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CHANGES IN THE LIVER OF AFRICAN CATFISH, *Clarias gariepinus* JUVENILES EXPOSED TO SUB-LETHAL CONCENTRATIONS OF GLYPHOSATE

[1]Cheikyula, J.O., [2]Umaru, J.A., and [1]Ategha, A.

[1]Department of Fisheries and Aquaculture, Joseph Sarwuan Tarka University, Makurdi Nigeria
[2]Department of Fisheries Technology, College of Agriculture Science and Technology Lafia Nigeria
***Corresponding Author:** *cheikyulaj@yahoo.com;* +234 703 382 2909

ABSTRACT

This study was undertaken to determine the histological changes in the liver of the African catfish, Clarias gariepinus exposed to sub-lethal concentrations glyphosate. One hundred (100) African Catfish (C. gariepinus) juveniles were exposed to increasing glyphosate concentrations of 0.00mg/l, 20.00mg/l, 34.00mg/l, and 56.00mg/l glyphosate with replicates for a period of 28 days in the General Purpose Laboratory of the Department of Fisheries and Aquaculture of Joseph Sarwuan Tarka University Makurdi, Nigeria. Histological changes observed in the livers were cell vacuolization, degeneration of hepatocytes, haemorrhage and ultimately necrosis. The severity of these changes increased with concentration of glyphosate from 0.00 – 56.00 mg. There was no significant difference (P>0.05) between temperature and pH in the water quality parameters of the treatments, but there was a significant difference (P<0.05), in Dissolved Oxygen (DO), Total Dissolved Solids (TDS), and Electrical Conductivity (EC) where they all decreased with increasing concentration of glyphosate.

Keyword: Glyphosate; *Clarias gariepinus*; Histology;

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INTRODUCTION

Histology is the microscopic examination of tissues of an organism and the determination of histological abnormalities after exposure to xenobiotics. Liver is the primary target organs for many chemicals because of its role in the detoxification of xenobiotics, accumulation of chemicals and susceptibility to damage by chemicals (Hedayati, 2016). Histological changes give valuable information about xenobiotic impacts (Yancheva *et al*., 2016).

Glyphosate is a non-selective herbicide used for the control of terrestrial, annual and perennial grasses, broad-leafed and aquatic weeds. It is the active ingredient of numerous commercial formulations of herbicides applied in different sectors in agriculture to aquaculture. Herbicides are used to control weeds, but they harm non - target organisms, especially in the aquatic environment like fish and small aquatic invertebrates important in the ecosystem. Glyphosate is one of the most popular herbicides used by farmers because it kills weeds without affecting the crops. Glyphosate has low toxicity to *C. gariepinus*, but persistent and indiscriminate application of glyphosate for weeds control may be toxic. Pure glyphosate is low in toxicity to fish and wildlife, but some products containing glyphosate may be toxic because of the other ingredients in them. Large quantities of glyphosate find their way into water bodies from agricultural land runoffs. This work attempts to provide information on the histological alterations in the liver of juveniles of the African catfish, *Clarias gariepinus* exposed to glyphosate at sub lethal concentrations in water.

The African catfish, *Clarias gariepinus*, belongs to the family Claridae. It is an air breathing, non-scaly fresh water C. gariepinus of commercial importance (Marioghae, 1991). It tolerates poorly oxygenated waters; and it is one of the most widely cultured African catfish, *C. gariepinus* in Nigeria (Nosakhare *et.al.*, 2013). *C. gariepinus* is also, used as biological indicator in ecotoxicological studies (Ayoola, 2008). Their use is due to their well-developed osmoregulatory, endocrine, nervous, and immune systems that are sensitive to environmental variables. This study will determine the histological changes in the liver of *Clarias gariepinus* exposed to sub lethal concentrations of a glyphosate herbicide.

MATERIALS AND METHODS

Juveniles of *Clarias gariepinus* were collected from the Fish farm of the Department of Fisheries and Aquaculture, Joseph Sarwuan Tarka University, Makurdi, Nigeria and acclimatized to laboratory conditions for the sub-lethal exposure to glyphosate. Ten (10*) C. gariepinus* were placed in three (3) 20 L tanks with glyphosate concentrations of 20.00, 34.00, and 56.00mg/l with replicates, for 28 days.

