Asian Journal of Environment & Ecology

Volume 23, Issue 8, Page 74-85, 2024; Article no.AJEE.119216
ISSN: 2456-690X

Investigating the Impact of *Senna alata* Extract on Hematology and Histopathology of Juvenile of *Clarias gariepinus*

Essien-Ibok, M. A. [a], George, U. U. [b*], Ajayi, O. O. [c] and Okokon, P. [a]

[a] Department of Fisheries and Aquatic Environmental Management, University of Uyo, Nigeria.
[b] Department of Fisheries and Aquaculture, Faculty of Agriculture, Akwa Ibom State University, Obio Akpa Campus, Akwa Ibom State, Nigeria.
[c] Department of Pathology, Faculty of Basic Medical Sciences, University of Uyo, Akwa Ibom State, Nigeria.

Authors' contributions

This work was carried out in collaboration among all authors. All authors read and approved the final manuscript.

Article Information

DOI: https://doi.org/10.9734/ajee/2024/v23i8585

Open Peer Review History:
This journal follows the Advanced Open Peer Review policy. Identity of the Reviewers, Editor(s) and additional Reviewers, peer review comments, different versions of the manuscript, comments of the editors, etc are available here: https://www.sdiarticle5.com/review-history/119216

Original Research Article

Received: 22/04/2024
Accepted: 24/06/2024
Published: 24/07/2024

ABSTRACT

This paper sheds light on the mechanism of action, potential effects and implication of *Senna alata* on fish health and ecosystem. *Senna alata* is renowned for its medicinal properties and ornamental value but is also recognized for its potential toxicity, particularly to aquatic organisms Understanding the toxicity of *Senna alata* to *Clarias gariepinus* fingerlings is essential for sustainable aquaculture

**Corresponding author: E-mail: ubonggeorge@aksu.edu.ng;*

Cite as: *M. A., Essien-Ibok, George, U. U., Ajayi, O. O., and Okokon, P. 2024. "Investigating the Impact of Senna Alata Extract on Hematology and Histopathology of Juvenile of Clarias Gariepinus". Asian Journal of Environment & Ecology 23 (8):74-85. https://doi.org/10.9734/ajee/2024/v23i8585.*

practices and environmental conservation efforts. 250 juveniles of *C. gariepinus* were procured from E. I farm Limited in Ikot Ekpene, Akwa Ibom State. Water quality tests were carried out to ensure that optimum water conditions were maintained and that the results obtained were solely due to the effects of the treatments (toxicants) on the fish samples. Chronic toxicity bioassay was done to determine the toxicity of the extract on the blood parameters and tissues of sensitive organs of the exposed fish. The bioassay was designed to be intermediate termed chronic test with 14 days exposure using sub-lethal concentrations of the treatment (0.0gm/, 0.18mg/L, 0.27mg/L. 0.4mg/L, 0.60mg/L, 0.90mg/L). Toxicology dose response (fish mortality) was subjected to detailed analysis using Statistical Package for Social Sciences (SPSS) 22.0. Data were analyzed using descriptive statistics. *Senna alata* exerted a toxic effect on the fish in the present study and toxicity increased with increased concentration. These findings underscore the potential adverse effects of prolonged exposure to *Senna alata* on the physiological and cellular integrity of the fish. Intriguingly, the absence of pathological changes in the control groups suggests a distinct response attributable to the treatment. This highlights the importance of continued monitoring and assessment of the impacts of *Senna alata* on aquatic organisms and ecosystem, emphasizing the need for further research to elucidate the underlying mechanisms and potential long-term consequences.

Keywords: Senna alata; Clarias gariepinus; toxicity; hematology; histopathology.

1. INTRODUCTION

The use of piscicide in treatment of ponds prior to stocking with desired fish species is a common practice in aquaculture. Use of synthetic piscicides for this purpose is not much appreciated by fish farmers as synthetic piscicides are non-biodegradable and persistent in the environment. Hence, the use of organic piscicides is embraced by fish farmers as they are biodegradable and non-persistent in the environment and fish killed are edible. This makes research on more plants of ichthyotoxic potential important to avail the famers more of such plants for this purpose.

Senna alata, commonly known as Candle bush or Candlestick senna, is a tropical shrub belonging to the Fabaceae family. It is renowned for its medicinal properties and ornamental value but is also recognized for its potential toxicity, particularly to aquatic organisms [1,2]. *Senna alata* is characterized by its distinctive appearance, featuring bright yellow flowers arranged in cylindrical clusters and a slender stem. The plants leaves are large, compound and ovate with a glossy texture and serrated edges. These leaves are alternately arranged along the stems and emit a strong odour when crushed or bruised, a trait often attributed to the presence of chemical compounds such as anthraquinones and saponins [2].

“The chemical composition of *Senna alata* includes various bioactive compounds, some of which include, phenolics (rhein, chryoasaphanol, kaempferol, aloeemodin, and glycosides), anthraquinones (alatinone and alatonal), fatty acids (oleic, palmitic, and linoleic acids), steroids, and terpenoids (sitosterol, stigmasterol, and campesterol)” [3], (Kare, 2007). “These secondary metabolites are reported to display numerous biological activities” [4].

“The flower, root, leaves, seed, and bark displayed diverse biological activities” [5,6]. These pharmacological activities include antimicrobial (Villasenor et al*.,* 2002), [7,8], antifungal [9], anticryptococcus [10], antibacterial [11], (Gaikward, 2014), antitumor (Olater et al*.,* 2010), anti-inflammatory (Manaogaran et al*.,* 2004), [12], antidiabetic [13], antioxidant [5], wound healing [14], and antihelmintic activities [15]. In recent times, the outbreak of drug-resistant diseases has led to several health issues. In an attempt to resolve these issues, pharmacological research has been tailored towards the discovery of innovative, potent, and safe drugs from natural compounds.

