	50.1	77.5	72.32 ± 0.166	NA
Depth (cm)	12	7.5	11	13	23	19	13.06 ± 1.647	NA
Transparency (cm)	12	7.5	11	13	23	19	13.06 ± 1.647	NA
Velocity (m/s)	0.5	0.7	0.3	0.5	0.5	0.2	0.436 ± 0.078	NA
T.S.S. (mg/l)	0.73	0.75S	0.71	0.63	0.64	0.67	0.678 ± 0.019	NA
Total .solids. (mg/l)	250.75	252	249	260.53	250.25	199	246.7 ± 17.27	NA
TDS**. (mg/l)	250.02	257.25	248.29	259.90	249.61	198.33	249.0 ± 15.15	1000
Total Alkalinity (mg/l)**	60.5	94	66	70.5	49.5	25	67.58 ± 8.516	100
DO (mg/l)**	0.99	1.74	2.99	3.67	5.51	0.31	2.45 ± 0.592	5.5 - 8.5
COD (mg/l)	1.57	1.59	1.58	1.58	1.6	1.69	1.621 ± 0.016	NA
BOD (mg/l)**	0.64	0.63	0.58	0.63	0.64	0.64	0.626 ± 0.007	2 ï 4
Ammonia (mg/l)	28	28	28	28	28	28	28.0 ± 0	NA
Ph	7.2	7.0	7.0	7.1	7.0	6.5	6.9 ± 0.056	6.5ï 8.5
Silica (mg/l)	0.05	0.05	0.05	0.05	0.05	0.05	0.05 ± 0	NA
Calcium (mg/l)**	13	13	13	13.5	13	13	13.13 ± 0. 114	100
Aluminium (mg/l)	0.03	0.03	0.04	0.03	0.04	0.03	0.0339 ± 0.002	NA
Magnesium (mg/l)	32.98	32.63	33.12	33.31	33.82	31.75	33.68 ± 0.763	NA
Nitrate (mg/l)**	0.2	0.25	0.2	0.3	0.25	0.2	0.247 ± 0.006	50
Hydrogen sulphide (mg/l)	2.0	2.0	2.0	2.0	2.0	2.0	2.0 ± 0	NA
Phosphate (mg/l)*	2.67	2.67	2.66	2.67	2.67	2.66	2.68 ±.0.108	0.5
Iron (mg/l)*	0.81	1.15	1.26	0.64	1.47	1.14	1.07 ± 0.0854	0.3
Lead (mg/l)*	0.46	0.05	0.04	0.02	0.3	0.03	0.0379 ± 0.004	0.01
Mercury (mg/l)**	0.25×10^{-5}	0.225×10^{-6}	0.275×10^{-6}	0.225×10^{-6}	0.275×10^{-6}	0.4×10^{-4}	$0.00035 \pm 2.89 \times 10^{-5}$	0.006
Zinc (mg/l)	0.4	0.4	0.5	0.4	0.5	0.5	0.0399 ± 0.-174	NA

NA = Not available * = Beyond WHO (2006) Standard

**** = Below permissible value**

4.4.2 Algal biodiversity of Asata at Ogbete market.

The composition of algae at Ogbete market is presented in the Table 8. Four divisions comprising Bacillariophyta (14%), Chlorophyta (61%), Cyanophyta (6%) and Euglenophyta (19%) were observed. Bacillariophyta was represented by *Caloneis* sp.*, Cyclotella* sp.*, Fragillaria* sp.*, Navicula* 1*, Navicula* 2*, Nitzschia dissipata* Kutzing*, Surirella* sp.*, Stephanodiscus* sp. *Pinnularia* sp. *and Tabellaria* sp., Chlorophyta was represented by *Ankistrodesmus* sp.*, Chlorella* sp.*, Closterium* sp.*, Cosmarium* sp.*, Oedogonium* sp.*, Cladophora* sp., *Scenedesmus curvatus, Scenedesmus obtutus, Desmodesmus quadricauda* and *Ulothrix* sp.. Cyanophyta were represented by *Gloeocapsa* Kutzing.*, Microcystis* sp.*, Oscillatoria* sp.*, Chroococus* sp. and *Nostoc* sp.. Euglenophyta was represented by *Euglena acus* Ehrenberg*, Euglena mutabilis* F. Schmitz *and Phacus curvicauda* Huebner.

Table 8: Composition of algae observed at Ogbete market

Class	Total Number Of Individual /Ml	% Composition
Bacillariophyta		
Caloneis sp.	3000	14
Cyclotella sp.	150	1
Fragillaria sp.	300	1
Navicula sp. 1	4250	20
Navicula sp. 2	4000	18
Nitzchia sp.	1500	7
Pinnularia sp.	300	1
Surirella sp	750	3
Stephanodiscus sp	2100	10
Tabellaria sp.	3300	15
Total	**21750**	**100 (11.95)**
Chlorophyta		
Ankistrodesmus falcatus Ralfs	4500	7
Chlorella sp.	30000	44.1
Closterium parvulum Nageli	2700	4
Cosmarium panamense Presc	7500	11
Cladophora sp.	2100	10
Oedogonium sp.	15000	22
Scenedesmus arcvatus	150	0.2
Scenedesmus obtusus	450	0.7
Desmodesmus qaudricauda	300	0.4
Ulothrix aequalis Kuetz	7500	11
Total	**68100**	**100 (37.43)**
Cyanophyta		
Gloeocapsa Kutzing	4500	11
Chroococus sp	600	1
Nostoc sp.	4500	11
Oscillatoria rubescens DeCand	750	2
Microcystis sp.	30750	75
Total	**41100**	**100 (22.59)**
Euglenophyta		
Euglena acus Ehrenberg	22500	73
Euglena mutabilis F. Schmitz	6000	20
Phacus curvicauda Huebner.	2250	7
Total	**30750**	**100 (28.03)**
GRAND TOTAL	**181950**	**100**

Note: The percetage in bracket indicates the percentage composition of each class of algae.

4.4.3 Monthly variations in algal density for Ogbete market

The monthly variations in algal density for Ogbete market are shown in Table 9. At Ogbete market, no alga was observed in the month of August. Bacillariophyta observed ranged from 1200 ï 11300 individuals/ml., recorded in the months of July and November respectively. They constituted 11.95 % of the total population of algae ncountered during the period of investigation. Chlorophyta observed ranged from 500 ï 68100 individuals/ml recorded in the months of September and November. They constituted 37.43 % of the total population of algae encountered during the period of study. Cyanophyta was not recorded in the months of August and September. The number recorded ranged from 7600 ï 18850 individuals/ml observed in the months of July and November. They constituted 22.59 % of the entire population. Euglenophyta was not recorded in the months August and September. The values observed ranged from 8500 ï 91750 individuals/ml. They constituted 28.03 % of the entire population of algae recorded at Ogbete market.

Table 9: Monthly variations in algal density (individual / ml) for Ogbete market

Division	June	July	August	September	October	November	Total	%
Bacillariophyta	2150	1200	0	3500	3600	11300	21750	11.93
Chlorophyta	6300	2800	0	500	19900	38600	68100	37.43
Cyanophyta	5000	7600	0	0	9650	18850	41100	22.59
Euglenophyta	2000	8500	0	0	17500	23000	51000	28.03
Total	15450	201000	0	4000	50650	91750	181950	100
%	8.49	11.05	0	2.19	27.84	50.43	100	

4.5 Ogui.

4.5.1 Physicochemical parameters of Asata River at Ogui.

The mean monthly variations of Asata River at Ogui are presented in Table 10. Air temperature was highest in the month of August and lowest in the month of June and rhe mean value was 27.92 ± 0.379^{o}C. The minimum water temperature value was obtained in October and the highest in August. and the mean value was 27.97 ± 0.269^{o}C. The value of colour was highest in October and lowest in September and. the mean value was 68.75 ± 5.671 Hazen. Depth was highest in September and lowest in November. The mean value was 23.27 ± 1.911 cm. Transparency was lowest in November and highest in September with a mean value was 23.27 ± 1.911 cm. Maximum velocity values was obtained in June and the minimum in October, the mean value was 0.55± 0.28 m/ sec. Total alkalinity was highest in November and the least in July. The mean value was 77.10 ± 3.838 mg/l. Dissolved oxygen was highest in October and lowest in September. The mean value was 2.733 ± 0.623 mg/l. Least biological oxygen demand value was obtained in June and the highest in October. The mean value was 0.616 ± 0.0181 mg/l. Chemical oxygen demand was highest in October and lowest in July with a mean value of 1.363 ± 0.0181 mg/l. pH was highest in October and lowest in September and. the mean value was 6.635 ± 0.0119. Total solids were highest in June and lowest in July. The mean value was 271.7 ± 25.52 mg/l.

