

# Histopathological Alterations in the Gills and Liver of *Clarias Gariepinus* Juveniles Exposed to Acute Concentrations of *Anogeissus Leiocarpus*

Bala Sambo Audu<sup>1,\*</sup>, Idris Audu Wakawa<sup>2</sup>, Omirinde Jamiu Oyewole<sup>3</sup> and Ponwa Zingfa Changdaya<sup>1</sup>

<sup>1</sup>Hydrobiology and Fisheries Unit, Department of Zoology, University of Jos, Jos, Nigeria; <sup>2</sup>Department of Biology, Umar Suleiman College of Education, Gashua, Yobe State, Nigeria; <sup>3</sup>Department of Anatomy, Faculty of Veterinary Medicine, University of Jos, Nigeria.

Received: August 12, 2020; Revised: November 25, 2020; Accepted: December 12, 2020

## Abstract

Over the years, people have used the stem bark extract of *Anogeissus leiocarpus* in traditional tanneries as a native agent in tendering hide and skin. The wastes from the processing plant are washed into aquatic environment and cause pollution. The acute toxicity (96hr-LC<sub>50</sub>) of the aqueous crude stem extract of *A. leiocarpus* on behaviour and histopathology of gills and liver of *Clarias gariepinus* juveniles (average weight 42.00±0.05 g and average total length 27.83±0.71 cm) were investigated in a static non-renewable bioassay to ascertain its toxicity. A total of five concentrations of aqueous crude stem extract of *A. leiocarpus* (450.00, 400.00, 350.00, 300.00, and 250.00 mg/L) and a control (0.00 g/L) were used. Ten (10) juveniles were stocked in each tank with dimension of 50x30x25 cm. Of the one hundred and twenty (120) mixed sex *C. gariepinus* juveniles (in duplicate replication) used, mortalities of 100, 70, 50, 20, and 10% were recorded in concentrations 450.00, 400.00, 350.00, 300.00, and 250.00 mg/L respectively while control recorded 0% after 96 hr. The 96 hr. LC<sub>50</sub> of *A. leiocarpus* on *C. gariepinus* resulted in 353.77 mg/L characterized by upper and lower confidence limits of 390.27 and 320.69 mg/L respectively. There was marked variation in the water quality parameters (total alkalinity and free carbon dioxide) in all the test tanks compared with the control. The behavioural signs exhibited by *C. gariepinus* exposed to concentrated grades of the plant material were erratic swimming, loss of stability, spiral movement, air gulping, restlessness and settling on tank bottom. Histopathological alterations such as lamellar vascular congestion, lamellar clubbing and partial to complete inter-lamellar space occlusion were recorded in the gills of *Clarias gariepinus* exposed to the graded concentrations of the extract. The liver of the catfish showed dose-related hepatic lesions such as portal congestion, periportal cellular degeneration and cellular infiltration. This study shows that 300.00 to 450.00 mg/L of *A. leiocarpus* is toxic to fish's health. Hence its indiscriminate disposal into aquatic environment should be discouraged or totally avoided to avert death of aquatic animals.

**Keywords:** Acute toxicity; Histopathology; *Anogeissus leiocarpus*; *Clarias gariepinus*

## 1. Introduction

*Anogeissus leiocarpus*, commonly known as African birch, is a deciduous tall plant found in the tropical Africa (Steentoft, 1988). It grows continually to attain a height of 15-18.0 m with the stem measuring about 1 m in diameter (Arbab, 2014). The bark of *A. leiocarpus* is grayish and rough while leaves are alternate, ovate to lanceolate shape with length-width dimensions of 2.0-8.0 cm and 1.3-5.0 cm respectively (Mbagwu, 2011). Small branches of *A. leiocarpus* are crushed to make dyes in tanning skin while decoction of the bark is reputed man and farm animals antihelminthics as well as potent antiprotozoans against malaria and trypanosomiasis in animals (Arbonnier, 2004; Okpekon, 2004). The sticks are chewed into fibrous brush to clean teeth by rural population in Nigeria (Rotimi, 1988).

Plants constitute an unlimited origin of a variety of biological active substances (Istvan, 2000) which have toxic effects on the aquatic biota. Artisanal fishermen use plant extract as part of their arsenal of fishing tools (Power *et al.*, 2010). Extracts of plants such as *Blighia sapda*, *Kigelia africana*, *Raphia, vinifera* (Omoitoyin *et al.*, 1999), *Derris elliptica*, *Tephrosia vogelli* (Oluwatoyin, 2011) and *Balanites aegyptiaca* (Wakawa *et al.*, 2018) have been reported to be used by fishermen as fishing tool. These plant extracts used in harvesting fish have toxic properties (Fafioye *et al.*, 2004) that paralyze or stupefy fish (Fafioye, 2011) in the aquatic environment. Examination of the phytochemicals of plants used as fish poison shows the presence of saponins, alkaloid and flavonoids (Fafioye, 2011). Others are tannins, resins, terpenes, cardiac glycosides and balsam (Wakawa *et al.*, 2018). Saponins affect haematology and oxygen uptake of fish (Roy and Munshin, 1989) while alkaloid and flavonoids have anaesthetic properties on fish (Tsuchiya,

\* Corresponding author e-mail: audusambo@yahoo.com.

