Evaluation of Metal Pollutants and Bioaccumulation Factors in *Macrobrachium macrobrachium* and *Sarotherodon melanotheron* from Porto-Novo Lagoon, Benin Republic

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Abstract: **- Significant distribution of metal pollutants and bioaccumulation factors in *Macrobrachium macrobrachium* and *Sarotherodon melanotheron* from Porto-Novo lagoon ecosystem was studied during two hydrological periods. Metal pollutants such as Hg, Cd, Cu, Zn, Cr, Fe, Mn, Pb, Ni, Va, and Methyl mercury were found in measurable quantities in the test animals using AAS equipment for analyses and are found to be above regulatory agency permissible limits. Tissues and organs of *M. macrobrachium* had less than 1.00 bioaccumulation factors in Cd, Fe, Mn, Ni, and MH_3Hg while bioaccumulation factors greater than 1.00 are found in Hg, Cu, Zn, Cr, Va, and Pb . Tissue and organs of *S. melanotheron* had less than1.00 bioaccumulation factor in Hg, Cd, Cu, Zn, Fe, Mn, Pb, Ni, and MH_3Hg while, Cr and Va had greater than 1.00 BF. There is a significant difference in the means of metal pollutants in the tissues and organs of *M. macrobrachium* during dry season at 5% level of significance. There is no significant difference in the means of metal pollutants in the tissues and organs of *S. melanotheron* at 5% level of significance in both seasons. The results indicate that *M. macrobrachium* and *S. melanotheron* from Porto-Novo lagoon bio-accumulate metal pollutants that are detrimental to human health s well as affecting the physiology of these test animals in the Porto-Novo lagoon ecosystem. It is therefore recommended that Porto-Novo lagoon should be put on National Priority List for adequate revamping.**

Keywords **– Metal, Pollutants, *Macrobrachium macrobrachium* , *Sarotherodon*, *melanotheron* Porto-Novo**

I. INTRODUCTION

One of the major environmental challenges the world is facing at the moment is that of ecological degradation with the attendant increase in the release of pollutants to the environment that is momentarily instigating irreversible damage to the earth. Environmental pollution has been on increase over the recent years as a result of population explosion, the industrial revolution and the aesthetic lifestyle brought about by innovation and technological advancement. These, however opened the floodgate of pollution and introduced lethal substances into the environment beyond the capacity the environment can curtail [2].

Aquatic pollution has been significantly on increase over the recent years and has become major problems in many developing countries. Urban and industrial activities in coastal areas introduce greater amount of metal pollutants into the aquatic environment causing permanent damages in aquatic ecosystems and thereby leading to environmental and ecological degradation and constitute a potential risk to a number of aquatic flora and fauna species, and humans through food chains [1]. Aquatic pollution takes place when there is an emancipation of pollutants into water bodies either directly or indirectly without sufficient treatment to remove detrimental compounds. The sources of these pollutants are majorly from effluents discharge from industries and metropolitan cities. These pollutants have been the leading worldwide cause of death and diseases in both human and animals [11].

The rate of bio-accumulation of metal pollutants in aquatic organisms depends on the ability of the organisms to digest the metal pollutants and the concentration of such metal pollutants in the water and in the surrounding soil sediments as well as the feeding habits of the organism. Aquatic animal bio-accumulates metal pollutants in considerable amounts and stay over a long period. Aquatic organisms are known as good accumulators of organic and inorganic pollutants [15] which can be determined by accumulation factor.

According to[27], a bioaccumulation factor greater than 1.00 shows that the bioaccumulation of metal pollutants in the tissues and organs are detrimental to both the health of the organisms and human thus, bioaccumulation Factor (BAF) is calculated as the ratio of the concentration of a pollutant (heavy metal) accumulated in the tissues and organs of an organism with respect to the concentration of that pollutant (heavy metal) in the water body [21].The effect of trace metals in aquatic environment could be beneficial or and can also lead to death of aquatic species. Some trace metals such as Cu, Mn, Fe and Zn are vital for good vigor and normal growth playing important roles in key metabolic activities of aquatic organisms such essential elements only become lethal when their concentrations surpass the trace amounts required for normal metabolism [20].