Despite the traditional use of *Senna alata* for medicinal purposes in humans and livestock, its impacts on aquatic life particularly fish species warrant investigation. In aquaculture, understanding the interactions between *Senna alata* and fish species like *Clarias gariepinus* (African catfish) is crucial due to the plant’s widespread presence in aquatic ecosystems and its potential impacts on fish health and growth.

“Biologically, the African catfish, *Clarias gariepinus* is the ideal aquaculture species in the world. It is widely distributed, thrives in diverse environments (temperate to tropical), and is

highly adoptable and an ecological pioneer species, principally as a major consequence of its air-breathing ability. It feeds on wide array of prey and can adopt its feeding habit depending on the food availability. It is able to withstand adverse environmental conditions highly fecund and spawn under captive conditions" [16-21]. "It has a wide tolerance of relatively poor water quality and possibly the most exciting features of its species is its potential for highly intensive culture without prerequisite pond aeration or high-water exchange rates and its excellent meat quality" [18-21].

Therefore, understanding the toxicity of *Senna alata* to *Clarias gariepinus* fingerlings is essential for sustainable aquaculture practices and environmental conservation efforts. Also, this paper will shed more light on the mechanism of action, potential effects and implication of *Senna alata* on fish health and ecosystem.

2. MATERIALS AND METHODS

2.1 Collection of Test Organisms (*C. gariepinus*)

250 juveniles of *C. gariepinus* were procured from E. I farm Limited in Ikot Ekpene, Akwa Ibom State. An average weight of 38.58g and length ranging from 20 to 22cm were transported to Vika Farms Limited in plastic containers filled with oxygenated water and drops of palm oil to reduce stress and suffocation before reaching the farm. Collection and transportation were done between 4pm to 6pm to further prevent stressful conditions.

2.2 Acclimatization of *Clarias gariepinus*

Test organisms were kept in a transparent plastic tank of length 18cm and width 13cm. The test organisms were acclimatized for 14 days (2 weeks) as recommended by APHA (2005). During this period, fish were fed twice daily according to APHA (2005) at 5% body weight with commercial feeds (blue crown). Water was changed twice daily to remove faecal materials and unconsumed feeds. The tanks were covered with netting material to prevent fish from jumping out from the plastic container and also protected from predators. Feeding was discontinued during the test period.

2.3 Collection and Identification of *S. alata*

Fresh leaves of *S. alata* were gathered from Ndueso's Farm, Ukpap, Ikot Idang, Ikono, Akwa Ibom State southern Nigeria. The plant was identified by Mr. Etefia of Pharmacology Department (Pharmacognosy unit, University of Uyo.

2.4 Preparation of Ethanol Leaf Extract of *Senna alata*

Extraction was done by solvent extraction technique according to Doughari (2012). Collected plant specimens were cleansed and mechanically cut into small pieces using knife and shade-dried to constant weight to obtain a dry matter. The dry matter is pulverized using a mortar and pestle to obtain finer particles of the dry matter.

After pulverizing a known weight of the pulverized dry matter was macerated in 3L of 70% ethanol concentration for 72hours, following the methods of Adewoye (2010). The macerated dry matter was filtered into a transparent plastic bucket carefully labeled using a cotton wool and filter paper to obtain the extract. Extract obtained was concentrated in a 1000ml beaker and was evaporated in a water bath at 40^0C. Concentrated extracts were transferred using spatula into a beaker and stored with a covered aluminum foil for bioassay immediately after the evaporation was completed.

2.5 Water Quality Tests

Water quality tests was carried out to ensure that optimum water conditions were maintained and that the results obtained were solely due to the effects of the treatments (toxicants) on the fish samples. The physico-chemical parameters of the test water with respect to temperature, dissolved oxygen (DO), pH, Total Dissolved Solids (TDS) and conductivity were determined using (Mercury in glass) thermometer, digital meters designed for the respective purposes. The measurements were taken before and after acclimatization of organisms, before and after the introduction of toxicant and continued twice daily throughout the 96 hours period of exposure of the bioassay.

2.6 Chronic Toxicity Bioassay

"Chronic toxicity bioassay was done to determine the toxicity of the extract on the blood parameters and tissues of sensitive organs of the exposed fish. The bioassay was designed to be intermediate termed chronic test with 14 days exposure using sub-lethal concentrations of the

treatment (0.0mg/L, 0.18mg/L, 0.27mg/L. 0.4mg/L, 0.60mg/L, 0.90mg/L)" [22].

The treatment was renewed every 24hours and physicochemical parameters of the test media measure before and after every renewal [23]. At the end of the 14 days exposure, blood and tissue samples were collected from randomly selected fish from each treatment for further analysis.

2.7 Collection of Blood and Tissues Samples

Blood samples were collected from randomly selected fished from each treatment. The sample was collected from the fish using syringe and needle and preserved in EDTA treated bottles for hematological analysis. Fish tissues (gill, gut and liver) were obtained by dissecting open the lower part of the operculum to access the internal organs of fish. The organs of interest were carefully removed and preserved in sample bottles containing 40% formalin and properly labelled against the respective organs and treatments. The samples were subsequently taken to the University of Uyo teaching hospital histology laboratory for histology analysis.

2.8 Blood and Tissue Analysis

Collected blood samples were subjected to auto-haem analysis using auto-haem analyser in the haematology laboratory of University of Uyo Teaching Hospital, Uyo, Nigeria. Blood parameters of interest (Hb, PCV, RBC, WBC and PLT) were analyzed. Histological analysis was carried out on the tissue samples using Haematoxylin and Eosin method.

2.9 Statistical Analysis

Data were analyzed using descriptive statistics. Comparison of data and physicochemical properties between the control and other treatments were carried out using Analysis of Variance (ANOVA). Effects of the extracts on the tissues of the test organism in the different treatments was compared using the photomicrographs.