Histology procedures

C. gariepinus samples were removed from each of the concentrations at end of the exposure, and dissected to remove their livers for histological examinations. The livers extracted were washed in saline water; fixed in a 10% dilute formalin solution; dehydrated through graded series of ethanol and embedded in paraffin wax. Blocks were then prepared and sectioned at 6-7 microns thickness, deparaffinized in xylene and stained with haematoxylin-eosin. Changes induced by glyphosate exposure in the liver tissue were observed in a photomicroscope, and compared with control.

Determination of Water Quality Parameters

Water quality parameters of exposure media were determined. Parameters of temperature (^{O}C), Total Dissolved Solids (TDS), Electrical Conductivity (EC) and pH were determined with Hanna multi parameter water checker model HI-98129. Dissolved oxygen (DO) was determined with Hanna DO Meter, model HI-9142.

Data Analysis

Data was analyzed using one-way Analysis of Variance (ANOVA) to compute the differences in the water quality parameters in the various concentrations of glyphosate observed in the experimental culture media.

RESULTS

Liver histology

Liver of the control *C. gariepinus* with 0.00mg/l glyphosate had normal appearance with polygonal structured hepatocytes with central spherical nucleus and a densely stained nucleolus under the photomicrograph at x100 magnification. (Figure 1). Livers from 20 mg/l exposure had mild cell vacuolization and degeneration different control (Figure 2). *C. gariepinus* liver in a higher concentration of 34.00 mg/l had severe cell vacuolization, degradation of hepatocytes and hepatic necrosis compared with the liver of the control (Figure 3). Livers in the exposures with the highest concentration of 56 mg/l glyphosate had more severe necrosis, haemorrhage and vacuolization. (Figure 4). Histological changes in the liver were more evident in the liver of the *C. gariepinus* exposed at this concentration than in concentrations of 20.00mg/l, 34.00mg/l and the control.

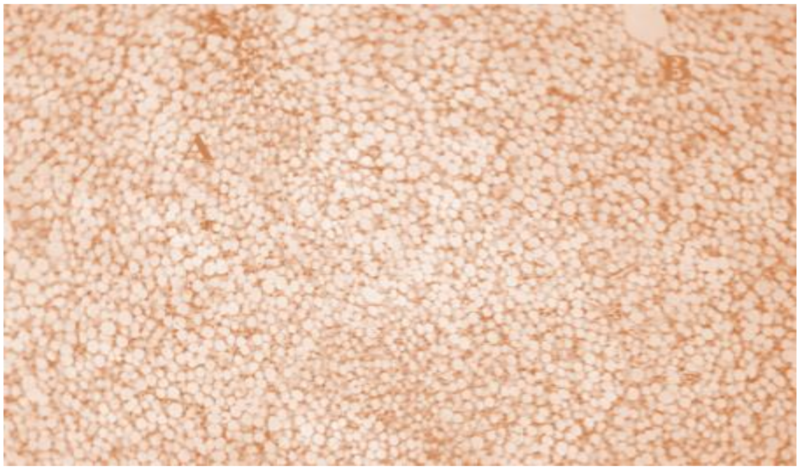

Plate 1: Photomicrograph of Liver of Control (0.00mg/l) *C. gariepinus* with normal liver structures (H & E stain x100)

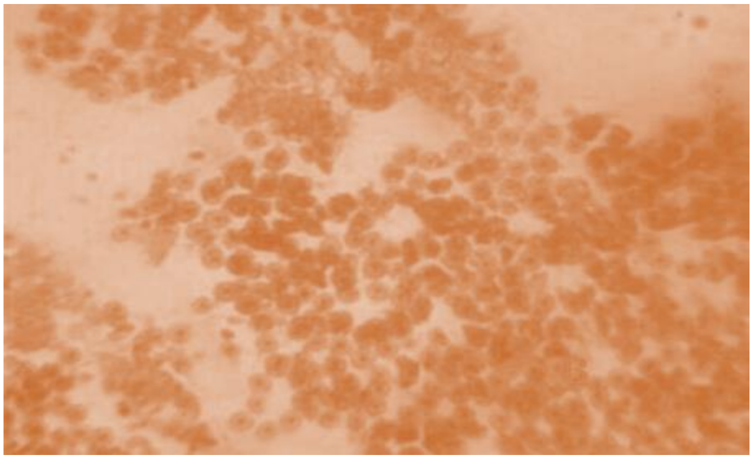

Plate 2: Photomicrograph of liver of *C. gariepinus* exposed to 20 mg/L glyphosate with cell vacuolization and degeneration of hepatocytes (H & E stainx100)