2017). Stem bark of *A. leiocarpus* contains phytochemicals such as tannins, flavonoids, terpenes and saponins with absence of alkanoids and anthraquinones (Salau *et al.*, 2013). Introduction of plant extracts containing these phytochemicals could result into physiological stress in aquatic biota which could ultimately reduce aquatic productivity (Oluwatoyin, 2011) or even death.

In view of the effect of the plant extracts used in harvesting fish, many workers have assessed the acute toxicity performance of biosynthetic chemicals of plant origin to cause disease conditions in tissues/organs, serum biochemistry and haematology of different fish species. Audu *et al.* (2020) studied histopathological effects of unrefined water fractions of the foliage of *Balanites aegyptiaca* on gills, kidney and liver of *Oreochromis niloticus* fingerlings. Similarly, Audu *et al.* (2017) examined histological changes in gills and liver of *C. gariepinus* intoxicated with acute concentrated grades of *Vernonia amygdalina*. Also, Adesina *et al.* (2013) evaluated the effect of acute toxicity of *Moringa oleifera* root extract on *O. niloticus*. Nasiruddin *et al.* (2012) investigated the histological alterations in organs of *Heteropneustes fossilis* intoxicated with extracts of three dry seed, while Oluwatoyin (2011) studied the *Ipomoea aquatica* leaf extract toxic potential on histopathology of *O. niloticus*. Shahi and Singh (2011) investigated the effects of extracts of euphorbia plants on serum biochemistry and haematology profiles of *Channa punctatus*. There is, however, paucity of data on acute toxicity of aqueous crude stem bark extract of *A. leiocarpus* on histopathology of *C. gariepinus* juveniles. Hence this study aimed at investigating the possibilities on behaviour and histopathology of gills and liver of *C. gariepinus* juveniles.

## 2. Materials and Methods

### 2.1. Collection and Preparation of Stem Bark of *Anogeissus leiocarpus*

*A. leiocarpus* stem bark (50 g) was collected from Hwol Yarje, Jos North Local Government, Plateau State, Nigeria. The plant was identified by a plant taxonomist as *Anogeissus leiocarpus* (DC.) Guill & Perr; voucher number=JUHN20000324, mounted and deposited in the Herbarium of Plant Science and Technology Department, University of Jos, Nigeria. The stem bark was sheared from the tree plant using an axe and shade-dried over seven days with outdoor relative humidity of 57% and ambient temperature of 28°C. The dried stem bark was pulverized in the laboratory using mortar and pestle into fine particles, sieved with a meshed utensil (30 $\mu$ m) and stored in airtight transparent polyethylene bag for subsequent use.

### 2.2. Laboratory Conditioning of Test Fish (*Clarias gariepinus*)

Outright, one hundred and twenty (120) juveniles (6-9 weeks old) of *C. gariepinus* (average weight 42.00 $\pm$ 0.05 g and average total length 27.83 $\pm$ 0.71 cm) were procured from a private fish farm at Kangang, Dadinkowa, Jos South were moved in three aerated cellophane bags (40 juveniles per bag) to the Zoology Department (Hydrobiology and Fisheries Laboratory Unit), University of Jos and stocked using 10 round plastic tanks of 20 L

capacity (10 fish/tank) each filled with 15 L of dechlorinated municipal water. Commercial diet (Multifeeds®) was given to fish twice daily on satiation basis and water in the holding tanks was changed once daily. Fish were allowed to acclimate for two weeks during this period; fish were held under natural photo regime (12 Light: 12 Dark) (Bala *et al.*, 2014).

### 2.3. Experimental Design

Sequel to acclimation period, fish were divided into experimental tanks which consists of twelve (12) rectangular plastic tanks (50x30x25 cm) and 120 *C. gariepinus* juveniles arranged in a randomized block design. All the tanks contained ten (10 L) liters each of chloric-free pipe borne water, with five (5) of the test tanks exposed to varying concentrations (450.00, 400.00, 350.00, 300.00 and 250.00 mg/L) of the aqueous stem bark extract of *A. leiocarpus*. Ten (10) *C. gariepinus* juveniles each were introduced into all the five (5) test and control tanks. The sixth tank served as the control and was not inoculated with the test material (0.00mg/L). The setup was replicated twice.

### 2.4. Acute toxicity test

Static non-renewal bioassay technique (USEPA, 1985) was used for the 96 hr. LC<sub>50</sub> experiment. The aqueous stem extract was obtained by macerating two (2) grams of the finely grinded particles of stem bark of *A. leiocarpus* in distilled water for 24 hours under room (25°C) condition from which graded concentrations of 450, 400, 350, 300, and 250.00 mg/L were obtained through serial dilution of the stock after range finding tests. The control tank (0.00mg/L) did not contain the test plant. The test tanks with the definitive concentrated grades of the plant extract and the control tanks were each duplicated replicated, stocked with ten (10) juveniles per tank and were devoid of artificial aeration and feeding throughout the 96 hours experiment.

