

Original Research Article

The Distribution and Seasonal Variation of Zooplankton Species of the Great Kwa River, Calabar, Nigeria: A Reassessment Approach

ABSTRACT

Background: Human activities adversely affect the distribution and diversity of zooplankton. They are diverse group of organisms with little or no locomotive ability and quickly respond to changes in their environment. This research was aimed at providing updated information on the distribution and seasonal variation of zooplankton in Great Kwa River.

Materials and Methods: Two sampling stations (S1- ObufaEsuk and S2- EsukAtu) were mapped along the river bank, samples were collected using plankton net of 55µm mesh size and preserved in 4% formalin. Species were identified using taxonomic keys. Data were analyzed using ecological indices.

Results: The results revealed 12 taxa; belonging to 38 species. *Tintinnida*, *Protozoa*, *Cladocera*, *Copepod* were 23.1%, 18.5%, 15.4% and 13.3% respectively. The lowest taxonomic groups were *Diptera*, *Foraminifera*, *Atenatadata* and *Trichoptera* having 1.85% for each order. The highest species was recorded in S2 having 31 species. In both Stations *Ascampbelliella acuta* was dominance over other species. Shannon-Weiner index (H) were 2.997 and 2.40 in S2 and S1 respectively. The evenness index were 0.576 and 0.547 for S1 and S2 respectively. Margalef's diversity index were 8.171 and 4.111 for Esuk Atu-S2 and Obufa Esuk-S1 respectively. Zooplankton species were abundant in wet season than dry season.

Conclusion: This present study provide updated information on the zooplankton distribution, diversity and seasonal variations of the Great Kwa River. The high dominance of *Tintinnida* in this study indicates a natural linkage between nano-planktons and macro-planktons in the food webs of the river.

Keywords: Distribution, Seasonal variation; Zooplanktonspecies; Kwa river; Calabar

Comment [R1]: The results presented are based much more on the analysis between the sample points than on the seasons. I suggest changing the name or modifying the discussion.

Comment [R2]: This subtitle would only be justified if data were presented from other campaigns carried out in the same location previously and the data presented in the results.

Comment [R3]: The authors present this information in the introduction and conclusion. However, at no time do they make an in-depth discussion about what the real impacts suffered by the study region are.

Comment [R4]: The keywords must be different from those contained in the title. In this way, the reach of the work will be increased in the search platforms. Therefore, the work will have greater visibility.

1. INTRODUCTION

Human activities through industrialization, deforestation, farming, oil exploration and transportation have resulted in climate changes which adversely affect the distribution, assembly structure of aquatic communities, especially the composition, richness and species diversity [1-3]. Activities of humans also lead to discharge of untreated animal wastes like releases from sewage and septic tanks, run-off from farm lands, laundering waste into surrounding bodies of water. Continuous subjecting these water bodies with high level of pollutants will consequently affect the health status of zooplanktons negatively; therefore influencing the distribution patterns of aquatic animals in the water [3-4].

Zooplankton species are essential constituent of the food chain in aquatic ecosystem. They feed on the phytoplankton, bacteria, aggregates of detritus and microorganisms and other zooplankton species. Almost all freshwater fish feed on zooplankton at different stage in their life cycle [5-7]. Zooplankton are heterotrophic planktonic animals. These are suspended in water with limited power of movement. Like phytoplankton, they are usually defused in water. Freshwater zooplankton are dominated by four major groups of animals namely *Protozoa*, *Rotifera* and two sub-classes of *Crustacea*- *Cladocera* and *Copepoda*[4,8,9]. Zooplankton is an important plankton component that regulates phytoplankton and microbial productivity by feeding on them. Feeding on phytoplankton and bacteria serve as main food for larval fishes, juvenile fishes and other carnivorous aquatic organisms [8]. The zooplankton feed on phytoplankton and facilitate the conversion of plant material into animal tissue and in turn constitute the basic food for higher animals including fishes, especially their larve [5,9], other aquatic vertebrates and man [10,11].

These fishes and some aquatic vertebrates are good source of protein, minerals, fat and oil for humans[9].