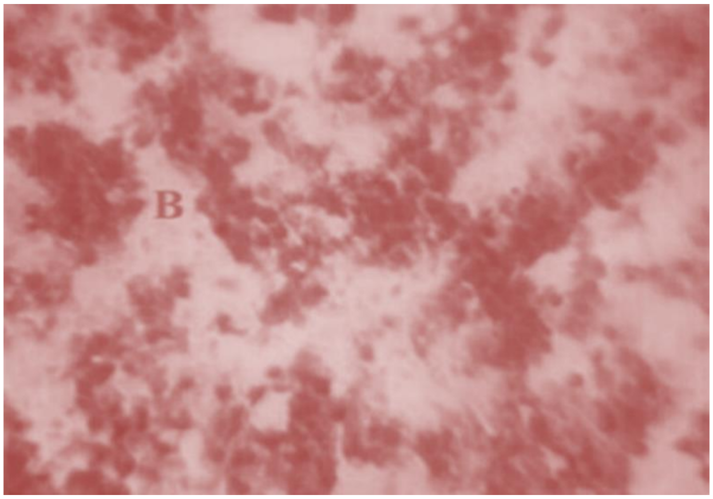

Plate 3: Photomicrograph of liver of *C. gariepinus* exposed to 34 mg/L glyphosate. High cellular vacuolization and degeneration of hepatocytes (H & E stain x100)

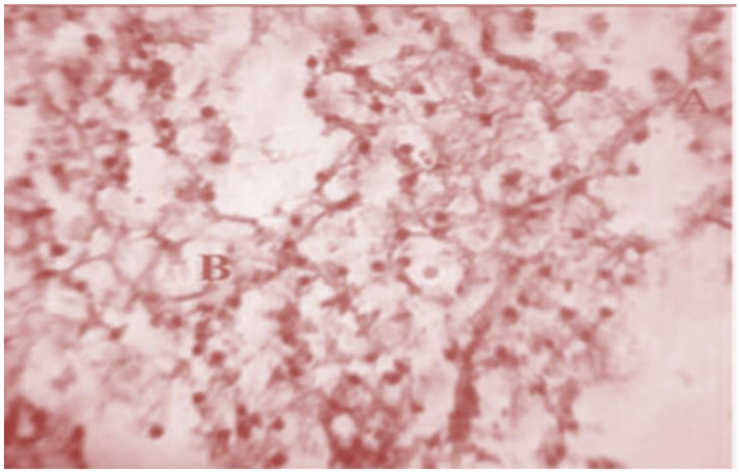

Plate 3: Photomicrograph of liver of *C. gariepinus* exposed to 56 mg/L glyphosate, with high Necrosis, Cell vacuolization and degeneration of hepatocytes (H & E stain x100)

Determination of water quality parameters

There was no significant difference (P<0.05) in temperature among the treatments and the control, with a range of 25.35±0.13 in the control to 25.50±0.14 in 56.00mg/l (Table 1). The pH of water from the exposures did not differ significantly (P<0.05) with the control (0.00mg/l) with a range of 8.06±0.04 in the control and 8.10±0.03 in the highest concentration of 56.00mg/l. Water quality was, however, altered in exposures with higher concentrations of 34 and 56 mg/L. these statistically different (P>0.05) from control and the lower concentration exposures.

Table 1: The Mean Water Quality Parameters in exposure of glyphosate to *Clarias gariepinus* for 28 days Water Quality

Conc.	Water Quality Parameters				
(mg/L)	Temp.(^{0} C)	DO (Mg/l)	TDS (Mg/l)	EC (µs/cm)	pH
0.00	25.35±0.13	4.77±0.35^a	678.25±9.10^a	856.00±1.72^a	8.06±0.04
20.00	25.45±0.15	4.55±0.18^a	385.75±7.94^b	768.30±1.42^b	8.08±0.03
34.00	25.55±0.08	3.37±0.21^b	374.75±6.42^b	756.50±1.69^b	8.10±0.03
56.00	25.50±0.14	2.50±0.17^c	365.50±9.75^b	755.80±1.14^b	8.10±0.03
P-Value	0.78^{ns}	<0.01	<0.01	<0.01	0.85^{ns}

Means on the same column with the same superscript are not significantly different (p<0.05); ns=not significant
Key: Temp. = temperature; DO = Dissolved oxygen; TDS = total dissolved oxygen; EC = electrical conductivity

