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ASSESSMENT OF PERIPHYTON BIOMASS AND WATER QUALITY BY CHLOROPHYLL DETERMINATION OF IFEWARA RESERVOIR, OSUN STATE, NIGERIA

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ABSTRACT: *This study explored the use of taxonomic composition of periphyton and chlorophyll to assess the Ifewara reservoir status. Glass slide was attached to wooden slab and submerge in water between 2 to 5days then removed from water and periphyton samples were scraped from glass slides with a spatula. The scraping was emptied into a sterilized plastic vial containing 20 ml of Lugol's solution for preservation. 2.0-5.0 liter of surface water-sample is often convenient for measurement of Chlorophyll in the biomass from established stations (inflow, middle and lower lentic basin). The recorded periphyton was microphotographed, counted and identified using various keys and guides. A total number of ninety (90) species of periphyton was recorded belonging to phytoplankton (73) and zooplankton (170 species. Spatially, bacillariophyta (124±52 Org/m3) recorded the highest mean abundance at the upper basin station. Chrysophyta (1.0±0.6 Org/m3) was higher at middle basin station and highest mean abundance for chlorophyta (30±12 Org/m3), chlorophyll a, b and c (3.14±0.39 Org/m3, 2.10±0.46 Org/m3 2.37±0.41 Org/m3) was observed in lower basin station. Temporally, bacillariophyta, cyanophyta, euglenophyta and copepod, cladocera, rotifer, chlorophyll a, b and c recorded the highest mean abundance during the dry season while chlorophyta and chrysophyta were dominant in the rainy season. Chlorophyta were the predominant group and account for 50.68% of total species composition followed by bacillariophyta (32.88%) and least was chrysophyta (1.36%). Among the periphyton fauna, rotifer was dominant with 76.47% and followed by protozoa (11.76%). The study revealed that reservoir is composed of highly diversified periphyton groups with great potential to support aquatic community and fishery production and portray the reservoir as unpolluted, dilute fresh water suitable for its intended primary purpose of water supply to the riparian communities.*

KEYWORD: Periphyton, Chlorophyll, Substrate, Reservoir, Biomass, Water Quality.

INTRODUCTION

Algal biomass serves as a sustainable raw material for producing pharmaceuticals, fertilizers, biofuels and food products. Algal density, abundance, and diversity are ideal indicators of the health of aquatic ecosystems and as a biomonitoring of water quality. Waterbody have submerged substrates that covered partially by periphyton community. Periphyton is a complex assemblage of algae, cyanobacteria, micro-invertebrates, heterotrophic microbes and detritus attached to submerged surfaces in most aquatic ecosystems [1]. Periphyton play a crucial and fundamental role in food web as the primary food source for small consumers such as fish, invertebrate and it an assembly of plant and animal species colonizing diverse substrates in aquatic environments [2, 3]. The utility of periphyton to expose ecological ramifications of restorative or deconstructive changes is due to its bearing several of the most desirable features of a reliable ecological indicator [2]. The interaction between periphyton and physico-chemical, biota and their surrounding environment affects many biological communities in aquatic ecosystem including nutrient concentrations, soil quality and dissolved gases [4]. Periphyton provides dissolved oxygen through photosynthesis to sustain the aquatic life in its surrounding [4]. They are organisms that attached directly to the substrate, as well as those that move freely between them includes algae, filamentous algae, bacteria, protozoans, free-swimming microorganisms and other minute invertebrates such as rotifers and cladocerans [1]. The periphytic algal growth and their development affected by both natural and artificial substrates that strongly influenced by abiotic factors such as type of water body, light availability, substratum types, depth water movement, current, water velocity, pH, alkalinity, water hardness, nutrient quantity (phosphorus, nitrogen and carbon), other dissolved nutrients (calcium, sulphur and silicon) temperature, salinity, oxygen and carbon dioxide [5].