### 2.5. Aquatic medium quality analysis

Physicochemical parameters including dissolved oxygen (DO), temperature, free carbon (iv) dioxide (CO<sub>2</sub>), hydrogen ion concentration (pH) and total alkalinity (TA) were monitored every day as described by the American Public Health Association (APHA) (1985) techniques for water quality for fish culture throughout the experimental phase.

### 2.6. Histopathological Examination

Fish from each of the test concentrations (450.00.00, 400.00, 350.00, 300.00 and 250.00 mg/L) were sacrificed and dissected to excise gills and liver. Excised organs were carefully washed of blood stains and kept in specimen bottles containing 0.005L formal saline (Audu *et al.*, 2017). Histopathological examinations were conducted at the central diagnostic unit of National Veterinary Research Institute (NVRI) Vom, Plateau State, Nigeria. Routine paraffin wax method and haematoxylin-eosin staining technique of tissue processing described by Drury and Wallington (1967) and Avwioro (2011) were adopted for the examinations of the excised organs (gills and liver) of *C. gariepinus* exposed to aqueous crude stem bark extract of *A. leiocarpus*.

### 2.7. Statistical Analyses

Water quality parameters were analysed using IBM SPSS (version 20) software. The Analysis of Variance (ANOVA) was used to establish significant differences. The Tukey test was used to separate treatment means while level of significance was determined at  $\alpha_{0.05}$ .

## 3. Results

### 3.1. Mean Water Quality Parameters of Tanks with *Clarias gariepinus* exposed to Acute Concentrations of *Anogeissus leiocarpus*

Water quality parameters in the 96 hr acute bioassay of *A. leiocarpus* extract on *C. gariepinus* juveniles are summarized in Table 1. Temperature, DO and pH decreased as the toxicant concentration increased while free CO<sub>2</sub> and TA correspondingly rose with elevated toxicant concentrations. P-value of significance ( $p < 0.05$ ) was obtained for TA and free CO<sub>2</sub> in the test tanks compared to the control group.

### 3.2. Behavioural Patterns of *Clarias gariepinus* Juveniles during 96 hr Acute Toxicity Test with Extract of *Anogeissus leiocarpus*

Fish exposed to the plant extract showed behavioural patterns such as erratic movement, loss of stability and spiral movement. Fish settled at the bottom of the tank and remained motionless for a while and sudden frequent swimming to the water surface to gulp air. After prolonged exposure (> 48 hr.) the fish skin peeled off and the fish gradually became weak, and finally death occurred. Fish was considered dead when there was permanent cessation of spontaneous movement and a failure to react to strong external stimulation probe made of glass rod. The death of the juveniles was directly concentration- dependent in relation to exposure time. At the highest plant extract concentration (400.00 mg/L), mortality was within 24 hr exposure while in the lower concentrations (300.00 and 350.00 mg/L) mortality resulted after 48 hr exposure of the juveniles to the plant extract. In the lowest concentrations (250.00 mg/L) of the plant extract, death of juveniles was recorded after 72 hr of exposure to the plant extract (Table 2).

**Table 1.** Mean Water Quality Parameters of Acute Bioassay of Crude Stem Bark Extracts of *Anogeissus leiocarpus* on *Clarias gariepinus* Juveniles

Water Quality Parameters	Concentration (mg/L)					
	0.00	250.00	300.00	350.00	400.00	450.00
Temp. (°C)	25.5±0.00	24.60±0.28	24.20±0.28	24.40±0.07	24.40±0.28	24.20±0.14
DO (mg/L)	2.20±0.00	1.90±0.00	1.90±0.00	1.70±0.00	1.50±0.14	1.45±0.07
TA (mg/L)	60.50±0.00	68.85±0.07*	74.90±0.71*	85.00±12.73*	114.50±0.71*	220.10±0.14*
pH	7.35±0.07	7.20±0.00	7.25±0.07	7.20±0.00	7.20±0.07	7.10±0.14
Free CO <sub>2</sub> (mg/L)	34.00±0.00	49.00±0.00*	49.60±0.85*	54.30±4.94*	60.00±2.83*	68.00±0.00*

Values with Asterisks (\*) in the same Row are Significantly Different Compared with the Control

TA= Total Alkalinity

**Table 2.** Behavioural Patterns shown by *Clarias gariepinus* Juveniles exposed to Aqueous Crude Stem Bark Extract of *A. leiocarpus*

Concentration mg/L	Behavioural Patterns	Exposure Period (Hours)		
		24	48	72
450	Erratic swimming	+++	++	+
	Loss of stability	+	+	+
	Air gulping	+++	++	++
	Spiral motion	-	+	+
	Inactivity	-	+	++
	Peeling of skin	+	++	+++
	Death	+	++	+++
400	Erratic swimming	+++	++	+
	Loss of stability	+	+	+
	Air gulping	++	++	++
	Spiral motion	-	+	+
	Inactivity	-	++	++
	Peeling of skin	-	+	+++
	Death	+	+	++
350	Erratic swimming	+	++	++
	Loss of stability	-	++	++
	Air gulping	+	+	++
	Spiral motion	-	++	++
	Inactivity	-	-	+
	Peeling of skin	-	-	+
	Death	-	-	+
300	Erratic swimming	+	++	++
	Loss of stability	-	++	++
	Air gulping	+	+	++
	Spiral motion	-	++	++
	Inactivity	-	-	+
	Peeling of skin	-	-	+
	Death	-	-	+
250	Erratic swimming	-	+	+
	Loss of stability	-	+	+
	Air gulping	-	+	+
	Spiral motion	+	+	+
	Inactivity	-	-	-
	Peeling of skin	-	-	-
	Death	-	-	-