Studies have shown that phytoplankton diversity increased zooplankton productivity [10,11], while the effect of phytoplankton evenness on resource-use efficiency(RUE) switched from negative at the producer level (phytoplankton) to positive at the consumer level (zooplankton) [12] in some lakes investigated in United State of America. Zooplankton species diversity, distribution and abundance in the aquatic world has a vital influence on the healthiness of the aquatic environment [13] and differs temporally and spatially from sea to sea. It may be influence by competition, predation, physical, biological and chemical parameters [14]. Biological factors include predation, breeding, phytoplankton concentration and vertical migration. The physical factors are mixing of the oceanic water (down welling and upwelling) that influences nutrients availability; and on the other hand affect phytoplankton production and subsequently in the future affect the zooplankton speciation [9-11,15,16-18].

The water bodies play crucial roles in the economy and aquatic food supply for the inhabitants of Calabar and its neighboring villages, serving as their major source of fish and employment to the local people. Despite these vital roles played by the Great Kwa River to the good people of Cross River State, little or no measures have been put in place to conserve the endangered species due to uncontrolled fishing and other detrimental human activities in the aquatic environment. Against this back drop, there is urgent need for an updated and current information on zooplankton species in the great Kwa River due to increased human activities (uncontrolled fishing, release of untreated sewage, etc) and industrialization; geared toward species

conservation and as a tool for foretelling the impact of human activities in the river. Therefore, this study aimed at investigating on current distribution and seasonal variation of zooplankton species in the Great Kwa River, Calabar south, Nigeria.

2. MATERIALS AND METHODS

2.1 Study location

This study was conducted in Great Kwa River, Cross River State. The area is located between latitude $8^{\circ} 15' E$ and $8^{\circ} 30' E$ and longitude $4^{\circ} 45' N$ and $5^{\circ} 15' N$. It has an estimated length of 56km and is about 2.8km wide at the mouth where it empties into the Cross River Estuary [17]. Two climatic seasons occur in the study area (wet and dry). The wet season is usually characterized by heavy rainfalls while the dry season experiences occasional downpours. The shore-lines are lined with dark plates usually exposed during low tides and the shore is brackish and rich with zooplankton. The banks are also surrounded by lush, evergreen, forest vegetation with different species of trees, shrubs and grasses.

2.2 Selection of sampling stations

Two sampling stations were mapped along the River bank. Station 1 (ObufaEsuk); designated as S1 located between Latitude $4^{\circ} 94' N$ and Longitude $8^{\circ} 35' E$, close to the Biological Sciences, University of Calabar and University of Calabar Teaching Hospital. Station 2 (EsukAtu); designated as S2 located between Latitude $4^{\circ} 95' N$ and Longitude $8^{\circ} 36' E$ close to the University of Calabar staff quarters (Figure 1) [17].

Comment [R5]: It remained to indicate the period of the work.

