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TOXICOLOGICAL EFFECTS OF AQUEOUS EXTRACT OF *PIPTADENIASTRIUM AFRICANUM* BARK ON *Clarias gariepinus* JUVENILES

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Abstract. A Laboratory study was carried out to assess the toxic effects of aqueous extracts of *Piptadeniastrium africanum* bark on *Clarias gariepinus* juveniles using the static bioassay with continuous aeration method. Phytochemical analysis of the *P. africanum* aqueous bark extract revealed the presence of tannins (1.95 %), saponin (2.95 %), flavonoid (5.63 %), Glycosides (1.05 %), Steroid (2.35 %), Terpens, (2.70 %).0 (control), 25, 50, 75, 100, 125 mg/L concentrations were then applied to *Clarias gariepinus* juveniles (Mean weight: 18.26± 0.04 g) to determine the acute toxicity. The result obtained revealed the 96 h LC_{50} values to be 42.21 mg/L with 95 % confidence limit of 30.37–58.67 mg/L. Fish exposed to acute toxicity test exhibited behavioural changes such as discolouration, air gulping, erratic swimming, aggression, excessive mucus secretion, and settling at the bottom motionless just before death. Histological assessment of the gills showed that acute exposure of *P. africanum* bark led to degeneration of primary and secondary lamellae, lamellae with marginal channel dilation, blanketing of the lamellae, hyperplasia of epithelial cells, epithelial lifting and haemorrhage, oedema, necrosis and malformed secondary lamellae of the African catfish. The liver also showed focal necrosis, bile stagnation, nuclear degeneration, hepatocytes with irregular shaped nucleaus, cytoplasmic vacuolation, vascular congestion, bile pigment disintegration and cytoplasic degeneration Hyperplasia of epithelial cells and fusion of secondary lamellae. The toxic effect of the aqueous extract of the plant on *C. gariepinus* juveniles was both time and dose dependent.
Keyword: *Piptadeniastrium africanum, Clarias gariepinus*, toxicity, Histology.

Introduction

The importance of Fish farming cannot be over–emphasised as it is one of the sources of protein to man. It is the fastest growing food sector in the world, accounting for an estimated 43 % of all fish consumed by humans globally [ALLSOPP *et al*., 2008].

One of the management practices in fish farming operations is the eradication of unwanted organisms (mollusc, insect larvae, fish weed and unwanted fish species) prior to the stocking of desirable fish species [AYOTUNDE *et al*., 2011, STAVARACHE, *et al.,* 2015, STEFAN, *et al.,* 2013].

Some Fish farmers get rid of these organisms by the use of certain synthetic chemicals (Sodium cyanide, malachite green antimycin). These chemicals are non-biodegradable and accumulate in the environment to pose serious toxic threat to other organisms in food chain, including humans [ABALAKA *et al*., 2014].

However, certain piscicidal Plants which are biodegradable and environmental friendly are widely used in aquaculture management practices including: tea seed cake and derris root powder as their toxicity to fish has been established [ORJI *et al.,* 2014, STOLERU, *et al.,* 2015].

These conventional plant piscicides are either not within the reach of the fish farmers or may be expensive especially for farmers in Nigeria. There is therefore need to search for other piscicidal plants that can be readily available for the fish farmers, affordable and biodegradable that can replace tea seed cake and derris root powder especially in countries like Nigeria where aquaculture is currently recording good growth [SOLBE, 1995].