The complex Porto-Novo lagoon is located in Republic of Benin and it is one of the largest lagoons in West Africa with high productivities and exploitation [29]. There are about 72 species of both fin and shell fishes in the Port-Novo Lagoon [19]. Several species are common to these lagoons (figure. 1).

Macrobrachium macrobrachium is common in fresh waters, brackish waters and in estuarine areas of the coastal zone of Southern Benin. Artisanal fisheries occur in the Oueme River, Mono River and their floodplains, Lake Nokoue, Lagoon of Porto-Novo, Lake Aheme, and the coastal lagoons where these prawns are intensively exploited for sales because of their high market demand and commercial value though underestimated. Yearly production in Benin exceeds 200 tons [3]. Prawns are known to feed on a wide variety of small epibenthic animals, especially polychaetes, mollusks and other crustaceans.

The black-chinned tilapia, *Sarotherodon melanotheron* (Cichlidae), is another major fish found in West African coastal waters. Cichlids are mostly freshwater species but *S. melanotheron* is generally found in estuaries and lagoons and occasionally in the mouth and the lower course of coastal basins. The fish is a filter feeder and also feeds on a wide range of plankton [19].

The geographical positioning and location of the complex Porto-Novo Lagoon makes it to become a receptacle in which all rejected water from gutter, sewage, waste water from surrounding cities such as Cotonou, Abomey-Calavi, So-Ava are emptied and also urban concentrations of population due to increased urbanization, anthropogenic activities and garbage deposits along the lagoon banks are the main cause of the complex pollution by trace metals and other toxic substances [29].

