Biochemical Changes in Mudskipper (*Periophthalmus papilio*) Exposed to Sodium Bromide

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ABSTRACT: One hundred and eighty (180) mudskipper (*Periophthalmus papilio*) of equal size (mean length 18.74 ± 2.64cm and mean weight 156.68 ± 1.81g) were exposed to different concentrations (0.0mg/l – control, 5.0, 10.0, 15.0, 20.0 and 25.0mg/l) of biocide, sodium bromide to determine the effects of this chemical on biochemical changes in this specie. The results obtained indicated that the values of lipids, cholesterol, glycogen and total protein analyzed in all organs reduced significantly with increase in biocide concentrations. The data obtained from this work will contribute to the base line biochemical parameters for use in health monitoring of aquatic organism.

Key Words: Fish, Toxicity, Mudskipper, Chemical, Sodium Bromide

I.INTRODUCTION

In response to the input of anti-fouling agents and series of toxicants into aquatic ecosystem, the sodium bromide contamination of water presents a very serious environmental concern [(1-2)]. Therefore, evaluation of the risk associated with the occurrence of the sodium bromide in the fresh and salt water environment and their monitoring in the brackish waters are necessary. This allows assessment of chemical toxicity to aquatic biota in these water bodies. However, in recent years there has been an increasing perception of the need to identify and evaluate the adverse effects of toxicants in aquatic environments. The biological responses (Biomarkers) of aquatic life may signal the comprehensive effects of contaminant impacts on the water body. This can also be used to compare relative alterations in water quality in different locations over a period of time[(3)]. In recent times, the use of biomarkers as veritable tools for assessing environmental quality has become important. And is generally acceptable in the assessments of aquatic pollution in many places around the world[(4-5)]. Biomarkers complement and enhance the reliability of the chemical analysis data, offering more integral and biologically relevant information on the potential impacts of toxic pollutants on the health of organisms especially fish [(6)]. Therefore, the use of biomarkers can offer an integrated evaluation of the effect of toxicant in both artificial and natural environment and give a clear picture of the "health status" of a system under investigation[(7)]. According to Akinrotimi *et al.* [(8)] knowledge of the physiological action of the toxicant helps to predict lethal and sub lethal effects of toxicants in fish

Mudskipper *Periophthalmus Papilio* is an amphibious fish that is very active during low tides and spend most of its time out of water in mangrove habitats. According to Akinrotimi *et al.* [(9)], they form a high density on tidal mud flats in the coastal areas of Niger Delta. This species is widely distributed and notably present in many riverine communities in this region, thus, it is essential to evaluate the toxicity of biocides in the fish, as many research activities in coastal environment have been focusing on other species.

II. EXPERIMENTAL WORK

EXPERIMENTAL PROCEDURE

The experiment was carried out at the brackish water research station of African Regional Aquaculture Centre, Buguma, Rivers State. Seven hundred and twenty (720) adult, mudskipper *P.Papilio* of equal size (mean length 18.74±2.64cm and mean weight 156.68±1.81g) was collected from the mangrove swamps in Buguma creek at low tide using locally made fishing trap. They were then transferred in six 50 litre plastic tanks to the laboratory for acclimation process. 10 Mudskippers each were introduced individually into 18, dug earthen pond of dimension 1.5m x 1m x 0.5m containing concentration of biocide in 0.0 (control), 0.5, 1.0, 1.5, 2.0 and 2.5m/L^{-1} of sodium bromide. Each treatment(s) and control was replicated three times and the experimental duration lasted for a period of 30 days. The solution for each concentration was renewed daily, with freshly prepared solution of biocide. The tanks were covered with netted materials and supported with heavy objects to prevent the mudskipper from escaping.

SAMPLE COLLECTION AND DETERMINATION OF BIOCHEMICAL PARAMETERS

At the end of the experiments, blood samples were collected from the fish. Also fish were sacrificed after blood collection and dissected to collect samples of the gill, liver, kidney, muscle and gastrointestinal tract for enzyme, electrolyte and metabolite analysis. The concentrations of total cholesterol, lipids, and glycogen were determined by enzymatic colorimetric test described by Begum .[(3)]. While the total protein level in serum was determined by the biuret method as described by Begum [(3)], using a commercial clinical investigation kit (Fortress Diagnostic Analysis, United Kingdom). All biochemical analyses were carried out Lively Stone Laboratory, Choba, Rivers State, Nigeria.