Total dissolved solid was highest in June and lowest in July. The mean value was 251.06 ± 24.841 mg/l. Total suspended solid was highest in August and lowest in June. The mean value was 0.637 ± 0.0186. Aluminium was highest in October and lowest in June. The mean value was 0.0327 ± 0.00158 mg/l. Ammonia was almost constant with a mean value was 28.0 ± 0. Calcium had the highest values in the

months of June, September and November, and the lowest values in the months of July, August and October. The mean value was 13.13 ± 0.128 mg/l. Magnesium had the highest value in August and the least in September. The mean value was 27.51 ± 0.2719 mg/l. Nitrate was almost constant with a mean value of 0.247 ± 0.00502 mg/l. Phosphate was constant with a mean value of 2.68 ± 0 mg/l. Silica was almost constant with a mean value was 0.05 ± 0 mg/l. Hydrogen sulphide was almost constant with a mean value of 2.0 ± 0 mg/l. Iron was highest in July and lowest in June with a mean value of 1.187 ± 0.0890 mg/l. Lead was highest in July and lowest in the months of September, October and November. The mean value was 0.0733 ± 0.00440 mg/l. Mercury was highest in September and lowest in July with a mean value of 0.000325 ± 0.0000446 mg/l. Zinc was almost constant with a mean value of 0.399 ± 0.0152 mg/l.

Mean values of pH, total alkalinity, calcium, nitrate, TDS and mercury were within the permissible limit of World Health Organization (2006) drinking water standard; BOD and dissolved oxygen were below WHO permissible limit; while the mean values of iron, phosphate and lead, were beyond WHO standards.

Table 10: Variations of Physicochemical Parameter of Asata River at Ogui

Parameter	June	July	Aug	Sept	Oct	Nov	Mean SE	WHO Standard
Air temperature (0C)	32.83	28.55	27.13	28.50	31.48	31.83	27.92 ± 0.3797	NA
Water temperature (0C)	28	27.5	26.5	27.6	29s	29	27.97 ± 0.269	NA
Colour (Hazen)	70.2	70.1	70.1	40	85.2	78	68.75 ± 5.671	NA
Depth (cm)	23	21	19	31	28	18	23.27 ± 1.911	NA
Transparency (cm)	23	21	19	31	28	18	23.27 ± 1.911	NA
Velocity (m/s)	1.6	0.8	0.3	1.0	0.3	0.4	0.55 ± 0.138	NA
T.S.S. (mg/L)	0.55	0.73	0.78	0.68	0.63	0.63	0.673 ± 0.0186	NA
T.S. (mg/L)	399.25	199.75	252	249	297	251	271.7 ± 25.52	NA
TDS* (mg/L)	398.70	199.02	251.2	248.2	296.37	250.7	251.06± 248.41	1000
Total Alkalinity. (mg/L)	82.1	88.5	72.5	67.1	79	57.1	77.10 ± 3.838	100
DO. (mg/L)	1.67	2.35	2.35	3.51	5.03	2.05	2.733 ± 0.623	5 ï 8.5
COD. (mg/L)	1.71	1.58	1.61	1.6	1.72	1.63	1.636 ± 0.018	NA
BOD. (mg/L)	0.58	0.59	0.6	0.64	0.65	0.63	0.66 ± 0.009	2 ï 4
Ammonia. (mg/L)	28	27.1	28	28	28.1	28.1	28.0 ± 0	NA
pH	6.2	6.6	6.5	5.9	6.7	6.2	6.633 ± 0.029	6.5 ï 8.5
Silica. (mg/L)	0.06	0.05	0.07	0.07	0.06	0.05	0.05 ± 0.0	NA
Calcium. (mg/L)	13.5	13	13	13.5	13	13.5	13.13 ± 0.126	100
Aluminium. (mg/L)	0.02	0.03	0.04	0.03	046	0.03	0.0327 ± 0.003	NA
Magnesium	33.01	32.79	33.29	30.45	32.99	32.57	27.57 ± 0.272	NA
Hydrogen sulphide. (mg/L)	2.0	2.1	2.1	2.0	2.0	2.1	2.0 ± 0	NA
Phosphate. (mg/L)	2.67	2.67	2.68	2.68	2.69	2.67	2.68 ± 0	0.5
Nitrate. (mg/L)	0.2	0.2	0.3	0.3	0.3	0.3	0.247 ± 0.005	50
Iron*. (mg/L)	0.67	1.37	1.31	0.98	1028	1.31	1.187 ± 0.089	0.3
Lead*. (mg/L)	0.05	0.06	0.05	0.02	0.02	0.02	0.0733 ± 0.004	0.01
Mercury. (mg/L)	0.825 × 106	0.175 × 106	0.225 × 106	0.85 × 106	0.216 × 106	0.4 × 106	3.25 × 10-4 ± 4.46 × 10-5	0.006
Zinc (mg/L)	0.4	0.4	0.4	0.5	0.5	0.4	0.399 × 0.015	NA

NA = NOT available * = Beyond WHO (2006) Standard
**** = Below WHO (2006) Standard**

4.5.2 Algal biodiversity of Asata River at Ogui

The composition of algae observed at Ogui is presented in the Table 11. Four divisions comprising Chlorophyta (57.06%), Bacillariophyta (5.63%), Cyanophyta (0.30%), Euglenophyta (37.02%) were observed. Bacllariophyta was represented *by Navicula* 1, *Navicula* 2, *Cyclotella* sp*, Tabellaria* sp.. Chlorophyta was represented *by Ankistrodesmus* sp., *Closterium* sp.*, Cosmarium* sp.*, Mougeotia* sp.*, Oedogonium* sp.*, Stegeoclonium subscecundum* and *Ulothrix* sp. *Cyanophyta* was represented by *Gloeocapsa* Kutzing. Euglenophyta was represented by *Phacus curvicauda* Huebner.

Table 11: Composition of algae encountered at Ogui during the period of investigation (June – November 2012).

Class	Total Number of Individual / Ml	% Composition
Bacillariophyta		
Navicula sp. 1	1000	35
Navicula sp. 2	800	28
Cyclotella sp.	600	21
Tabellaria sp.	450	16
Total	**2850**	**100 (5.63)**
Chlorophyta		
Ankistrodesmus falcatus Ralf.	300	1
Closterium parvulum Nageli	450	2
Cosmarium panamense Presc	750	3
Mougeoetia sp.	1750	6
Oedogonium sp.	3000	10
Stegeoclonium subscecudum	150	1
Ulothrix aequalis Kuetz	22500	78
Total	**28900**	**100 (57.06)**
Cyanophyta		
Gloeocapsa Kutzing	150	100
Total	**50650**	**100 (0.30)**
Euglenophyta		
Euglena acus Ehrenberg	3000	16
Phacus curvicauda Huebner	15750	84
TOTAL	**18750**	**100 (37.02)**
GRAND TOTAL	**50650**	**100**

Note: The percentage in bracket indicates the percentage composition of each class of algae.

4.5.3 Monthly variations in algal density of Asata River at Ogui

No algae were recorded in the months of August and September. Bacillariophyta recorded ranged from 400 - 1150 individuals/ml. observed in the months of June and November respectively. They constituted 5.63% of the total algae encountered during the period of study. Chlorophyta recorded ranged from 450 ï 19650 individuals/ml observed in the months of June, July and November. They constituted 57.06% of the total population of the algae encountered. Cyanophyta was recorded only in the month of July with a value of 150 individuals/ml. It constituted 0.03% of the total population of algae encountered. Euglenophyta recorded ranged from 850 ï 32300 individuals/ml. They constituted 37.02% of the total population of algae encountered during the period of study.