Many factors have influenced the growth of phytoplankton, generally represented by the concentration of chlorophyll-a, such as physical variables, nutrients, organic substances, and metal ions [6, 7].The deleterious proliferation of planktonic algae is a main cause of death of aquatic life and damage to aquatic ecosystems and water functions in lakes [8]. Chlorophyll is a major key in biochemical component of molecular apparatus that is responsible for photosynthesis, the critical process in which the energy from sunlight is used to produce life-sustaining oxygen. Chlorophyll in plants, algae and some bacteria is vital to the survival of the plant, animal and other kingdoms in nature. Chlorophyll ‘a’ is a blue-green microcrystalline solid, while Chlorophyll ‘b’ is green black microcrystalline solid [5]. Chlorophyll ‘a’ is of universal occurrence in the green plants; Chlorophyll ‘b’ occurs in higher plants and green algae. All plant-life contains the primary photosynthetic pigment Chlorophyll ‘a’. Chlorophyll concentration is an indirect estimation of the biomass and the photosynthesis-rate of the primary producers [5]. Chlorophyll is present in many organisms including algae and some species of bacteria while a good dietary source of Chlorophyll includes dark green leafy vegetables, algae, chlorella, wheat-grass and barley grass. It is the only precursor molecule that was developed to evaluate the trophic level of waters. Chlorophyll itself is not a single molecule but a family of related molecules, designated as chlorophyll a, b, c and d (only present in red algae). Chlorophyll pigments are major supplement source in the production of nutraceutical products [9], Chlorophyll a is most abundant in nature, although chlorophyll b and c are also common in fresh water phytoplankton. The quantification of chlorophyll a is easier than the algal biomass itself and can be used as an indirect method of biomass quantification [10],

Therefore, algal biomass measurement is important in many biological and ecological studies and in microalgae industry. The main objective of the study is to determine the taxonomic composition, abundance and diversity of the periphyton community and chlorophyll concentrations of Ifewara reservoir with respect to seasonal and spatial variations.

MATERIALS AND METHODS

A Description of area of Study

Ifewara Reservoir was impounded in 1996 with main primary purpose of water storage in order to supply potable water for people Ifewara community and its environs. It is an impoundment of River Ejipade, a tributary in upper Owena River in Ifewara Atakumosa West Local Government Area of Osun State and the reservoir is within the Osun River sub basin of the Ogun-Osun River Basin of Nigeria. The reservoir catchment area is approximated within Latitudes 07° 28' 50" to 07° 29' 0" North and Longitudes 004° 41' 0" to 004° 41' 20" East with surface area of about 3km^2 (300ha). Ifewara is a tropical savanna climate prevailing with the average annual temperature of 33ºC and there is about 580nm of rain in a year. It is dry 82 days a year with an average humidity of 76% and an UV-index of 6. Samples were collected bi-monthly for period of a year to covering both dry and rainy seasons. The grid co-ordinates of each station were determined using a Global Positioning System (GPS) handset while rubber floaters were used to indicate the permanent location of the sampling stations for subsequent recognition. Four sampling stations (stations 1, 2, 3 and 4) were established on the reservoir along its horizontal axis for the study. Station 4 was located within the lower basin of the reservoir close to the dam wall, Stations 3 and 2 were in the middle basin while Station 1 was located at the inflow basin of the reservoir (Figure 1).

Periphyton Collection

Periphyton samples were collected from all sampling stations by scraping glass slides attached to wooden slabs with a spatula. The scrapings were emptied into a sterilized plastic vial containing 20 ml of Lugol's solution that was allow to stand for a week. They were then microphotographed, counted and the taxonomic composition of periphyton determined using identification keys by [11], [12] as applicable.

Determination of Chlorophyll Concentration

Chlorophyll concentration was determined using the trichromatic method [13]. Surface water was collected from the reservoir into 5 liters sampling bottles. Constrains should be placed on filtering of the water-samples that are to be used for the extracted analysis. From the time of collection to the measurement, the sample should be stored in the dark, on ice. Water-sample can be held up to 2 weeks in the dark at 4ºC by Using opaque bottle, because even exposure to light during storage will alter the Chlorophyll values. 200 ml of water sample was filtered through glass-fiber filters (Whatman GF/A). Filtration assembly was attached with suitable source of reduced pressure until the sample was dry. Glass-fiber filter containing green pigment should be immediately analyzed for Chlorophyll. Placed the glass-fiber filter in beaker and added pure acetone for approximately 3 ml acetone to the one cuvette and measured the optical density at 750 nm and adjusted the zero. Similarly, clarified extract was added to the second cuvette and the absorbance measured at same wavelength. Same measurements were then repeated at 665 nm. Extract was then acidified with 1 ml of $MgCO_3$ suspension during final few ml of filtering to prevent acidification of chlorophyll content, the filter paper was folded into quarters and wrapped in aluminum foil after which they were left to freeze.

