



Antioxidants Activities in The Plasma of *Clarias Gariepinus* Juveniles Exposed to Atrazine

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Abstract

Atrazine is a pre-emergence herbicide that is widely used in Nigeria to control weeds. Given that herbicides could harm non-target species like fish, this study examined the effects of atrazine on antioxidant activities which includes Catalase (CAT), Lipid peroxidation (LPO), Glutathione (GSH), Superoxide dismutase (SOD), and Glutathione - S - Transferase (GST), in the plasma of *Clarias gariepinus*. Juveniles of *C. gariepinus* of mean length (11.74±2.64cm) and mean weight (256.68±1.81g) were exposed to different concentrations of atrazine containing 0.00 (control), 0.05, 0.10, 0.15, and 0.20mg/l of atrazine for a period of 96 Hours. Results obtained in the study indicated that atrazine caused significant increase (p<0.05) in the activities of antioxidant enzymes, CAT and LPO in all experimental groups compared with control exposures. There was also a significant decrease in the values of GSH, SOD and GST in all experimental groups compared with control exposure. The study therefore revealed that atrazine exposure had a toxic effect on *C. gariepinus* in a dose-dependent manner; hence caution should be applied in its application, and the usage must be monitored and controlled especially when in use close to any aquatic ecosystem.

Keywords: Antioxidants; pollution; toxicity; fish; aquatic environment

Introduction

The ever-increasing world population and the attendant increase in food demand necessitated those new ways of increasing agricultural output be sought [1]. In an attempt to increase agricultural output, man relies heavily on the use of chemicals to protect crops from pests, right from the time of dressing of seeds before planting, through fighting weeds and other pests on the farm, to the preservation of already harvested products. On one side, benefits derived from the use of pesticides in agriculture are immense, but on the other side, environmental pollution and/or degradation is one major problem that is linked to their application [2]. The com

petition for survival between humans and other organisms in the environment dates back to the beginning of history. Insects, rodents and generally pests have taken a serious liking for cultivated crops [3], or the conducive environments in which they are grown. This competition became more intense as humans continued to modify the environment, leading to the evolution of an organized pattern of pest control through the development of pesticides [4].

At its onset, the development of pesticides was actually slow and limited in application. In such instances, their use was highly restricted to small acreage, hence small and restricted impacts on

the environment [5]. The increase in the world population led to the increase in the use of pesticides, resulting in higher and more pronounced impacts on the environment [6]. The presence of pesticides in the environment has caused significant social and scientific development anxiety worldwide, as their all-over-the-world extensive usage can create potential risks to the environment and human health, and easily pollute bodies of water thereby resulting in extensive damage to non-target species, including fish [7]. Be it by intentional or unintentional application, water is contaminated through direct application into the aquatic system, drifts during spray, atmospheric fallout as rain and dust, soil erosion, sewage, industrial effluent and occasionally by spillage [8]. The aquatic environment is particularly one vulnerable area as it is the ultimate recipient of pollutants due to basin drainage. The aquatic ecosystems have been known to receive a wide spectrum of pollutants, which may be introduced to them directly or indirectly.

The indiscriminate use of chemicals has resulted in large scale reduction in aquatic productivity. Pesticides have different diverse impacts on aquatic animals especially fishes which are of economic importance and high value from the point of biological conservation [9]. Environmental pollution by pesticides has become a serious problem in terms of global conservation and animal and human health [9]. Besides overexploitation and habitat loss, pollution is ranked third on the list of main causes of fish species loss [10]. Atrazine and simazine are stable in pure solution, with an estimated half-life for hydrolysis of atrazine in sterile, neutral water of 1800 years [11]. Atrazine is persistent in water and together with its metabolites (which are often more hazardous than the parent product) can accumulate in drinking water resources downstream from farms [12]. Atrazine in soil breaks down through interaction with environmental compounds, particularly soil bacteria. Because the metabolizing bacteria are rare or nonexistent in surface water and groundwater, atrazine can persist for months to as long as a year once it enters surface or ground water [12].

