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PHYSICOCHEMICAL CHARACTERISTICS AND GREEN MICROALGAE COMPOSITION OF SELECTED RIVERS IN OGUN STATE, NIGERIA

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ABSTRACT

The river system is the most complex of the freshwater bodies in the world and is prone to pollution especially from anthropogenic activities and the quality of water, as partially determined by physicochemical properties, is very crucial for primary productivity in the aquatic environment. This study determined the physicochemical characteristics and green microalgae of water collected from Majopa, Ogunpa and Uren rivers in Ogun State, Nigeria. Surface water samples were collected from three different rivers using direct collection method into the labelled bottles for physicochemical characteristics determination, culturing and ecological studies. Water samples for ecological studies were fixed in 4% unbuffered formalin while sample for culturing were not fixed but growth stimulated using Bold Basal Medium (BBM) and Blue- Green Medium (BG-11). The physicochemical parameters varied widely with the profile of water sample from Ogunpa River being the most favourable for widest diversity of green microalgae of all three. The pH of the water samples were slightly acidic ranging between 6.3 and 6.5 for the rivers, while the lower dissolved oxygen (2.61-3.01 mg/L) recorded pointed to a stressed environment which probably accounted for lower number of microalgae taxa observed, especially in Majopa River. The ecological studies showed the presence of ten taxa from the division Chlorophyta with nine taxa from Ogunpa River and *Scenedesmus* sp. being dominant genus across the rivers. The microalgae diversity of the river water was Ogunpa River>Uren River>Majopa River. BBM supported growth of the microalgae much better. The physicochemical properties portrayed Majopa and Ogunpa River water as unfit for drinking, while that of Uren River is relatively fit for drinking. It is highly recommended that the microalgae reported therein be further characterised for various possible economic benefits.

Keywords: Growth, Microalgae, Nutrient, Ogun State, River, WaterDepth, Geothermal Energy.

INTRODUCTION

In September 2015, the 2030 Agenda for sustainable development goals (SDGs) was voted by the United Nations, to ultimately transform our world and make it more habitable now and in the future. Goal six, specifically is for people to have access to clean water and good sanitation, while goal fourteen is for life below water. The achievement of these goals would require continuous monitoring of water bodies that consumers depend on for drinking and other uses. According to Dimowo (2013), the sources of freshwater for human consumption and other domestic purposes include the rivers, lakes, streams, wells, and springs and the river system is the most complex and one of the oldest of the freshwater bodies in the world (Higler, 2012). Rivers are prone to pollution due to the huge anthropogenic activities which are suspected to be related to a large number of epidemic and chronic diseases suffered by humans (Gbola *et al*., 2017; Titilawo *et al*., 2019) and the quality of water can be determined by its physical, chemical and microbiological properties (Oluseyi *et al*., 2011). The physicochemical parameters including pH, turbidity, dissolved salts, and hardness can affect the biological activities of the river and the presence of nutrients such as nitrates, sulphates and phosphates are equally crucial for primary productivity in the aquatic environment (Annalakshmi and Amsath, 2012).

Microalgae are minute photosynthetic organisms that are found mostly in aquatic systems such as marine, fresh or brackish waters as well as in the soil and air (Ratha and Prasanna, 2012). Aquatic microalgae (phytoplankton) can be found living suspended close to the lighted surface growing above the high tide in deep oceans (supralittorial) or within the waves (intertidal) or extreme low and clear water level of about 200 m in depth (sublittorial). Microalgae can also be found living within sediments (benthonic) which includes those growing on sand (epipelic), attached to stones (epilithic), on other algae or plants (epiphytic) or on animals (epizoic) (Perumal *et al*., 2012; Barsanti and Gualtieri, 2014).

Green microalgae are photosynthetic eukaryotes containing double membrane bounded plastids enclosing both chlorophylls *a* and *b* (Brown and Jeffrey, 1992) and accessory pigments which include beta carotene and xanthophyll (Lewis and McCourt, 2004; Kirchman, 2012). According to Chapman (2013), about 90% of the green algae are found in freshwater habitats with an estimate of 6,000 - 8,000 species. Although green microalgae and higher plants are monophyletic (Lewis and McCourt, 2004), the rate of growth of terrestrial plants is comparably slow to microalgae, and hence, an estimation of only 3-6% of flue gases can be captured by the green plants (Mondal *et al*., 2017).