The filter was later grinded in 10ml of 90% aqueous solution of acetone after which 2ml was added to make the solution up to 12ml. It was then transferred into 15ml centrifuge tubes and centrifuged at 4000rpm for 15 minutes. The supernatant was gently decanted into a 1 cm path length cuvette and measured the optical density at 750nm, 665nm, 645nm and 630nm respectively for chlorophyll-a, b and c within 90s. while that of 750nm was to account for background turbidity losses [5, 13]. Following formulas was used for the calculation of Chlorophyll a and Pheophytin a.

$$\text{Chlorophyll a, mg/m}^3 = \frac{26.7(664_b - 665_a) \text{ x} V_1}{V_2 \text{ x } L}$$

$$\text{Phenophytin a, mg/m}^3 = \frac{26.7[1.7(665_a - 664_b) \text{ x} V_1]}{V_2 \text{ x } L}$$

Where: V_1 = Volume of extract, L

V_2 = Volume of sample, m

L = width of cuvette (cm)

664b, 665a = Optical densities of extract before, and after acidification respectively

The value 26.6 is the absorbance correction and equals A x K

Where: A = Absorbance coefficient for chlorophyll a at 664 nm = 11.0 and K = Ratio expressing correction for acidification

The trichromatic equations below were then used to calculate

Chlorophyll a =11.6 E6650 – 1.31 E6450 –0.14E6300

Chlorophyll b =20.7 E6650 – 4.34 E6450 –4.42E6300

Chlorophyll c = 55E6300 – 4.64 E6650 –16.3E6450

Statistical Analysis

Data obtained were subjected to appropriate statistical analysis that includes descriptive statistics, correlation analysis, Analysis of variance (ANOVA as applicable to show spatial and seasonal variations in periphyton community and chlorophyll value.