Table 12: Monthly variations in algal density (individual /ml) of Asata River at Ogui

Division	June	July	August	September	October	November	Total	%
Bacillariophyta	4\00	0	0	0	1300	1150	2850	5.63
Chlorophyta	450	450	0	0	8350	19650	28900	57.06
Cyanophyta	0	150	0	0	0	0	150	0.3
Euglenophyta	0	1500	0	0	5750	11500	18750	37.02
Total	850	2100	0	0	15400	32300	50650	100
%	**1.68**	**4.15**	**0**	**0**	**30.40**	**63.77**	**100**	

4.6 **Checklist of algae in Asata River, Enugu**

Check list of the algae encountered during the period of study are presented in Table 13. Thirty nine taxa of algae belonging to 4 divisoins ï Cyanophyta - (Cyanobacteria) ,Chlorophyta, Bacillariophyta and Euglenophyta were encountered in the study. The photo ï micrograph of algae encountered are presented in plates 1 ï 4. Cyanophyta was represented by *Nostoc* sp., *Spirulina laxa* G. M. Smith*, Gleocapsa* sp*., Chroococcus* sp. and *Microcystis* sp.. Bacillariophyta was represented by *Achnanthes* sp., *Caloneis* sp., *Cyclotella* sp., *Fragillaria* sp., *Gomphonema* sp., *Nitzschia dissipata* Kutzing, *Navicula* 1, *Navicula* 2, *Navicula* 3, *Pinnularia* sp., *Surirella* sp. and *Tabellaria* sp. Chlorophyta was represented by *Spirogyra ellipsopora* Traneau*, Spirogyra webori* Kuetzing*, Spirogyra crassa* Kuetzing, *Chlorella* sp,, *Cosmarium* sp,, *Cladophora* sp., *Microspora* sp,, *Scenedesmus obtusus, Scenedesmus curvatus, Desmoedesmus qaudricauda*, *Oedogonium* sp,, *Oocystis* sp,, *Ankistrodesmus* sp., *Tolypothrix distorta* Kutzing, *Ulothrix* sp,, *Cladophora* sp,, *Oscillatoria* sp,, *Stigeoclonium subsecundum* and *Zygonema* sp, Euglenophyta was represented by *Phacus curvicauda. Euglena mutabilis* F. Schmith *and Euglena acus* Ehrenburg. Twenty three were recorded at Okpara coal mine, eighteen for UNTH Old site, twenty four for Ogbete market and twelve for Ogui. *Navicula* 1, *Navicula* 2, *Cosmarium* sp., *Ankistrodesmus* sp, *Ulothrix* sp, *Chroococcus* sp and *Phacus curvicauda* Huebner were observed in all locations. *Achnanthes* sp., *Gomphonema* sp., *Spirogyra ellipsopora, Spirogyra webori*, *Spirogyra crassa and Tolypothrix distorta* were found only at Okpara Coal mine, *Microspora* sp, was observed only at UNTH Old Site, *Mougeoetia* sp., *Scenedesmus obtusus***,** *Scenedesmus curvatus, Desmodesmus qaudricauda*, *Cladophora* sp,, *Nostoc* sp. and *Microcystis* sp. were observed only at Ogbete market. *Stigeoclonium subsecundum* was found only at Ogui.

Table 13: Check list of algae on Asata River at different locations

Class	Okpara Coal mine	UNTH	Ogbete market	Ogui
Bacillariophyta	+	+	+	++
Navicula 1	+	+	+	+
Navicula 2	+	+	+	_
Navicula 3	+	+	+	+
Nitzschia dissipata Kutzing	+	+	+	
Cyclotella sp.	+	_	+	+
Tabellaria sp	+	+	+	+
Pinnularia sp	_	_	+	_
Gomphonema sp	+	_	_	_
Surirella sp	+	+	+	_
Fragillaria sp	_	+	+	_
Achnanthes sp	+	_	_	_
Stephanodiscus sp	_	_	+	_
Calonies sp.	_	_	+	_
Chlorophyta				
Spirogyra ellipsopora Traneau	+	_	_	_
Spirogyra webori Kuetzing	+	_	_	_
Spirogyra crussa Kuetzing	+	_	_	_
Chlorella sp	_	+	+	_
Cosmarium sp	+	+	+	+
Closterium sp.	+	+	+	+
Microspora sp	_	+	_	_
Scenedesmus obtusus	_	_	+	_
Scenedesmus curvatus	_	_	+	_
Desmodesmus qaudricauda	_	_	+	_

Oedogonium sp	_	+	+	+
Ankistrodesmus sp	+	+	+	+
Tolypotrix sp	+	_	_	_
Ulothrix sp	+	+	+	+
Cladophora sp	_	_	+	_
Stigeoclonium subscecundum	_	_	_	+
Cyanophyta				
Nostoc sp.	_	_	+	_
Spirulina laxa G. M. Smith	+	+	_	_
Gloeocapsa Kutzing.	+	+	+	+
Chroococcus sp.	+	+	+	+
Microcystis sp .	_	_	+	_
Oscillatoria sp	+	+	+	_
Euglenophyta				
Phacus curivicanda Huebner.	+	+	+	+
Euglena mutabilis F.Schmitz.	+	_	+	+
Euglena acus Ehrenburg.	+	_	+	.+

4.7 Photomicrograph of some algae in Asata River

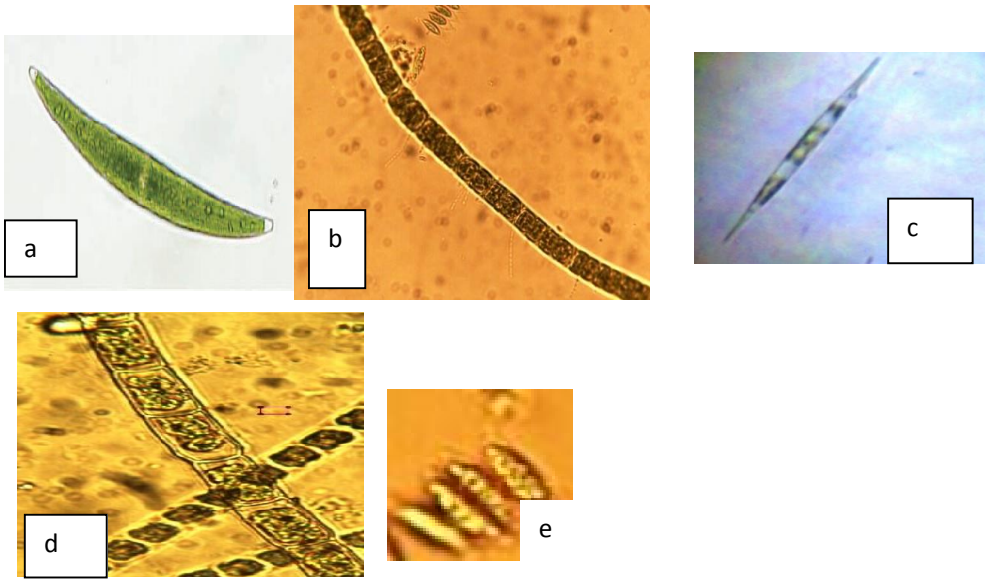

Plate 1 Photomicrographs of Some chlorophyta encountered during the study. (a*) Closterium* sp. X 40 (b) *Ulothrix* sp.. X 40 (c*) Ankistrodesmus* sp. X 40. (d) *Odogonium* sp. X 40 (e).*Desmodesmus quadricuada* X 40

.

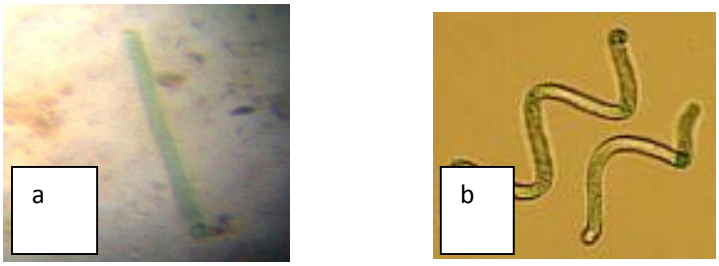

Plate 2 Photomicrograph of Cyanophyta encountered during the study. (a*) Oscillatoria* sp. X 40 (b) *Spirulina* sp.. X 40

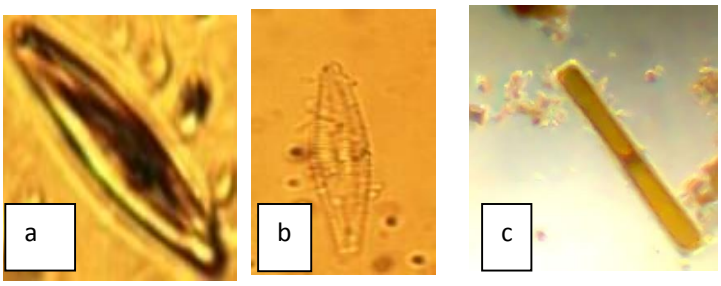

Plate 3 Bacillariophyta encountered during the study period (a*) Navicula* sp.X 40 sp. (b) *Achnanthes* sp.. X 40 (c) *Pinnularia* sp. X 40

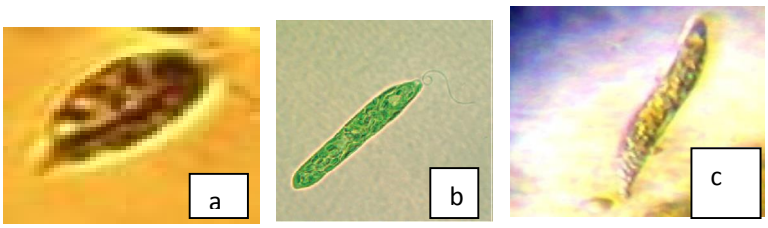

Plate 4 Photomicrographs of some Euglenophyta encountered during the study period (a*) Phacus curvicauda* Huebner X 40 (b) *Euglena mutabilis* F. Schmitz X 40 (c) *Euglena acus* Ehrenberg X 40

4.8 Percentage composition of algae encountered during the study period June – November 2012

Of the total population of algae encountered during the study, among the study areas, the highest population was observed at Ogbete market (39 %), followed by Okpara coal mine (32 %), followed by UNTH (old site 18 %) and Ogui (11 %) (fig 4).