Atrazine has also been associated with adverse health effects in humans and animals [13]. The rise in agricultural operations is mostly to blame for the increased usage and application of pesticides in Nigerian agriculture today, which significantly lowers water quality by contributing to aquatic pollution [14]. Fish are among the most common aquatic creatures, and their susceptibility to environmental contamination may be an indicator of the severity of the biological impacts of environmental pollution in water [15]. Aquatic ecosystem health is frequently assessed using fish, and biochemical alterations in fish serve as indicators of environmental contamination. Fish are often used as sentinel organisms for ecotoxicological studies because they play a number of roles in the trophic web, accumulate toxic substances and respond to low concentrations of mutagens [16]. Therefore, the use of fish biomarkers as indices of the effects of pollution are of increasing importance and can permit early detection of aquatic environmental problems [17]. Toxicants accumulated in tissues of animals catalyze redox reactions that generate ROS which may lead to oxidative stress and therefore cause biochemical alterations.

Knowledge on the nature of these alterations would be pivotal to accurate diagnosis of pesticide toxicity especially in an environment that is becoming increasingly prone to pesticide contamination, and hence the possibility of increased incidence of pesticide poisoning [18]. The nature of the biochemical, and antioxidants alterations associated with atrazine poisoning remains to be clearly defined in aquatic animals, hence the need for this study. Fish are widely used to evaluate the health of aquatic ecosystems, and biochemical changes observed in fish serve as biomarkers of environmental pollution [19]. Bioassay experiments in general present the most preferred way to evaluate the ecological influence of toxic compounds as their effects on fish and ecological risks cannot be determined by chemical analysis [20]. Knowledge of the sub-lethal effects of toxic compounds at the biochemical levels is very important for delineating fish health status and for understanding future ecological impact. The results of this study will offer baseline data that will be important for establishing environmental control on the use of atrazine. The aim of this study was to evaluate the effects of atrazine on antioxidant activities in juveniles of the African catfish, *Clarias gariepinus*.

Materials and Methods

Experimental Location

The experiment was carried out at the Wet Laboratory in the Department of Fisheries and Aquaculture Management, Faculty of Agriculture, Nnamdi Azikiwe University, Awka, Anambra State, Nigeria.

Source of Experimental Fish

One Hundred and Fifty (150) *Clarias gariepinus* of equal size (mean length 11.74 ± 2.64 cm and mean weight 256.68 ± 1.81 g) were sourced from House Tully Fish Farms, Okpuno, Awka, Anambra State, Nigeria. They were transferred in two 50 litre plastic tanks to the laboratory for acclimation process.

Acclimation and Feeding of Fish

The experimental fish were acclimated in four 150L capacity circular plastic tanks containing 150L de-chlorinated water, for 7 days to experimental conditions at room temperature. Netted materials with central slits was tied to the tops of the tanks to prevent escape of fish. Water renewal was done every two days. The fish were fed with a commercial feed at 5% body weight throughout this period.

Experimental Design

The experimental design was a completely randomized design (CRD) with four treatments levels and a control with each level having three replicates.

Procurement of Test Solution

A commonly used selective herbicide Vestrazine (Atrazine 100.0%) was purchased off shelf, from "Analytical" chemical shop, Eke-Akwa Market, Akwa, Anambra State, Nigeria.

Preparation of Test Solution

The solution of the chemical in water was prepared by serial dilution protocols using the dilution formula of Reynolds [21]

$$N_1 V_1 = N_2 V_2$$

Where N_1 = is the manufacture concentration of sodium bromide

V_1 = Volume of original solution added

N_2 = Concentration of the test solution desired

V_2 = Volume of test solution

Exposure of Fish to Atrazine

Ten *C. gariepinus* each were introduced individually into 15 aquaria tanks of 1.5m x 1m x 0.5m dimension, containing 0.00 (control), 0.05, 0.10, 0.15, and 0.20 of Atrazine. Each treatment(s) and control were replicated three times and the experimental duration lasted for a period of 96 Hours. The tank was covered with netted materials and supported with heavy objects to prevent the fish from escaping.

Evaluation of Physico-Chemical Parameters of Water

During the experiment, the following water quality parameters namely: Temperature, pH, Dissolved Oxygen, Nitrate and Ammonia levels of control and other treatment exposures were determined and the readings taken at 0, 24, 48, 72 and 96hr intervals in three replicates. Temperature was determined using the mercury-in-glass thermometer, which was inserted in water and the temperature (°C) reading was taken after four minutes. pH was determined using a Jenway® type pH meter (Model 3015). The probe was first inserted in the buffer for 5 minutes to standardize the meter to pH 7, thereafter, it was dipped into the water and the static pH was read 60 seconds later. Dissolved Oxygen was measured by Winklers method APHA, [22]. Ammonia and nitrate were determined by automation using a multi-parameter photometer (Hanna instrument H183200).