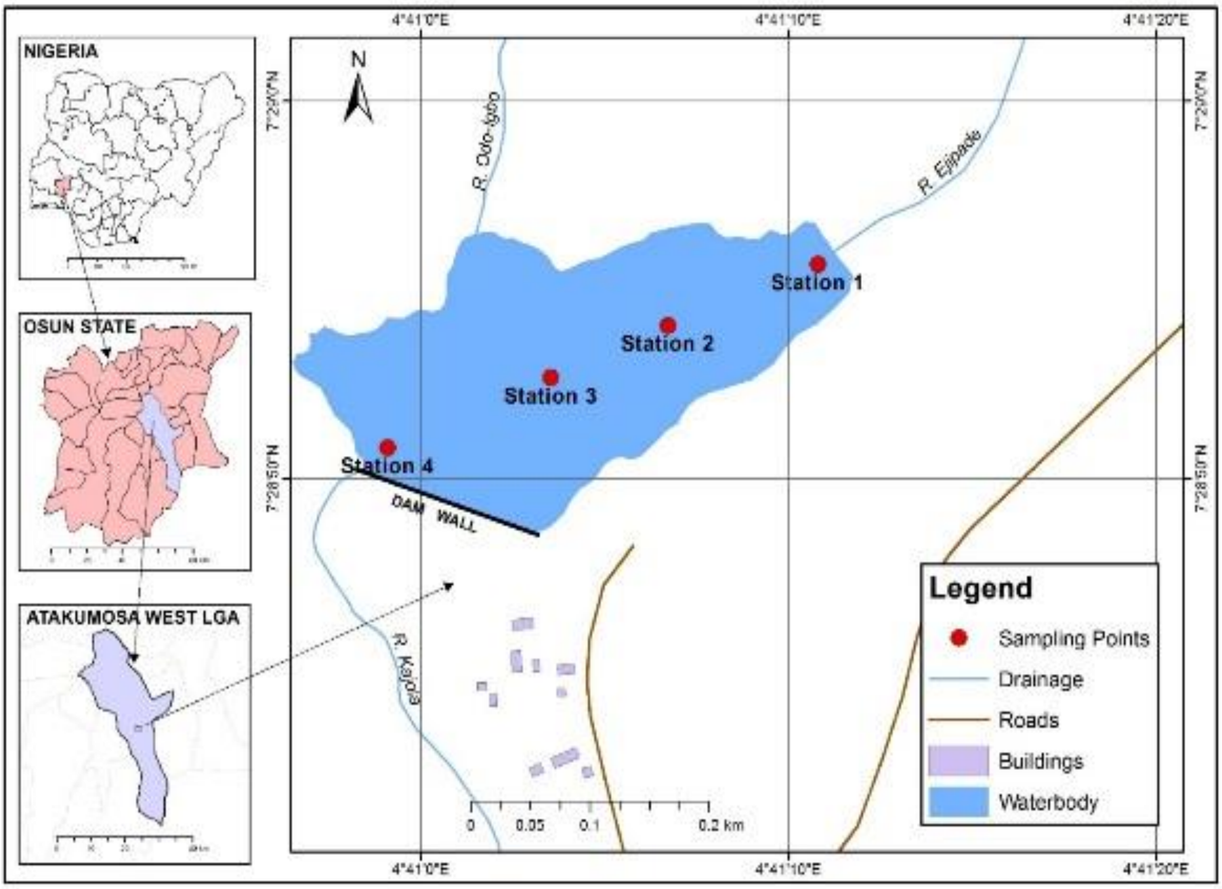

Figure 1: Map Showing the Sampling Stations Located within Ifewara Reservoir, Atakumosa West Local Government, Osun State, Nigeria.

RESULTS

A total number of ninety (90) species of periphyton belonging to 11 classes were identified from the investigated sampling stations of Ifewara reservoir during the period of study. The biota identified comprised seventy-three (73) species of periphyton flora (phytoplankton) and seventeen (17) species of periphyton fauna (zooplankton). The 73 periphyton recorded species of periphyton flora belonged to fifty seven (57) genera, thirty-nine (39) Families, thirty-two (32) Orders, six (6) Classes of algae while seventeen (17) species of periphyton fauna belonged to five (5) classes, five (5) orders, seven (7) families, ten (10) genera as presented in Table 1. Chlorophyta had the highest percentage occurrences among the phytoplankton flora while chrysophyta had the lowest occurrence and rotifer had the highest percentage occurrences among zooplankton fauna while copepod and cladocera had the lowest percentage as presented in Table 1. Spatially, bacillariophyta (124±52 Org/m^3) recorded the highest mean abundance at the upper basin station. Chrysophyta (1.0±0.6 Org/m^3) was higher at middle basin station and highest mean abundance for chlorophyta (30±12 Org/m^3), chlorophyll a, b and c (3.14±0.39 Org/m^3, 2.10±0.46 Org/m^3 2.37±0.41 Org/m^3) was observed in lower basin station

(Table 2). Among the periphyton fauna, copepod (0.1±0.1 Org/m^3) record the highest mean abundance at middle basin station while high mean abundance of cladocera (0.2±0.2 Org/m^3), protozoa (0.2±0.2 Org/m^3) and rotifer (16±15 Org/m^3) occurred at lower basin station as shown in Table 2. There was a highly significant difference ($P \leq 0.01$) between mean abundance of rotifer along the horizontal axis of the reservoir. The highest mean abundance of bacillariophyta (78 ±24 Org/m^3), chrysophyta (11±6.42 Org/m^3), copepod (0.3±0.1 Org/m^3), chlorophyll a, b and c (2.57±0.39 Org/m3, 2.25±0.46 Org/m3 and 2.42 ±24 Org/m3) were recorded from surface water. Cyanophyta and cladocera had the higher mean abundance at mid-depth water and chlorophyte, euglenophyta and rotifer at bottom water (Table 3). Seasonally, bacillariophyta, cyanophyta, euglenophyta and copepod, cladocera, rotifer, chlorophyll a, b and c recorded the highest mean abundance during the dry season while chlorophyta and chrysophyta were dominant in the rainy season (Table 4). There are two major clustering diagrams showing the relationship between periphyton groups identified with chlorophyll concentration. (i) There is strong relationship between protozoa, bacillariphyta, chlorophyll a, b, c, chrysophyta and copepod. (ii) There is correlation between cyanophyta with cladocera, rotifer, chlorophyte and euglenophyta as presented in figure 2.