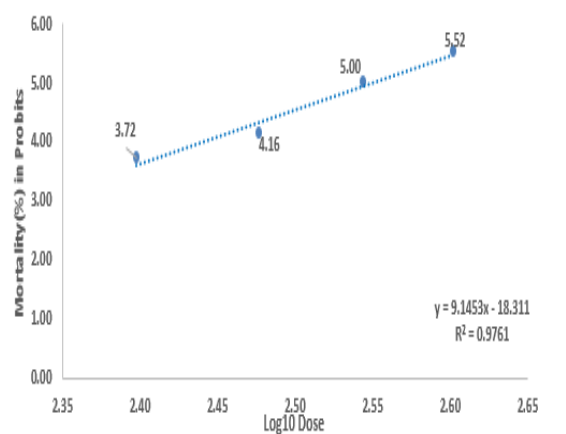
No recorded pattern (-), slight (+), moderate (++), severe (+++)

### 96 hr (LC<sub>50</sub>) Acute Bioassay of Extract of *Anogeissus leiocarpus* on Survival of *Clarias gariepinus* Juveniles

The effect of 96 hr LC<sub>50</sub> acute bioassay of the extract of *A. leiocarpus* on survival of *C. gariepinus* juveniles is shown in Table 3. Survivals of the juveniles depend on the concentration of the extract. The rate of survival increased with decrease in the aqueous crude stem extract concentration. Recorded mortalities were 100, 70, 50, 20 and 10% of the plant extract concentrations 450.00, 400.00, 350.00, 300.00 and 250.00 mg/L respectively. The control tank (0.00 mg/L) recorded 0% mortality.

### 3.3. Linear Relationship Between Mean Probit Mortality Versus Log Concentration of *C. gariepinus* Juveniles Exposed to Extract of *Anogeissus leiocarpus*

Figure 1 shows the linear relationship between mean probit mortality and log concentration of *C. gariepinus* juveniles exposed to the extract of *A. leiocarpus*. The 96hr LC<sub>50</sub> of *A. leiocarpus* on *C. gariepinus* resulted in 353.77 mg/L characterized by upper and lower confidence limits of 390.27 and 320.69 mg/L respectively.

**Figure 1.** Linear Relationship between Mean Probit Mortality and Log Concentration of *Clarias gariepinus* Juveniles Exposed to Aqueous Crude Stem Bark Extract of *Anogeissus leiocarpus***Table 3.** Effects of Acute Bioassay of Aqueous Crude Stem Bark Extract of *A. leiocarpus* on Survival of *Clarias gariepinus* Juveniles

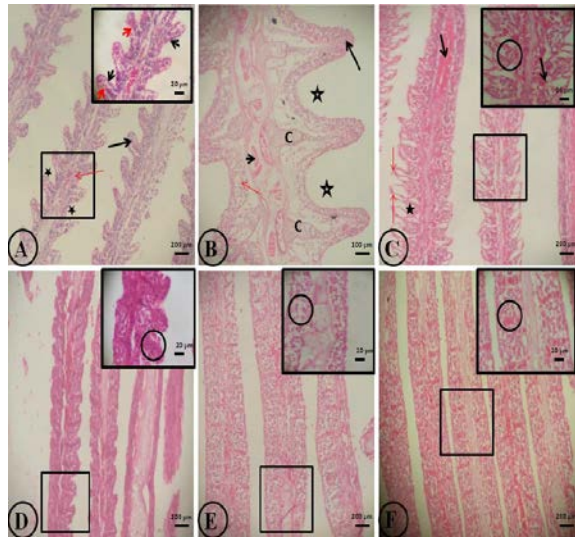
Conc. (mg/L)	Log Conc.	No. of fish	Mortality Time (Hours)	Mortality Time (Hours)								Total Mortality (%)	Probit Mortality
				12	24	36	48	60	72	84	96		
450.00	2.6532	10	3.50	2.50	1.50	1.00	1.00	1.00	0.50	0.00	10	100	8.7190
400.00	2.6020	10	2.00	1.50	0.50	1.50	0.50	0.50	0.00	0.50	7	70	5.5244
350.00	2.5440	10	0.00	0.00	1.00	0.50	0.50	0.50	0.00	1.00	5	50	5.0000
300.00	2.4771	10	0.00	0.00	0.00	1.50	0.00	0.00	0.00	0.50	2	20	4.1584
250.00	2.3979	10	0.00	0.00	0.00	0.00	0.00	0.50	0.5	0.00	1	10	3.1784
0.00	0.0000	10	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0	0	0.0000