DISCUSSION

Livers of Juveniles of *C. gariepinus* exposed to varying concentration of glyphosate herbicide in this study had cellular rupture, aggregation of inflammatory cells, and vacuolar degeneration in the hepatocytes, necrosis, and rupture of blood vessels. These results indicate that *C. gariepinus* is sensitive to the herbicide because it presented higher incidence of liver anomalies compared to the control. The liver is the site of detoxification of all chemicals and toxins that have entered the body, the alterations observed may be the effect of the glyphosate on the fish hepatocytes. The findings of this work is similar to that of Fanta *et.al*., (2003) who reported irregular shaped hepatocytes, cytoplasmic vacuolization in *C. gariepinus* contaminated by organophosphate pesticides. Degeneration of the hepatocytes and focal necrosis in the liver of *Clarias gariepinus* exposed to lead were also reported (Olojo *et.al.*, (2005).

Vacuolation of hepatocytes and necrosis were observed by in the liver of *Oreochromis niloticus* after 4 days of exposure to a glyphosate herbicide Ayoola (2008). Ayanda and Egbamuno (2012) reported varying degrees of liver damage in *C. gariepinus* exposed to glyphosate for 10 – 15 days, ranging from mild portal to congestion of central veins. The liver is the site of detoxification of all chemicals and toxins, the changes in the liver observed in this may be attributed to direct effect of glyphosates on hepatocytes (Sani and Idris, 2016). These results also indicate that C. gariepinus may be more sensitive species to the environmental changes because it presented higher incidence of liver anomalies compared with the control.

The alterations in the water quality parameters of Dissolved Oxygen (DO), Total Dissolved Solids (TDS), and Electrical Conductivity (EC) the in the exposures at higher concentrations indicates that glyphosate can cause alterations not only in the test fish, but also in the culture media. These authors also observed a dose dependent reduction of these parameters in exposures of *Clarias* to acute concentrations of glyphosate. Glyphosate may affect fish and wildlife indirectly because killing the plants and small invertebrates alters the animals' habitat (Henderson *et. al*., 2010).

CONCLUSION

Histological changes observed in the livers of Juveniles of *Clarias gariepinus* exposed to glyphosate in this study were cellular rupture, aggregation of inflammatory cells, and vacuolar degeneration of the hepatocytes, necrosis, and rupture of blood vessels. The changes observed in the liver of C. gariepinus indicate that the fish were responding to the effects of glyphosate (360g/L).

REFERENCES

Ayanda, O.I., Egbamuno, E.A. (2012): Histological examination of the liver and gills of *Clarias gariepinus* treated with glyphosate. *Environmental Research Journal* 6 (3): 228 – 234)

Ayoola S.O. (2008): Toxicity of glyphosate herbicide on Nile tilapia (*Oreochromis niloticus*) juveniles. *African Journal of Agricultural Research* Vol. 3 (12), pp. 825-834

Fanta, E., Rios, F. S., Romão, S., Vianna, A. C. C. and Freiberger, S. (2003). Histopathology of the fish *Corydoras paleatus* contaminated with sub lethal levels of organophosphorus in water and food. *Ecotoxicology and Environmental Safety*, 54: 119-130.

Hedayati, A. (2016): Liver as a Target Organ for Eco-Toxicological StudiesJournal of Coastal Zone Management, 19(3) 1000e118

Henderson, A. M.; Gervais, J. A.; Luukinen, B.; Buhl, K.; Stone, D.; Cross, A.; Jenkins, J. (2010): Glyphosate General Fact Sheet; National Pesticide Information Center, Oregon State University Extension Services. http://npic.orst.edu/factsheets/glyphogen.html.

Marioghae, I. E. (1991) Culturable fish PP. 3–10 in Anyinla O. A. (Ed) proceeding of fish seed propagation course, ARAC. 14^{th}-28^{th} August, 1991.

Nosakhare O. E., Alohan, F.I., Alex Enuneku, Isioma Tongo, Martins Ainerua, Stefano Idugboe, (2013).

Pathological alterations in the liver of post-juvenile African Catfish (*Clarias gariepinus*) exposed to sub lethal concentrations of the herbicide Glyphosate. *Journal of Natural Sciences Research Vol.3, No.15*

Sani, A., Idris, M.K. (2016): Acute toxicity of herbicide (glyphosate) in *Clarias gariepinus* juveniles. Toxicology Reports 3 (2016) 513–515

Yancheva, V., Velcheva, I., Stoyanova, S., Georgieva, E. (2015): Histological Biomarkers in Fish as a toll in Ecological Risk Assessment and Monitoring Programs: A preview *Applied Ecology and Environmental Research* 14 (1): 47-75