Table 1: Outline Classification and Taxa Composition of the Periphyton Flora and Fauna (Org/m^3) of Ifewara Reservoir

Division	Class	Order	Family	Genus	Species	Occurrence (%)
Periphyton flora (Phytoplankton)						
Bacillariophyta (Diatoms)	1	18	19	22	24	32.88
Chlorophyta (Green Algae)	1	9	11	24	37	50.68
Chrysophyta (Golden Algae)	1	1	1	1	1	1.36
Cyanophyta (Blue-green Algae)	1	4	6	7	9	12.33
Euglenophyta (Euglenoids)	1	1	1	1	2	2.74
Periphyton fauna (Zooplankton)						
Copepod	1	1	1	1	1	5.88
Cladocera	1	1	1	1	1	5.88
Protozoa	1	1	1	2	2	11.76
Rotifera	2	2	4	6	13	76.47
Total Phytolankton	**6**	**32**	**39**	**57**	**73**	**100**
Total zooplankton	**5**	**5**	**7**	**10**	**17**	**100**
Total Periphyton (Flora and Fauna)	**11**	**37**	**46**	**67**	**90**	

Table 2: Mean Abundance (Org/m^3) of Chlorophyll, Periphyton Flora and Fauna along Horizontal Variation of Ifewara Reservoir

Taxonomic Group	Upper Basin Mean ± Sem	Middle Basin Mean ± Sem	Lower Basin Mean ± Sem	ANOVA F	ANOVA P
Periphyton flora (Phytoplankton)					
Bacillariophyta	124.0 ± 52.0	57.0 ± 10.0	51.0 ± 13.0	2.331	0.106
Chlorophyta	23.0 ± 3.0	27.0 ± 4.0	30.0 ± 12.0	0.104	0.901
Chrysophyta	0 ± 0	1.0 ± 0.6	0.11 ± 0.01	0.263	0.770
Cyanophyta	9.0 ± 3.0	12.0 ± 4.0	12.0 ± 6.0	0.040	0.961
Euglenophyta	4.0 ± 3.0	3.0 ± 1.0	4.0 ± 2.0	0.114	0.892
Periphyton fauna (Zooplankton)					
Copepoda	0.01±0.01	0.1 ± 0.1	0 ± 0	0.590	0.558
Cladocera	0 ± 0	0.13 ± 0.1	0.2 ± 0.2	5.130	0.009
Protozoa	1.0 ±0.7	0.2 ± 0.1	0.2 ± 0.2	1.289	0.283
Rotifera	10.0 ± 8.0	4.0 ± 2.0	16.0 ± 15.0	1.708	0.190**
Chlorophyll values					
Chlorophyll-*a*	2.80 ± 0.39	1.63 ± 0.14	3.14 ± 0.20	0.632	0.535
Chlorophyll-*b*	1.34 ± 0.46	1.56 ± 0.18	2.10 ± 0.29	0.551	0.579
Chlorophyll-*c*	1.03 ± 0.70	1.87 ± 0.28	2.37 ± 0.41	0.045	0.956

***Highly Significant difference (P≤ 0.01)*

Table 3: Mean Abundance ((Org/m^3) of Periphyton Flora and Along Vertical Variation of Ifewara Reservoir

