

Toxicity Of Buta Force[®] On Juveniles Of *Clarias Gariepinus* And Its Impact On The Haematological Parameters

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Abstract: The toxicity test of Buta Force[®] on juveniles *Clarias gariepinus* and its impact on the haematological parameters was investigated. The fish were exposed to lethal concentration of 0.00 μ l/l, 0.15 μ l/l, 0.20 μ l/l, 0.25 μ l/l, and 0.30 μ l/l of Buta force in the experimental procedure. The behavior and mortality was used as the measure of toxicity. Loss of equilibrium, loss of reflexes, erratic swimming, gasping of breath, gill movement, float ventral side-up, frequent surfacing and lethargy was observed as behavioral response. For the determination of 96 h LC₅₀ of Buta-force[®], a static renewal acute bioassay test was conducted. Each group of catfish was subjected separately to a daily single dose of Buta-force[®]; 0.00mg/l, 0.15mg/l, 0.20mg/l, 0.25mg/l and 0.30mg/l. Haematological tests were conducted at the end of the 96/hours experiment. Blood samples were collected from the fish by cardiac puncture with the aid of a hypodermic needle and syringe (2ml) and taken to a Laboratory for analysis. No adverse behavioral responses recorded throughout the period of the experiment on the control tank. But the chemically exposed fish show some signs of Loss of equilibrium, loss of reflexes, erratic swimming, gasping of breath, gill movement, float ventral side-up, frequent surfacing and lethargy was observed as behavioral response. Dead was observed as mortality in the exposed fish and these varied greatly with differences in the concentration of the Buta Force and this shows that mortality increases with increase in concentration. There was no mortality recorded in the control tank while 71.42%, 75.71%, 80.74% and 85.71% were recorded in 0.15 μ l/l, 0.20 μ l/l, 0.25 μ l/l and 0.30 μ l/l respectively. The differences observed in the mortality of *C. gariepinus* juveniles at varying concentration were significant ($P < 0.05$), an indication that mortality could be a factor of concentration and time of exposure. Haematological parameters such as RBC, HB decreased with increased in concentration of Buta Force. WBC fluctuates with increased in concentration. The study has been able to established the fact that, exposure of *C. gariepinus* juveniles to even low concentration (0.15 μ l/l) of Buta Force can induce various toxicological and haematological effects.

Keywords: Buta Force, toxicity, haematology, mortality, water pollution and *Clarias gariepinus*

I. INTRODUCTION

Buta force is a protein synthesis inhibitor. It is selective systemic agricultural herbicide, used to control grasses and some broad-leaved weeds, absorbed primarily by the germinating shoots and secondarily by the roots, with translocation throughout the plant, giving higher concentrations in vegetative part than in reproductive part. The use of synthetic chemicals such as Buta-force as a means of increasing agricultural productivity has posed a serious treat and great consequences to the water bodies. Fish and other organisms are affected by pesticides which pollute the natural

water. The contamination of aquatic ecosystem by pesticides can cause acute and chronic poisoning of fish and other organism [8]. Unfortunately, due to extensive use and/or misapplication, Buta-force can diffuse into the aquatic ecosystem through surface runoff on reaching water bodies, this toxicant is easily absorbed by organisms through consumption, respiration and skin [14] and gets accumulated. Fish species serve as major test organisms in ecotoxicological assessment because of their link to man in food chain. The blood of fish, gill, liver and kidney are the major target of pesticidal action where changes in the activities of enzymes and eventual pathological damage can

easily be detected. The main portal of entry of any Pesticide is through the gills from where it is transported to various parts of the body through the blood stream. Activity depends on the availability of water such as rainfall following treatment, overhead irrigation or application to standing water as in rice culture. The widespread use of herbicides has resulted in steady increase in water pollution, evoking considerable damage of phytoplankton and zooplankton, thus depleting essential sources of the food chain [11]. The aim of this research work is to determine the toxicity of Buta force on juvenile of *Clarias gariepinus* and its impact on the haematological parameters.

II. MATERIALS AND METHODS

STUDY AREA

The study was carried out at the Department of Fisheries and Aquaculture, Adamawa State University Mubi. Adamawa state is located at the north eastern part of Nigeria, it lies between latitude 7° and 11° N of the equator and between longitude 11° and 14° E of the Greenwich meridian [1].

TEST ORGANISM

One hundred and thirty juveniles *C. gariepinus* juveniles with 31.07 ± 0.12 g mean body weight and 18.23 ± 0.27 cm mean total length were obtained from the Fisheries and Aquaculture Department of Adamawa State University Mubi, Nigeria. The fish were visible free of any deformities. Preliminary test was carried out to determine the concentration range as described by [19].