STATISTICAL ANALYSIS

Data obtained from the experiments were subjected to ANOVA using Statistical Package for the Social Sciences, (SPSS) version 10 and differences among means were separated by Duncan Multiple Range test at 0.05%. The dependent variables in the trials biochemical indices were compared based on the concentration of the toxicants.

III.RESULTS AND DISCUSSION

Variations in some biochemical parameters such as cholesterol, lipids, glycogen and total protein in the kidney of *P. Papilio* are highlighted in Table 1. All the biochemical parameters reduced considerably with increasing concentration of the chemical. In the muscle, the same trend of significant reduction were observed in all biochemical parameters, lower values were however, recorded in glycogen and total protein (Table 2). In the liver, consistent reduction with increasing concentrations of the chemical were recorded for all parameters, with the lowest reduction value 6.42±0.81 to 0.12±0.01 in glycogen (Table 3). In the gill and GIT, the biochemical parameters also reduced considerably as the concentrations of the chemical increased with lower values observed in glycogen and total protein (Table 4 and 5). The relative levels of lipid in the organs of *P.Papilio* treated with sodium bromide revealed that the highest levels of lipid was found in GIT closely followed by liver and muscle, while the lowest was in the kidney (Figure 1). The cholesterol level in various organs of *P.Papilio* exposed to sodium bromide was shown in Figure 2. Lipid levels reduced consistently in all the organs, except in kidney where lipid levels were found to be elevated. The glycogen and total protein levels in various organs also reduced significantly as the concentrations of the chemical increased (Figures 3 and 4).

Exposure to sublethal concentration of Sodium Bromide caused significant alterations in the biochemical parameters of the fish *P.papilio*. Cholesterol level decreased considerably in all the tissue and organs except in the kidney. Sodium bromide toxicity causes damage and blockage of enzyme system for steroidogenesis in ovary and also caused damage in liver . Due to this damage, the cholesterol level decreased in the entire organ. Similar results were observed by Oruc and Usta [(10)] in common carp (*Cyprinus carpio*) exposed to diazinon. Also Remia *et al.* [(11)] observed a decrease in cholesterol level of *Tilapia mossambica* treated with insecticide

Moreover, James *et al.* [(12)] equally studied the effect of sublethal doses of lead on the blood, and organs of fresh water fish *Oreochromis niloticus* and observed decreased level of cholesterol. Lipid profile analysis has been reported as a major contribution to the loss of cell function under oxidative stress condition, and is usually indicated as lipid in tissues and organs of fish [(13)]. Considering the typical reaction during toxicant induced damage of cells in fish system involve peroxidation of unsaturated fatty acids, the results of this study clearly showed that sodium bromide exposure to fish for 30days led to oxidative stress, with lipid levels reducing constituently in all the organs. This result is in tandem with that of Remyla *et al.* [(14)] in *Catla catla* after long-term exposure to cadmium. The reduction which was more pronounced in the muscle may be due to direct attacks on protein which lead to the formation of carbonyl. As the formation of carbonyl protein is non-reversible, resulting in normal protein metabolism disrupted and accumulation of damaged molecules.

Table 1: changes in the level of some Biochemical Constitutes in the Kidney of Mudskipper treated with Sodium Bromide *P.pailio* (Mean± S D)

Concentration of (mg/L)	Biochemical Indices (MMOI/L)			
	Cholesterol	Lipids	Glycogen	Total Protein
0.00	110.64±8.61[a]	32.61±2.12[c]	6.12±0.84[d]	6.24±0.64[f]
0.50	114.21±6.01[a]	30.71±1.21[c]	5.71.±0.61[c]	5.11±0.81[e]
1.00	141.62±4.63[ab]	26.11±2.66[b]	5.01±0.81[c]	4.71±0.22[d]
1.50	193.74±2.61[c]	21.61±1.82[b]	4.91±0.62[c]	3.61±0.14[c]
2.00	186.76±1.12[c]	20.88±2.11[b]	3.78±0.82[b]	2.79±0.22[b]
2.50	150.81±2.61[ab]	16.61±3.12[a]	2.62±0.61[a]	1.71±0.32[a]

Means with the same superscript in the column are not significantly different (p›0.05).