Evidences indicating the presence of biological activities and chemical pollutants in Majopa River, making it unfit for drinking and other hygiene-related anthropogenic activities was reported by Alabi *et al*. (2013) and Fagbohun *et al*. (2017) stated the presence of high dissolved inorganic phosphate in the river, acceding to it as unfit for consumption. Akin-Oriola (2003) reported the zooplankton abundance in Ogunpa River, while Ajibade *et al*. (2010) did morphometric analysis on the river. Several major flooding (from 1951 to 2011) have been reported for Ogunpa River as reviewed by Adewale *et al*. (2010) and Agbola *et al*. (2012), which could have negatively affected the hygienic status of the river water. Furthermore, Ogunpa River is occasionally being used for fishing, but has various domestic and industrial activities around it, which negatively impact the water quality over the years (Iyiola, 2015). Currently, there is paucity of data on the quality of water from Uren River in Ikenne Remo. The river serves mainly as the town's drinking water and other municipal activities excluding fishing. Personal interview revealed that the town's culture prohibits fishing in that particular river due to some cultural beliefs (Unpublished data).

Several studies have previously identified and characterized microalgae in water collected from some rivers within south-western Nigeria (Adesalu and Nwankwo, 2009; Adesalu, 2015; Adesalu *et al*., 2016; Kunrunmi *et al*., 2016; Oyewumi and Olukunle 2018). However, there is paucity of data on microalgae from rivers in Ogun State. Therefore, this study was designed to assess the physicochemical characteristics and green microalgae composition of Majopa, Ogunpa and Uren Rivers in Ogun State, Nigeria.

MATERIALS AND METHODS

Study Sites

Majopa River, is a fast flowing freshwater river that exists in Ilaye area (Isale Oko) in Sagamu Local Government Area of Ogun State, south-western Nigeria, while Ogunpa River, another fast flowing freshwater, takes its source from Ashi village in the Orita Bashorun area of Ibadan (Adewale *et al*., 2010; Ajibade *et al*., 2010) and flows through Bodija in a North-South direction with multiple channels (Ogidiaka *et al*., 2012), and one of the channels flows through Irolu and Ago Iwoye and empties into the Epe Lagoon, Lagos State. Uren River exists to the north of Ikenne Remo, with an accessibility point along Odogbolu-Aiyepe road.

Sample Collection

Duplicate surface water samples were collected systematically at three different spots from each of Rivers Majopa, Ogunpa, and Uren (Figure 1). The samples from different spots were pooled into a two 2.5 L labelled sterile plastic bottles for each river with the sample for ecological analysis preserved in 4% unbuffered formalin and transported to the laboratory in an ice chest for further analysis. At the laboratory, unpreserved samples were divided into two (1.25 L) for each river and used for culturing and physicochemical analysis while the samples for ecological study were allowed to settle and decanted after 24 h.