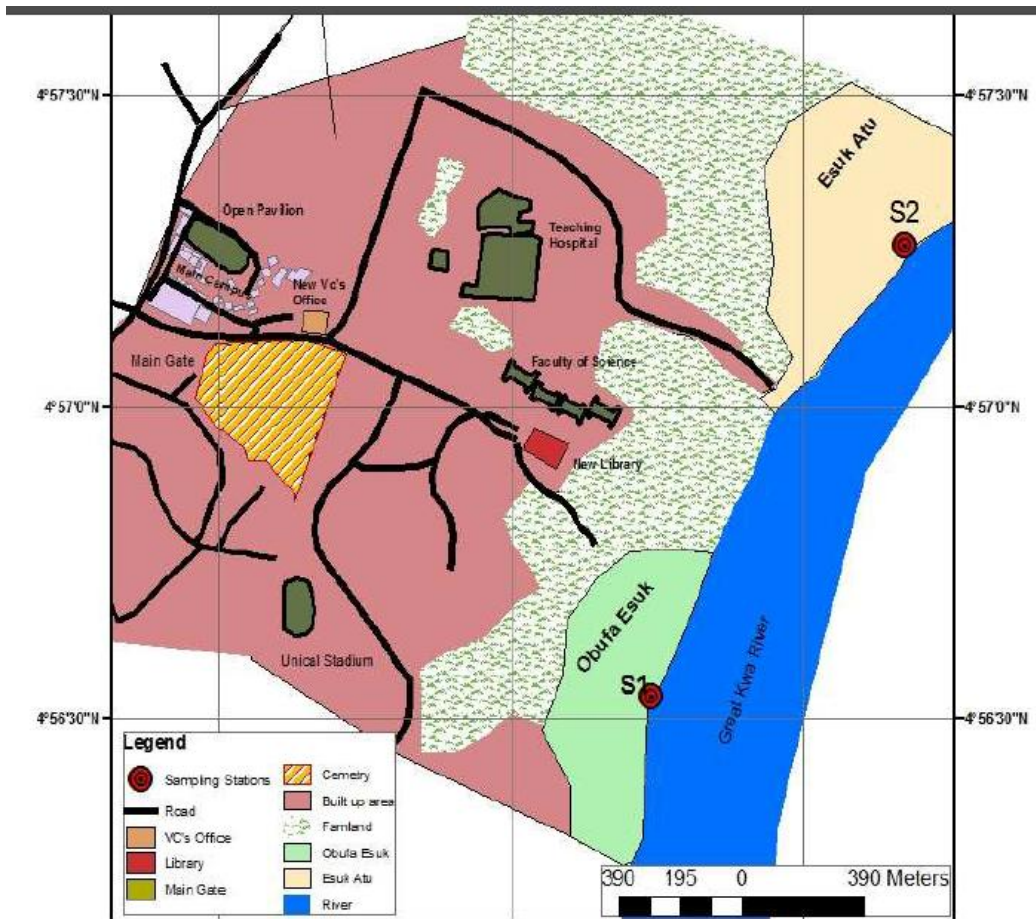


Figure 1: Map showing the study location of S1 and S2

Source: Okorafor, K. A. *et al.*, [17].

Comment [R6]: The figure caption must be self-explanatory.

Suggestion: Location of the Great Kwa River, Calabar, Nigeria. In red, the collection points S1 and S2.

Present a more comprehensive map of the region. Showing the location of the sea. As this is an international magazine, readers must be able to locate themselves as easily as possible.

The map presented by "Okorafor, K. A. *et al.*, [17]" presents more information than that contained in the present manuscript.

2.3 Zooplankton sample collection

A 55µm mesh standard plankton hydrobios net was used to collect zooplankton samples by towing method. Three replicates were collected at each station. After filtration, the zooplankton was collected in sampling bottles and was preserved in 4% formalin solution before transferring to Institute of Oceanography Laboratory, University of Calabar, Calabar. Some identification of zooplankton species was also carried out in the Department of Zoology and Environmental Biology, Faculty of Biological Sciences, University of Calabar, Calabar. Samples were taken and all individual taxa present were examined and counted. Species were sorted and counted using dissecting microscope at different magnifications. The abundance of zooplankton was determined using ecological indices.

Comment [R7]: What are the months of each collection?
This information only appears in the results.

Were 3 replicates performed for each point or 3 collections in total?
What is the year of collection?
What is the time or area swept by the drag?

The information contained in the methodology is important for other researchers to be able to repeat the same study in the future.

2.4 Procedure for zooplankton analysis

In the laboratory, zooplankton samples were allowed to settle for two days, thereafter some liquid were decanted to get concentrated samples which were stored in 50ml bottles. A homogenate of the sample fixed with 4% formalin solution was put in a 1ml plankton chamber (AJAH Model 001) and allowed to settle after covering it with a glass slide. Examination was carried out at 100x magnification; using the x10 magnification lens. They were identified to species levels using identification key guide or taxonomic keys [19-20]. Also relevant materials were utilized for species identification [21-26].

Comment [R8]: The zooplankton identification procedure was very detailed, unlike the collection procedure.

2.5 Ecological indices and statistical analysis

Analysis was carried out using software packages. Software package XLSTAT and JMP were used to analyze biological parameters in order to determine the zooplankton abundance and

Comment [R9]: I suggest adding the calculation of Dajoz's constancy values. This information helps to verify if the species always appear in the collections or if only in some events they are collected.

Comment [R10]: I suggest the authors carry out an analysis comparing the stations and the collection points to verify if there were significant differences between them.

diversity of the Great Kwa River. Ecological indices of zooplankton was estimated using margalef's index (D), Shannon-weiner (H), Pielou's evenness (E) and Simpson's dominance indices as documented [27]. Simple percentage was also used.