Collection of Blood Samples

Blood samples were collected at 0hrs, 24 hours, 48 hours, 72 hours and 96 hours of the experimental period. Each blood collection was completed within 5 minutes of fish removal from the experimental tank. 5ml samples were drawn once and poured into Eppendorf tubes containing 500U of sodium heparin used as an anticoagulant. The blood samples were put in ice chest box and transported within 6 hours of collection to biochemistry laboratory for analysis.

Analytical Procedure

Blood samples were centrifuged immediately for 15 minutes at 5000 rpm. Plasma specimens were separated, pipetted into eppendorf tubes and stored in a refrigerator at -20°C until assayed [23]. The results were read using a universal microplate reader on a Jenway visible spectrophotometer (Model 6405). The activities of CAT, SOD, GST, GSH and LPO in centrifuged plasma was determined spectrophotometrically using the method of Bebianno, [24].

Statistical Analysis

Date obtained from the experiments were collated and subjected to ANOVA using Statistical Package for the social Sciences, (SPSS) version 22, differences among means were separated by Turkey's Comparative Test at 0.05%.

Results

Physico-Chemical Parameters of Water in the Experimental Tanks

Table 1 shows the results for the physiochemical parameters of water in tanks of *C. gariepinus* exposed to different concentrations of vestrazine (0.00, 0.05, 0.10, 0.15, and 0.20 mg/l) respectively for 96hrs. The results indicated a significant reduction ($p < 0.05$) in the values of dissolved oxygen from 6.67 ± 0.25 in the control to 4.03 ± 0.99 at 0.20mg/l concentration of the chemical. Also, significant ($p < 0.05$) increase with increasing concentration of the chemical were however recorded in the values of nitrite and ammonia. While other parameters such as temperature and pH were within the same range comparable to the control in all concentrations of the chemical.

Changes in the Level of Antioxidants Enzymes in the Plasma of *C. gariepinus* Exposed to Different Concentrations of Atrazine for 96hrs

The antioxidants in the plasma of *C. gariepinus* exposed to acute concentrations of Atrazine for 0 Hours are presented in Table 2. Generally, the values of all the antioxidants (CAT, GSH, SOD, GST and LPO) in the plasma of the exposed *C. gariepinus* were within the same range with no significant differences in all concentrations. At 24 hours of exposure (Table 3), slight reductions were observed in the values of GSH, SOD, and GST while the values of CAT and LPO were slightly elevated. At 48, 72, and 96 hours of exposure of *C. gariepinus* to varying concentrations of Atrazine (Tables 4-6), there was significant reduction in the values of GSH, SOD, and GST While the values of CAT and LPO increased significantly with increasing concentrations of the chemical.

Table 1: Physicochemical Parameters of Water in Tanks of *C. gariepinus* exposed to acute concentrations of Atrazine for 96 Hours.

Concentrations (mg/L)	Physico - Chemical Parameters of Water				
	Temperature	pH	DO	Nitrite	Ammonia
0.00	28.33 ± 0.77 ^a	6.53 ± 0.06 ^a	6.67 ± 0.25 ^a	0.00 ± 0.00 ^a	0.09 ± 0.02 ^a
0.05	28.34 ± 0.40 ^a	6.63 ± 0.06 ^a	6.17 ± 0.21 ^a	0.05 ± 0.00 ^b	0.24 ± 0.06 ^b
0.10	28.30 ± 0.92 ^a	6.70 ± 0.10 ^a	5.03 ± 0.51 ^b	0.05 ± 0.00 ^b	0.31 ± 0.01 ^c
0.15	28.29 ± 0.51 ^a	6.77 ± 0.06 ^a	5.00 ± 0.78 ^b	0.07 ± 0.00 ^c	0.32 ± 0.05 ^c
0.20	28.45 ± 0.99 ^a	6.80 ± 0.10 ^a	4.03 ± 0.99 ^c	0.07 ± 0.00 ^c	0.36 ± 0.017 ^c

Means within the same column with different superscript are significantly different ($P < 0.05$).

Table 2: Antioxidants in the Plasma of *C. gariepinus* Exposed to Atrazine for 0 Hours (Mean ± S.D).