3. RESULTS

3.1 Initial Physicochemical Parameters of Test Water

Physicochemical parameters of the test water were observed to be within acceptable limit for survival and growth of *C. gariepinus* juveniles prior to commencement of the experiment [24]. Physicochemical parameters of the test water prior to commencement of the bioassay were tested and the F values recorded. The values of the basic water quality parameters prior to stocking were, dissolved oxygen (5.60 mg/l), temperature (26.5^{o}C), pH (7.16), TDS (7.1 mg/l) and EC (12.6 µS/cm) (Table 1).

3.2 Variation of in Physico chemical Parameters of Test Media

The effect of *S. alata* leaf extract on the physico-chemical properties of the culture medium was assessed in the study (Table 2). During the 14-day chronic toxicity bioassay physicochemical parameters of the culture medium showed variations in values. No significant change ($p<0.05$) in temperature was observed. DO and pH were observed to reduce significantly ($p<0.05$) while TDS and Electric Conductivity were observed to increase significantly ($p<0.05$). The variations were all observed to be concentration related. Duncan's statistical test revealed that changes in pH of test media were significant ($p<0.05$) only at higher concentrations (0.60mg/L and 0.90mg/L).

Table 1. Initial Physico-chemical parameters of the test water prior to stocking of test organism

Fish Species	Initial physico-chemical parameters prior to stocking				
	DO (mg/l)	Temp (^{o}C)	pH	TDS (mg/l)	EC (µS/cm)
***Clarias gariepinus* juveniles**	**5.60**	**26.5**	**7.16**	**7.1**	**12.6**

Table 2. Initial Physico-chemical parameters of the test water prior to stocking of test organism

Parameter / Conc. (mg/L)	0.0	0.18	0.27	0.40	0.60	0.90
DO	5.50 ± 0.58^{a}	5.20 ± 0.28^{a}	4.80 ± 0.64^{a}	4.65 ± 0.67^{ab}	4.5 ± 0.85^{b}	4.4 ± 0.78^{b}
pH	7.16 ± 0.12^{a}	5.25 ± 0.16^{b}	5.10 ± 0.04^{b}	4.93 ± 0.18^{b}	4.89 ± 0.32^{bc}	4.86 ± 0.32^{bc}
TDS	7.0 ± 0.71^{a}	13.0 ± 0.00^{b}	20.5 ± 0.35^{bc}	27.0 ± 2.13^{bc}	35.0± 2.13^{c}	44.0 ± 1.42^{d}
EC	12.5 ± 0.35^{a}	26.5 ± 0.35^{b}	38.0 ± 2.84^{bc}	58.0 ± 7.80^{bc}	70.5 ± 3.90^{c}	91.0 ± 4.96^{d}
Temp.	26.2 ± 0.14^{a}	26.2 ± 0.14^{a}	26.2 ± 0.14^{a}	26.2 ± 0.14^{a}	26.3 ± 0.07^{a}	26.2 ± 0.14^{a}

Mean with different superscripts along the same row are significantly different (Duncan's test) p<0.05

Table 3. Effects of *S. alata* on the blood parameters of *Clarias gariepinus* juveniles

Parameter / Conc. (mg/L)	0.0	0.18	0.27	0.40	0.60	0.90
Hb (g/L)	9.0 ± 0.24^{a}	8.8 ± 0.75^{a}	8.65 ± 0.45^{a}	8.45 ± 0.59^{b}	8.45 ± 0.59^{b}	8.1 ± 0.35^{b}
RBC	3.6 ± 0.31^{a}	2.9 ± 0.35^{a}	2.85 ± 0.17^{a}	2.80 ± 0.21^{a}	2.75 ± 0.04^{ab}	2.65 ± 0.11^{b}
PCV	34.1 ± 2.05^{a}	26.5 ± 3.19^{ab}	26.0 ± 1.42^{b}	25.5 ± 1.77^{b}	25.5± 0.35^{b}	24.5 ± 1.03^{bc}
PLT	28.5 ± 3.19^{a}	31.0 ± 0.70^{a}	32.5 ± 1.77^{ab}	36.0 ± 1.42^{b}	55.0 ± 4.25^{bc}	68.0 ± 3.74^{c}
WBC	35.8 ± 0.99^{a}	76.9 ± 0.78^{b}	98.15 ± 11.75^{b}	105.95 ± 5.85^{bc}	131.4 ± 0.21^{c}	146.4 ± 1.06^{c}

Mean with different superscripts along the same row are significantly different (Duncan's test) p<0.05