3.RESULTS

3.1 Zooplankton taxa and species distribution

The results of this study revealed a total of 12 taxa belonging to 38 species. The order *Tintinnida* was represented by three species (*Ascampbelliella acuta*, *Coxiella longa* and *Coxiellamariana*) consisting of 23.1%, followed by *protozoa*; which was represented by 10 species (like *Blephanirisma* species, *Loxodes* species, *Nebella caudate*, *Trigonopyxis arcula*, *Plagiopyxis callids*, *Arcelladiscoides*, etc) with 18.5%, while *Cladocera* was 15.4% of the total abundance of zooplankton species. The lowest taxonomic groups were *Diptera*, *Foraminitera*, *Atenadata* and *Trichoptera* having only one representative with 1.85% for each order as displayed on Table 1 and figure 2. The highest number of species were recorded in EsukAtu (S2) station with 31 species represented during the research period. In both Stations (ObufaEsuk and EsukAtu), *Ascampbelliella acuta* displayed high dominance over other species. The following taxonomic groups namely: *Diptera*, *Foraminitera*, *Atenadata* and *Trichoptera* where only found in station two (EsukAtu) (Table 1).

3.2 Zooplankton diversity

The trend of Shannon-Weiner index for both stations in the Great Kwa River can be illustrated as $S2 > S1$. The highest value of 2.997 (Shannon-Weiner index) was recorded in S2 (EsukAtu) whereas the minimum of 2.40 was detected in S1 (ObufaEsuk) station. Margalef's

diversity index were 8.171 and 4.111 for Esuk Atu-S2 and Obufa Esuk-S1 respectively depicting richness in diversity of zooplankton species in the study area. The Equitability or evenness index was slightly high in S1 (0.576) than in S2 (0.547) as shown on Table 2. In Obufa Esuk (S1), 8 individuals belonging to 3 taxa were recorded in February; while 21 individuals from 5 taxa were recorded in July. In S2 (EsukAtu), 20 individuals belonging to 6 taxa were recorded in February, while 57 individuals belonging to 5 taxa were recorded in July. In summary, zooplankton species were relatively abundant in wet season (June and July) than during dry season (February and March) (Table 3).

Table 1: Composition and Distribution of zooplankton species in the Great Kwa River during the study period

Comment [R11]: The information in the table shows only the comparison between the collection sites.
Suggested tabulation of data in the following table

Taxonomic group	Species	DrySeason			WetSeason		
		Nº Ind			Nº Ind		
		S1	S2	Totally	S1	S2	Totally
Copepoda	<i>Copepod nauplii</i>	-	-	-	-	-	-
	<i>Tropocyclops prasinus</i>	-	-	-	-	-	-
	<i>Eurytemora</i>	-	-	-	-	-	-
	<i>Bryocaptus beisteinii</i>	-	-	-	-	-	-
	<i>Paracyclops fimbriata</i>	-	-	-	-	-	-

Taxonomic group	Species	No. of species	Individual	
			S1	S2
Copepoda	<i>Copepodnauplii</i>	2 (1.03)		2 (1.03)
	<i>Tropocyclops prasinus</i>	3(1.54)		7(3.59)
	<i>Eurytemora</i>	2(1.03)		2(1.03)
	<i>Bryocaptus beisteinii</i>	1(0.51)		5(2.56)
	<i>Paracyclops fimbriata</i>	1(0.51)		1(0.51)
Nemata	<i>Ethmolaimus americanus</i>	1(0.51)		2(1.03)
	<i>Anonchus monhystera</i>	5(2.56)		7(3.59)
	<i>Bastianaexilis</i>	4(2.05)		4(2.05)
Ostracoda	<i>Ostracod spp.</i>	1(0.51)		8(4.10)
	<i>Limnocythere verrucosa</i>	2(1.03)		3(1.54)
Rotifera	<i>Notholca acuminata</i>	2(1.03)		5(2.56)
	<i>Aspladinaprodonta</i>	1(0.51)		1(0.51)
	<i>Keratella longispina</i>			2(1.03)
	<i>Keratella quadrata</i>			1(0.51)
	<i>Platylabus quadricornis</i>			1(0.51)
Oligochaeta	<i>Pentanueraspp</i>	1(0.51)		2(1.03)
Protozoa	<i>Blephanirisma spp.</i>	1(0.51)		3(1.54)
	<i>Protozoa spp.</i>	4(2.05)		6(3.08)
	<i>Arthrodesmum incus</i>	2(1.03)		5(2.56)
	<i>Centropyxis spp.</i>	1(0.51)		2(1.03)
	<i>Loxodes spp.</i>	1(0.51)		4(2.05)
	<i>Arcella spp.</i>			1(0.51)
	<i>Nebella caudate</i>			2(1.03)
	<i>Trigonopyxis arcuata</i>			1(0.51)
	<i>Plagiopyxis callidis</i>			2(1.03)
	<i>Arcelladiscoides</i>			1(0.51)
	Cladocera	<i>Daphnia magna</i>		
<i>D. rosa</i>		3(1.54)		6(3.08)
<i>D. pulex</i>		1(0.51)		3(1.54)
<i>Conchoecia elegans</i>		2(1.03)		3(1.54)
<i>Gyrinus spp</i>		2(1.03)		7(3.59)
Tintinnida	<i>Ascampbelliella acuta</i>	8(4.10)		29(14.87)
	<i>Coxiella longa</i>			4(2.05)