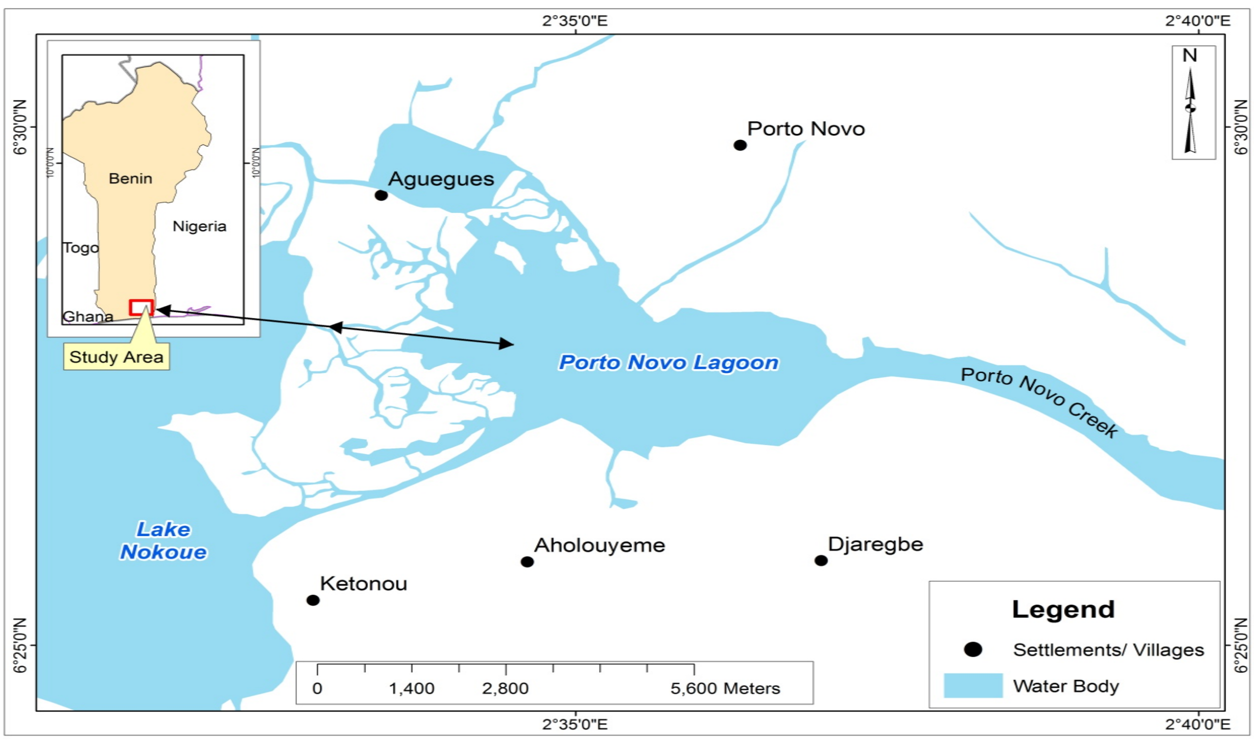

Figure 1: Location of Porto-Novo lagoon showing the study area. Source: [7]

II. MATERIALS AND METHODS

Study Site

The Lagoon of Porto-Novo is located in the South-East of the Republic of Benin (6º 27'N, 2º 36'E) and a basin of up to 6 meters. Porto-Novo lagoon has a surface area of 17.52 km^2 and maximum length of 6km and maximum width of 4km (Figure. 1) and it is linked to Nokoue lagoon in the east through tatche where it receives the major inflow which characterizes the main hydrological regime of Porto-Novo lagoon seasonal tidal and salinity range [29].

Sampling Collection

Data were collected for 12 months, between July 2014 and June 2015 representing a complete hydrological cycle. A stratified random sampling method was adopted in collection of mixed sexes of *Macrobrachium macrobrachium* and *Sarotherodon melanotheron* of mean weight of 5.5g and 10.5g respectively from Djassin main fish landing site together with fish market. Test animal samples were kept in a tagged polythene bags respectively and stored in an iced cooler (<4°c) for the analyses of selected metal pollutants as cited by [2]. Metal pollutants that were investigated during the period in view are Hg, Cd, Cu, Zn, Cr, Fe, Mn, Pb, Ni, Va, and methyl mercury. All chain of custody was duly observed [10].

Determination of metal pollutants

The methods of [14] and [6] were used to determine levels of trace metals in fish tissues. Digestions of test animal tissues and organs for metal pollutants were carried out separately using Atomic Absorption Spectrophotometer (Model Thermo Electron Corporation, S series AA Spectrometer with Gravities furnace, UK) as described by (AOAC, 1990). Twelve samples each of *Macrobrachium macrobrachium* and *Sarotherodon melanotheron* of average weight of 5.5g and 10.5g respectively were collected from commercial landing site and transported in iced box cooler for digestion and AAS laboratory procedures. Tissues and organs of interest are weighed. 5g each of tissue/organ sample from all the samples were weighed and put in a conical flask of about 250ml capacity then added 25ml of digesting reagent (Nitric acid and Hydrogen peroxide) in ratio 1:1 and place on a hot plate under the fume cupboard and heat at a temperature not more than 120^0c .When the sample is about 5ml in the conical flask and allow to cool filter the sample using the whatman 1 filter paper into a 50ml standard volumetric flask then make up to mark and Set the sample for AAS (calibrated) for Trace Metals reading.

Statistical analyses:

The bioaccumulation of metal pollutants in the test animals during two hydrological periods were subjected to two-way analysis of variance (ANOVA). The differences between the means were determined using randomized complete block design (RCBD) with the aid of SPSS version 22 statistical package. All the hypotheses are tested at 5% level of significance (i.e $\alpha = 0.05$).