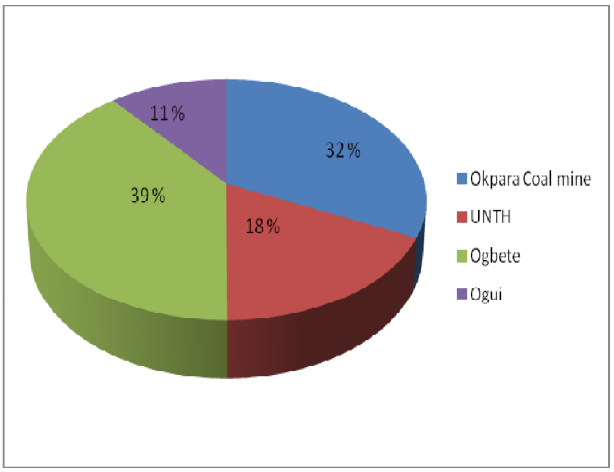

Figure 4 Percentage populations of algal classes from Asata River by location

The algae encountered during the study period belong to four (4) divisions as follows; Bacillariophyta 33 %, Chlorophyta 32 %, Cyanophyta 14 % and Euglenophyta 21 % (fig 5).

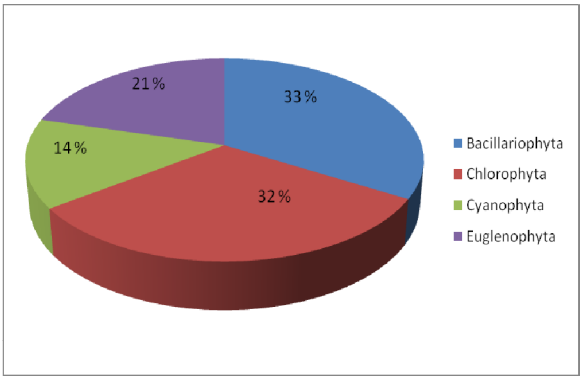

Figure 5 Percentage compositions of algae encountered in the study area

4.9 ALGAL SPECIES DIVERSITY AND EVENNESS INDICES

Algal species diversity indices in Asata River are presented in Table 14. Shannon Wiener index ranged from 0.8234- 1.322, the lowest value was obtained at Okpara Coal mine while the highest value was obtained at UNTH (old site). Evenness index ranged from 0.5696 ï 0.9378, the lowest value was recorded at Okpara Coal mine and the highest value at UNTH (old site).

Table 14 Values of species diversity indices for Asata River

Location	Shannon Wiener index	Evenness index
Okpara Coal mine	0.8284	0.5696
UNTH (old site)	1.322	0.9378
Ogbete market	1.314	0.9305
Ogui	0.8672	0.5951

4.10 **MICROBIAL ANALYSES**

4.10.1 Characterization of bacteria

The characterization and identification of the bacteria isolated from Asata River are presented in Table 15. Both gram negative and gram positive bacteria were isolated. The following bacteria were isolated from Asata River; *Salmonella* sp., *Escherichia* sp., *Enterobacter* sp., *Micrococcus* sp. and *Proteus* sp.. *Escherichia* sp. was isolated from all locations. *Enterobacter* sp. was isolated from UNTH Old site, Ogbete market and Ogui. *Salmonella* sp. was isolated from Okpara coal mine and Ogui. *Micrococcus* sp. was isolated from UNTH old site and Ogbete market; and *Proteus* sp. was isolated from Ogui.

Table 15: Characterization and identification of the bacteria isolated from Asata River

Isolate from water sample	Gram morphology	Colony morphology	Urease	Catalas	Oxidase	Coagulase	Voges proskauer	Methyl red	Citrace	Indole	Glucose	Lactose	manitole	Endospore stain	Tentative organism
Okpara coal mine A	-short rod	Colourless colonies	_	_	_	_	_	+	+	_	+	_	+	_	*Salmollena* sp
Okpara coal mine B	Short rod	Non-mucoid red colonies	_	_	_	_	_	+	_	+	+	+	_	_	*Escherichia* sp
UNTH A	Short rod	Round pink colonies	_	_	_	_	+	_	+	_	+	+	_	_	*Enterobacter* sp
UNTH B	+cocci in pair	Round colourless colonies	_	+	_	_	_	+	_	_	+	_	_	+	*Micrococcus* sp
UNTH C	short rod	Non mucciod red colonies	_	_	_	_	_	+	_	+	+	+	_	_	*Escherichia* sp
Ogbete market A	short rod	Non-mucciod red colonies	_	_	_	_	_	+	_	+	+	+	_	_	*Escherichia* sp
Ogbete market B	+cocci in pair	Round colourless colonies	_	+	_	_	_	+	_	_	+	_	_	_	*Micrococcus* sp
Ogbete market C	short rod	Round pink colonies	_	_	_	_	+	_	+	_	+	+	_	_	*Enterobacter* sp
Ogui A	short rod	Grey colourless	+	_	_	_	_	+	+	_	+	_	_	_	*Proteus* sp
Ogui B	short rod	Round pink colourless	_	_	_	_	+	_	+	_	+	+	_	_	*Enterobacter* sp
Ogui C	short rod	Non-mucoid red colonies	_	_	_	_	_	+	_	+	+	+	_	_	*Escherichia* sp
Ogui D	short rod	Colourless colonies	_	_	_	_	_	+	+	_	+	_	+	_	*Salmorella* sp

4.10.2 Monthly variations in microbial count

The mean monthly variations in the microbial count measured as colony forming unit per millilitre (cfu/ml) are presented in Table 16. At Okpara Coal Mine, the microbes ranged from 1.1 to l1.45 cfu/ml with the highest value obtained in August and the value in November; the mean value was $1.273 \times 10^4 .\pm 0.183$ At UNTH, cfu/ml value ranged from 1.1 to 9.95l with the maximum and minimum values obtained in the months of September and October respectively; the mean value was $6.8 \times 10^4 \pm 1.276$ cfu/ml. The cfu/ml value of Ogbete ranged from 1.1 to 9.95 with the maximum and minimum value obtained in the months of November and October respectively; the mean value was $3.37625 \times 10^4 \pm 1.451$ cfu/ml. At Ogui the cfu/ml value ranged from 1.335 to 1.7 with the maximum and minimum value obtained in the months of August and September respectively; the mean value was $1.505 \times 10^4 \pm 0.237$ cfu/ml.

The highest monthly microbial count was obtained at Ogui in August; at UNTH in September and October amd at Ogbete in November.The mean monthly value was highest in the month of November ($5.04 \times 10^4 .\pm 1.472$ cfu/ml), followed by September ($3.506 \times 10^4 .\pm 1.487$ cfu/ml), and October ($3.029 \times 10^4 .\pm 1.174$ cfu/ml). The least was observed in the month of August ($1.386 \times 10^4 .\pm 0.084$). (Table 18).

Table 16: Monthly variation in microbial count of Asata River

Location/Date	August	September	October	November	Mean ± SE
Coal mine	**1.45×10^4**	**1.325×10^4**	**1.19×10^4**	**1.1×10^4**	**$1.274 \times 10^4 \pm$ 0m.183**
UNTH	**1.1×10^4**	**9.95×10^4**	**8.4×10^4**	**7.75×10^4**	**$6.8 \times 10^4 \pm 1.276$**
Ogbete	**1.27×10^4**	**1.185×10^4**	**1.1×10^4**	**9.95×10^4**	**$3.376 \times 10^4 \pm 1.051$**
Ogui	**1.7×10^4**	**1.565×10^4**	**1.42×10^4**	**1.335×10^4**	**$1.505 \times 10^4 \pm 0.217$**
Mean ± SE	**1.386×10^4 ± 0.0835**	**3.506×10^4 ± 1.407**	**3.029×10^4 ± 1.174**	**5.04×10^4 ± 1.472**	

4.11 Correlation of algae and physicochemical parameter

Result of correlation analysis (P Ò 0.05) presented in Table 17 showed that there were significant positive and negative correlations between the classes of algae and physicochemical parameters. Bacillariophyta had significant negative correlations with calcium (-0.350*), lead (-0.327**) total suspended solids (-0.312**) and pH (-0.139***) but positive correlation with colour (0.388**), Chlorophyta (0.712***) and Cyanophyta (0.318**). Chlorophyta had significant negative correlations with pH (-0.265) but significant positive correlation with Cyanophyta (0.603), Euglenophyta (0.420), and Bacillariophyta (0.712). Cyanophyta had significant negative correlations with mercury (-0.297) and pH (-0.274), but it had significant positive correlations with magnesium (0.329), Bacillariophyta (0.361), Chlorophyta (0.603), and Euglenophyta (0.420). Euglenophyta had significant negative correlations with total dissolved solids (-0.313), pH (-0.306) and total solids (-0.332). There were also significant positive and negative correlations among the physicochemical parameters investigated.