Taxonomic group	Surface Mean ± Sem	Mid-depth Mean ± Sem	Bottom Mean ± Sem	ANOVA F	ANOVA P
Periphyton flora (Phytoplankton)					
Bacillariophyta	78.0 ± 24.0	34.0 ± 5.0	77.0 ± 14.0	2.344	0.105
Chlorophyta	18.0 ± 3.0	23.0 ± 5.0	39.0 ± 9.0	3.148	0.050
Chrysophyta	11.0 ± 6.42	0 ± 0	1.0 ± 0.3	0.712	0.495
Cyanophyta	7.0 ± 2.0	15.0 ± 5.0	13.0 ± 7.0	0.633	0.535
Euglenophyta	2.0 ± 1.0	1.0 ± 0.3	7.0 ± 3.0	3.348	0.042
Periphyton fauna (Zooplankton)					
Copepoda	0.3 ± 0.1	0 ± 0	0.1±0.1	2.978	0.059
Cladocera	0.1 ± 0.1	0.24 ± 0.20	0 ± 0	1.000	0.374
Protozoa	0.3 ± 0.2	0.1 ± 0.1	0.3 ± 0.2	0.606	0.549
Rotifera	1.0 ± 1.0	7.0 ± 5.0	8.0 ± 5.0	1.035	0.362
Chlorophyll values					
Chlorophyll-*a*	2.87 ± 0.39	1.94 ± 0.14	1.87 ± 0.39	0.632	0.535
Chlorophyll-*b*	2.25 ± 0.46	1.98 ± 0.18	1.80 ± 0.46	0.551	0.579
Chlorophyll-*c*	2.42 ± 0.70	2.53 ± 0.28	2.32 ± 0.70	0.045	0.956

Table 4: Seasonal Mean Abundance (Org/m^3) of Periphyton Flora and Fauna

Taxonomic Group	Dry Season Mean ± Sem	Rainy Season Mean ± Sem	St Anova F	P
Periphyton flora (Phytoplankton)				
Bacillariophyta	77.0 ± 24.0	56.0 ± 8.0	1.135	0.291
Chlorophyta	25.0 ± 5.0	28.0 ± 5.0	0.086	0.771
Chrysophyta	0 ± 0	1.0 ± 1.0	1.079	0.303
Cyanophyta	13.0 ± 7.0	12.0 ± 3.0	0.025	0.875
Euglenophyta	6.0 ± 3.0	2.0 ± 1.0	2.548	0.116
Periphyton fauna (Zooplankton)				
Copepod	0.2 ±0.1	0.1±0.1	0.594	0.444
Cladocera	0.1 ±0.1	0 ± 0	2.035	0.159
Protozoa	0.1 ±0.1	0.3 ± 0.1	0.523	0.473
Rotifer	6.30 ± 4.0	6.1 ± 3.0	0.002	0.961
Chlorophyll value				
Chlorophyll-*a*	2.47 ± 0.23	1.78 ± 0.10	9.855	0.003**
Chlorophyll-*b*	2.44 ± 0.29	1.84 ± 0.16	3.912	0.053*
Chlorophyll-*c*	2.87 ± 0.48	2.28 ± 0.22	8.174	0.206