### 3.4. Histopathology of Gills of *Clarias gariepinus* Juveniles

Photomicrograph of effects of 96hr acute concentrations of extract of *A. leiocarpus* on gills of juveniles of *C. gariepinus* are presented in Plate 1 A-F. The gills of the control (Plate 1A) appear morphologically normal with typical structural organization of primary and secondary lamellae. There were progressive dose-

dependent gill branchial-lamella injuries typified by slight (Plate 1B) to heavy vascular congestion and clubbing of the lamellae, partial to complete interlamellar space occlusion and lamellar cell hyperplasia. The gravity of the tissue distortion seemed to be more visible in the gill anatomical structure of juveniles treated with concentrations of 300.00, 350.00, 400.00, and 450.00 mg/L of the plant extract (Plates C-F). The histo-

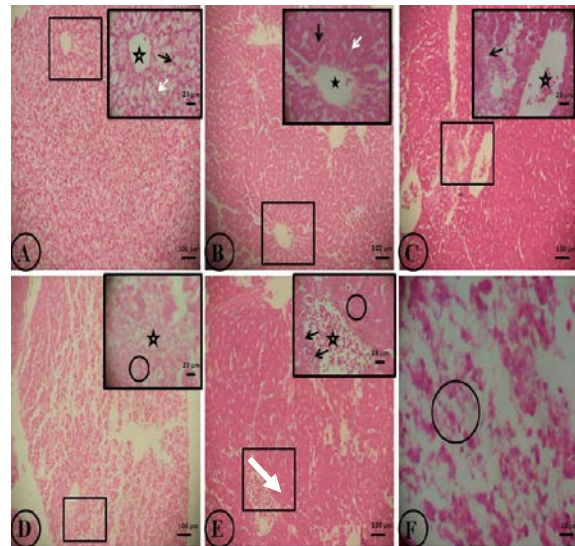
architecture of *C. gariepinus* intoxicated with the 250.00 mg/L of the extract appeared to be normal when compared to the control.



**Plate 1.** Light micrographs of the Gills of *Clarias gariepinus* Exposed to Acute Concentrations of Aqueous Crude Stem Bark Extract of *Anogeissus leiocarpus*. **A. Control (0.00 mg/L):** normal gill parenchyma typified by occurrence of structures like primary (red arrow) and secondary lamellae (black arrow), distinct epithelial (short black arrow) and pillar cells (short red arrow) of the secondary lamellae and patent inter-lamellar space or water channel (star). **B. 250.00 mg/L:** Normal parenchyma as revealed by intact primary and secondary (red and black arrows respectively) lamellar epithelium and water channels (star) except for mild lamellar vascular congestion (short black arrow) **C. 300.00 mg/L:** Apical lamellar clubbing (red arrow), moderate lamellar cell fusion with partial to complete interlamellae space occlusion (star and circle outline (within inset) respectively), severe lamellar vascular congestion (black arrow) **D. 350.00 mg/L:** Severe hyperplastic lamellar cell with total disappearance of water channels (oval-outline). **E. 400.00 mg/L; F. 450.00 mg/L:** Severe lamellar cell hyperplasia with complete inter-lamellar space occlusion (oval outlines within inset).

### 3.5. Histopathology of Liver of *Clarias gariepinus* Juveniles

Histopathological lesions noticed in the *C. gariepinus* liver exposed to acute concentrated grades of crude extract of *A. leiocarpus* are presented in Plate 2 A-F. The histopathological features of liver of *C. gariepinus* juveniles in the non-exposed (control) group include normal hepatic histo-architecture characterized by polyhedral outline and roundish nuclear, sinusoidal space and central veins; while, in *C. gariepinus* groups exposed to various concentrations of *A. leiocarpus*, the liver displayed increase in severity of histopathological lesions as concentration of the plant extract increased. Histopathological lesions observed in the treatment groups include moderate to severe portal congestion, periportal cellular degeneration, cellular infiltration and complete hepatocellular depletion.



**Plate 2.** Light micrographs of the Liver of *Clarias gariepinus* Exposed to Acute Concentrations of Aqueous Crude Stem Bark Extract of *Anogeissus leiocarpus*. **A. Control (0.00 mg/L):** normal hepatic histo-architecture as revealed by hepatocytes with typical polyhedral outline and roundish nuclear (black arrow), sinusoids space (white arrows) and central vein (star). **B. 250.00 mg/L:** no visible lesion as evidenced by liver with intact hepatocyte (black arrow), sinusoids space (white arrow) and central vein (star) **C. 300.00 mg/L:** visible lesion except for moderate sinusoidal congestion (black arrow). **D. 350.00 mg/L:** moderate portal congestion (star) and peri-portal cellular degeneration (circle outline). **E. 400.00 mg/L:** severe portal congestion (star), marked peri-portal cellular infiltration (arrow) and peri-portal cellular degeneration (circle outline) **F. 450.00 mg/L:** Severe hepatocellular depletion (circle outline).