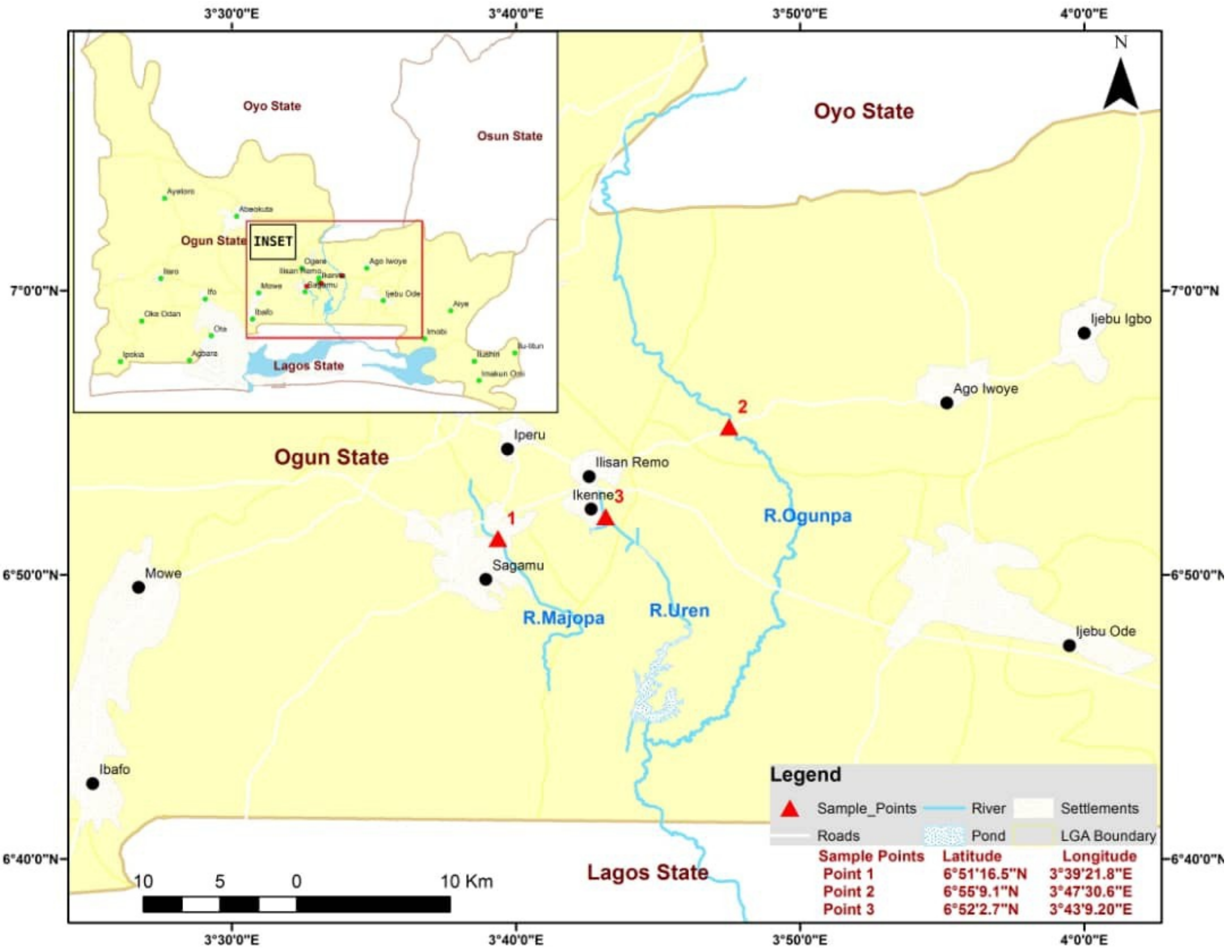

Figure 1: Map Showing the Collection Points of the Three River Water Samples

Physicochemical Characteristics of Water Samples

Some of the parameters (pH, conductivity, temperature, dissolved oxygen) were analysed *in situ* using handheld equipment. The pH was determined using pH meter and the electrical conductivity was measured using a potable combined Electrical conductivity/TDS/Temperature meter (HM Digital COM-100). Dissolved oxygen and turbidity were determined using portable dissolve oxygen meter (Orion 3) and spectrophotometric method, respectively. Total alkalinity and total acidity were measured with potentiometric end point using an automatic titrator (Hanna HI 84430-2) (Sadler and Murphy, 2010). Calcium and magnesium levels were determined using complexometric method (Rathore *et al.*, 1997), while sodium and chloride contents were determined by sodium meter (Laqua twin) and argentometric method (Food Safety and Standards Authority of India (FSSAI), 2016), respectively. The nutrients: nitrates (EPA, 1971), phosphate (FSSAI, 2016) and sulphates (APHA, 2005) were determined spectrophotometrically.

Ecology

River water samples, preserved with formalin and viewed under the light microscope to identify the cells present. Identification of biological samples was done using the reports of Prescott (1962); Patrick and Reimer (1975); and Adesalu *et al.* (2016).

Microalgae Cultivation and Identification

A generous amount of the fresh water sample was poured into a 250 ml conical flask and 5 ml of nutrient media (bold basal medium (BBM) and blue-green-11 (BG-11)) was added in separate flasks and kept close to the window for sunlight penetration to aid photosynthesis. Media were prepared from stock solutions of macronutrients, trace metals, and vitamins which were added to a large proportion of the final volume of water in order to avoid precipitation (Blinova *et al.*, 2015). Identification of biological samples was also done using the reports of Prescott (1962); Patrick and Reimer (1975); and Adesalu *et al.* (2016).