**= Very Highly Significant difference (P ≤ 0.05)*
***=Highly significant difference (P ≤ 0.01)*

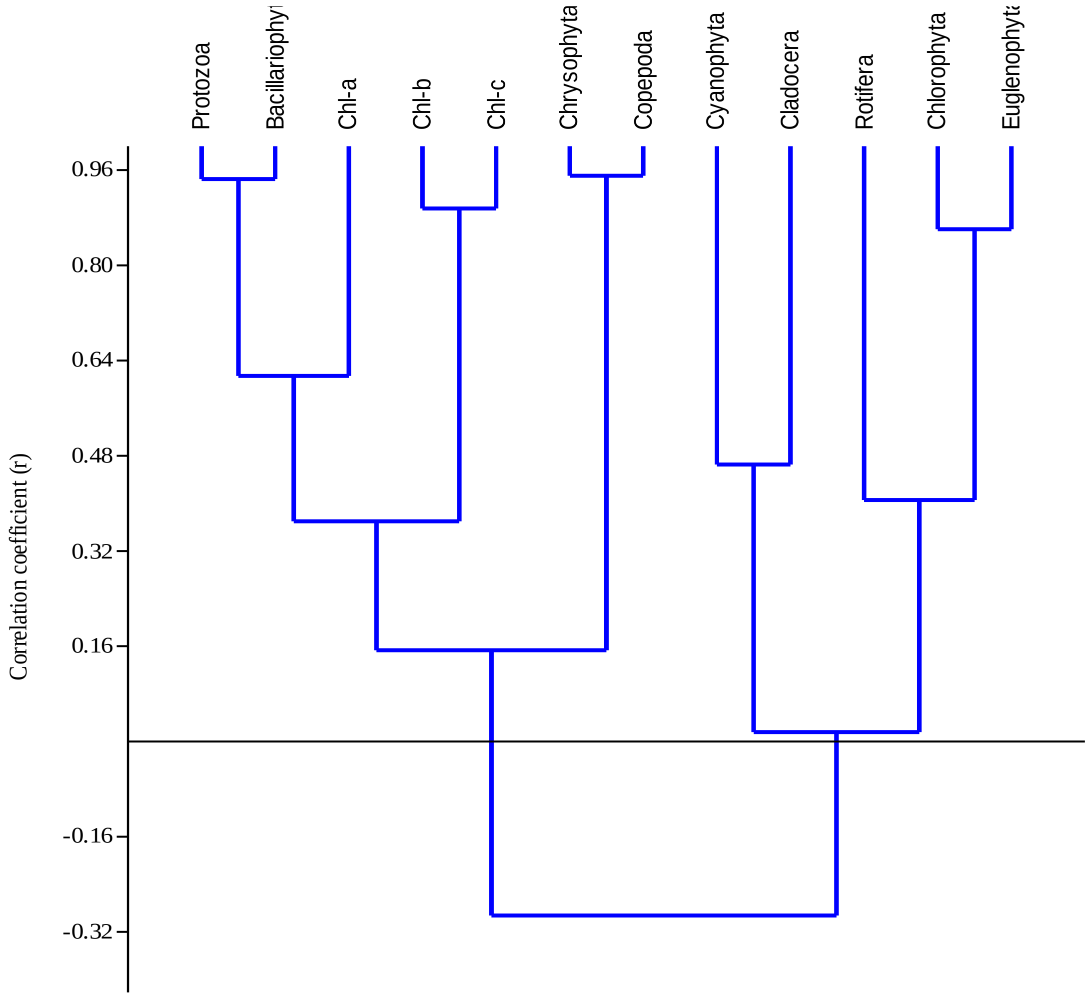

Figure 2: Cluster Diagram Showing the Relationship Between Groups of Periphyton and Chlorophyll

