



## Activities of Electrolytes in Kidney and Liver of *Clarias Gariepinus* Exposed to Fluazifop-P-Butyl

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**Abstract:** The study evaluated the effect of fluazifop-p-butyl on the kidney and liver electrolytes of *Clarias gariepinus*. Adult fish sample was purchased from biotechnology resource centre, Odi, Bayelsa State, Nigeria. The fishes were transported to the wet laboratory of the Department of Fisheries and Animal Science, Niger Delta University, Wilberforce Island, Bayelsa State, Nigeria. Adult *Clarias gariepinus* were exposed in various concentrations of the toxicants (0.00, 0.01, 0.02 and 0.03ppm) in a 30 day semi static bioassay. Thereafter, the fish was dissected and kidney and liver obtained. They were processed and analyzed using standard protocol. The electrolytes in the kidney and liver were respectively observed in the range of 0.02 – 0.15mmol/L and 0.10 – 0.15mmol/L (calcium), 3.90 – 13.10 mmol/L and 3.40 – 5.51mmol/L (potassium) and 65.50 – 150.00mmol/L and 61.00 – 177.50 mmol/L (sodium). The result trend showed significant decline and elevation of potassium content in the kidney and liver respectively. The sodium content showed significant decline in the liver and fluctuation in the kidney. While the calcium content showed significant fluctuation in both kidney and liver of the fish. The study found that electrolytes are useful parameter in determining the sublethal effect of fluazifop-p-butyl on potential non-target organism in the aquatic ecosystem.

**Keywords:** Aquatic ecosystem; Fish; Herbicides; Toxicity.

### 1. Introduction

Fishes (including shelled and fin) is a major source of animal protein [1-3]. Oladejo [4] reported that about 20% of animal protein is met by fish. Due to high demand for fish most countries in the world especially in Africa export fish [5]. Similarly several countries are recipient of the fish exported in Africa and Nigeria is one of them despite being an exporter. Nigeria fish importation which is mainly herring, mackerel and stock-fish are used to balance the deficit in domestic production [6]. Statistics on money spent on fish importation in Nigeria between 2000 to 2008 have been documented by Olaoye, *et al.* [5], Oluwemimo and Damilola [7], Angaye, *et al.* [6].

In several communities especially in the coastal region of Nigeria, fish culturing is a major business [8-10]. As such, it's a source of employment and livelihood [11]. Fish are also harvested from the wild in Nigeria in-land water especially in the Niger Delta, where there are several tributaries resulting from River Niger. Due to depletion of fish in their natural stock rearing at homestead a major panacea to the issue.

Several fish species are reared at homestead and harvested from the wild in Nigeria in-land water. But the genus, *Clarias* is one of the most widely cultured in Nigeria [12]. Several species of *Clarias* exist in Nigeria especially in the Niger Delta. But *Clarias gariepinus* is one of the species are that common in the Niger Delta. Ogamba, *et al.* [13], Ohimain, *et al.* [14], described *Clarias gariepinus* as a common Niger Wetland fish.

The aquatic ecosystem that harbors fisheries often gets polluted in Nigeria as result of improper wastes management and the use of some toxic chemicals (especially xenobiotics) that could affect non target organisms indirectly in the ecosystem. Also improper discharged of materials containing pesticides and heavy metals could also lead to their presence in the environment. These substances could be washed to the aquatic ecosystem via runoff leading to water pollution. As such the toxicant could bioaccumulate in the organisms in such water. Typically, toxicants are substances that could alter the physiology and metabolism of living organisms.

Pesticides are substances applied to the environment with the intention to mitigate, control, prevent pest [12, 15]. The type of pest intended to wade off determine the class of pesticides to be applied. Due to the nature of pesticides, some class/group is recalcitrant to degradation. Also, most pesticides including herbicides, fungicides,

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insecticides, fumigants, rodenticides etc end up in the aquatic ecosystem via run-off, especially during rainy season and when they are applied in area/field close to water bodies.

On the aquatic ecosystem, pesticides can bioaccumulate and biomagnify in the tissue/organs of fisheries. Pesticides have the tendency to cause alteration in metabolism and other relevant parameters such as enzymes, electrolytes, haematology and growth response. Several studies have been conducted on the effect of some common pesticides on the electrolytes of fisheries. Ogamba, *et al.* [12] reported the effect of 2,4-dichlorophenoxyacetic acid in *Clarias gariepinus*. Ogamba, *et al.* [16] reported effect of 2,2-dichlorovinyl phosphate on *Clarias gariepinus*. Inyang, *et al.* [17] reported effect of glyphosate in *Heterbranchus bidorsalis*. Adewale and Adeyemi [18] reported effect of ionoregulation in *Clarias gariepinus*. Singh [19] reported effect of dimethoate (EC 30%) on the electrolytes of *Cyprinus carpio*. Ogamba, *et al.* [20] reported effect of 315EC (dimethoate) and Lambda-cyhalothrin in *Clarias gariepinus*. Erhunmwunse and Ainerua [21] reported effect of dizensate on *Clarias gariepinus*. Inyang and Patani [22] reported effect of rthonasate 360sl containing glyphosate in *Heterobranchus bidorsalis*.