EXPERIMENTAL DESIGN

The concentrations of Buta force were prepared in the following order: 0.00mg/l (Control), 0.15mg/l, 0.20mg/l, 0.25mg/l and 0.30mg/l. One hundred and thirty (130) juveniles of *C. gariepinus* were used for this research. Ten (10) juveniles of *C. gariepinus* per treatment were placed in rubber tanks with size 80 X 40. The treatment was replicated three times. The rubber tanks were covered with nets and fastened with rubber bands in order to prevent the fish from escaping. The test lasted for 96hours and was observed initially at 1-2hours interval followed by 12hour intervals.

BEHAVIORAL RESPONSE OF *C. GARIEPINUS* EXPOSED TO BUTAFORCE®

The behavior and mortality was used as the measure of toxicity. Loss of equilibrium, loss of reflexes, erratic swimming, gasping of breath, gill movement, float ventral side-up, frequent surfacing and lethargy was observed as behavioral response and those that does not respond to touch were considered dead and removed immediately.

DETERMINATION OF THE 96HRS LC₅₀ AND MORTALITY OF *C. GARIEPINUS* EXPOSED TO BUTAFORCE®

Ninety one pre-acclimated catfish were held in thirteen rubber container (80 by 40 cm) each containing seven *C. gariepinus* juveniles which represent a group. For the determination of 96 h LC₅₀ of Buta-force®, a static renewal acute bioassay test was conducted. Each group of catfish was subjected separately to a daily single dose of Buta-force®; 0.00mg/l, 0.15mg/l, 0.20mg/l, 0.25mg/l and 0.30mg/l. the concentrations which resulted in 50% mortality (LC₅₀) for 96 h exposure was calculated accordingly.

DETERMINATION OF HAEMATOLOGICAL PARAMETERS OF *C. GARIEPINUS* EXPOSED TO BUTAFORCE®

Haematological tests were conducted at the end of the 96/hours experiment. Blood samples were collected from the fish by cardiac puncture with the aid of a hypodermic needle and syringe (2ml) and taken to a Laboratory for analysis using method described by [19]. The haematological parameters analyzed in this experiment were: Haemoglobin (HB), Total red blood cell (RBC) and white blood cell (WBC) count using the formula by [15]. A relatively small drop of blood from each sample was placed on a disposable test strip which interfaces with a digital meter.

STATISTICAL ANALYSIS

The Data obtained in this study were subjected to Analysis of variance (ANOVA) at 0.05 level of significance. Least Significant Difference was used to separate mean [5].

III. RESULTS

BEHAVIORAL RESPONSES OF *C. GARIEPINUS* TO DIFFERENT PERCENTAGE (%) CONCENTRATION OF BUTA FORCE®

Behavioral Responses of *C. gariepinus* to different percentage (%) concentration of Buta force is presented on table 1. No adverse behavioral responses recorded throughout the period of the experiment on the control tank. But the chemically exposed fish show some signs of a loss of reflexes, erratic movement, over secretion of mucus, gasping of breath, regular gill movement, frequent surfacing, lethargy and those that could not tolerate the changing environment gradually became weak and settled at the bottom of the plastic container and later died.

parameters	0.00ul/l	0.15ul/l	0.20ul/l	0.25ul/l	0.30ul/l
Loss of Reflexes	-	+	+++	+++	+++
Erratic Swimming	-	+	+++	+++	+++
Over secretion of mucus	-	+	++	++	+++
Gasping of breath	-	+	++	++	+++

Gill Movement	-	++	+++	+++	+++
Frequent Surfacing	-	++	++	+++	+++
Lethargy	-	++	+++	+++	+++

+ indicates an increase in responses; -indicates no response;

Table 1: Behavioral Responses of *C. gariepinus* to different percentage (%) concentration of Buta force®

DETERMINATION OF THE 96HRS LC₅₀ AND MORTALITY

The LC₅₀ of *C. gariepinus* juveniles exposed to various concentrations of Buta Force R is 0.10µl/l with lower and upper limits of 0.12µl/l and 0.15µl/l. The regression equation of the relationship was calculated to be probit: $Y = 11.11x - 12.70$, log concentration and on R – square value, $R^2 = 0.991$. The expression, R^2 value indicates that mortality rate of fish increased with increased in concentration of the Buta Force® herbicide. There was no mortality recorded in the control tank while 71.42%, 75.71%, 80.74% and 85.71% were recorded in 0.15µl/l, 0.20µl/l, 0.25µl/l and 0.30µl/l respectively. The result of the mean mortality is presented in the table 2.

Concentration (ul/L)	Log concentration	Percentage mortality	Probit value
0.00(control)	0.00	0.00	0.00
0.15	0.8239	71.42	7.71
0.20	0.6989	75.71	6.90
0.25	0.6020	80.74	6.28
0.30	0.5228	85.71	5.77

Table 2: Mortality rate of *C. gariepinus* juveniles exposed Buta Force®

DETERMINATION OF HAEMATOLOGICAL PARAMETERS OF *C. GARIEPINUS* EXPOSED TO BUTAFORCE®

The result of haematological parameters of juveniles of *C. gariepinus* exposed to Buta force® is presented on table 3. The result obtained in this research work showed that the concentration of Buta-force at 0.30µl/l has the least Red blood cells of 1.18 ± 0.00 compare to the control 1.33 ± 0.00 . The White blood cells are highest at 0.30µl/L with value of 4.06 ± 0.5 while it is lowest at the control. Haemoglobin also recorded lowest value 14.24 ± 0.05 at 0.30µl/L concentration when compared to the control. All the haematological parameters were significantly different ($P < 0.05$) significant difference among the concentrations in term of the HB values.