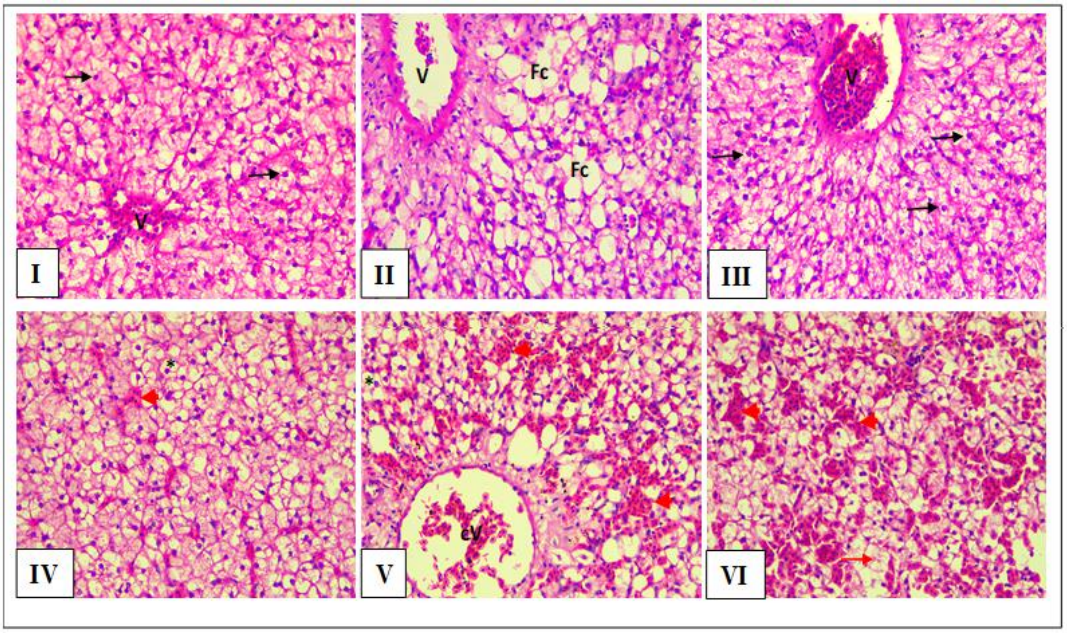

Fig. 1. The photomicrograph of the liver section across the groups. (I) Normal liver parenchymal showing hepatocytes (arrow), normal sized blood vessel (V), no lesion seen. (II) showed diffused ballooning degeneration and focal area of macrovesicular fatty changes (Fc). (III) showed normal section with dilated congested blood vessel (CV). (IV) showed enlarged hepatocytes and mildly congested sinusoid. (V) depicts diffused dilated congested blood vessels and sinusoid, ballooning degeneration and necrotic cells. (VI) showed diffused congested sinusoid and necrotic hepatocytes. Haematoxylin and Eosin (H&E) stain, x400 magnification

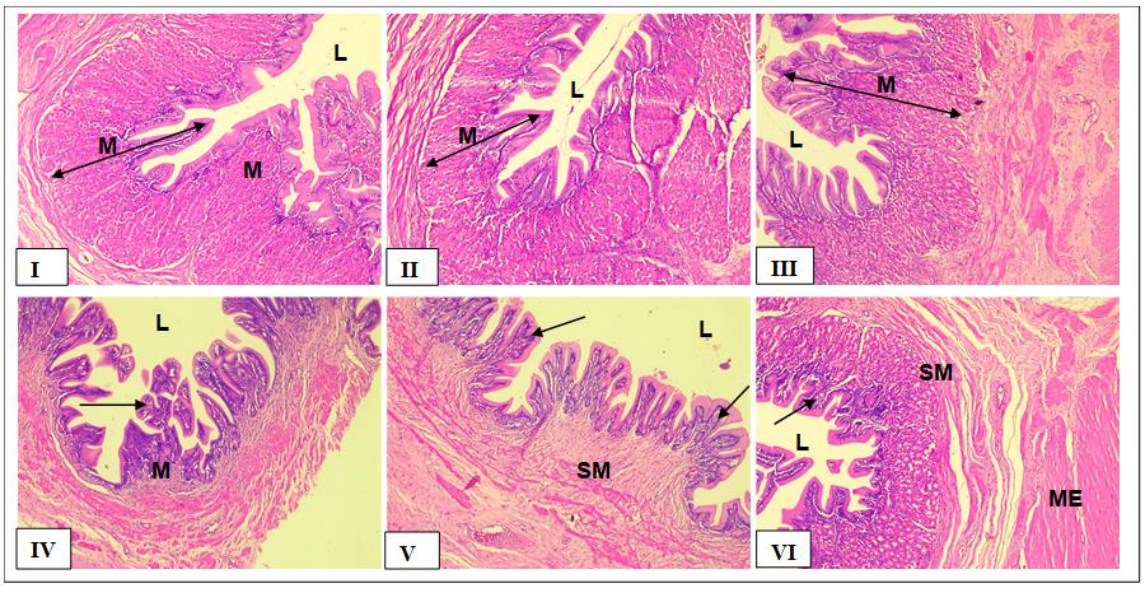

Fig. 2. The section intestinal tissue across the groups (I-VI). The section showed lumen (L), the mucosa (M) showed increased villi (Thin arrows) and the submucosa (SM). Also seen was muscular externa (ME); No pathologic changes seen in the control group (I) and the experimental groups (II-VI). Haematoxylin and Eosin (H&E) stain, x100 magnification

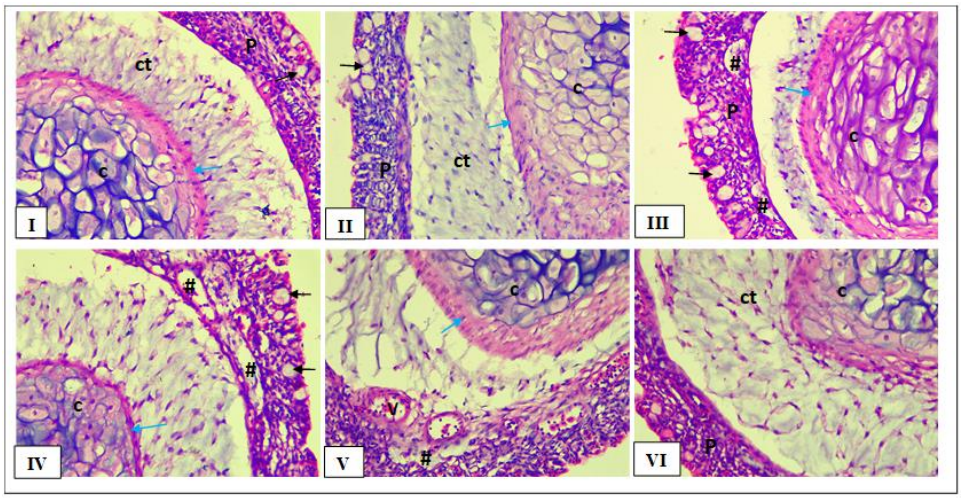

Fig. 3. Photomicrograph of the transverse section of the fish gill arch of the Control (I) and the treatment groups (II-VI). (I) showed normal gill histology with centrally supporting cartilaginous tissue (C) surrounded by skeletal muscle (blue arrow) and basophilic connective tissue (Ct). The primary filaments (P) were vascularized and showed sporadic fatty changes (black arrow). (II) showed few fatty changes (black arrow) in the primary filament. (III)& (IV) showed diffused fatty changes across the primary lamella and multiple area of epithelium degeneration (#) and congested blood vessels (red arrowhead). V depict a focal epithelium degeneration, while VI showed increase fatty lobules (black arrow) in the primary filaments