Fluaxifop-p-butyl is highly toxic to aquatic animals such as fish when compared to toxicity level to mammals and avian fauna. Information about toxicity of Fluaxifop-p-butyl, a post emergence phenoxy herbicide on fisheries is scares in literature. Inyang, *et al.* [23] has recently reported the effect of Fluaxifop-p-butyl on tranferases and phosphatase in plasma and organs of *Clarias gariepinus*. Inyang and Thomas [24] reported the effect of Fluaxifop-p-butyl on the metabolites and haematological parameters of *Clarias gariepinus*. Hence, this study focused on the kidney and liver electrolytes of *Clarias gariepinus* exposed to Fluaxifop-p-butyl.

## 2. Materials and Methods

### 2.1. Experimental Stock

Fish samples used in this study were purchased from biotechnology resource centre, Odi, Bayelsa State, Nigeria. The fishes were transported to the wet laboratory of the Department of Fisheries and Animal Science, Niger Delta University, Wilberforce Island, Bayelsa State, Nigeria, where the experiment was carried out. Thirty (30) healthy adult *Clarias gariepinus* purchased with mean length  $16.04 \pm 0.23$ cm and mean weight  $94.04 \pm 0.6$ g were acclimatized individually in a rectangular aquaria for seven days. Fish were fed once a day at 10.00 – 11.00hrs with 35% crude protein.

### 2.2. Experimental Design

The experimental design employed in this study is completely randomized design (CRD). There are four treatment with each having three replicates. A range finding test (trial test) was carried out using the toxicant fluazifop-p-butyl. Four (4) concentrations of the toxicant were prepared from the original (150g/l).

### 2.3. Test Chemical

Sublethal concentrations of fluazifop-p-butyl for the assay (0.01, 0.02 and 0.03ppm) was determined based on the range finding test [25]. These were prepared by transferring 0.02, 0.04, 0.06mls respectively from the original concentration (150g/l) of the toxicant and making it up with borehole water in the test aquaria, 30L of the diluents was used as control.

### 2.4. General Bioassay

The fishes were introduced into the aquarium individually. The exposed media were renewed every 2 days, the physiochemical properties of the water used for the bioassay was carried out using standard methods of APHA (American Public Health Association) [26] and the following values were obtained: Temperature 24.05 – 24.20°C, pH 6.15 – 6.24, Conductivity 97.43 – 135.02 $\mu$ /Scm, Alkalinity, 11.31 -17.13mg/l, Dissolved Oxygen, 5.21 – 7.13mg/l, and Turbidity, 103 – 146NTU.

At the end of the 30 days experiment, the fish were dissected and the kidney and liver was collected. About 0.5g of each organ was macerated (grounded) with pestle and mortar, physiological saline and deionized water was used for preservation and stabilization. Samples were centrifuged at the rate of 3000rpm for 10 minutes. The supernatants were then removed and store in plain bottles at -20°C for analysis. The activities electrolytes i.e calcium was assayed based on the modified method of Gitelman [27] and sodium and potassium was based on the method of Logawarny, *et al.* [28] and APHA (American Public Health Association) [26].

### 2.5. Statistical Analysis

Statistical Package for Social Sciences software version was used for the statistical analysis. Data were expressed as mean  $\pm$  standard error. One-way analysis of variance was carried out at  $\alpha = 0.05$  and Duncan Multiple range test was used to determine the source of observed difference.

## 3. Results and Discussion

The concentration of the electrolytes (sodium, potassium and calcium) in the kidney and liver of *Clarias gariepinus* exposed to different concentrations of Fluaxifop-p-butyl is presented in Table 1 and 2 respectively. In the kidney the concentration of calcium were highest 0.15mmol/L at 0.02ppm and least (0.10mmol/L) at 0.00 and

0.01ppm (Table 1). While in the liver, calcium level was highest (0.15mmol/L) at 0.03ppm and least (0.02mmol/L) at 0.01ppm (Liver) in the control with (0.00ppm) (Table 2). There were significance difference ( $P<0.05$ ) among the various concentration and the control. The concentration of calcium was least in the liver compared to the kidney.