DISCUSSION

In this study, the periphyton algal community observed were taxonomically important in the order Chlorophyta > Bacillariophyta > Cyanophyta > Euglenophyta > Chrysophyta in the number of species indicated above. Most of the observed phytoplankton species in this study seem to be similar to the report of previous researchers on most tropical African freshwaters investigated especially Nigerian reservoirs based on the work of [14] and [15]. The predominant abundance of chlorophyta in the flora is in line with the work of [14], [16], [17] and [12] who reported a high diversity of green algae, particularly desmids, in Western Africa and attributed this to the high prevalence of rainfall in the area. Numerically, Bacillariophyta have higher mean abundance more than Chlorophyta might be due to the ability of diatoms to colonise substrates in a short time [18], [19]. Members of the Division Bacillariophyta are also been widely reported to dominate quantitatively the phytoplankton population of Nigerian freshwater and to a large extent tropical waters as reported by [20], [21], [22], [23], [24], [25] and the possession of raphe in pennate diatoms has been characterized as an asset [26]. It is more likely to attach to the slide surface if favorable conditions exist and can adapt successful in fresh / saline water habitats. It was observed that there was a high abundance of periphyton at the dam site and bottom water could be because of high availability of nutrients and less disturbances in these regions compared to the surface level and inflow section of the reservoir. The low organisms' abundance recorded from inflow and surface water due to excessive nitrogen and phosphorus inputs are important factors to shift lakes from oligotrophic to hypertrophic conditions [27], and lead to dramatic increases in harmful cyanobacteria blooms, which would create a serious threat to aquatic ecosystems [28]. The excessive amount of organic substances and metal ions in freshwaters generally originate from domestic sewage, urban run-off, industrial effluents and farm wastes, which are main causes of water pollution. Dissolved organic matter in lakes would absorb light and alter the light environment at depth, which would subsequently affect phytoplankton [29] and could also be consumed directly or indirectly by aquatic life and have widespread effects on zooplankton, benthic invertebrates, and fish [30]. The higher occurrence of some divisions over others as observed in this study may be attributed to substratum preference as substratum type and some environmental attributes are vital to periphyton development [31]. Periphyton play a vital role in the provision of food to fish and many other aquatic animals in a natural and controlled environment [32]. Although, the effects of physicochemical factors such as temperature, light intensity, transparency, pH, dissolved nutrients may be equally important in this distribution [13], [8]. The species richness of the periphyton fauna was in the order: Rotifera > Protozoa > Copepoda > Cladocera. The Phylum Rotifera was the most abundant quantitatively and qualitatively among the zooplankton species recorded. The result is similar to previously studies carried out on other water bodies in Nigeria [33], [34], [35], [36]. The Rotifers dominance among the zooplankton in this reservoir could be attributed to their resistance to a broad variety of impacts that makes them adaptable to several environmental conditions hence their wide geographical distribution. The Copepods and cladocerans were the least abundant zooplankton in this study with a percentage of 5.88% occurrence each from 17 species. This is in accordance to the earlier works on tropical waters where it was reported that cladocerans are rare [37] as well as [38] report of *Bosmina* sp. being the only cladoceran representative in Lake Baikal.

Chlorophyll *a*, *b* and *c* concentrations in this reservoir followed an increasing trend from the inflow section to the dam site. This might be as a result of higher water clarity and less disturbances in the dam site section of the reservoir compared to the inflow basin. The light conditions in any waterbody influence the growth of the plankton community, and the eutrophication caused by the high concentration of chlorophyll-a would impact light availability in aquatic ecosystem [39, 40]. The concentrations were also higher in the surface water sample compared mid-depth and bottom water sample, which is also high during dry season than during the rainy season. The dry season period is known to be associated with high amount of light energy from the sun reaching almost to the bottom of the reservoir and this is necessary for photosynthesis. The results of chlorophyll a disagreed with report of Suzuki *et al* [41], low chlorophyll *a* values reflecting limited phytoplankton growth in an investigation of a Mexican lagoon was associated to dark water, which reduced adequate light penetration into the lagoon. According to Kadiri [42], the typical seasonal pattern of variation in chlorophyll *a*, has a single major peak usually occurring in the dry season and a minor peak occurring in the late rainy season of Ikpoba reservoir, Edo State, Nigeria. Consequently, higher chlorophyll-*a* values recorded in the dry season in this study is probably a pointer to improved water clarity in the dry season that allowed greater light penetration required for photosynthesis. According to Sakamoto [43]and Vollenweider [44] lakes can be classified as Eutrophic lakes, with Chlorophyll ‘a’ concentration 5-140 mg/m3, Mesotrophic lakes with 1-15 mg/m3 and Oligotrophic lakes with 0.3-2.5 mg/m3. Chlorophyll has been used to ameliorate bad breath, as well as to reduce the odors of urine, feces and infected wounds. Ifewara reservoir can be classified as oligotrophic waterbody as result of mean ranged of chlorophyll-a (1.63-3.14) recorded from this reservoir.

CONCLUSION

From this study, it can be concluded that Ifewara Reservoir is characterised with richness and diverse periphyton community dominated mostly by desmids and diatoms especially species that are indicators of clean water system suggesting that the reservoir if not pristine is relatively clean and/or unpolluted. The water body can be said to be oligotrophic as a result of the abundance of desmids and diatoms as well as the chlorophyll concentrations. It would require low chemical dosage for treatment for potable use, the primary purpose for which the reservoir was impounded.

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