Conc. (mg/l)	Antioxidants (U/mg protein)				
	CAT	GSH	SOD	GST	LPO
0.00	2.15 ± 0.01 ^a	3.85 ± 0.03 ^b	0.25 ± 0.04 ^a	13.25 ± 0.01 ^a	7.11 ± 1.01 ^a
0.05	2.16 ± 0.02 ^a	3.83 ± 0.04 ^b	0.24 ± 0.01 ^a	13.24 ± 0.01 ^a	7.13 ± 0.05 ^a
0.10	2.15 ± 0.01 ^a	3.82 ± 0.07 ^b	0.25 ± 0.03 ^a	13.81 ± 0.01 ^a	7.11 ± 0.01 ^a
0.15	2.17 ± 0.03 ^a	3.84 ± 0.07 ^b	0.24 ± 0.02 ^a	13.14 ± 0.01 ^a	7.21 ± 0.02 ^a
0.20	2.16 ± 0.01 ^a	3.85 ± 0.02 ^a	0.26 ± 0.03 ^a	13.18 ± 0.81 ^a	7.11 ± 0.81 ^a

Means within the same column with different superscript are significantly different ($P < 0.05$).

Table 3: Antioxidants in the Plasma of *C. gariepinus* Exposed to Atrazine for 24 Hours (Mean ± S.D).

Conc. (mg/l)	Antioxidants (U/mg protein)				
	CAT	GSH	SOD	GST	LPO
0.00	2.15 ± 0.07 ^a	3.85 ± 0.03 ^a	0.25 ± 0.08 ^a	13.27 ± 0.09 ^a	7.19 ± 1.77 ^a
0.05	2.66 ± 0.05 ^a	3.73 ± 0.07 ^a	0.24 ± 0.07 ^a	13.00 ± 0.09 ^a	7.99 ± 0.87 ^a
0.10	2.75 ± 0.03 ^a	3.72 ± 0.04 ^a	0.23 ± 0.01 ^a	12.75 ± 0.05 ^b	8.06 ± 2.54 ^b
0.15	2.77 ± 0.06 ^a	3.64 ± 0.07 ^a	0.22 ± 0.08 ^a	12.05 ± 1.44 ^b	8.71 ± 1.77 ^b
0.20	2.70 ± 0.06 ^a	3.60 ± 0.09 ^a	0.21 ± 0.07 ^a	11.14 ± 1.99 ^b	9.88 ± 2.08 ^b

Means within the same column with different superscript are significantly different ($P < 0.05$).

Table 4: Antioxidants in the Plasma of *C. gariepinus* Exposed to Atrazine for 48 Hours (Mean ± S.D).

Conc. (mg/l)	Antioxidants (U/mg protein)				
	CAT	GSH	SOD	GST	LPO
0.00	2.18 ± 0.09 ^a	3.84 ± 0.12 ^a	0.25 ± 0.12 ^a	13.26 ± 0.11 ^a	7.20 ± 1.69 ^a
0.05	2.76 ± 0.65 ^a	3.70 ± 0.61 ^a	0.22 ± 0.07 ^a	12.85 ± 0.11 ^b	8.01 ± 0.55 ^b
0.10	2.81 ± 0.31 ^a	3.69 ± 0.09 ^a	0.20 ± 0.01 ^a	12.66 ± 0.27 ^b	8.99 ± 1.00 ^b
0.15	2.94 ± 0.77 ^a	3.60 ± 0.11 ^a	0.19 ± 0.08 ^b	12.37 ± 1.81 ^b	9.12 ± 1.77 ^b
0.20	2.99 ± 1.11 ^a	3.57 ± 0.11 ^a	0.18 ± 0.07 ^b	11.02 ± 1.38 ^c	10.47 ± 3.17 ^c

Means within the same column with different superscript are significantly different ($P < 0.05$).

Table 5: Antioxidants in the Plasma of *C. gariepinus* Exposed to Atrazine for 72 Hours (Mean ± S.D).