Parameters	0.00(control)	0.15(ul/l)	0.20(ul/l)	0.25(ul/l)	0.30(ul/l)
RBC	1.33 ± 0.00^a	1.29 ± 0.00^b	1.28 ± 0.00^c	1.25 ± 0.00^d	1.18 ± 0.00^e
WBC	3.50 ± 0.5^d	3.50 ± 0.5^d	3.62 ± 0.5^c	3.80 ± 0.5^b	4.06 ± 0.5^a
HB	14.34 ± 0.05^a	14.30 ± 0.05^b	14.26 ± 0.05^c	14.24 ± 0.05^d	14.20 ± 0.05^e
L.S.D	0.00	0.00	0.00	0.00	0.00

Means on the row with the same superscript are not statistically significant ($P > 0.05$)

Table 3: Haematological parameters of *C. gariepinus* exposed to ButaForce®

Where; WBC = White Blood Cell, RBC = Red Blood Cell, HB = Hemoglobin.

IV. DISCUSSION

Buta Force (protein inhibitor) herbicide which is widely used to control grasses, some broad leaved weeds and sedges in agriculture, however once it enters surface waters, it may affect other organism such as fish as a non-target organism either in natural or culture conditions. In this study, the chemically exposed fish show some signs of a loss of reflexes, erratic movement, over secretion of mucus, gasping of breath, regular gill movement, frequent surfacing, lethargy and those that could not tolerate the changing environment gradually became weak and settled at the bottom of the plastic container and later died. The LC₅₀ derived from the toxicity test revealed that *C. gariepinus* was sensitive to Buta Force protein inhibitor herbicide. Also the result of the present study clearly showed that Buta Force® exposure results in significant haematological alterations in *C. gariepinus* juveniles. The ecotoxicological studies suggest that Buta Force® may be harmful to aquatic invertebrates [7], microbial communities [9] and also a possible carcinogen in animals and humans [3]. It is reported as a neurotoxin [18], genotoxin [12] and carcinogen [6]. The Results obtained is similar with the 96/hours LC₅₀ for the African catfish exposed to formalin by [13]. Following exposure to various concentrations of Buta Force, the RBCs and WBCs count decreased; the hemoglobin also reduced of it natural level. All the mentioned alterations indicate that exposed fish slightly suffered from anemia induced by the herbicide. This is an indication of the disruptive effects of the Buta Force on erythropoietic tissue as well as cell viability. This agrees with the report of [17]. It is also possible that the Buta Force adversely suppressed fish osmoregulation. The disturbed osmoregulation may result in dilution of blood as reported by [10]. [16] and [2] carried out similar research for some pesticides. Erythrocytes and their hemoglobin contents are responsible for oxygen transportation within the body. Low number of red blood cells or insufficient amount of their hemoglobin content could influence energy balance of the body. In this case fish may suffer from oxygen deficiency as reported by [4] which ultimately prohibit its normal growth. Moreover it seems that reduction in red blood cell is a key factor which could be responsible for productivity reduction. This study showed that following 96/hours of Buta Force exposure, all exposed fish experienced lower white blood cells count, this varied significantly compared to the control media. The white blood cells play a vital role in fish immunology. Generally, fishes with low white blood cells count are more susceptible for bacterial or fungal infections. It has been demonstrated due to leukocyte reduction [20]. The result of this study showed that Buta Force reduced water quality which makes it inhabitable for the organism and cause adverse effect on their behavioural changes and death even at slightly high concentration.

V. CONCLUSION

Buta Force[®] caused a general disorder in the haematology of the fish which is an indication that it is toxic to the fish and so can cause dead after prolonged exposure. The study has been able to establish the fact that, exposure of *C. gariepinus* juveniles to even low concentration (0.15µl/l) of Buta Force can induce various toxicological and haematological effects. In view of the toxicity effects of this chemical, it can be inferred that indiscriminate discharge of chemical insecticide could induce damages as well as respiratory disturbance which might make all the living entities in polluted environment vulnerable to disease and eventually lead to death.

VI. RECOMMENDATIONS

- ✓ The study showed that Buta Force[®] is toxic to *Clarias gariepinus*.
- ✓ Its use as herbicide in agriculture should be strictly regulated to prevent its adverse consequences on aquatic ecosystem and man.
- ✓ The chemical should not be used indiscriminately and Government agencies should regulate the distribution and usage.
- ✓ There is a need for caution in application of the herbicide.

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