Calcium had significant negative correlation with iron (-0.380**), microbes (-0.497***) and zinc (-0.474***) but it had significant positive correlation with lead (0.530***), velocity (0.327**), total solids (0.272**) and pH (0.324**). Chemical oxygen demand had significant negative correlation with zinc (-0.258**) and pH (-0.371**) .Colour had significant positive correlation with iron (0.281**) but significant negative correlation with depth (-0.298**), nitrate (-0.314**) and transparency (-0.298**). Depth had significant negative correlation with lead (-0.309**). Dissolved oxygen had significant negative correlation with magnesium (-0.452***) but positive correlation with zinc

(0.449***). Lead had significant negative correlation with the microbes (-0.643***). Magnesium had significant negative correlation with transparency (-0.118*). Mercury had significant positive correlation with total dissolved solids (0.309**) and total solids (0.286**). Total dissolved solids had significant positive correlation with total solids (0.820***). Transparency had significant negative correlation with total suspended solids (-0.416***). Total solids had significant negative correlation with pH (-0.034**). Zinc had significant positive correlation with pH (0.094*).

4.12 ANOVA RESULT FOR INTER – LOCATION AND MONTH ANALYSES

Test for significance for results of all the analyses were at P Ò 0.05.ANOVA (Table 18) showed that there were significant differences among some physicochemical parameters, such as depth, transparency, alkalinity, dissolved oxygen, lead, total suspended solids and pH.

From the ANOVA (Table 19) showed that there were significant differences among the values of some physico ï chemical parameters at different sampling months. These include air temperature, velocity, alkalinity, aluminium,colour, BOD, COD, calcium, nitrate, pH, TSS, iron, lead and zinc.

Table 18: Table of analysis of Variance of physicochemical parameters of Asata River at different locations showing sources of variation, sum of square, mean square and F- ratio ($P \leq 0.05$).

Variate	Sum of square	df	Mean square	F	Sig
Air Temperature (^oC)	59.652	3	19.854	3.21	Ns
Water Temperature (^oC)	17.053	3	5.684	1.97	Ns
Colour (Hazen)	1127.1	3	375.7	1.36	Ns
Depth (cm)	1429.06	3	476.36	5.86	Ns
Transparency (cm)	1296.98	3	432.33	6.65	**
Velocity (m/s)	0.31752	3	0.105884	2.56	Ns
Alkalinity (mg/l)	5193.7	3	1731.2	8.93	***
Aluminium (mg/l)	0.00007006	3	0.00002336	1.20	Ns
Ammonia (mg/l)	0	3	0	_	_
DO (mg/l)	11.7456	3	3.9152	5.96	**
BOD (mg/l)	0.000020521	3	0.0006840	0.79	Ns
COD (mg/l)	0.007973	3	0.002658	1.36	Ns
Calcium (mg/l)	0.00106	3	0.00036	0.01	Ns
Magnesium (mg/l)	6.650E+07	3	2.217E+07	1.00	Ns
Nitrate (mg/l)	0.0029977	3	0.0009992	1-00	Ns
pH (mg/l)	1.80229	3	0.6000076	6.82	**
Phosphate (mg/l)	8.333E-06	3	2.77778E-06	0.65	
Silica (mg/l)	9.244E-33	3	3.0815E-033	_	_
Hydrogen Sulphide (mg/l)	0	3	6	_	_
Iron (mg/l)	0.21687	3	0.07229	2.44	Ns
Lead (mg/l)	0.00001386	3	0.00000462	0.10	Ns
Mercury (mg/l)	2.297E-08	3	7.658E-09	0.42	Ns
T.S (mg/l)	14967	3	4989	1.36	Ns
T.S.S (mg/l)	0.011814	3	0.003938	2.83	*
T.D.S (mg/l)	5.571E+09	3	1.857E+09	1.00	Ns
Zinc	0.0142	3	0.000491	0.20	Ns

. * = significant ** = Highly significant *** = Very highly significant Ns = Not significant

Table 19: Table of Analysis of Variance of physicochemical parameters of Asata River at different sampling months showing sources of variation, sum of square, mean square and F- ratio ($P \leq 0.05$).

Sourece of variation	Sum of square	Df	Mean square	F	Sig
Air Temperature (^oC)	299.729	11	27.248	4.40	***
Water Temperature (^oC)	31.347	11	6.269	2.17	Ns
Colour (Hazen)	10822.9	11	983.9	3.37	***
Depth (cm)	1326.84	11	120.80	1.48	Ns
Transparency (cm)	1177.23	11	107.02	1.65	Ns
Velocity (m/s)	490052	11	0.44550	10.58	***
Alkalinity (mg/l)	21025.1	11	1911.4	9.85	***
Aluminium (mg/l)	0.00082523	11	0.00007502	3.84	***
Ammonia (mg/l)	0	11	0	_	_
DO (mg/l)	133.4568	11	12.1324	18.46	***
BOD (mg/l)	0.0276492	11	0.0025136	2.92	***
COD (mg/l)	0.164423	11	0.014948	7.63	***
Calcium (mg/l)	9.66229	11	0.87839	12.83	***
Magnesium (mg/l)	2.439E+08	11	2.217E+07	1.00	Ns
Nitrate (mg/l)	0.0038596	11	0.0003509	0.66	Ns
pH (mg/l)	6.14052	11	0.55824	6.34	***
Phosphate (mg/l)	4167E-06	11	3.728E-06	0.88	Ns
Silica (mg/l)	0.0000E+00	11	0.0000E+00	_	_
Hydrogen Sulphide (mg/l)	0	11	0	_	_
Iron (mg/l)	2.92167	11	0.26561	8.96	***
Lead (mg/l)	0.00888190	11	0.00080745	18.17	***
Mercury (mg/l)	2.740E-07	11	2.419E-08	1.37	Ns
T.S (mg/l)	73967	11	6724	1.84	Ns
T.S.S (mg/l)	0.157184	11	0.014289	10.36	***
T.D.S (mg/l)	9.260E+09	11	1.852E+09	1.00	Ns
Zinc	0.089801	11	0.008164	3.26	***

* = significant ** = Highly significant *** = Very highly significant Ns = Not significant

4.12.1 Comparism of water quality assessment at the various locations sampled.

4.12.1.1 Mean values of some physicochemical parameters at different locations on Asata River

ANOVA test showed that there were significant differences among some physicochemical parameters, such as depth, transparency, alkalinity, dissolved oxygen, lead, total suspended solids and pH.

Table 20: Mean values of some physicochemical parameters at different locations of Asata River

Parameter	Coal mine	UNTH	Ogbetes	Ogui	F-LSD (0.05)
Air Temperature (^{o}C)	30.33	30.83	27.33	27.92	NS
Water Temperature (^{o}C)	27.9	28.45	28.22	27.97	NS
Colour (Hazen)	60.0	64.2	72.9	68.8	NS
Depth (cm)	25.6	14.4	13.1	23.3	7.49
Transparency (cm)	28.8	14.4	13.1	23.3	6.70
Velocity (m/s)	0.554	0.666	0.436	0.550	NS
Alkalinity (mg/l)	52.8	52.4	67..6	77.0	11.57
Aluminium (mg/l)	0.03508	0.03192	0.03382	0.03287	NS
Ammonia (mg/l)	28.0	28.05	28.0	28.0	NS
DO (mg/l)	3.20	3.76	2.45	2.73	0.673
BOD (mg/l)	0,6090	0.6225	0.6262	0.6258	NS
COD (mg/l)	1.6567	1.6425	1.6202	1.6258	NS
Calcium (mg/l)	13.138	13.131	13.127	13.127	NS
Magnesium (mg/l)	33	32.	33	27.51	NS
Nitrate (mg/l)	0.2648	0.2457	0.2475	0.2467	NS
pH (mg/l)	6.600	7.05	6.942	6.633	0.2424
Phosphate (mg/l)	2.68083	2.6808	2.68000	2.68000	NS
Silica (mg/l)	0.5	0.05	0.05	0.05	NS
Hydrogen Sulphide (mg/l)	2.0	2.0	2.0	2.0	NS
Iron (mg/l)	1.084	0.998	0.70	1.187	NS
Lead (mg/l)	0.0387	0.0376	0.0379	0.0373	1.826
Mercury (mg/l)	0.0000306	0.000292	0.0000350	0.0000325	NS
T.S (mg/l)	233.3	225.0	246.7	271.7	NS
5T.S.S (mg/l)	0.695.4	0.712.7	0.6779	0.6729	0.3086
T.D.S (mg/l)	211.1	222.4	249.0	251.66	NS

NS = Not significant

4.12.1.2 Mean values of the physicochemical parameters at different sampling months

ANOVA test showed that there were significant differences among the values of some physico ï chemical parameters at different sampling months. These include air temperature, velocity, alkalinity, aluminium, BOD, COD, calcium, nitrate, pH, TSS, iron, lead and zinc.