Conc. (mg/l)	Antioxidants (U/mg protein)				
	CAT	GSH	SOD	GST	LPO
0.00	2.19 ± 0.04 ^a	3.85 ± 0.88 ^a	0.24 ± 0.03 ^a	13.24 ± 0.88 ^a	7.21 ± 1.77 ^a
0.05	2.80 ± 0.77 ^a	3.65 ± 0.77 ^a	0.20 ± 0.12 ^a	11.78 ± 0.32 ^b	9.03 ± 1.82 ^b

0.10	2.94 ± 0.87 ^a	3.60 ± 0.88 ^a	0.19 ± 0.01 ^b	11.51 ± 0.79 ^b	9.99 ± 1.76 ^b
0.15	2.98 ± 0.82 ^a	3.45 ± 0.82 ^a	0.17 ± 0.04 ^b	11.02 ± 1.51 ^b	10.15 ± 1.62 ^c
0.20	3.05 ± 1.11 ^b	3.00 ± 0.75 ^b	0.17 ± 0.02 ^b	10.59 ± 1.88 ^c	12.04 ± 2.98 ^c

Means within the same column with different superscript are significantly different (P<0.05).

Table 6: Antioxidants in the Plasma of *C. gariepinus* Exposed to Atrazine for 96 Hours (Mean ± S.D).

Conc. (mg/l)	Antioxidants (U/mg protein)				
	CAT	GSH	SOD	GST	LPO
0.00	2.20 ± 0.12 ^a	3.84 ± 0.41 ^a	0.25 ± 0.07 ^a	13.23 ± 0.73 ^a	7.22 ± 1.03 ^a
0.05	2.99 ± 0.55 ^a	3.42 ± 0.41 ^a	0.17 ± 0.43 ^b	10.03 ± 0.44 ^b	11.72 ± 1.02 ^b
0.10	3.05 ± 0.96 ^b	3.01 ± 0.90 ^a	0.15 ± 0.39 ^b	9.77 ± 0.63 ^b	13.01 ± 2.97 ^b
0.15	3.66 ± 0.71 ^b	2.83 ± 0.77 ^b	0.14 ± 0.05 ^b	8.75 ± 1.83 ^b	18.66 ± 1.88 ^c
0.20	3.76 ± 1.02 ^b	2.24 ± 0.55 ^b	0.13 ± 0.01 ^b	7.07 ± 1.33 ^c	20.88 ± 4.07 ^c

Means within the same column with different superscript are significantly different (P<0.05).

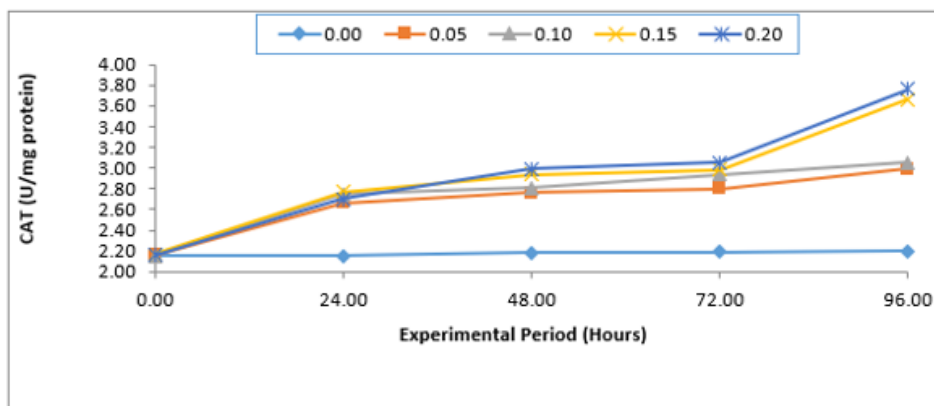


Figure 1: Variations in the values of Catalase (CAT) in the plasma of *C. gariepinus* exposed to Atrazine for 96 Hours.