Bold basal medium (BBM)
BBM comprised 10 ml each of following: 25 g/L $NaNO_3$, 2.5 g/L $CaCl_2.2H_2O$, 7.5 g/L $MgSO_4.7H_2O$, 7.5 g/L K_2HPO_4, 17.5 g/L KH_2PO_4, 2.5 g/L NaCl; and 1 ml each of the these four solutions: alkaline EDTA solution (50 g/L EDTA & 31 g/L KOH), acidified iron solution (4.98 g/L $FeSO_4.7H_2O$ & 1 ml conc. H_2SO_4), boron solution (11.42 g/L H_3BO_3) and trace metals solution (8.82 g/L $ZnSO_4.7H_2O$, 1.44 g/L $MnCl_2.4H_2O$, 0.71 g/L MoO_3, 1.57 g/L $CuSO_4.5H_2O$, 0.49 g/L $Co(NH_3)_2.6H_2O$). The components were made up to 1 L with distilled water and the final pH made up to 6.6; thereafter, sterilised with an autoclave at 121 °C for 15 mins (Andersen *et al.*, 2005).

Blue-green 11 (BG-11)
Blue green-11 medium comprised 1 ml each of the following solutions: Fe citrate solution (6 g/L citric acid & 6 g/L ferric ammonium citrate), 40 g/L $K_2HPO_4.3H_2O$, 75 g/L $MgSO_4.7H_2O$, 36 g/L $CaCl_2.2H_2O$, 20 g/L Na_2CO_3, 1 g/L $MgNa_2EDTA.H_2O$, trace metals solution (2.86 g/L H_3BO_3, 1.81 g/L $MnCl_2.4H_2O$, 0.22 g/L $ZnSO_4.7H_2O$, 1 ml of 79 g/L $CuSO_4.5H_2O$, 0.391 g/L $Na_2MoO_4.2H_2O$, 1 ml of 49.4 g/L $Co(NO_3)_2.6H_2O$) and 1.5 g $NaNO_3$. The components were made up to 1 L with distilled water and final pH made up to 7.4. The medium was then sterilised with an autoclave at 121 °C for 15 mins (Andersen *et al.*, 2005).

Statistical Analysis
This analysis was done with the aid of Graphpad prism version 6.0. Data was reported as mean of duplicate values.

RESULTS
Physicochemical Parameters
The physicochemical parameters of the surface water samples collected from the three rivers are shown in table 1. The pH of the river water ranged from 6.30 to 6.50. Ogunpa River recorded the highest values for calcium, magnesium, chloride, total hardness and salinity, whereas Uren River had the lowest values for phosphates, conductivity, calcium, magnesium, chloride, total hardness and salinity; and Majopa River had the highest values for nitrates, sulphates, phosphates and conductivity (Table 1). Comparing with the other physicochemical properties of some rivers in Ogun State, the total acidity, sulphate and chloride levels from other studies were low compared to results obtained from this study while the levels of pH and calcium were higher from other studies (Table 2).

Table 1: Physicochemical Parameter Obtained from River Water Samples

Physicochemical Parameter	Majopa	River Ogunpa	Uren
pH	6.50	6.30	6.40
Conductivity (µS/cm^3)	94	92	74
DO_2 (mg/L)	2.6	2.8	3.0
Turbidity (NTU)	0.05	0.04	0.05
Total Hardness (mg/L)	30	35	25
Total Alkalinity (mg/L)	50	150	100
Total Acidity (mg/L)	25.0	12.5	25.0
Calcium (mg/L)	12.0	14.0	10.0
Magnesium (mg/L)	4.4	5.1	3.6
Sodium (mg/L) – Salinity	117.4	175.8	63.2
Chloride (mg/L)	65.0	125.0	35.0
Nitrate (mg/L)	12.6	10.7	12.2
Sulphate (mg/L)	97.2	90.3	92.6
Phosphate (mg/L)	1.7	1.6	1.4

Values represent mean of values obtained from duplicate analyses.