III. RESULTS

Macrobrachium macrobrachium

Table 1 shows metal pollutants in the tissues and organs of *Macrobrachium macrobrachium* and presented in descending order of magnitude as follows; Fe=125.450±31.818 mg/kg, Cr=64.872±16.233 mg/kg, Cu=52.241±32.127 mg/kg, Zn =50.102±30.397 mg/kg, Pb =47.565±9.684 mg/kg, Ni= 16.618±2.567 mg/kg, Mn = 8.069±7.907 mg/kg, Cd=0.228±0.026 mg/kg, Va=0.005±0.002 mg/kg, Hg=0.002±0.001 mg/kg, MH_3Hg=0.000±0.000 mg/kg. The means ranged between 19.637±24.895 mg/kg - 48.507±54.49 mg/kg of metal pollutant as observed in *Macrobrachium macrobrachium.* However, between July and December there was no catch on *Macrobrachium macrobrachium* due to zero salinity. There is a significant difference in the means of metal pollutants in the tissues and organs of *Macrobrachium macrobrachium* between January and June, at 5% level of significance. During the study periods, *Macrobrachium macrobrachium* had less than 1.00 bioaccumulation factors in the following metal pollutants; Cd, Fe, Mn, Ni, and MH_3Hg while Hg, Cu, Zn, Cr, Va, and Pb had bioaccumulation factors greater than 1.00 in their tissues and organs. It was observed from the metal pollutants bioaccumulation factors in *Macrobrachium macrobrachium* had the bioaccumulation factors greatly influenced by fish ecology, season and types of metal pollutants.

TABLE 1: Mean value of metal pollutants in *Macrobrachiu macrobrachium* between January and June 2015

Metal pollutants (mg/Kg)	Fish tissue/organ						Agency's permissible levels			
	Gill	Hepato - Pancreas	Bone	Gut	Muscle	Mean(±SD)	WHO /FAO (mg/day)	USFDA Limit (mg kg-1)	EU regulation (mg kg-1)	NRS (mg kg-1)
Hg	0.003	0.003	0.001	0.001	0.002	0.002±0.001^c	0.5	0.5	0.5	0.5
Cd	0.240	0.260	0.193	0.210	0.237	0.228±0.026^c	0.1	0.5	0.05	N/A
Cu	31.763	96.867	38.230	74.283	20.060	52.241±32.127^b	3.0	70	N/A	0.5
Zn	22.400	93.767	46.347	65.600	22.397	50.102±30.397^b	60.0	30	N/A	N/A
Cr	60.643	82.280	78.203	61.717	41.517	64.872±16.233^b	50	N/A	N/A	N/A
Fe	114.923	162.157	124.137	146.783	79.250	125.450±31.818^a	43.0	N/A	N/A	N/A
Mn	2.467	17.993	15.237	3.517	1.133	8.069±7.907^c	2.0-9.0	5.4	N/A	N/A
Pb	47.650	59.937	53.720	40.600	35.917	47.565±9.684^b	0.214	0.5	0.4	0.5
Ni	14.123	20.303	18.213	14.967	15.483	16.618±2.567^c	2.45	N/A	N/A	N/A
Va	0.005	0.007	0.002	0.005	0.007	0.005±0.002^c	0.5	N/A	N/A	N/A
MH_3Hg	0.001	0.001	0.000	0.000	0.001	0.000±0.000^c	0.0033	0.001	0.0016	0.0005
Mean (±SD)	26.75 ±36.06^{bc}	48.51 ±54.49^a	34.03 ±39.81^b	37.06 ±46.85^{ab}	19.64 ±24.90^c	33.20 ±41.26				