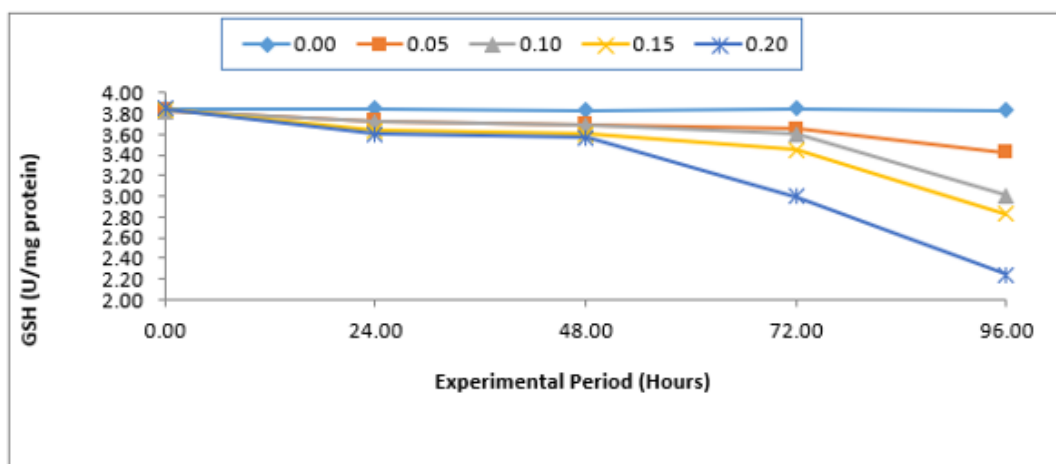


Figure 2: Variations in the values of Glutathione in the plasma of *C. gariepinus* exposed to Atrazine for 96 Hours.

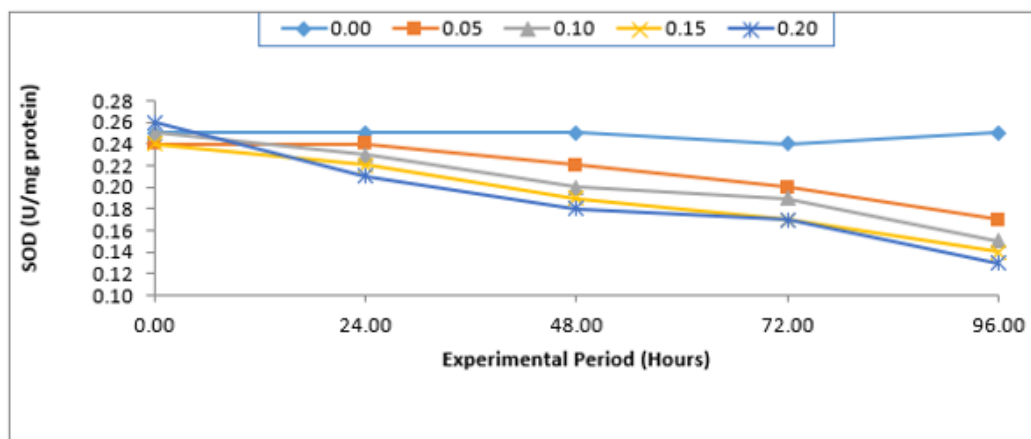


Figure 3: Variations in the values of Superoxide dismutase (SOD) in the plasma of *C. gariepinus* exposed to Atrazine for 96 Hours.

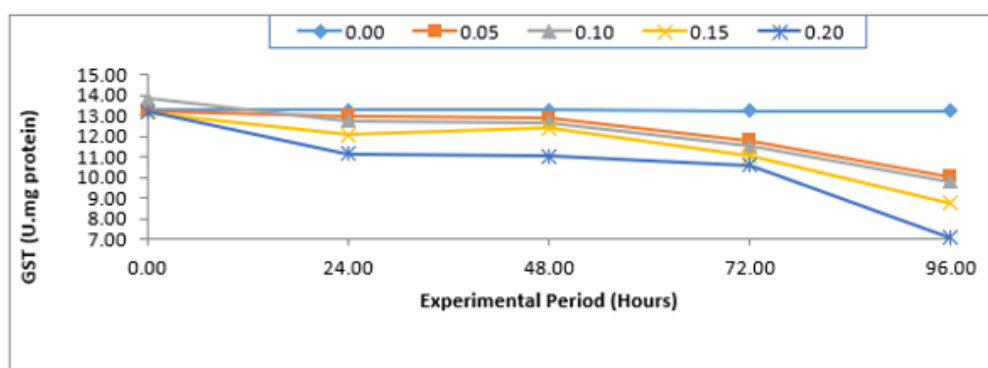


Figure 4: Variations in the values of Glutathione S-Transferases (GST) in the plasma of *C. gariepinus* exposed to Atrazine for 96 Hours.