3.3 Blood Parameters of Test Media

Results of blood parameters of the respective test media during the period of exposure (14 days) is presented in Table 3. Results from blood analysis show that all the parameters measured, reduced in value when compared with the control. The reductions were observed to be related to the concentration of the extracts. Statistical analysis of data from blood parameters revealed that the observed reduction in the respective parameters (Hb, RCB and PVC) were statistically significant ($p<0.05$).

3.4 Tissue Analysis

The photomicrographs of liver sections from the control group (I) and treatment groups (II– VI) reveal varying degrees of histopathological changes induced by *Senna alata* extract. The control group (I) displayed normal liver parenchyma, characterized by intact hepatocytes, normal-sized blood vessels, and the absence of lesions. In contrast, group II showed diffuse ballooning degeneration and focal areas of macrovesicular fatty changes, indicative of early hepatic distress. Group III mostly maintained normal histology, but with dilated and congested blood vessels, signaling some circulatory disturbance. Group IV exhibited enlarged hepatocytes and mild sinusoidal congestion, suggesting initial hepatic stress. Group V presented significant pathology with diffuse dilated and congested blood vessels and sinusoids, ballooning degeneration, and necrotic cells, indicating severe hepatic damage. Finally, group VI showed extensive damage with widespread congested sinusoids and necrotic hepatocytes, reflecting the most severe hepatic injury (Fig. 1).

The intestinal histology across all groups (I-VI) displayed the lumen (L), mucosa (M) with increased villi (thin arrows), submucosa (SM), and muscularis externa (ME). There were no pathological changes observed in either the control group (I) or the experimental groups (II-VI) (Fig. 2).

The photomicrograph of the transverse section of the fish gill arch revealed distinct histological features across the control group and treatment groups. In the control group, the gill histology appeared normal, with centrally supporting cartilaginous tissue surrounded by skeletal muscle and basophilic connective tissue. The primary filaments were vascularized and exhibited sporadic fatty changes, indicating normal physiological variation. In treatment group II, the gill sections showed few fatty changes in the primary filament, suggesting minimal impact from the extract. However, treatment groups III

and IV exhibited more pronounced pathological alterations, including diffused fatty changes across the primary lamella, multiple areas of epithelium degeneration, and congested blood vessels, indicating significant tissue stress and damage. Group V depicted focal epithelium degeneration, while group VI displayed increased fatty lobules in the primary filaments, reflecting extensive fat deposition and potential dysfunction in gill tissue. Group V depicted focal epithelium degeneration, while group VI displayed increased fatty lobules (black arrow) in the primary filaments, reflecting extensive fat deposition and potential dysfunction in gill tissue (Fig. 3).

4. DISCUSSION

Senna alata exerted toxic effect on the fish in the present study and toxicity increased with increased concentration. Abnormal behavior such as restlessness, incessant jumping and gulping for air, loss of equilibrium, increase in opercular activities, surface to bottom movement, sudden quick movement and resting at the bottom of the aquaria and death observed in this study were similar to the findings of George et al., [25-27], Essien-Ibok et al., [28].

The abnormalities observed prior to mortality are an indication of depleted oxygen content due to higher demand for oxygen. Consequently, it was observed in this study that the abnormal behavior and mortality rate of the test organism's increased with increase in concentrations of toxin. This corresponds to the findings of Shobha (2007) that "the behaviour and mortality rate of *C. carla* during experiment was found to depend on both duration of exposure and concentration of the toxicant". Similar observations were also, reported by George, et. al., [29-32] in a related study.

The introduction of the leaf extract at different concentrations impairs the swimming pattern, skin colouration and general behaviour of fish. Also, the variation in the behavioural responses and mortality in the sublethal test can be attributed to the low level of accumulation of the toxicant. This suggests that fish can tolerate low concentrations of pollutants with reduced mortality.

The reductions in blood cell indices and tissue deformation observed from the chronic bioassay are in line with findings of Okogwu et. al., [33] during their "study on behavioural, haematological and histopathological changes in *C. gariepinus* exposed to 2,4-dichlorophenoxyacetic acid". Similar observations have also been reported "in a study on exploitation of ethanol extract of *Adenium obesum* stem bark as a potent organic piscicide" [22]. "In a study conducted on haematological responses, Serum Biochemistry and Histology of *C. gariepinus*, exposed to sublethal concentrations of cold water fresh root bark extract of *Plumbago zeylanica* (Leadwort), it was reported that, exposure of fish to the toxicant for 21 days resulted in remarkable anaemic condition in *C. gariepinus* resulting from reductions in values of haematological parameters (RBC, Hb, PCV) in the juveniles of *C. gariepinus* exposed to *P. zeylanica* extract" [34]. Olusegun and Adadayo [34] also reported "histopathological alterations observed in the brain, gills, liver, intestine and muscle/flesh of the *C. gariepinus* juveniles which align with the results of the present findings".