### 4. Discussion

Determination of water quality parameters in fish culture during an experiment is necessary owing to the complete dependent nature of the whole life process of fish on the quality of the immediate surroundings (Bolorunduro and Abdullahi, 1996). Water quality is determined to confirm whether it plays any role in the alterations observed during the experiment as decline in water quality influences stress and disease in fish (Devi *et al.*, 2017). The concentration-dependent decrease in DO in this study corroborates the findings of Makori *et al.* (2017). The minimum DO requirement of fish is 3.00 mg/L (Makori *et al.*, 2017); therefore, the minimum mean DO ( $1.45 \pm 0.07$  mg/L) in this study could be attributed to the presence of the plant extract in the water (Adebola and Ayo, 2014). The pH range ( $7.10 \pm 0.14$ - $7.25 \pm 0.07$ ) in treatment tanks of this study is within the tolerable limits of catfish since the optimum pH for their survival is between 5 and 8 (Nobre *et al.*, 2014); therefore, the pH in this study could not have affected the recorded mortalities in the test animal. Similarly, the recorded temperature range ( $24.20 \pm 0.14$ - $24.60 \pm 0.28$  °C) in treatment tanks falls within the standard range (20-35 °C) documented by Ngugi *et al.* (2007), hence temperature could not have influenced the observed distortion in the histology of organs (gills and liver) of *C. gariepinus*. The TA which measures water productivity shows that the water tainted with the plant extract in this study is productive since the TA range ( $68.85 \pm 0.07$ -

220.10±0.14 mg/L) is within the productive (50-500 mg/L) level (Devi *et al.*, 2017).

The abnormal behavioural patterns such as erratic movement, loss of stability, spiral movement and air gulping displayed by *C. gariepinus* juveniles in the higher treatment concentrations (350.00, 400.00 and 450.00 mg/L) of the aqueous stem bark extract could be due to the fish's deliberate effort to overcome the toxic plant bioactive substances and the hypoxic condition possibly caused by the aqueous stem bark extract of *A. leiocarpus*. The manifested behavioural patterns have earlier been linked to derangement in the biochemical and nervous systems of the stressed fish (Fadina *et al.*, 1991; Fafioye *et al.*, 2005). In addition, the dose-related increase in the mortality rate seen in this study further established the toxic nature of *Anogeissus leiocarpus*. The trends of behavioural signs and the mortality rate shown by fish exposed to graded concentrations of *A. leiocarpus* are similar to those documented for *Trephosia vogelii* (Adewoye, 2010), *Parkia biglobosa* (Ojutiku *et al.*, 2012), *Carica papaya* (Eyo *et al.*, 2013), and *Vernonia amygdalina* (Audu *et al.*, 2017).

Histological distortions have been extensively optimized as biomarkers of pollutants in fish (Naeemi *et al.*, 2013). Gill of fish plays important function including respiration, osmoregulation and excretion (Camargo and Martinez, 2007; Jalaludeen *et al.*, 2012; Audu *et al.*, 2017) due to its contact with the immediate water environment (Olojo *et al.*, 2005). This proximity with the external environment predisposes it to histological damages such that the fish becomes vulnerable to respiratory and osmoregulatory difficulties (Olusegun and Adedayo, 2014) especially when toxicants enter the body and cause damage to gill membranes and affect its physiological functions (Bala and Malachy 2020). Succinctly put, fish exposed to toxicants die when their gill lamella epithelia and blood vessels are adversely affected (Hinton and Lauren, 1990).

Therefore, the observed moderate to severe gill histo-architectural alterations (lamellar vascular congestion, lamellar clubbing, partial to complete inter-lamellar space occlusion and lamellar cell hyperplasia) in this study further established the toxic potential of *A. leiocarpus* extract. The gill histopathological profiles in this study corroborate lesions earlier reported in similar studies conducted by Camargo and Martinez (2007) in Neotropical fish caught from stream laden with toxicant and the histopathological report of Nasiruddin *et al.* (2012) on *Heteropneustes fossilis* exposed to three dried leaves extracts.

The detoxification and biotransformation processes are reputed functions of the liver which has directly placed it as most morpho-physiologically disrupted organ by contaminants in the water (Hadi and Alwan (2012). With respect to these physiological roles, the histopathological alterations (moderate to severe portal congestion, periportal cellular degeneration, cellular infiltration and complete hepatocellular depletion) shown by *C. gariepinus* exposed to graded concentrations of *A. leiocarpus* could precipitate serious hepatic dysfunction. The dose-related disruption in the liver parenchyma of *C. gariepinus* juveniles has earlier been credited to extreme physiological activities needed by the fish to excrete the toxic substances out of its body during detoxification and

biotransformation processes (Adebola and Ayo, 2014). However, further studies that will incorporate liver enzymes profiles and anti-oxidant assays will be necessary to reveal the impact of the extract (aqueous stem bark) of *A. leiocarpus* on *C. gariepinus* liver physiology.

## 5. Conclusion

This study has demonstrated that aqueous crude stem bark extract of *A. leiocarpus* has deleterious and piscicidal effects on *C. gariepinus* juveniles, hence its washing, processing and indiscriminate disposal into aquatic environments should be discouraged or totally avoided to preserve aquatic biodiversity and abundance particularly of fish species such as tropical freshwater African catfish *C. gariepinus*.

## Acknowledgement

The authors are appreciative of the laboratory space provided by the Department of Zoology, University of Jos for the conduction of the experiment and for the expertise rendered by the technical staff of the Central Diagnostic Unit of National Veterinary Research Institute, Vom, Plateau State, Nigeria.