Comment [R12]: Indicate that all species of Tintinnida found here are marine

	<i>Coxiella mariana</i>	4(2.05)
Diptera	<i>Chironomus spp.</i>	1(0.51)
Foraminifera	<i>Globigerina bulloides</i>	1(0.51)
Atentadata	<i>Bolinopsiscumis</i>	1(0.51)
Trichoptera	<i>Glossosoma spp.</i>	1(0.51)
TOTAL	12	38
		51
		144

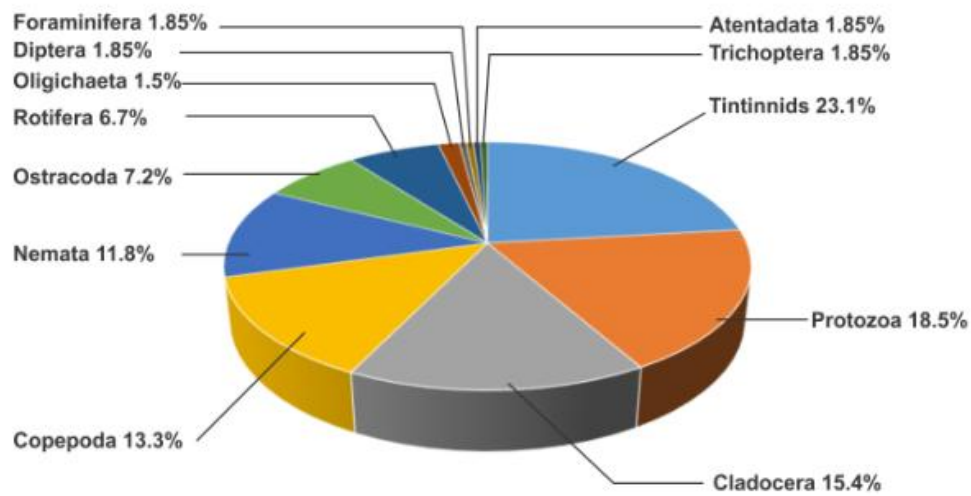


Fig. 2: A chart showing the relative abundance of zooplankton during the period of study

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Table 2: Species diversity indices of zooplankton taxonomic group in the Great Kwa River in relation to sampling stations

Parameters	Sampling stations		Total
	ObufaEsuk (S1)	EsukAtu (S2)	
No of taxonomic group	12	8	
No of individual	51	144	
Margalef's Index (d)	4.111	8.171	12.282
Shannon-wiener Index (H)	2.40	2.997	5.397
Pielou's evenness Index (E)	0.576	0.547	1.123
Simpson's Dominance Index (D)	0.403	1.553	1.956

Comment [R14]: Calculate these indices for seasons

Comment [R15]: Add the values for calculating the constancy of Dajoz.

Comment [R16]: These indices cannot simply be added up. They must be recalculated with all the data, as the number of individuals and their quantities influence the final calculation.