"Stressors evoke non-specific responses in fish which enables the fish to cope with the disturbance and maintain its homeostatic state" [35]. "If severe or long lasting, the response then becomes maladaptive and threatens the fish health and well-being. Therefore, in the presence of stressors (contaminants / pollutants), blood parameters and blood chemistry can be employed as standard laboratory test to determine diseased conditions and metabolic disturbances in fish" [35]. "Blood is a tissue fluid and serves as transport medium whose primary function is to supply oxygen and nutrients as well as constitutional elements to tissues and to remove waste products. Blood also enables hormones and other substances to be transported between tissues and organs. Blood is basically composed of the plasma, red blood cells, white blood cells and platelets, each with sub constituents which collectively contribute to the overall functioning of the blood" [35]. "The WBCs help the immune system and as well remove toxins, waste, foreign bodies and abnormal or damaged cells by phagocytosis. Platelets secrete factors that increase local platelet aggregation e.g., Thromboxane A, enhance vasoconstriction e.g., serotonin and promote blood coagulation e.g., Thromboplastin" [36].

"These observations in blood parameters confirm concentration related toxicity of the toxicant to the test organism" [37]. "The observed reduction in the respective blood parameters results in diseased conditions such as haemolytic

anaemia, leukemia and lymphopaenia in exposed fish resulting from interference with blood cells formation process (erythropoiesis) and destruction of blood cells by the toxicants" [35]. Haematolgical changes as recorded in this study are attributed to rapid destruction of the red blood cells by the toxicant and the increase in white blood (WBC) was probably a defense mechanism deployed by the fish to protect itself from assault of *Senna alata*.

"Fish gills are the prime target organ of all pollutants due to their extensive surface in contact with the external medium and the reduced distance between the external and internal medium. Gill morphology and morphometric are important biomarkers providing a rapid method for detection of the effects of pollutants" [38]. "In histopathological studies, gills have also been reported to act as storehouse for bio accumulating toxicants" [38]. The general morphological changes in the gills recorded in this study have been reported in *Astyanax sp*. The histological examination of gill sections from juvenile *Clarias gariepinus* treated with varying concentrations of *Senna alata* extract demonstrated a clear dose-dependent response. The control group maintained normal gill architecture with minimal fatty changes, while the treatment groups exhibited a progression of pathological features with increasing extract concentration. Group II's minor fatty changes contrasted sharply with the severe alterations in groups III and IV, including widespread fatty changes, epithelial degeneration, and vascular congestion. These changes indicate significant gill tissue stress and compromised function. Group V's focal epithelial degeneration and group VI's extensive fatty lobules further underscore the potential toxicity of higher extract concentrations. These findings suggest that while lower doses of *Senna alata* extract may be relatively safe, higher doses pose a risk of significant gill damage, highlighting the need for careful dose optimization in aquaculture applications.

Vascular changes in gills of exposed fish could be attempts by the fish to supply more blood to the gills to increase oxygen uptake and supply to the internal organs. The gill of fish is a multipurpose organ that, in addition to providing for aquatic gaseous exchange, plays a dominant role in osmotic and ionic regulation, acid-base regulation and excretion of nitrogenous waste. Hence, impairment of the gill functions by the overall effect of the pathological changes in the gill of exposed fish will have grave consequences for the fish with respect to the normal functions of the gills [37,39,40].

The intestine tissue is responsible for storage of food and package of food items in the fish. In the present study, it is evident that the gut tissue without treatment shows the normal mucosa, submucosa and muscular externa layer with no pathologic changes while that of the treatment revealed photomicrographs of the intestine showing the lumen with the mucosa and a dilated congested blood vessel in the submucosa.

The liver is the primary organ for metabolism, detoxification of xenobiotics and excretion of harmful substances. Glucose is stored in the liver as glycogen and is returned to the blood via circulation when needed to maintain stable blood sugar levels. Other than glycogen, the liver stores fat soluble vitamins. The liver has the ability to degrade toxic compounds but its regulatory mechanism can be overwhelmed by elevated concentration of toxic substance which could subsequently result in structural damage. The histopathological findings demonstrate a dose-dependent hepatotoxic effect of *Senna alata* extract on juvenile *Clarias gariepinus*. While the control group maintained normal liver structure, the treatment groups exhibited varying degrees of hepatic alterations with increasing severity correlating with higher extract concentrations. Group II's early signs of hepatotoxicity progressed to moderate hepatic stress in groups III and IV, as evidenced by vascular congestion and cellular hypertrophy. The most severe damage in groups V and VI, marked by extensive congestion, ballooning degeneration, and necrosis, underscores the extract's potential hepatotoxic effects at higher doses. These results highlight the necessity for careful dosage determination of *Senna alata* extract to avoid liver toxicity in aquaculture applications. Further research is essential to understand the mechanisms driving these changes and to establish safe and effective usage guidelines for *Senna alata* in fish health management. The liver of freshwater fish is known to accommodate a high level of toxicants [41,42].

Physicochemical parameters such as temperature, dissolved oxygen, pH, total dissolved solids and electric conductivity are vital aquatic indices which determine the fish health, growth and reproduction. pH decreases because the extract is biodegradable and when it is much

in water biological activities degrades it and microbial activities will occur which uses oxygen and more of the oxygen present in the water would be used by the microorganisms to decompose the extract and that is why there is vast depletion of dissolved oxygen, because of the aerobic process. The pH decreases because the extract is acidic pushing the pH of the treatment to acidity and reducing the alkalinity. TDS and electronic conductivity are increasing with increased concentration because of the presence of more particles in the water while the temperature did not get affected because there was nothing to influence the atmospheric temperature in the room [43].