## References

- Adebola K and Ayo F. 2014. Histological changes in liver, gills and kidney of catfish (*Heterobranchus bidorsalis*) exposed to cypermethrin concentration. *Int. J Histol .Cytol.*, **1**: 31-36.
- Adesina BT, Omitoyin BO, Ajani EK and Adesina OA. 2013. Acute-lethal toxicity (LC<sub>50</sub>) of *Moringa oleifera* (Lam.) fresh root bark extract on *Oreochromis niloticus* juveniles under renewal toxicity exposure. *Int. J Appl. Agric. Apicu. Res.*, **1**: 182-188.
- Adewoye SO. 2010. Haematological and biochemical changes in *Clarias gariepinus* exposed to *Trephosia vogelii* extract. *Adv. Appl. Sci. Res.*, **1**(1): 74 – 79.
- APHA (American Public Health Association) 1985. **Standard Methods for Examination of Water and Wastewater**. Washington. USA. Pp: 258-259.
- Arbab AH. 2014. Review on *Anogeissus leiocarpus* a potent African traditional drug. *Int. J Res. Pharm. Chem.*, **4**: 496-500.
- Arbonnier M. 2004. **Trees, Shrubs and Lianas of West African Dry Zones**. The Netherlands: Cirad Margraf. Pp. 573.
- Audu BS, Omirinde JO, Gosomji IJ and Wazhi PE. 2017. Histopathological changes in the gill and liver of *Clarias gariepinus* exposed to acute concentrations of *Vernonia amygdalina*. *Ani. Res. Int.*, **14**: 2576-2587.
- Audu BS, Wakawa IA, Omirinde JO, Garba U and Damshit M. 2020. Histopathological alterations in organs of Nile tilapia fingerlings exposed to sublethal concentrations of aqueous crude leaves extract of desert date. *Pan Afri. J Life Sci.*, **4**: 59-67.
- Bala SA, Kabir MA and Paul CO. 2014. Biochemical Parameters of Common Carp (*Cyprinus carpio*) exposed to Crude Leaf Extract of *Cannabis sativa*. *Jordan J Biol Sci.*, **7**, 147 – 151. Cite score (0.47)
- Bala SA and Malachy NOA. 2020 Metabolic enzyme profile, behavioural changes and morpho physiological parameters of African catfish *Clarias gariepinus* juveniles in response to burnt waste tyres. *Comp Clin Pathol.*, **29**:787–797.