Table 2: changes in the level of some Biochemical Constitutes in the Kidney of Mudskipper treated with Sodium Bromide *P.pailio* (Mean± S D).

Concentration of (mg/L)	Biochemical Indices (MMOI/L)			
	Cholesterol	Lipids	Glycogen	Total Protein
0.00	112.63±8.91[e]	65.67±2.19[e]	6.99±0.26[d]	6.49±0.26[c]
0.50	100.91±6.21[e]	59.61±1.34[d]	5.09±0.14[c]	6.02±0.11[c]
1.00	89.61±7.31[d]	47.88±2.14[c]	4.16±0.22[c]	5.78±0.19[b]
1.50	64.71±6.21[c]	33.65±1.29[b]	3.18±0.35[b]	5.01±0.21[b]
2.00	147.89±7.22[b]	27.17±2.10[a]	2.16±0.28[b]	4.72±0.17[a]
2.50	31.79±7.14[a]	24.60±2.16[a]	0.49±0.11[a]	4.18±0.19[a]

Means with the same superscript in the column are not significantly different (p›0.05).

Table 3: changes in the level of some Biochemical Constitutes in the liver of Mudskipper treated with Sodium Bromide *P.pailio* (Mean± S D).

Concentration of (mg/L)	Biochemical indices (MMMOI/L)			
	Cholesterol	Lipids	Glycogen	Total Protein
0.00	110.49±6.12[c]	70.88±2.01[c]	6.42±0.81[c]	6.42±0.41[c]
0.50	96.41±3.19[b]	62.71±0.81[b]	4.02±0.11[b]	6.31±0.21[c]
1.00	84.71±2.66[b]	51.61±0.81[b]	3.11±0.21[b]	5.71±0.81[b]
1.50	78.61±1.69[b]	42.71±1.21[a]	2.19±0.11[a]	5.06±0.21[b]
2.00	61.72±1.78[a]	638.66±1.08[b]	1.81±0.11[a]	4.71±0.14[a]
2.50	51.91±1.96[a]	31.71±1.78[a]	0.12±0.01[a]	4.02±0.22[a]

Means with the same superscript in the column are not significantly different (p›0.05).

Table 4: changes in the level of some Biochemical Constitutes in the Gill of Mudskipper *P.pailio*

Concentration of (mg/L)	Biochemical indices (MMMOI/L)			
	Cholesterol	Lipids	Glycogen	Total Protein
0.00	101.66±11.61[d]	40.81±3.16[d]	4.61±1.11[d]	4.99±0.64[c]
0.50	90.71±7.86[c]	36.11±2.14[c]	3.66±1.01[c]	4.71±0.24[c]
1.00	82.66±6.84[b]	29.61±1.21[b]	2.81±1.01[b]	3.61±0.18[b]
1.50	62.71±7.14[b]	27.48±2.11[b]	2.01±1.02[b]	3.88±0.27[b]
2.00	51.62±7.61[a]	24.62±3.10[b]	1.97±0.08[a]	3.64±0.14[b]
2.50	40.71±8.66[a]	19.71±2.14[a]	1.90±0.18[a]	2.01±1.11[a]

Means with the same superscript in the column are not significantly different (p›0.05).

Table 5: changes in the level of some Biochemical Constitutes in the Gill of Mudskipper *P.pailio* (Mean± S D).

Concentration of (mg/L)	Biochemical indices (MMMOI/L)			
	Cholesterol	Lipids	Glycogen	Total Protein
0.00	126.71±21.68[d]	86.41±6.33[d]	8.78±1.11[c]	9.66±0.81[b]
0.50	108.94±19.61[c]	69.71±4.81[c]	6.11±0.94[b]	8.71±0.11[c]
1.00	86.71±7.64[b]	51.78±7.81[b]	5.11±0.18[b]	7.76±0.44[b]
1.50	61.77±8.41[b]	40.61±6.66[b]	3.99±0.17[a]	5.68±0.31[b]
2.00	41.91±6.19[a]	30.45±4.71[a]	2.94±0.14[a]	3.64±0.14[a]
2.50	34.71±7.62[a]	24.78±5.11[a]	2.06±0.89[a]	1.81±0.01[a]

Means with the same superscript in the column are not significantly different (p›0.05).