5. CONCLUSION

In conclusion, the chronic toxicity studies on *Senna alata* conducted on juvenile *Clarias gariepinus* have unveiled notable alterations in both haematological parameters and histopathogical features, particularly evident in the gills, liver and intestine of treated groups. These findings underscore the potential adverse effects of prolonged exposure to *Senna alata* on the physiological and cellular integrity of the fish. Intriguingly, the absence of pathological changes in the control groups suggests a distinct response attributable to the treatment. This highlights the importance of continued monitoring and assessment of the impacts of *Senna alata* on aquatic organisms and ecosystem, emphasizing the need for further research to elucidate the underlying mechanisms and potential long-term consequences. Such insights are crucial for informed decision-making regarding the management and regulation of *Senna alata* and its interaction with aquatic environments.

DISCLAIMER (ARTIFICIAL INTELLIGENCE)

Author(s) hereby declare that NO generative AI technologies such as Large Language Models (ChatGPT, COPILOT, etc) and text-to-image generators have been used during writing or editing of manuscripts.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

REFERENCES

1. Abo KA, Fred-Jaiyesimi AA, Jaiyesimi AEA. Ethnobotanical studies of medicinal plants used in the management of diabetes mellitus in South Western Nigeria. Journal of Ethnopharmacology. 2008;115(1):67–71.
2. Kumar A, Shukla R, Singh P, Prasad CS, Prasad NK. Assessment of *Thymus vulgaris* L. essential oil as a safe botanical preservative against post-harvest fungal infestation of food commodities. Innovative Food Science & Emerging Technologies. 2008;9(4):575–580.
3. Liu A, Xu L, Zou Z. Studies on chemical constituents from leaves of *Cassia alata*. China Journal of Chinese Matters on Medicine. 2009;34:861–863.
4. Aviello G, Rowland I, Gill CI. Anti-proliferative effect of rhein, an anthraquinone isolated from Cassia species, on Caco-2 human adenocarcinoma cells. Journal of Cellular and Molecular Medicine. 2006;14:2006–2014.
5. Makinde AA, Igoli JO, TA'Ama L. Antimicrobial activity of *Cassia alata*. African Journal of Biotechnology. 2007; 6(13):1509–1510.
6. Tarkang PA, Agbor GA, Armelle TD. Acute and chronic toxicity studies of the aqueous and ethanol leaf extracts of *Carica papaya* Linn in Wistar rats. Journal of Natural Product and Plant Resources. 2012;2(5): 617–627.
7. Khan MR, Kihara M, Omoloso AD. Antimicrobial activity of *Cassia alata*. Fitoterapia. 2001;72(5):561–564.
8. Somchit MN, Reezal I, Nur IE, Mutalib AR. *In vitro* antimicrobial activity of ethanol and water extracts of *Cassia alata*. Journal of Ethnopharmacology. 2003;84(1):1–4.
9. Donatus EO, Fred UN. Cannabinoid Dronabinol alkaloid with antimicrobial activity from *Cassia alata* Linn. Der Chemica Sinica. 2010;2(2):247–254.
10. Ranganathan S, Balajee SA. Anti-cryptococcus activity of combination of extracts of *Cassia alata* and *Ocimum sanctum*. Mycoses. 2000;43(7-8):299–301.
11. Otto RB, Ameso S, Onegi B. Assessment of antibacterial activity of crude leaf and root extracts of *Cassia alata* against Neisseria gonorrhea. African Health Science. 2014;14:840–848.
12. Sagnia B, Fedeli D, Casetti R. Antioxidant and anti-inflammatory activities of extracts from *C. alata, E. indica, E. speciosa, C. papaya* and *P. fulva* medicinal plants collected in Cameroon. Plos One. 2014; 9(8):1-10.