Sarotherodon melanotheron

The bio-concentration of metal pollutant in *Sarotherodon melanotheron* during the two hydrological periods from July to December,2014 and from January to June, 2015 are shown in Tables 2 and 3respectively and it follows this trend, Fe >, Zn> , Cr>, Pb>, Ni >, Cu >, Cd >, Mn >, Va >, Hg >, MH3Hg (between July and December) while it follows this trends, Cr >, Fe >, Mn >, Ni >, Zn >, Cu >, Pb >, Cd >, Va >, Hg >, MH_3Hg.(between January and June). *Sarotherodon melanotheron* tissues and organs had the highest bioaccumulation of Fe (127.552±101.705 mg/kg) between July and December while the highest bioaccumulation recorded from January to June was in Cr (1238.046±2257.93 mg/kg). MH_3Hg had the lowest concentrations with the mean value of 0.001±0.000 mg/kg in the organs and tissues of *Sarotherodon melanotheron* during the two hydrological cycles. There is no significant difference in the means of metal pollutants in the tissues and organs of *Sarotheredon melanotheron* between July and December at 5% level of significance. Also, there is no significant difference in the means of metal pollutants in the tissues and organs of *Sarotheredon melanotheron* between January and June at 5% level of significance. The bioaccumulation factors during the investigation revealed the relationship that exist between the three factors variance of metal pollutant species, the ecology of *Sarotherodon melanotheron,* and season of the year. *Sarotherodon melanotheron* had less than1.00 bioaccumulation factor in Hg, Cd, Cu, Zn, Fe, Mn, Pb, Ni, and MH_3Hg while, Cr and Va had greater than 1.00.

TABLE 2: Mean value of metal pollutants in *Sarotheredon melanotheron* between July and December 2014

Metal pollutants (mg/Kg)	Fish tissue/organ						Agency's permissible levels			
	Gill	Liver	Bone	Gut	Muscle	Mean (±SD)	WHO /FAO (mg/day)	USFDA limit (mg kg-1)	EU regulation (mg kg-1)	NRS (mg kg-1)
Hg	0.0047	0.0052	0.0029	0.0039	0.0031	0.004±0.001[b]	0.5	0.5	0.5	0.5
Cd	1.21	1.126667	0.986667	0.843333	0.925	1.018±0.149[b]	0.1	0.5	0.05	N/A
Cu	0.806667	14.06	0.643833	6.66	0.756667	4.585±5.885[b]	3.0	70	N/A	0.5
Zn	54.62667	37.16	27.49167	26.69333	16.5667	32.508±14.352[b]	60.0	30	N/A	N/A
Cr	21.665	38.125	32.30167	27.99667	16.41667	27.301±8.557[b]	50	N/A	N/A	N/A
Fe	258.7733	148.2933	23.797	180.2583	26.64	127.552±101.705[a]	43.0	N/A	N/A	N/A
Mn	1.099	0.378333	0.151667	0.126667	0.154667	0.382±0.414[b]	2.0-9.0	5.4	N/A	N/A
Pb	15.245	27.25	32.58333	19.31833	30.25667	24.931±7.376[b]	0.214	0.5	0.4	0.5
Ni	4.241667	4.253333	12.43333	10.7	12.77	8.880±4.301[b]	2.45	N/A	N/A	N/A
Va	0.017917	0.016833	0.011667	0.008167	0.011667	0.013±0.004[b]	0.5	N/A	N/A	N/A
MH_3Hg	0.0012	0.0013	0.001	0.001	0.001	0.001±0.000[b]	0.0033	0.001	0.0016	0.0005
Mean (±SD)	32.52 ±76.837[a]	24.61 ±43.73[a]	11.86 ±14.27[a]	24.78 ±52.69a	9.50 ±11.58 a	20.65 ±45.83				

***N/A= Not available**

TABLE 3: Mean value of metal pollutants in *Sarotheredon melanotheron* between January and June 2015