Table 3: Summary of Seasonal distribution of zooplankton in the Great Kwa River for S1 and S2 stations

Month of sampling	Number of individual		Relative Abundance (%)	
	S1	S2	S1	S2
Feb	8	20	15.7	13.9
March	6	18	11.8	12.5
June	16	49	31.4	34.0
July	21	57	41.2	39.6
Total	51	144	100	100

S1- ObufaEsuk, S2- EsukAtu, February and March represent dry season, June and July represent wet season

Comment [R17]: The aim of the work was to present variations related to seasonality. Thus, these data must be separated into seasons. In addition, statistical tests should be carried out to check if there is a difference between the seasons

4. DISCUSSION

Urban development, industrialization around coastal areas and high levels of anthropogenic contaminants from catchment area affect water quality globally [28] and in-turn influences zooplankton availability and diversity. The Great Kwa River displays a typical tropical zooplankton species composition [4,18, 32]. Overall zooplankton species distribution, diversity and abundance in the study sites are influenced by various environmental and biological factors such as water colour, surface area, depth and trophic level, predation, breeding and vertical migration [9-11,15,16-18]. The results of this present study revealed that some common types of freshwater zooplankton are *Cladocera*, *Copepoda*, *Protozoa*, *Tintinnida*, *Nemata*, *Rotifera*; which forms the bulk of zooplankton groups in previous studies [2-4,13-17,29-35] and are in agreement with our present findings. The submissions of other researchers suggested that the availability of these zooplankton species could be as a result of genetic adaptation; coupled with other relevant environmental factors [3,4,36]. The zooplankton composition (abundance and diversity) in this study revealed that the various zooplankton species varied seasonally and spatially. The highest number of zooplankton species were recorded during the wet season (June and July), while the low zooplankton species were recorded in the dry season (February and March) in both study locations. This result is in harmony with other findings in River Ossiomo [10], Opobo river, Rivers State [11], Okhuo River, Edo state [37] and Ehoma lake [38]. The seasonal difference in zooplankton species abundance as observed in this study maybe due to the chemical composition of the water; although not examined in this research. Also, flooding during wet season (June and July) because of high rainfall may contribute positively by recruiting zooplankton from other water bodies; thereby increasing the zooplankton community during wet season. Mixing of high water masses causes high turbidity, this may affect the availability and distribution of zooplankton species [33-35]. The water level in Great Kwa River can be categorized as low to

Comment [R18]: All species in this group found in the present study are marine.

Comment [R19]: What chemical change causes this variation?

moderate during the study period. This same level of water was documented in Ehomalake, Nigeria [38]. The condition may be due to the discharge and draining of water from the mainland of Parliamentary extension (the central dump site in Calabar metropolis), finally ending up in Great Kwa River. This condition may be related to relatively low diversity of zooplankton species. Reports from different studies supported that low zooplankton species availability was affected by discharge of water from mainland, and therefore collaborates with our study [40-45].

Comment [R20]: Indicate this location on the map.
How much discharge and drainage water is released?
Do these waters go through a treatment process?

This information is important for the discussion that the authors present in the manuscript.

The results of this study showed that *Tintinnids* are the most abundance zooplankton (23.1%), followed by other protozoans (18.5%). *Tintinnids* is a type of protozoa found in marine environments belonging to the order Tintinnida [46]. *Tintinnids* represent a group of ciliates belonging to the subclass *Choreotrichida*. *Tintinnids* are essential link between nano-planktons and macro-planktons in the food webs of the marine environment. It is also known that *Tintinnids* are one of the major components of marine planktonic ciliates and has a cosmopolitan character [46]. In Western Philippines, South China Sea, the taxonomic group named *Tintinnids* was reported; although was relatively rare and contributed to only 1% of the total zooplankton identified [31], but in our study high occurrences of *Tintinnids* (23.1%) was documented. Also in Manila-bay, Philippines the taxonomic groups of *Tintinnids* and *Trichoptera* were documented [30], which is in tandem with our present finding in the Great Kwa River. Therefore, *Tintinnids* distribution in our study area formed an important linkage between nano-planktons and macro-planktons in the food webs of the marine environment in Great Kwa River, Calabar. Rotifers species like *Keratella* species (typical of oligotrophic to mesotrophic in nature) were also observed in the study, collaborating with the documented research conducted in Ikwori Lake, central Cross River State, Nigeria. The occurrences of *Protozoa*, *Cladocera*, *Copepoda* and *Rotifer* were observed, which is similar to different documented results [11,47-48]. Specifically,

Comment [R21]: This information is of great relevance. However, in the way the results are presented, it contributes little. To really understand the dynamics that lead a marine species to be found in a freshwater environment, data must be presented more clearly.