Table 21: Mean values of the physicochemical parameters at different sampling months

Parameter	June	July	August	September	October	November	$FLSD_{(0.05)}$	WHO Standard
Air Temperature (^oC)	30.25	29	26.25	30.25	30.62	30.50	3.579	NA
Water temperature (^oC)	28.0	26.73	27.71	27.86	28.23	29.46	NS	NA
Colour (Hazen)	73.75	70	70	55	56.25	73.75	NS	NA
Depth (cm)	26.34	12.77	14.65	21.12	23.19	16.45	NS	NA
Transparency (cm)	25.01	12.77	14.65	21.12	23.19	16.45	NS	NA
Velocity (m/s)	0.537	0.834	0.374	0.389	0.878	0.346	0.2915	NA
Alkalinity (mg/l*)	98.25	84.50	55.62	51.75	57	36.50	20.04	100
Aluminium (mg/l)	0.0285	0.0309	0.0403	0.0350	0.0344	0.0314	20.04	NA
Ammonia (mg/l)	28	28	28	28	28	28	NS	NA
DO (mg/l)	1.775	2.544	3.119	4.006	5.315	1.456	1.166	5 ï 8.5
BOD (mg/l)*	0.595	0.608	0.601	0.633	0.643	0.631	0.04222	2 ï 4
COD (mg/l)	1.686	1.6	1.63	1.639	1.615	1.664	0.06368	NA
Calcium (mg/l)*	13.72	13.31	13.32	12.87	12.62	12.75	0.3794	100
Magnesium (mg/l)	32.01	33.32	41.10	32.62	32.73	33.62	NS	NA
Nitrate (mg/l)*	0.244	0.245	0.257	0.251	0.258	0.244	NS	50
pH (mg/l)	6.95	6.95	7.12	6.913	6.688	6.225	0.4269	5 ï 8.5
Phosphate (mg/l)*	2.68	2.68	2.681	2.681	2.68	2.68	NS	0.5
Silica (mg/l)	0.05	0.05	0.05	0.05	0.05	0.05	NS	NA
Hydrogen Sulphide (mg/l)	2.0	2.0	2.0	2.0	2.0	2 .0	NS	NA
T.S (mg/l)	290	225	250	250	225	225	NS	NA
T.S.S (mg/l)	0.633	0.736	0.792	0.655	0.653	0.639	0.05345	NA
T.D.S (mg/l)*	273.6	216.5	246.1	375.04	225.2	217.2	NS	1000
Iron (mg/l)*	0.730	1.292	1.297	1.024	0.985	1.244	0.2472	0.0
Lead (mg/l)*	0.0494	0.0546	0.0493	0.0251	0.0235	0.0244	0.03591	0.01
Mercury (mg/l)*	0.000362	0.0003	0.000312	0.000362	0.000283	0.000309	NS	0.006
Zinc (mg/l)	0.320	0.374	0.417	0.422	0.436	0.396	0.07199	NA

NS = Not significant NA = Not available * = Beyond WHO (2006) Standard

4.13 Comparism of water quality of Asata River with World Health Organization (WHO) standard

A comparison of observed water quality parameters for Asata River with World Health Organization standard (WHO, 2006) standards for drinking water for developing countries water is presented in Table 22.

Alkalinity, calcium, magnesium, BOD, DO and TDS were below WHO standard; phosphate, iron and lead were beyond WHO standard while nitrate and pH within WHO standard.

Table 22: Comparison of mean values of physicochemical parameters with WHO Standards

Parameters	Min – Max	Mean ± SE	WHO (2006)
Air Temperature (^{o}C)	26.25 ï 30.62	29.60 ± 2.487	NA
Water temperature	28.73 ï 29.46	28.18 ± 0.965	NA
Colour (Hazen)	55 ï 75.75	68.6 ± 16.60	NA
Depth (cm)	12.77 ï 26.34	19.1 ± 9.02	NA
Transparency (cm)	12.77 ï 25.01	18.9 ± 8.06	NA
Velocity (m/s)	0.346 ï 0.834	237.6 ± 55.26	NA
Alkalinity (mg/l)**	36.5 ï 89.25	62.4 ± 2.398	100
Aluminium (mg/l)	0.0285 ï 0.0403	0.03340 ± 0.004418	NA
Ammonia (mg/l)	28 ï 28	28 ± 0	NA
DO (mg/l)**	1.456 ï 5.314	3.04 ± 0.811	5 ï 9.5
BOD (mg/l)**	0.595 ï 0.643	0.6185 ± 0.02935	2 ï 4
COD (mg/l)	1.6 ï 1.686	1.6390 ± 0. 04428	NA
Calcium (mg/l)**	12.62 ï 13.72	13.13 ± 0.2637	100
Magnesium (mg/l)**	32.01 ï 41.10	32.46 ± 1.485	150
Nitrate (mg/l)	0.242 ï 0.258	0.2012 ± 0.02310	50
pH (mg/l)	6.225 ï 7.12	6.806 ± 0.2967	6.5 ï 9.5
Phosphate (mg/l)*	2.68 ï 2.681	2.68 ±0.00207	0.5
Silica (mg/l)	0.05 ï 0.05	0.05 ± 0	NA
Hydrogen Sulphide (mg/l)	2 ï 2	2 ± 0	NA
T.S (mg/l)	225 ï 290	244.2 ± 60.46	NA
T.S.S (mg/l)	0.633 ï 0.792	0.6898 ±0.03718	NA
T.D.S (mg/l)**	216.5 ï 375.04	273.6 ± 55.26	1000
Iron (mg/l)*	0.730 - 1.292	1.096 ± 0.1509	0.3
Lead (mg/l)*	0.0235 ï 0.0546	0.03879 ± 0.035	0.01
Mercury (mg/l)	0.000283 ï 0.000362	0.000318 ± 0.0001346	0.006
Zinc (mg/l)	0.320 - 0.436	0.3944 ±0.05004	NA

NA = Not Available *** = Beyond WHO Standards**

**** = Below acceptable WHO standards**

CHAPTER FIVE

DISCUSSION

It is obvious from this study that the algal flora of Asata River is rich in species. This according to Kadiri (2000) can be attributed to the different environments or habitats created by the river and its users which influenced some factors like nutrient (Mustapha, 2008), hydrology and physicochemical parameter (Kadiri 2000, Jafari and Gunale 2006, Nweze and Chumbo 2006). The cosmopolitan distribution of some taxa show that they can survive in varied conditions both chemical and physical. Patterns of abundance in algal biomass have been needed to provide a fundamental tool in understanding the driving forces in ecosystem productivity in relation to changing environmental function (Steve *et al*., 2007).

At Okpara Coal mine, Bacillariophyta had the highest taxa with some species such as *Navicula*, *Nitzschia*, *Gomphonema*, *Synedra* and *Fragillaria* species observed which was in line with the findings of Jafari and Gunale (2006) at Mutha River Pane city South West of India. They noted that these taxa are among the algae used as indicator of water pollution. Moreover these were also recorded by Zakariya *et al.,* (2011), on lower Niger River in Kogi State and Opute (1990) on Warri River. The abundance of Bacillariophyta over Chlorophyta (*Ankistrodesmus, Scenedesmus and Oedogonium*) is also in line with the findings of Zakariya *et al*., (2011).These algae according to Jaferi and Gunale (2006) are also classified as pollution tolerant algae. The population of Chlorophyta was followed by Cyanophyta and Euglenophyta in decreasing order. The dominance of diatom at Okpara Coal mine can be attributed to mixing of the water body as observed by Kadiri, 2006 whom opined that diatom associations develop in well mixed water. At UNTH Old Site the abundance in decreasing order was as

follows: Chlorophyta > Bacillariophyta > Cyanophyta > Euglenophyta. The highest population of Chlorophyta can be attributed to high content of magnesium, phosphate, nitrate and sulphate. Also washing of cow dungs and bathing with phosphate based detergent and soap on the river could have caused the high concentration of these ions (Mustapha, 2008). This favoured the Chlorophytes

At Ogbete market the population in decreasing order was Chlorophyta > Euglenophyta > Bacillariophyta > Cyanophyta. This could be as a result of imput of waste from the market into the river. At Ogui the population of algae in decreasing order was Chlorophyta > Euglenophyta > Cyanophyta > Bacillariophyta. Chlorophyta still dominated at this point though its population at Ogbete marrdt was higher.