Metal pollutants (mg/Kg)	Fish tissue/organ						Agency's permissible levels			
	Gill	Liver	Bone	Gut	Muscle	Mean(±SD)	WHO /FAO (mg/day)	USFDA Limit (mg kg-1)	EU regulation (mg kg-1)	NRS (mg kg-1)
Hg	0.0016	0.0017	0.0012	0.0013	0.0011	0.001±0.000[b]	0.5	0.5	0.5	0.5
Cd	0.0025	0.0011	0.0036	0.0046	0.0122	0.005±0.004[b]	0.1	0.5	0.05	N/A
Cu	1.109	1.6989	0.234	1.6484	0.302	0.998±0.71[b]	3.0	70	N/A	0.5
Zn	2.08	2.16	1.63	1.67	1.89	1.886±0.237[b]	60.0	30	N/A	N/A
Cr	1.733	1.002	0.2461	982.25	5205	1238.046±2257.93[a]	50	N/A	N/A	N/A
Fe	102.77	1906.25	16.275	196.48	914.42	627.239±798.79[a]	43.0	N/A	N/A	N/A
Mn	49.207	18.538	9.058	1.709	6.251	16.953±19.05[b]	2.0-9.0	5.4	N/A	N/A
Pb	0.1067	1.01	0.076	0.00	0.0184	0.242±0.43[b]	0.214	0.5	0.4	0.5
Ni	1.079	1.063	0.079	63.286	5.563	14.214±27.51[b]	2.45	N/A	N/A	N/A
Va	0.006	0.006	0.004	0.0027	0.004	0.005±0.001[b]	0.5	N/A	N/A	N/A
MH_3Hg	0.0004	0.0004	0.0003	0.0003	0.0003	0.000±0.000[b]	0.0033	0.001	0.0016	0.0005
Mean (±SD)	14.37 ±32.74 [a]	175.61 ±574.01[a]	2.51 ±5.295[a]	113.37 ±294.29[a]	557.59 ±1565.5[a]	172.69 ±756.80				

*N/A= Not available

IV. DISCUSSION

The rapid industrialization arising from modern and sophisticated technology have introduced some pollutant materials into our environment especially our aquatic ecosystem which can cause some health challenges to both human and aquatic organisms. Some of these pollutants are trace metals which are found in various lethal concentrations in the lagoon water and in some economically important fisheries resources such as: *Macrobrachium macrobrachium* and *Sarotheredon melanotheron*.

The elevated mean concentrations of Fe (Iron) (125.450 mg/kg) from all the metal pollutants investigated in *Macrobrachium macrobrachium* might be the heterogeneous nature of the lagoon sediment. Sediments that are rich in textural clay posseses high tendency of Fe (Iron) attraction and sedimentation that will eventually released into surrounding water for the uptake of *Macrobrachium macrobrachium,* this corroborate the finding of [21] on the study of heavy metals in the environment.

Porto-Novo lagoon being repository of water from the gutter, sewage, wastewater from surrounding cities, the biodegradable organic pollutants and non-biodegradable organic pollutants in these polluted water are one of the materials responsible for the prominent bioaccumulation of some metal pollutants found in *Macrobrachium macrobrachium* tissues and organs, this agreed with the report of [17] on trace elements trans-bioaccumulation in the environment.

In the present study, Hepatopancreas in *Macrobrachium macrobrachium* tissues and organs had the highest means of all the metal pollutants, this parhaps might be due to physiological function of this particular organ has reiterated by [16] on the presence of sulfide bonds and the digestive enzymes in hepatopancreas that are needed for the detoxification process. The secretion of sulfide bonds and the digestive enzymes possess greater propensity to attract some metal pollutants species from the animal's environment. The relationship between metal pollutants in the sediment and the organs of *Macrobrachium macrobrachium* is positively correlated and significant at 5% level of probability, this is as a result of food and feeding habit of *Macrobrachium macrobrachium* which feeds on macro- invertebrates benthos such as: epibenthic animals, especially polychaetes, mollusks and other crustaceans that are benthic organisms. These organisms accumulate pollutants from the sediments [12] and [3].