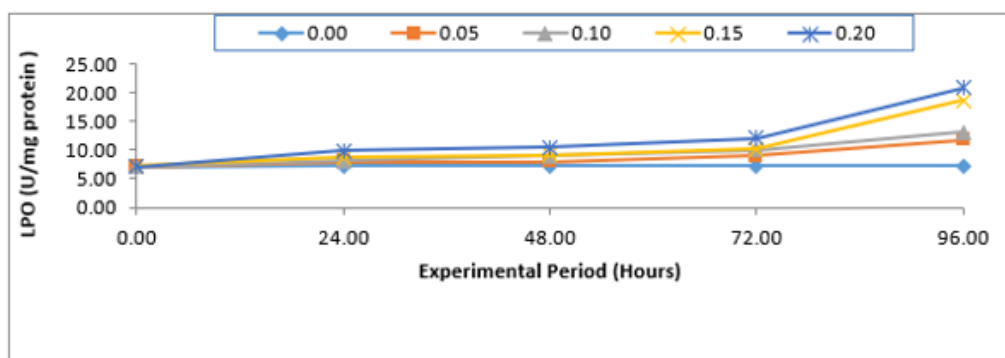


Figure 5: Variations in the values of Lipid peroxidation (LPO) in the plasma of *C. gariepinus* exposed to Atrazine for 96 Hours.

Comparative Values of Antioxidants in the Plasma of *C. gariepinus* Exposed to Acute Concentrations Atrazine for 96 Hours

Comparative values of catalase (CAT) in the plasma of *C. gariepi-*

nus exposed to Atrazine for 96 hours is shown in Figure 1. The values of CAT increased as the experimental period increased, with the highest value of 3.76 observed at the 0.20mg/L and 2.20 in the control at 96 hours. Comparatively, the values of GSH as shown in

Figure 4.2, indicated that the values of GSH in *C. gariepinus* exposed to varying concentrations of Atrazine were reduced progressively as the experimental period increased with the lowest values at 96 hours for all concentrations. The lowest value of 2.24 was recorded in the fish exposed to 0.20mg/L of the chemical at 96 hour, while the value of 3.84 was observed in the control (Figure 2). The values of SOD (Figure 3) reduced when compared to the control value in all concentrations of exposure. The values of GST (Figure 4) reduced considerably as the experimental period increased, this was more pronounced at the concentration of 0.10, 0.15 and 0.20mg/l concentrations of the chemical. Comparatively, the value of LPO is shown in Figure 5. The value of LPO increased as the experimental period progressed from 24 to 96 hours. However, a sharp increase was observed in the concentration of 0.20mg/l at 96 hours.

Discussion

At exposure of the toxicant for 96hours, the recorded increase in nitrite, ammonia, and decrease in dissolved oxygen and the fluctuations that occur between the various concentration and time could be due to the fact that these parameters are highly unstable. Although herbicides cause changes in the quality of water in and around sprayed areas and decrease the dissolved oxygen in the water, along with an increase in temperature which may pose a threat to the survival of fish species, the result of the present study indicates that Atrazine application does not result in significant changes in the physicochemical parameter to a point that is capable of causing visually observable negative impacts in fish. The water quality parameters under study are within the standard meant for aquaculture purposes. This study is in line with the work of Akinrotimi *et al.* [25], who reported that pH of 6.5 – 9.0 supports fish life. Ajani *et al.* [26] also reported that catfish and other air breathing fish can tolerate low Dissolved oxygen concentration of 4 mg/l. Hence the dissolve oxygen content is within the values necessary for fish life. Similar findings were also reported by Akinrotimi *et al.* [27].

Basically, in aquatic ecosystem temperature, pH and other physiochemical properties of water are very essential for the survival of fish through metabolism. As such inability of fish to adapt to the environment could cause a change in their physiological responses which could lead to mortality. The temperature, pH, Dissolved oxygen and other parameters are within the ambient values of the area and values of surface water resources in Nigeria [28,29]. Hence, the fishes may not have died due to temperature or other of the physiochemical parameters recorded in this study. The values recorded in this study have been reported to be within the tolerance ranges of warm water fish species. Therefore, the water quality parameters may not have caused the observed changes in the antioxidant's enzymes in plasma of *Clarias gariepinus* in this study. Oxidative stress is a situation when steady state ROS concentration is transiently or chronically enhanced, disturbing cellular metabolism and its regulation and damaging cellular constituents [30]. A state when antioxidant defenses are overcome by pro-oxidant forces [31].