Table 2: Comparison of Physicochemical Results Obtained from other rivers in Ogun State

Physicochemical Parameter	River					
	Majopa[a]	Ogunpa[a]	Uren[a]	Akin-Olugbade[b]	Lafenwa[b]	Iju[c]
pH	6.5	6.3	6.4	7.6	7.2	7.2
Conductivity (µS/cm^3)	94	92	74	n.a	n.a	757
DO_2 (mg/L)	2.6	2.8	3.0	n.a	n.a	n.a
Turbidity (NTU)	0.05	0.04	0.05	n.a	n.a	n.a
Total Hardness (mg/L)	30	35	25	n.a	n.a	n.a
Total Alkalinity (mg/L)	50	150	100	44	43	57
Total Acidity (mg/L)	25.0	12.5	25.0	4.6	3.6	n.a
Calcium (mg/L)	12.0	14.0	10.0	22.1	33.1	n.a
Magnesium (mg/L)	4.4	5.1	3.6	14.1	5.1	n.a
Chloride (mg/L)	65.0	125.0	35.0	9.0	9.9	n.a
Sodium (mg/L)	117.4	175.8	63.2	n.a	n.a	n.a
Nitrate (mg/L)	12.6	10.7	12.2	n.a	n.a	1.2
Sulphate (mg/L)	97.2	90.3	92.6	n.a	n.a	18.3
Phosphate (mg/L)	1.7	1.6	1.4	n.a	n.a	3.3

n.a: value not available
a: This study
b: Osunkiyesi (2012)
c: Alabi *et al.* (2013)

Ecology

A number of green microalgae (Division: *Chlorophyta*) were identified across the three rivers with Ogunpa river having the highest number of different green microalgae. The abundant green microalgae identified in Majopa River are *Scenedesmus* sp., while Uren River contained *Pediastrum complex*, *Coelastrum* sp., and Coccoidal algae. Ogunpa River contained *Pediastrum* sp., *Coelastrum* sp., *Scenedesmus* sp., *Chlorella* sp., *Chlorococcus* sp. and *Desmodesmus* sp.

The surrounding vegetation around the rivers includes a number of different species of green plants. Majopa River was bounded by *Sacciolepis africana* with *Nymphaea lotus* floating on top of the river. *Panicum maximum* surrounded Ogunpa River, while the following plants surrounded Uren River: *Sacciolepis africana, Ipomoea cairica, Costus afer* and *Ludwig* sp.

The green microalgae identified in the standard media, with Bold Basal Medium (BBM) and Blue-Green 11 Medium (BG-11) being used in culturing the algae, are listed in table 3. *Scenedesmus* sp. were present in all the rivers under study when cultured in BBM but present only in Ogunpa River when cultured in BG-11. *Pediastrum* sp. were present in both Ogunpa and Uren Rivers, but not in Majopa River. Ogunpa River contained the highest varieties of green microalgae. Coccoidal algae were also present in both Ogunpa and Uren Rivers. No microalgal growth occurred in Majopa and Uren Rivers, when cultured in BG-11.

Table 3: Some Species of Green Microalgae Identified in Water Obtained from Rivers in Ogun State

Green Microalgae	River		
	Majopa	Ogunpa	Uren
Coelastrum **sp.**	-	✓	✓
Pediastrum **sp.**	-	✓	-
Scenedesmus bicaudatus	-	✓	-
Scenedesmus quadricauda	-	✓	-
Chlorococcus **sp.**	-	✓	-
Chlorella **sp.**	-	✓	-
Scenedesmus **sp.**	✓	✓*	✓
Pediastrum duplex	-	-	✓
Oedogonium **sp.**	-	✓	-
Coccoidal algae	-	✓	✓

✓: algae identified in Bold Basal Medium (BBM)
*: algae identified growing in Blue Green 11 Medium (BG-11)

DISCUSSION

The aquatic environment especially in the tropics changes with physical and chemical factors contributing to the diversity of the phytoplankton abundance, which are highly dependent on these physicochemical parameters that are crucial to their optimum growth, reproduction and survival (Odulate *et al*., 2017). These water bodies were sampled during the dry season (March, 2018), when the level of rainfall was far reduced, and this tends to have impacted most of the physicochemical parameters evaluated. The pH values of the water from the three rivers showed that they were all slightly acidic, during the period of study, with Ogunpa River having the highest acidity. This reflected in the total alkalinity levels of the samples showing the highest value in this river. Ogunpa River sample also had the highest value for total hardness compared with the samples from other rivers and this can be attributed to its higher level of calcium and magnesium as shown in table 1. On another hand, water from Uren River showing least level of total hardness could be a reflection of its usefulness for drinking, as primarily relied upon by some of the community dwellers.