13. Varghese GK, Bose LV. Habtemariam S. Antidiabetic components of *Cassia alata* leaves: Identification through α-glucosidase inhibition studies. Pharmaceutical Biology. 2013;51(3):345–349.
14. Midawa SM, Ali BD, Mshelia BZ. Cutaneous wound healing activity of the ethanolic extracts of the leaf of *Senna alata* L (*Caesalpiniaceae*). Journal of Biological Science and Bioconservation. 2010;2:63-68.
15. Kundu S, Roy S, Lyndem LM. *Cassia alata* L: Potential role as anthelmintic agent against *Hymenolepis diminuta*. Parasitology Research. 2012;111(3):1187–1192.
16. Otoh AJ, Udo MT, George UU. Comparative effect of inducing broodstock with natural and artificial hormones on reproductive performances of *Heterobranchus longifilis*. Tropical Freshwater Biology. 2022;31:95-102.
17. Otoh AJ, Udo MT, George UU. Comparative growth performance and sex ratio of *Heterobranchus longifilis* and its offspring induced with synthetic hormone and pituitary gland of *Heterobranchus longifilis.* Journal of Wetlands and Waste Management. 2023;5(1):106- 111.
18. Otoh AJ, Okoko AC, Ekanem IE, George UU, Asangusung PS. Effects of multiple batches of stripped eggs on the reproductive performances *of Heterobranchus longifilis*. Asian Journal of Research in Zoology. 2024a;7(2):9-16.
19. Otoh AJ, George UU, Asangusung PS, Okoko AC, Ekanem IE, Umaha MN. Investigation into the impacts of varied batches of stripped eggs on the reproductive success of *Clarias gariepinus*. Asian Journal of Research in Zoology. 2024 b;7(2):28-35.
20. Otoh AJ, Ekanem IE, Okoko AC, Asangusung PS, George UU. Comparative study of inducing broodstock with natural and artificial hormones on reproductive performances of *Clarias gariepinus*. Asian Journal of Fisheries and Aquatic Research. 2024c;26(3):31-38.
21. Otoh AJ, Okoko AC, Asangusung PS, Ekanem IE, George UU, Idiong TE. Effect of different ages of african catfish (*Clarias gariepinus*) broodstock on reproductive performance and fries production. Asian Journal of Fisheries and Aquatic Research. 2024d;26(3):39-47.
22. Abalaka SE, Fatihu MY, Ibrahim ND, Ambali SF. Exploitation of ethanol extract of Adenium obesum stem bark as a potent organic piscicide. Res J Biol Sci. 2013; 8:143-149.
23. Ayuba VO, Ofojekwu PC. Acute toxicity of the root extract of Jimsons weed. *Datura innoxia* to the African catfish *Clarias gariepinus*. Journal of Aquatic Sciences. 2002;17(2):131133.
24. Interafrican Bureau for Animal Resources (IBAR). African catfish *Clarias gariepinus*. FAO, Rome. 2015;6.
25. George UU, Essien-Ibok MA, Ajayi OO, Abiaobo NO. Mbong EO. Assessment of the toxicity and histo-morphological effects of clint car wash liquid soap on african catfish (*Clarias gariepinus*) fingerlings. Journal of Aquatic Science and Marine Biology. 2024a;5(1):1-10.
26. George UU, Essien-Ibok MA, Ajayi OO, Abiaobo NO, Mbong EO. Investigating the toxicity and histopatho-morphological alterations induced by *Cana indica* extract on *Clarias gariepinus* fingerlings. Annals of Ecology and Environmental Science. 2024b;6(1):46-57.
27. George UU, Ajayi OO, Essien-Ibok MA, Abiaobo NO, Mbong EO. Assessment of the toxicity and histopathological effects of launaea taraxocifolia extract on fingerlings of *Clarias gariepinus*. Mathews Journal of Cytology and Histology. 2024c;8(1):1-10.
28. Essien-Ibok MA, George UU, Abiaobo NO, Mbong EO. Evaluation of the toxic potentials and histopathological variations in *Clarias gariepinus* fingerlings exposed to ethanolic extract of *Costus afer*. Mathews Journal of Cytology and Histology. 2024d; 8(1):1-12.
29. George UU, Ajayi OO, George IE, Vincent UE. Establishing a dose-response toxicity for *Clarias gariepinus* fingerlings exposed to ethanolic extract of *Latana camara*. Asia Journal of Fisheries and Aquatic Research. 2023a;24(1):1-10.
30. George UU, Mbong EO, Abiaobo NO, Akpan II. *In Vivo* studies on mortality and histopathological indices of *Phragmenthera capitata* (Mistletoes) on *Clarias gariepinus* fingerlings in aquarium. Mathews Journal of Cytology and Histology. 2023b;7(2):1-10.
31. George UU, Otoh AJ, Ajayi OO, George IE. Studies on mortality and histopathological alteration on the gills of *Oreochromis niloticus* juveniles following

exposure to ethanolic extract of *Phramenthera capitata* under laboratory conditions. Asian Journal of Fisheries and Aquatic Research. 2023c;24(3):23-34.
32. George UU, Otoh AJ, Ajayi OO, George IE. Dose-response relationship and histo-morphological alterations on *Oreochromis niloticus* juveniles following exposure to ethanolic extract of *Latana camara*. Asian Journal of Research in Zoology. 2023d;6(4):71-83.
33. Okogwu OI, Anionwo Q, Anoke DC, Ugwuezi PO. Behavioural, haematological and histopathological changes in the African catfish, *Clarias gariepinus* exposed to 2,4-dichlorophenoxyacetic acid (2,4-D). Niger J Biotechnol. 2015;30:26-35.
34. Olusegun AA, Adedayo OO. Haematological responses, serum biochemistry and histology of *Clarias gariepinus* (Burchell, 1822) exposed to sublethal concentrations of cold-water fresh root back extracts of *Plumbago zeylanica* (Leadwort). J Aquac Res Dev. 2014;5:282-288.
35. Crook MA. Clinical biochemistry and medicine. 8th edition, Hodder & Stoughton Ltd. 2012;74-81
36. Aqra A. Human medical physiology: Blood physiology. John Wiley & Sons Inc., USA. 2012;365-371.
37. Gabriel UU, Amakiriand EU, Ezeri GNO. Haematology and gill pathology of *Clarias gariepinus* exposed to refined oil, kerosine under laboratory conditions. J. Anim Vet Adv. 2007;6:461-465.
38. Fafioye OO, Adebisi AA, Fagade SO. Toxicity of parkia biglobosa and raphia vinifera extracts on *Clarias gariepinus* Juveniles. African Journal of Biotechnology. 2012;3:627-630.
39. George UU**,** Joseph A, Andy JA. Histopathological alterations in gills of fingerlings of *Clarias gariepinus* (Burchell, 1822) following sublethal acute exposure to *Hevea brasiliensis.* International Journal of Scientific & Technology Research. 2014a;3(9):252- 255.
40. George UU, Urom SE, Etanketuk N. Acute toxic effect of qua iboe light crude oil on the gills of *Clarias gariepinus* juveniles. International Journal of Environment and Pollution Research. 2014 b;2(2):16-30.
41. Musa SO, Omeregie E. Haematological changes in the mud fish, *Clarias gariepinus* (Burchell) exposed to malachite green. J. Aquat Sci. 1999;14:37-42.
42. Ojutiku RO, Asuwaju FP, Kolo RJ, Obande RA, Agbelege OO. Haematological effect of acute concentration of cypermethrin on juveniles of *Claria gariepinus*. Int J Eng Sci Inven. 2013;2:33-41.
43. Udo PJ, Ekanem AP, Eze EE. Toxicity of crude oil to early life stages of *Heterobranchus longifilis* (Cruveier and Valiennces) Pisces: Bagridae). Tropical Environmental Research. 2006;1:450-459.

Peer-review history:
The peer review history for this paper can be accessed here:
https://www.sdiarticle5.com/review-history/119216