The activation of oxidative manifestations leads to the response of antioxidants activation in expression of genes encoding antioxidant enzymes. Nevertheless, there are considerable gaps in the

knowledge on response to oxidative stress, particularly in aquatic animals. Antioxidant enzymes are included in the environmental pollution assessment because of their inducibility under conditions of mild oxidative stress and their potential role in adaptation to pollutant-induced stress. It is expected that they may be more sensitive at detecting initial insult [32]. Laboratory studies confirmed that the measurement of changes in the expression of a large number of specific genes or activities of certain enzymes of antioxidant defense can be explored in an early warning system of toxicant exposure [33]. Obviously, the early warning can be used when temporal effect of pollution is expected. In the field studies, fish is frequently subjected to long-term exposure of number of factors.

Therefore, the observed difference in the activity of antioxidant enzymes between two sites may be attributed both to their activation under mild stress conditions of the location or to their suppression due to strong oxidative damage [34]. Typically, the array of oxidative stress parameters in fish includes the activities of superoxide dismutase (SOD), catalase and glutathione (GST). The assessment of catalase, GST and SOD activities has most often been used in biomonitoring programmes for fish [35]. In this study, elevated CAT and LPO activities in combination with the decrease in GSH, SOD and GST were observed in the plasma of *C. gariepinus* exposed to different concentrations of atrazine in the experimental tanks. A result was observed in the field study of *C. carpio* exposed to toxicants. The results showed that SOD and GST activities were higher, while CAT activities were lower when compared to the control farms [36]. Moreover, the concerted elevation of SOD and GST activities was equally indicated in the plasma of starlet fish (*Acipenser ruthenus*) collected from the Danube-oil refinery site compared to that from the reference site, while no differences were found in catalase [37].

In the study of three populations of brown trout (*S. trutta*) exposed to elevated Cd and Zn or Cu levels in their natural environment, both metal-exposed groups had higher activities of SOD in their plasma compared to unexposed trout from reference site, and catalase activity in the plasma was the same in all three populations [38]. Conversely, misbalanced antioxidant activities were shown in the various oxidative stress biomarkers in the Indian freshwater fish, *Wallago attu* sampled from seemingly polluted River site. It showed higher activities of SOD and GST in the serum, whilst CAT activity was found to be significantly lower when compared with values in tissues of fish collected from clean site [39]. The depletion of catalase activity or its stability along with increment of SOD and GST activities have been reported by some authors [40,41]. Despite this, Dorval *et al.* [42] observed that in plasma, hepatic and adrenal tissues of white sucker (*C. commersoni*) exposed to toxicants. The same trend was observed in this study, where catalase activities in fish sampled from control were higher than those from other farms.

However, Falfushynska and Stolyar [43], attributed the low catalase activity in fish sampled from relatively polluted site to low production of oxygen, which has been reported to inhibit catalase in the case of excess of production of pollutants [44]. Catalase depreciation and activation can be considered as a last refuge of antioxidant

defense in Teleost fish. The catalase role in the antioxidant defense of fish was reported by Shalaby *et al.* [45], based on the information on its activation by hydrogen peroxide at high concentrations. They suggested that catalase normally plays a relatively minor role in hydrogen peroxide catabolism at low rates of peroxide generation, but it becomes indispensable when the rate of hydrogen peroxide production is enhanced, for example, at oxidative stress. Low intensity, but prolonged effect of spontaneous sources of pollution from aquaculture activities can activate SOD activity in fish tissues. That is exemplified by some studies [46-48]. The increased SOD activity along suggests the strength of antioxidant defenses is common in fish exposed to toxicants. In some cases, increase in catalase activity is often observed in the model experiments and can occur without relation to SOD responses, due to high pollutant impact. Higher values of catalase activity were reported in the liver and plasma of red mullet, *Mullus barbatus* exposed to toxicants in the laboratory [49].

Conclusion and Recommendations

Based on the data and evidence recorded in this study, atrazine is found to be toxic and posed stress to *Clarias gariepinus* and the effect increased with increase concentration of atrazine. The results of the biochemical parameters assayed showed that *C. gariepinus* was seriously affected by atrazine. The results of the present study showed that vestrazine herbicide significantly lowered the dissolved oxygen level of the test water and induced detrimental behavioral changes in *C. gariepinus*. The study contributes to knowledge on the toxicity of herbicides to non-target species and has shown that vestrazine herbicide contributes to environmental pollution and could decline biodiversity. To check this, reasonable restraints backed up by law must be meted to those in the habit of discarding untreated wastes into our water bodies.

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