Moreover, the sodium and chloride levels of the water samples were relatively higher for the freshwater bodies, especially the chloride levels in comparison with other rivers in Ogun state as shown in table 2. This can also be attributed to the limited rainfall at the time of study. Furthermore, the relatively highest levels of sodium and chloride in water from Ogunpa River when compared with samples from other rivers under study could be because the river is somewhat undisturbed, compared with Majopa and Uren Rivers, where various municipal activities (like washing of clothes and food items as well as abattoir and car cleaning services) occur.

The level of nitrates, sulphates and phosphates in the water samples were highest in sample from Majopa River probably due to the municipal activities previously stated, thus making the water unsafe for drinking as equally reported by Alabi *et al.* (2013). The lower levels of these parameters in water from Ogunpa River could be due to minimal activities in and around the river. The same trend was observed in the lower level of turbidity in sample from Ogunpa River, which would enable better light penetration, compared with other rivers; thus, aiding photosynthesis in phytoplankton.

Although some of these physicochemical parameters are higher in the rivers when compared to some other rivers in Ogun state, they still vary within the acceptable limits for drinking water:

conductivity <1000 μS/cm, turbidity <5 NTU, total hardness <150 mg/L, sodium <200 mg/L, chloride <240 mg/L, nitrates <50 mg/L, sulphates <100 mg/L (Nigerian Industrial Standard (NIS), 2007). The other parameters that did not fall within limit may be due to the presence of heavy rocks in the river and its surroundings, which could have leached into the rivers overtime in addition to the minimal amount of rainfall available at the time of sampling.

This study reports for the first time the presence of *Scenedesmus* sp. in water from Majopa River, presence of *Coelastrum* sp., *Pediastrum* sp., *Scenedesmus bicaudatus*, *Scenedesmus quadricauda*, *Chlorococcus* sp., *Chlorella* sp., *Scenedesmus* sp., *Oedogonium* sp., and Coccoidal algae in water from Ogunpa River and *Coelastrum* sp., *Scenedesmus* sp., *Pediastrum duplex*, and Coccoidal algae in water from Uren River. The ecological study revealed that water sample from the rivers showed richness in biodiversity of green microalgae with Ogunpa River being the richest, in comparison with Majopa and Uren Rivers. These outcomes must have been influenced partly by the pattern of the physicochemical properties of the different water samples. The number and type of plants growing around the river may have a role to play in the biodiversity of the green algae in the rivers as they may compete with the phytoplankton for nutrients. Also, in areas with narrow rivers or floating plants, the plants may shield the algae from sunlight thus limiting light penetration required for photosynthesis.

The standard media that supported growth of green microalgae across all rivers was the bold basal medium (BBM). It has also been reported that BBM is the medium that best supports the growth of most green algae (Andersen *et al*., 2005). Although blue-green 11 medium can still support the growth of some green algae, it is mainly used to grow the blue-green algae (cyanophyta division).

CONCLUSION

The physicochemical parameters of the water collected from these rivers seem to have been affected by seasonal variation. The parameters obtained from Majopa River showed the water as unfit for drinking, and with very limited biodiversity of green microalgae. Ogunpa River had the highest amount of total hardness, sodium and chloride with relatively low levels of nitrates, sulphates and phosphates that though unfit for drinking, it ultimately favoured the thriving of very rich biodiversity of green microalgae, compared with Majopa and Uren Rivers. The physicochemical properties of Uren River showed the river water as fit for drinking, and with some biodiversity of microalgae. It is highly recommended that the microalgae identified should be further characterized to enable the discovery of those with potentials for nutraceutical, biofuel, and agricultural production; for remediation, air pollution control, and so on.

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