From the monthly variations of algae observed during the study period, the total algal population was greater during the early dry season. Reduced population of algae in the wet season could be due to increased cloud cover which reduced insolation and consequently photosynthesis and increased senescence of algae (Nweze and Domrufus, 2006). Moreover high turbidity from runoff may have reduced photosynthesis (Ezra and Nwankwo 2000, Nweze 2003). Heavy rainfall as reported by USEPA (2002), can disrupt or disintegrate floating mats and increase the habitat space for zooplankton which graze on the phytoplanktonic algae (USEPA, 1999).

Diatoms have been reported to be the dominant algae in rivers as reported in this study (Zakariya *et al*., 2011). The percentage of algae encountered during the study period showed that Bacillariophyta had 33 %, Chlorophyta 32 %, Euglenophyta 21 % and Cyanophyta 14 %. The quantitative and qualitative dominance of diatom in Asata River is in line with the report of Opute (1990) on Warri River, Delta State Nigeria who opined that the species of diatoms such as *Surirella, Fragillaria* and many others

as fresh water forms. It is also in agreement with the findings of Zakariya *et al.,* (2011) on lower Niger River, Kogi State and Tanimu *et al.,* (2012) on wet land of Hadejia, Nguru all in Nigeria. Chlorophyta was next in abundance and the least represented division was Cyanophyta. The observed difference in biomass between stations could be explained on the basis of changes in the natural population which could be due to different magnitude of causative factors such as pollution level, wash out, sinking, consumption by animals and parasites (Kadiri, 2006).

Significant positive and negative correlations between algal population and physicochemical parameters were recorded in all of the locations suggesting that some of the chemicals are nutrients for the algae (Mustapha, 2008). Bacillariophyta had significant positive correlation with colour, Chlorophyta, Euglenophyta and Cyanophyta but negative correlation with calcium, lead and pH. The significant negative correlation of calcium and lead ion with Bacillariophyta indicates that the algae take up these ions. This is in line with the findings of Sumita *et al.,* (2010) on River Gomti and Varuna, India where they noted that higher concentration of calcium favored the growth of diatoms. The significant negative correlation between the algal classes and nutrients showed that algae utilize the nutrient (Nweze and Domrufus, 2006), perhaps more than the nutrients supplied. The significant negative correlation of all the groups of algae and pH may be as a result of anthropogenic impact.

The result showed that the quality of water varied among the months. Temperature fluctuations both diurnal and seasonal are more evident in fresh water habitat although flowing water lack wide fluctuation in temperature (Elloit and Linday, 2008). In this study there were fluctuation in both water and air temperatures. There was decline in the air temperature with relatively higher values in the dry months. This is in line with the findings of Onyema (2007) on Estuarine Creek in Lagos, Nigeria. He attributed

this to increased cloud cover and consequent reduction in solar radiation. The surface water temperature range (27.9^{o}C and 28.45^{o}C) was similar and compared well with the ranges reported for other African water bodies (Kadiri 2006; Chima *et a*l., 2009). The significant difference seen between the seasons might be as a result of meteorological conditions such as trade winds, sunshine duration and absorption of the solar radiation by the shallow water body (Mustapha, 2008).

Air temperatures were generally higher than the water temperature in all the locations sampled except at Ogbete market where water temperature was higher than the air temperature. Heat generated by the decomposition of waste from the market and milling activities may have contributed to increase in water temperature. Water temperature increased during the warmer month and decreased during the colder months. This is in line with the findings of Garg *et al.,* (2009) at Ramsagar Reservoir, Deltia, Madhya Pradesh.

The colour of Asata River was sometimes cloudy and sometime clear. The significant variation in the mean colour between months showed that run ï offs into the river in the rainy months brought in debris, sediments or silt which increased the colour value (USEPA, 2002; Nweze and Chumboh, 2006). There was no significant variation in the mean colour between locations. This might be as a result of uniform soil properties along the investigated stretch of the river. Colour had negative correlation with dissolved oxygen, nitrate, transparency and depth but positive correlation with iron. This might be as a result of high level of biological activities.

The depth of water decreased progressively during the dry months in all the locations sampled possibly as a result of decreased rainfall, increased temperature, increased relative humidity and high evaporation (Nweze and Chumboh, 2006), which were

characteristic of dry season. The depth value of 12.77 - 26.34 cm recorded in this study might be as a result of low input of water from the catchment area which according to Nweze and Chumboh (2006) may result in low water volume. Depth of water showed significant negative correlation with lead and total suspended solids.

Garg *et al.,* (2009) noted that transparency of water is mainly due to biological productivity, suspended particle and water colour. The value of transparency 12.77 ï 25.01 cm recorded in the study might be as a result of low depth of the water corroborating the findings of Davies (2009), at Okpoka Creek Niger Delta, Nigeria. Transparency had a significant positive correlation with depth but significant negative correlation with total suspended solids.

The significant variation in the mean rate of flow between the sampling months can be attributed to the fact that many rivers have a relatively low rate of flow at certain times of the year. The biological implication of rate of flow is that high rate of flow can wash away some algae biomass, thereby disrupting their growth and subsequent removal from the system (Bellinger and Sigee, 2010).

The fluctuation in the surface pH indicates the buffering capacity of the system. The pH range of 6.23 ï 7.12 and mean value of 6.8 recorded during the study period indicates that the water is slightly acidic. This was also observed on Oyun Reservoir Offa and was associated with high carbon dioxide concentration occurring from organic decomposition (Mustapha, 2008). In Oyun Reservior, pH was found to be high during the rainy season. Also Zakariya *et al.,* (2011) noted that significant difference in the monthly mean pH might be due to the influence of fresh water influx, dilution by rain, low temperature and organic matter decomposition. Significant differences observed at different sampling points might be as a result of

anthropogenic impact at the different sampling points. The mean pH 6.8 recorded in this study is within WHO permissible standard of 6.5 ï 9.5.

Dissolved oxygen level recorded in this study (1.76 ï 5.32 mg/l) with a mean value of 3.04 ± 0.811 mg/l is comparable with 0.5 - 1.6 mg/l recorded for Asa River, Kwara state, by Eletta and Adekola (2005) and 3.4 ï 7.0 mg/l recorded for Calabar River by Akpan and Offem (1993). The observed values are low compared to 5.9 ï 16.9 mg/l recorded by Mohammed and Saminu (2012), on Solanta River, Kano, Nigeria. The dissolved oxygen value of 1.76 ï 5.32 mg/l recorded in this study is below WHO permissible limit. Statistical difference for mean dissolved oxygen was observed between locations and seasons. In this study, dissolved oxygen was high during the rainy season and this agreed with the findings of Mustapha (2008) on Oyun reservoir and Onyema (2007), on Estuarine Creek in Lagos, Nigeria. High temperature coupled with high rate of decomposition in the dry season may explain the low dissolved oxygen recorded in the dry season (Kadiri, 2000; Mustapha, 2008; Garg *et al*., 2008).

BOD values for both seasons are similar in all the locations and ranged between 0.595 ï 0.643 mg/l suggesting a uniform condition associated with the hydrodynamics of the system and possible mixing effect and perhaps the ability of the system to considerably cleanse itself due to tidal flushing nature of the system despite the difference in the quality and magnitude of discharge received. at the different locations (Ogamba *et al*., 2005). In this study, BOD was found to be high during the dry season. This was also observed by Ogamba *et al.,* (2005) at Elechi Creek Niger Delta, Nigeria but contradicts the work of Onyema (2007) at Estuarine Creek in Lagos, Nigeria. There was no statistical difference between mean values of BOD between locations but there was statistical difference between the mean values of the

different sampling months. BOD value 0.6185 ± 0.02935 mg/l recorded in this study is below WHO permissible limit of drinking water standard.

Total alkalinity values (35.5 ï 89.25 mg/) were in agreement with 22 - 47 mg/l recorded by Kadiri (2006) in some waters at the Eastern Niger Delta area of Nigeria. The statistical difference in mean alkalinity between the wet and dry months could be attributed to the erosion of the soil with bicarbonate, carbonates, or hydroxide compounds during the wet season into the river (APHA, 2005). Low alkalinity recorded in this study during the dry season is in line with the results of Offem *et al.,* (2011), who reported that alkalinity of Ikwori Lake, South ï Eastern Nigeria was low during the dry season. The mean alkalinity value 62.4 ± 2.398 mg/l recorded in this study was below WHO standard of 100 mg/l.