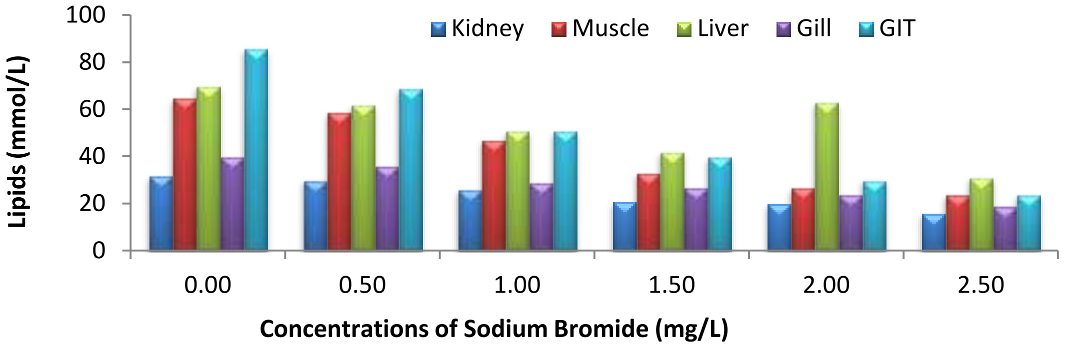

Figure 1: Relative levels of Lipids inthe organs of *P.papillio* exposed to Sodium Bromide

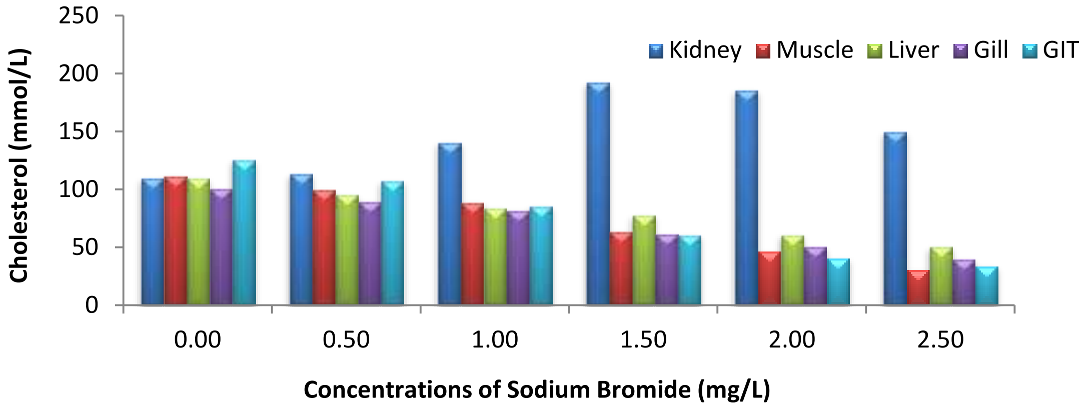

Figure 2: Relative levels of Cholesterol in the organs of *P.papillio* exposed to Sodium Bromide