- Bolorunduro PI and Abdullah AY. 1996. Water quality management in fish culture, national agricultural extension and research liaison services, Zaria, Extension Bulletin No. 98.
- Camargo MMP and Martinez CBR. 2007. Histopathology of gills, kidney and liver of a neotropical fish caged in an urban stream. *Neotrop Ichthyo.*, **5**: 327-336.
- Devi PA, Padmavathy P, Aanand S and Aruljothy K. 2017. Review on water quality parameters in freshwater cage fish culture. *Int. J App. Res.*, **3**: 114-120.
- Drury R Wallington E. 1967. **Histological Technique**. 4th edition, Oxford University Press, USA, pp 279-280.
- Eyo JE, Levi CA, Asogwa CN, Odii EC, Chukwuka CO, Ivoke N, Onoja US and Onyike CC. 2013.
- Toxicity and effect of Carica papaya seed aqueous extract on liver biomarkers of *Clarias gariepinus*. *Int. J Indig. Med. Plant.*, **46**(3): 1301 – 1307.
- Fadina OO, Taiwo VO. and Ogunsanmi AO. 1991. The effects of single and repetitive oral administration of common pesticides and alcohol on rabbits. *Trop Vet.*, **17**: 97 – 106.
- Fafioye OO, Fagade S O and Adebisi AA. 2005. Toxicity of *Raphia vinifera*, P. beauv fruit extracts on biochemical composition of Nile tilapia (*Oreochromis niloticus*, Trewavas). *Biokemistri.*, **17**: 137 – 142.
- Fafioye OO. 2011. Lethal and sublethal effects of plant extract on some freshwater fauna. PhD thesis, University of Ibadan, Nigeria.
- Fafioye OO, Adebisi AA and Fagade SO. 2004. Toxicity of *Parkia biglobosa* (locust bean) and *Raphia vinifera* extracts on *Clarias gariepinus* juveniles. *Afri. J Biotech.*, **3**: 627-630.
- Hadi AA and Alwan SF. 2012. Histopathological changes in gills, liver and kidney of fresh water fish, *Tilapia zillii*, exposed to aluminium. *Int. J Pharm Life Sci.* **3**: 2071-2051.
- Istvan U. 2000. Semi-natural products and related substances as alleged botanical pesticides. *Pest Manage. Sci.*, **56**: 703-705.
- Jalaludeen MD, Arunachalam M, Raja M, Nandagopal Bhat SA, Sundar S and Palannimuthu D. 2012. Histopathology of the gill, liver and kidney tissues of the freshwater fish *Tilapia mossambica* exposed to cadmium sulphate. *Int. J Adv. Biol. Res.*, **2**: 572-578.
- Hinton DE and Laurén DJ. 1990. Liver structural alterations accompanying chronic toxicity in fishes: Potential Biomarkers of Exposure. In: McCarthy JF and Shugart LR (Eds.). **Biomarkers of Environmental Contamination**, Boca Raton: Lewis Publishers. pp. 51-65.
- Makori AJ, Aboum PO, Kapiyo R, Ayona DN and Dida GO. 2017. Effects of water physico- chemical parameters on tilapia (*Oreochromis niloticus*) growth in earthen ponds in Teso North Sub-county, Busia county. *Fish. Aqua. Sci.*, **20**: 2-10.
- Mbagwu ES. 2011. Acute toxicity of *Persea americana* crude seed extracts on the physiology and histopathology of *Clarias gariepinus* fingerlings under laboratory conditions. BSc project, University of Jos, Jos, Nigeria.
- Naeemi A, Jamili S, Shabanipour N, Mashinchian A and Shariati FS. 2013. Histopathological changes of gill, liver, and kidney in *Caspian kutum* exposed to linear alkylbenzene. *Iran J Fish. Sci.*, **12**: 887-897.
- Nasiruddin M, Azadi MA and Jahan A. 2012. Histopathological changes in gill, liver and intestine of *Heteropneustes fossilis* (bloch) treated with three dry seed extracts. *J Asia Soc Bangladesh Sci.*, **38**: 217-226.
- Ngugi CC, James RB, and Bethuel OO. 2007. **A New Guide to Fish Farming in Kenya**, Oregon State University, USA. 95pp.
- Nobre MKB, Lima FRS and Magalhaes FB. 2014. Alternative liming blends for fish culture. *Acta Sci. Ani. Sci.*, **36**: 11-16.
- Ojutiku, RO, Avbarefe EP, Kolo RJ and Asuwaju FP. 2012. Toxicity of *Parkia biglobosa* pod extract on *Clarias gariepinus* juveniles. *Int. J Fish. Aquac.*, **4**(7): 133 – 138.
- Okpekon T. 2004. Antiparasitic activities of medicinal plants used in Ivory Coast. *J Ethnopharm.*, **90**: 91-97.
- Olojo EAA, Olurin KB, Mbaka G and Oluwemimo AD. 2005. Histopathology of Gills and Liver Tissues of the African Catfish (*Clarias gariepinus*) exposed to Lead. *Afri. J Biotech.*, **4**: 117-122.
- Olusegun AA and Adedayo OO. 2014. Haematological responses, serum biochemistry and histology of *Clarias gariepinus* (Burchell, 1822) exposed to sublethal concentrations of cold water fresh root bark extracts of *Plumbago zeylanica* (leadwort). *J. Aqua. Res. Dev.*, **5**: 282-288.
- Oluwatoyin AS. 2011. Histopathology of Nile tilapia (*Oreochromis niloticus*) juveniles exposed to aqueous and ethanolic extracts of *Ipomoea aquatic* leaf. *Int. J Fish. Aquac.*, **3**: 244-257.
- Omoitoyin BO, Ogunsami AO and Adesina BT. 1999. Studies on acute toxicity of piscidal plants extract on tilapia (*Sarotherodon gallicus*) fingerlings. *Tropic J Ani. Sci.*, **2**: 191-197.
- Power DM, Fuentes J, Harrison AP. 2010. "A noninvasive monitoring device for anaesthetics in fish" *Dove press*, <http://www.dovepress.com> (May 5, 2014).
- Rotimi VO. 1988. Activities of Nigerian chewing stick extracts against *Bacteroides gingivalis* and *Bacteroides melaninogenicus*. *Antimicrob Agents Chemothera.*, **32**: 598-600.
- Roy RK and Munshin DJ. 1989. Effects of saponin extracts in oxygen uptake and haematology of air breathing climbing perch (*Anabastes titudines*) (Bloch) *J Fresh Water Biol.*, **1**: 167-172.
- Salau AK, Yakubu MT, Oladiji AT, 2013. Cytotoxic activity of aqueous extracts of *Anogeissus leiocarpus* and *Terminalia avicennioides* root barks against ehrlich ascites carcinoma cells. *Indian J Pharm.*, **45**: 381-385.
- Shahi J and Singh A. 2011. Effect of bioactive compounds extracted from euphorbious plants on haematological and biochemical parameters of *Channa punctatus*. *Rev Inst. Med. Tropic. Sao Paulo.*, **53**: 259-263.
- Steenoft M. 1988. **Flowering plants in West Africa**. Cambridge University Press. London UK. Pp234.
- Tsuchiya H. 2017. Analgesic agents of plant origin. A review of phytochemicals with anaesthetic activity. *Molecules.*, **22**: 1369-1402.
- USEPA 1985. Disciplinary Review Ecological Effects Profile. US Environmental Protection Agency, Office of Pesticide Programs, 401 M St. S.W., Washington DC 20460, USA.
- Wakawa AI, Audu BS, Sulaiman Y. 2018. Phytochemistry and proximate composition of root, stem bark, leaf and fruit of desert date, *Balanites aegyptiaca*. *J. Phytopharm. Pharmacog. Phytomed. Res.*, **7**: 464-470.