The low silica content (0.05 mg/l) observed in Asata River contradicts the result of Mustapha (2008), on Oyun Reservoir (10 ï 60 mg/l) and Nweze (2006) on Ogeleube Lake, Opi, Enugu State, Nigeria (5.18 ï 14.33 mg/l) The value of phosphate 2.68 mg/l recorded in this study was higher than 0.1113 ï 0.1313 mg/l recorded by Nweze (2006) at Nike Lake Enugu, Nigeria and the 2.2 mg/l recorded by Mustapha (2008) at Oyun Reservoir. High concentration of phosphate ion might be due to bathing and washing with phosphate based detergent and soap (Mustapha, 2008). The mean value of phosphate 2.68 ± 0.00207 mg/l recorded in this study is beyond WHO drinking water standard of 0.5 mg/l. The values of nitrate 0.242 ï 0.257 mg/l recorded in this study was within the range 0.10 -0.50 mg/l recorded by Kadiri (2006), on some waters in the Eastern Niger Delta area of Nigeria. It contradicts the value (6.9 mg/l) recorded by Mustaphta (2008), at Oyun Reservoir. According to Kadiri (2006), low nitrate level observed is a typical of most African waters. There was no significant difference

between the value of nitrate observed both within the locations and seasons. The mean value of nitrate 0.2012 ± 0.02310 mg/l recorded in this study is within WHO permissible limit of 50 mg/l.

In this study, TDS and TSS were highest in the month of September which is the peak of rainy season, suggesting that flood from the rain dissolves solids from land surrounding the river to increase TDS values during rainy months as noted by Mustapha (2008) and Akubuko and Duru (2011). This is contrary to the findings of Onyema (2007), on an estuarine creek in Lagos, Nigeria, where he recorded high TDS and TSS values during the dry months. It was suggested that high TDS and TSS values during the dry months might be due to intense pressure from pollution such as untreated sewage, sawdust, petrochemical materials, detergent and industrial effluents. There was no significant difference in the mean TDS values in both locations and the different sampling months. The mean value of TDS 273.6 ± 55.26 mg/l recorded in this study was below WHO drinking water standard limit of 1000 mg/l.

Calcium level (12.62 ï 13.72 mg/l) recorded in this study is within the range (5.5 ï 14.8 mg/l) recorded by Kadiri (2000) on Ikpoba Reservoir and Singh *et al.,* (2010) on Manipur River, India (6.01 ï 17.63 mg/l) Maximum mean value recorded in June, which is a wet month is in line with the findings of Imoobe and Oboh (2003) on River Jamieson, Niger Delta, Nigeria and Joshi *et al* (2009) on Ganga River but contradicts the findings of Singh *et al.,* (2010), who reported minimum calcium level during the wet season. Imoobe and Oboh (2003) attributed calcium peak during wet season to influx of flood water that brings in calcareous materials. There was significant difference in the mean values of calcium recorded in the different sampling months

but no significant difference between the mean values of calcium recorded at different locations sampled. The mean value of calcium 13.13 ± 2637 mg/l recorded in this study is below WHO (2006) drinking water standard.

Magnesium value 32.01 ï 40.10 mg/l recorded in this study is higher than 10 ï 22 mg/l recorded by Mustapha (2008), on Oyun Reservoir, 1.17 ï 5.50 mg/l recorded by Garg *et a*l., (2009) on Ramsagar Reservoir, Datia, India. High value of magnesium ion might be due to dilution from heavy rain, weathering from rock and runoff from surrounding water shed (Mustapha, 2008). The mean value of magnesium 32.46 ± 1.485 mg/l recorded in this study is below WHO acceptable drinking water standard.

The lead in Asata River ranged from 0.0244 ï 0.0546 mg/l, which is above the range 0.0557 ï 0.0618 mg/l recorded by Nweze and Domrufus (2006) on Nike Lake Enugu, Nigeria and in contrast with undetected levels by Akubuko and Duru (2011) on Otamiri River, Imo state, Nigeria. The presence of lead in water is associated with change in water chemistry (e.g reduced pH or ionic composition) which can cause sediment lead to become mobilized and potentially bioavailable to aquatic organism (Weber, 1993). The primary form of lead in fresh water at low pH (Ò 6.5 mg/l) is predominantly Pb^{2+} and less abundant inorganic forms include Pb ($HCO_{)}$, PbCl, $Pb(SO_4)$ and $Pb_2(OH)_2CO_3$ (UNEP, 2010). Conversely, at low pH, lead is repelled from the adsorbent surfaces (Gao *et al*., 1993). Low pH value in Asata River might have induced the release of lead from sediments while high values could have been derived from biogenic material, Aeolian particles, erosion and fluvial particles (Ritson *et al*., 1994). The mean lead 0.03879 ± 0.035 mg/l recorded in this study was beyond the WHO (2006) standard of 0.01 mg/l.

Iron level of 0.730 - 1.292 mg/l recorded in this study was in agreement with 0.2 ï 0.8 mg/l recorded by Trivedi *et a*l., (2009) on Ganga river India, and higher than 0.004 - 0.01 mg/l recorded by Akubuko and Duru (2011) at Ottamiri Imo state, Nigeria. The mean value of iron 1.096 ± 0.1509 mg/l recorded in this study is beyond the WHO (2006) standard of drinking water of 0.3 mg/l. The high iron level might have come from runoffs from the surrounding land carrying loose earth (Nweze, 2003). The value of mercury 0.000283 ï 0.000362 mg/l recorded in this study is in contrast to the undetected levels by Akubuko and Duru (2011) on Ottamiri River Imo state, Nigeria. However this mean value 0.000318 ± 0.0001346 mg/l recorded was below WHO (2006) limits for drinking water (0.006 mg/l).

The value of zinc (0.320 ï 0.436 mg/l) recorded in this study was lower than 0.34 ï 1.52 mg/l recorded by Nweze and Domrufus (2006) on Nike Lake, Enugu, Nigeria and 10.50 ï 59.90 mg/l recorded by Fikrat *et al.,* (2010) on Euphrate River, Iraq. The concentration of zinc in water might have been influenced by factors such as the flow of the dredged material from upper region of the river, dilution and increase of water flow, direct drainage from farm lands, factories, sewage disposal plants, dissolution of sediments, chemical adsorption on sediments and complexes with organic matter (Fikrat *et a*l., 2010).

Shannon Weiner index value of less than one are interpreted as heavily polluted, Values between 1 -3 as moderately polluted and more than 3 as clean water (Whitton, 1975). Shannon Wiener index showed that the values of diversity index at Okpara coal mine and Ogui are equal while the values observed at UNTH and Ogbete were similar. An evenness index 1 (one) shows complete evenness in any community (Begon et al., 1996) and none of the locations recorded 1, so there was no complete evenness in the river. The locations with high evenness values were UNTH (0.9378)

and Ogbete (0.9305) showing that species were more distributed there more than the rest of the locations. Okpara coal mine was least in both Shannon Wiener index and evenness indices of 0.834 and 0.5696 respectively, meaning that the location had fewer species number and distribution, Based on this classification, the water at Okpara Coal Mine and Ogui can be classified as eutrophic (heavily polluted),while that of UNTH and Ogbete can be classified as mesotrophic (moderately polluted).

The microbial analysis of Asata river indicated the presence of *Salmonella* sp., *Escherichia* sp., *Enterobacter* sp., *Microccocus* sp., and *Proteus* sp. Nweze (2006) stated that these organisms are known human pathogens. They are the causative organisms of some diseases like diarrhoea (*E.coli),* typhoid fever (*Salmonella* sp.), and Gastroenterities (*Salmonella* sp.), urinary tract infection (*Proteus* sp.), and opportunistic urinary tract infection (*Enterobacter* sp.). Bacteria count of water sample of Asata River revealed a substantial number of aerobic heterotrophic bacteria.

Entero bacteria Gram ï negative shot rod were isolated. Among the Gram ï negative isolates are *Enterobacter* sp. and *Escherichia coli. Escherichia coli* was found to be dominant among the Gram ï negative bacteria indicating the presence of fecal pollution of the river. Saha *et al* (2009) stated that the coliform group of bacteria in general and *E. coli* in particular are universal indicators for faecal contamination. The load of aerobic heterotrophic bacteria, the presence and abundance of *Enterobacter, Salmonella, Escherichia and Microccocus*. in the water clearly indicate significant level of pollution in the river.

CONCLUSION

The physicochemical characteristics of water quality analysed during the period of study from the four locations along Asata River; Okpara Coal Mine, UNTH (old site), Ogbete market and Ogui revealed that due to anthropogenic activities the water quality had deteriorated. The mean values of iron and lead were beyond the permissible limit as indicated by World Health Organization (WHO, 2006) standard; other parameters were within WHO (2006) standard.

The high diatom density showed that the river was not healthy and its nutrient content was very high. The recorded physicochemical parameters favoured diatom growth. Some of the observed genera: *Gomphonema, Nitzschia, Navicula* sp, *Surirrella, Euglena* and *Oscillatoria* indicated organic pollution. Moreover the load of aerobic heterotrophic bacteria, the presence and abundance of *Enterobacter, Salmonella, Escherichia and Micrococus*, in the water clearly indicated significant level of pollution in the river. Also the values of diversity index and evenness showed that there was no evenness in the river. It is therefore the opinion of the author that the Asata River should be classified as eutrophic (highly polluted). Discharge of untreated domestic and industrial wastes into the river should be discouraged.

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