The bioaccumualtion factors(BAF) of the metal pollutants in *Macrobrachium macrobrachium* are greater than 1.00 [27]. The elevated occurrence of Copper which is an essential metal *Macrobrachium macrobrachium* contradicts the findings of [25] on various metal pollutants interactions in shellfish, the author proposed that Copper, an essential metal is thought to undergo regulatory concentration mechanism in the tissue of shellfish and this could be for tissue bio-mineralization processes. There is no catch of *Macrobrachium macrobrachium* from Porto-Novo Lagoon hence no calculated value for BAF between July-December due zero salinity in the Lagoon water during the study period because Macrobrachium *macrobrachium* cannot withstand zero salinity therefore migrated from Porto-Novo lagoon water. [13]. The result showed that metal pollutants concentration in the tissues and organs of *Macrobrachium macrobrachium* from Porto-Novo lagoon ecosystem are higher than the surrounding water and it is therefore not fit for human consumption.

The seasonality and inconsistency in the sequential order of occurrence in metal pollutants bioaccumulation in the tissues and organs of *Sarotheredon melanotheron* could be associated with the heterogeneous nature of complex Porto-Novo Lagoon ecosystem [8]. The seasonal variation of salinity in the complex Porto-Novo lagoon ecosystem might account for the abundance or scarcity of some euryhaline and stenohaline zooplankton and algae such as: Cyanophyceae, Chlorophyceae, Rotifera, Cladocera, Bacillariophyceae, and Cyanophyceae that forms major food basis for *Sarotheredon melanotheron*. The ingestion of these metal pollutants-laden food items by this filter feeder that is, *Sarotheredon melanotheron* which feeds on a wide range of plankton at different trophic levels might be responsible for these different concentrations of metal pollutants in the organs and tissues as reiterated by[12] and [23] respectively.

Sarotheredon melanotheron had the highest concentrations of all the metal pollutants in the gill during rainy season, this might not be unconnected with filter feeding capability of *Sarotheredon melanotheron* this filtration is enhanced by high volume of water in the Lagoon that are gotten from precipitation and run-off into the Lagoon. Also, [24] as cited by [16] reiterated that the first point of entry of metal pollutants occurs through the gills.

The concentrations of metal pollutants in the muscle of *Sarotherodon melanotheron* during the dry season might be due to strong speciation influence of geochemistry and bioavailability of trace metals in the Lagoon due to high temperature and low level of water as reported by [9].

There is a significant positive correlation at 5% level of probability in metal pollutants in the Lagoon water and organs of *Sarotherodon melanotheron* during the rainy season but not significant during the dry season though positive. This is an indication that during the rainy season, there is an increase in the metal pollutants concentration due to massive inland run-off to the Lagoon [4].

There are similarities in the values of BAF in tissues and organs of *Sarotherodon melanotheron* during the two hydrological periods, this could be due to the feeding habitat of *Sarotherodon melanotheron* and concentrations of metal pollutants in the surrounding water. The result from Bioaccumulation factors of *Sarotherodon melanotheron* in the sediment and water from Porto- Novo lagoon ecosystem proposed that Cichlids family could be used as a biomonitor agent in monitoring accumulation of some metal pollutants in the aquatic environment [30]. In addition, *Sarotherodon melanotheron* has been known to be a good accumulator of organic and inorganic pollutants which can be influenced by age of the fish, lipid contents and concentration in the tissues and organs, and interaction of these metal pollutants in the environment as reported by [18]. Bioaccumulation of essential and non- essential metal pollutants are greater than BAF 1.00 in *Sarotherodon melanotheron*, therefore *Sarotherodon melanotheron* is considered unhealthy for human consumption as recommended by [28].

V. CONCLUSION AND RECOMMENDATION

Macrobrachium macrobrachium and *Sarotherodon melanotheron* from complex Porto-Novo lagoon ecosystem are heavily polluted with heterogeneous species of metal pollutants, although some metal pollutants were found to be in traces that might overtime become higher in concentrations than recommended standard by regualtory bodies which could be detrimental to human consumers of the fisheries resources from such aquatic environment. Consiquently, each metal species concentrations are dependent on the three factors variance: metal pollutant , the ecology of test animals, and season of the year. It is therefore recommended that close season should be declared by the government regulatory agency on Environmental Protection on Porto-Novo Lagoon for adequate environmental revamping and subsequent enforcing of environmental protection regulations.

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