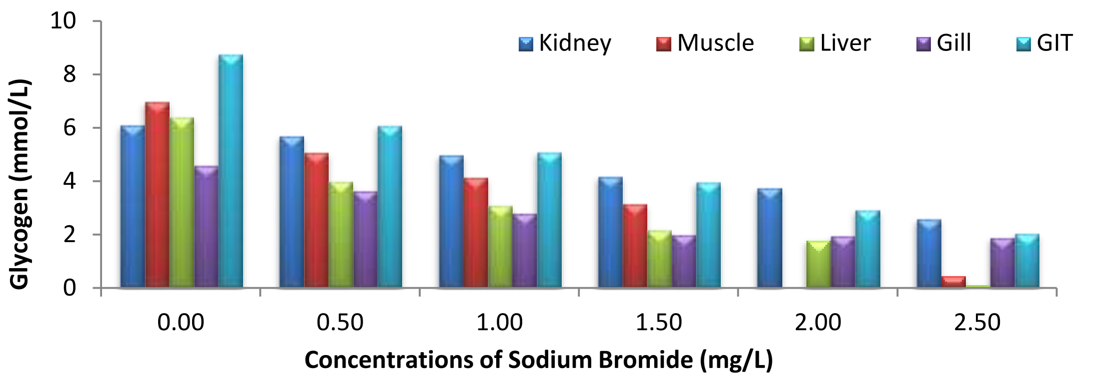

Figure 3: Relative levels of Glycogen in the organs of *P.papillio* exposed to Sodium Bromide

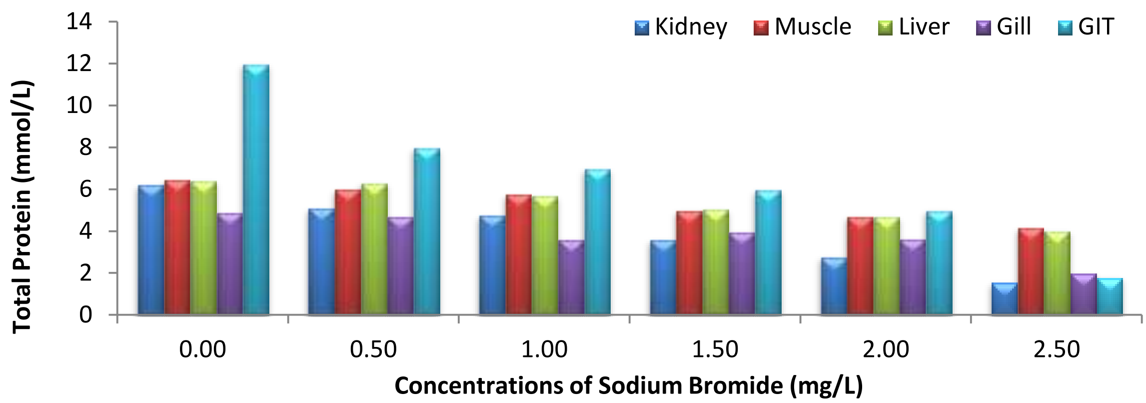

Figure 4 : Relative levels of Total protein in the organs of *P. papillio* exposed to Sodium Bromide

The glycogen level decreased significantly in the tissues of the fish during exposure to Sodium Bromide. Highest reduction was observed in the muscle and the liver. Decreased glycogen levels observed in the present study might be due to increased energy production by supplying them as keto acids into TCA cycle through aminotransferases to contribute energy needs during toxic stress [(15)]. The reduction in the glycogen has functional relevance for meeting energy demands and is involved in osmoregulation as well. The reduced glycogen may be due to depletion of reserved glucose, so the fish can try to yield metabolic energy by gluconeogenesis process [(26)]. Similar findings were observed by Swetha *et al*. [(15)] in various animals during different toxic conditions. The lowering of proteins and glycogen are apparently inter-related and are indicative of metabolic utilization driving a possible source of energy to meet the energy demand under stress [(16)].

Protein plays a major role as energy precursors for fish under stress conditions changes in each of these blood components have been employed as useful general indicators of stress in teleosts [(17)]. In the present study a significant decrease ($P < 0.05$) in protein concentration compared with the control group in all the concentrations of the toxicant involved may be an indication of protein catabolism, the process converting blood and structural protein and carbohydrate reserves to energy, to meet the higher energy demand during the prevailing stress caused by Sodium Bromide. Some of the other possible reasons for the observed protein reduction in response to the chemical might be due to hemolysis and shrinkage of erythrocytes which must have caused dilution of the plasma volume contributing to some extension in such a reduction. [(18)]. Similar significant reduction in plasma protein level was reported in *Cirrhimus mrigala* exposed to cypermethrin [(19)].

CONCLUSION

This study demonstrated that exposure of *P.papilio* to various concentrations of sodium bromide resulted in a stress response manifested in alterations of biochemical profile of the fish. Based on the results of this study these alterations, although not leading to lethal outcome might result in metabolic imbalance and impair the fish ability to withstand adverse environmental conditions.

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