**Table-1.** Activities of Electrolytes in kidney of *Clarias gariepinus* exposed to fluzafop-p-butyl for 30 days

Conc of Fluzafop (ppm)	Calcium, mmol/L	Potassium, mmol/L	Sodium, mmol/L
0.00	0.10±0.00a	5.51±0.06d	177.50±9.58d
0.01	0.10±0.00a	4.20±0.0c	58.50±4.29a
0.02	0.15±0.04c	3.65±0.01b	109.17±3.13c
0.03	0.11±0.00b	3.40±0.02a	61.00±2.17b

Means with the same superscript within column are not significantly different ( $p>0.05$ ) according to Duncan multiple range test statistics.

**Table-2.** Activities of Electrolytes in liver of *Clarias gariepinus* exposed to fluzafop-p-butyl for 30 days

Conc of Fluzafop (ppm)	Calcium, mmol/L	Potassium, mmol/L	Sodium, mmol/L
0.00	0.11±0.01c	3.90±0.05a	150.33±0.85d
0.01	0.02±0.02a	6.95±0.01b	113.33±5.03c
0.02	0.08±0.00b	7.65±0.1c	87.00±1.48b
0.03	0.15±0.02d	13.10±0.10d	65.50±1.83a

Means with the same superscript within column are not significantly different ( $p>0.05$ ) according to Duncan multiple range test statistics.

The concentration of potassium in the kidney ranged from 3.40 – 5.51mmol/L, being significantly different ( $P<0.05$ ) among the various treatments. The potassium concentration decrease as the concentration of the toxicant increases (Table 1). In the liver, the potassium concentration ranged from 3.90 – 13.10mmol/L, being significantly different ( $P<0.05$ ) among the various treatment (Table 2). The potassium concentration was generally higher in the liver compared to the kidney.

In the kidney, the concentration of sodium was least (58.50mmol/L) at 0.01 ppm and highest (177.50) at 0.00ppm. There was significance difference ( $P<0.05$ ) among the various concentration of the toxicant. Like the calcium concentration in both liver and kidney, sodium in the kidney fluctuates (Table 1). In the liver, the concentration of sodium significantly decreases ( $P<0.05$ ) as the concentration of toxicant increases. However the concentration ranged from 65.50 – 150.00mmol/L (Table 2). Sodium concentration was higher in the liver compare to the kidney.

The findings of this study showed that fluzafop-p-butyl alters the electrolytes (calcium, potassium and sodium) in organs (liver and kidney) of *Clarias gariepinus*. This alteration suggests that the toxicant have altered the cell membrane permeability by causing ionic variation due to ionic imbalance in the muscular activity of the experimental animal [29]. As such, elevation or decline in values may lead to hyper or hypo function in the kidney and liver [17]. The significance variation in the electrolytes among the different experimental group suggests damage in the kidney and liver of the fish. Prolong exposure could lead to death

The fluctuation trend in sodium content in the kidney and calcium in both kidney and liver in this study is comparable to the work of other authors Ogamba, *et al.* [12], Ogamba, *et al.* [16], Inyang, *et al.* [17], Inyang and Patani [22]. However, the higher concentration of potassium and sodium in the liver could be associated to their role in detoxification. Generally electrolytes aid in the maintenance of ionic balance to enhance normal function of the cells [29]. Specifically, sodium and potassium are vital extra cellular fluids that are essential for the transportation of ATP [17, 21, 29, 30]. Some specific function of electrolytes are maintenance of heart contraction and involuntary muscles and other metabolic activities (sodium) [21], intracellular physiological (nerve and muscle) functions, acid-base balance and osmotic pressure, enzymatic transfer of phosphate from ATP to pyruvic acid [31] (potassium), blood coagulation [31, 32] and regulation of permeability in the cell membrane (calcium) [32].

The concentration of the electrolytes were typically in the order; sodium> potassium> calcium. This trend is in consonance with the work of Ogamba, *et al.* [12], Ogamba, *et al.* [16]. The alteration in these parameters under study could lead to death of the fish exposed to toxicant such as fluzafop-p-butyl.

## 4. Conclusion

This study evaluated the effect of fluzafop-p-butyl on the electrolytes in the kidney and liver of *Clarias gariepinus*, a common Niger Delta wetland fish. The study found that significance variation in the electrolytes (sodium, potassium and calcium) content exposed to the toxicant and the control. As such, the electrolyte is a potential biomarker in monitoring toxicity of post emergence phenoxy herbicide such as fluzafop-p-butyl in fisheries. The use of herbicides containing fluzafop-p-butyl should be used with caution especially close to in-land water and or fish ponds.

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