For that I suggest:
Present Dajoz's constancy - this way we can see if that happens at specific events or throughout the year.
What stations does it appear the most?

Comment [R22]: It is important that the authors discuss in a more critical way the possible causes that led the *Tintinnids* to be the predominant group in this state.

Copepod nauplii in the *Copepoda* taxonomic group was identified in this study, agreeing to previous reports [31] and they are among the free-living filter feeder zooplankton utilized as in bio-monitors for pollutants in the water bodies [39]. The variations in the number of individual species recorded in these findings may be due to the sampling method and sampling time which are supported by other investigations from different aquatic populations [10,11,18].

The ecological diversity indices in this present study indicated richness and abundance in the distribution of zooplankton (Margalef's index and Shannon-Wiener index) in both stations (ObufaEsuk and EsukAtu), with relatively low evenness or equality in both station; especially in station one from the Pielous evenness index values. The relatively low zooplankton species evenness maybe attributed to high abundance and dominance of some species like *Copepods*, etc [49]. Evenness indices of the zooplankton species in this study was lower than 0.99 – 0.993 values documented by [51] in a Nigerian forest river. The values of the diversity indices in our study showed that zooplankton at station 2 (EsukAtu) were more diverse than station one (ObufaEsuk). The diversity shown by EsukAtu may be due to the presence of phytoplankton at that station; constituting rich food web in the location. This study is similar to other documented researches [32,50] but lower than the value obtained by Ekwu and colleagues [51] in lower Cross River estuaries. Although, the zooplankton community showed diversity; the different species of zooplankton that make up the community were relatively unevenly distributed. This finding is similar with the findings in Opobo River (precisely Opobo town station); that displayed diversity of zooplankton species, but evenness index was 0.825 relatively higher than the values in our study.

Comment [R23]: Explain what causes this point to have a greater amount of phytoplankton.

Numerous mechanisms are associated with zooplankton occurrence, distribution and diversity in an aquatic ecosystem. These mechanisms include intense predation [52], anti-predator refuge effect [53], increased niche availability, food sources [48], and the physical complexity habitat provided by banks of macrophytes; which are positively associated with zooplankton diversity [53]. These mechanisms may have contributed to the distributions and diversity of these zooplankton species identified in this study during the duration of the study.

5 CONCLUSION

This present study provide updated information on the zooplankton distribution, diversity and seasonal variations of the Great Kwa River. The results revealed 12 major taxonomic groups including *Tintinnida* consisting of 23.1%, followed by other *protozoa* (18.5%), *Cladocera* (15.4%), *Copepoda* (13.3%) and the lowest taxonomic groups were *Diptera*, *Foraminitera*, *Atenadata* and *Trichoptera* having only one representative with 1.85% for each order of total abundance of zooplankton species. The highest number of species was recorded in Esukatu (S2) station with 31 species represented during the research period. In both Stations, *Ascampbelliella acuta* displayed high dominance over other species. The abundance of Zooplankton species in the river varies seasonally, with more species recruited during the wet season. The high dominance of the *Tintinnida* (*Ascampbelliella acuta*) in our current study indicates that the river is a natural important linkage between nano- planktons and macro-planktons in the food webs of the marine environment.

6. RECOMMENDATIONS

Comment [R24]: Very superficial results to cover this claim

The following recommendations are suggested:

- a. The government of Cross River State should establish a waste treatment facility to control and reduce the level of waste disposed in open surface areas (especially in the Calabar central dump site located at Parliamentary Extension); which finally ends-up in the aquatic ecosystem causing contaminations.
- b. Proper environmental monitoring should be put in place to check sporadic behavior of direct dumping of untreated waste into water bodies; to reduction aquatic/water pollution in the Great Kwa River.

Comment [R25]: This information did not appear in the discussion. What are the reasons that led the authors to indicate these problems as causing changes in the structure of the zooplankton of Rio Great Kwa.

Limitations of the study

Genetic and molecular characterization of zooplanktons were not carried out in this study.

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