Commonly used piscicidal plants in Nigerian Inland waters are well described in Lake Kainji basin by Reed, Obot and Udolisa[*REED et al;* 1967; UDOLISA *et al*; 1994 and OBOT 1996]; these include such species as: *Cassia alata, Erythrophloem ivorensis, Piptadeniastrum africanum, Albizia ferruginea, Strychnosaculeate,Tetrapleura tetraptera*, *Tephrosia vogelii, Blighia*

sapida among others. *Piptadeniastrum africanum* (African green heart) is reported to be one of the piscicidal plants. *P. africanum* is a large buttress tree called variously as: 'African green heart' in English, 'kiryarkurmi' in Hausa, 'Ofie' in igbo and 'agboin' in Yoruba, *P. africanum* belongs to the family Mimosaceae and it is represented by only one species–*P. africanum* [AKINLAMI *et al;* 2012, STEFAN, *et al.,* 2013, STOLERU, *et al.,* 2012]. It is widely distributed in the West and Central Africa [BURKILL, 1995].

In Nigeria, it occurs in the rainforest belt of Southern Nigeria. It is used for various purposes including: fish poisoning, construction, soap substitute and in traditional medicine, mostly the bark, sometimes also roots and leaves [BETTI; 2002., ITA, OFFIONG; 2013, STOLERU, *et al.,* 2016].

The African catfish, *Clarias gariepinus* belongs to the family clariidae. It is a hardy fish species because it possesses an accessory breathing organ [HENGSAWAT *et al*., 1997].

It is frequently and widely cultured in ponds and it also occur freely in Nigerian natural fresh waters as reported by Fagbenro [FAGBENRO.,1992].

Although, *P. africanum* has been used for fish poisoning in the South–south and South–eastern part of Nigeria, information about its toxic effect on *Clarias gariepinus* is not been established.

Hence, the study aimed to evaluate the toxic effect of aqueous extract of *P. africanum* stem bark on African catfish, *Clarias gariepinus* juveniles.

Material and methods

Experimental Site

The study was carried out at the General Laboratory complex, Department of Fisheries and Aquaculture, University of Agriculture Makurdi, Benue state.

Collection of Experimental Samples

The stem bark of *Piptadeniastrium africanum* was collected by peeling off the stem bark of the plant with a knife from selected forest site in Ugep, Yakurr L.G.A, Cross River state. It was identified and authenticated by Mr. Emmanuel Abam of Forestry Commission Calabar, Cross River State. The fresh samples of *P. africanum* bark were cut into pieces and air dried under shade to constant weight. It was pulverized to powder using a mortar, sifted with 0.25 mm Sieve and then stored in an air tight bottle according to Ayuba [AYUBA *et al.*, 2012].

180 healthy, active juveniles of African catfish *Clarias gariepinus* were collected from Finite fish farm, Makurdi. The fish was transported in plastic containers that are well aerated to the

General-purpose laboratory, Department of Fisheries and Aquaculture, University of Agriculture, Makurdi. The fish were acclimatized in the laboratory for two weeks during which they were fed with commercial floating feed (coppens) at 5% of their body weight. Unconsumed feed and faeces were removed and water replenished twice a week as recommended by Oyelese and Faturoti [OYELESE and FATUROTI1995].

Phytochemical screenings

Qualitative and quantitative phytochemical screening was carried out to establish the presence or absence of saponins, tannins, hydrocyanic glycoside, steroids, terpens, flavonoids, alkaloid as well as their concentrations using Standardized chemical method described by HARBONE and BAXTER [1993] as well as AOAC; [2000] and AOAC; [2005] methods respectively.

Preparation of Aqueous Extract of *piptadeniastrium africanum* Bark

A quantity of 250 g of the resultant fine powder of *Piptadeniastrium africanum* bark was weighed and dissolved in one (1) L of distilled water at a room temperature (25±5°C) in 1 L sample bottle separately. The mixture was shaken and allowed to stand for 24 h after which it was decanted and filtered using Whatman filter paper (125 mm). The filtrate was stored in air tight bottle [AYUBA *et al.,* 2012] and was used for the bioassay tests.

Physicochemical analyses

The temperature, pH, electrical conductivity and the total dissolved solids of the fish culture water were monitored with Hanna "Combo" portable hand instrument (Hi 98129, Hanna Instruments, Mauritius) while their dissolved oxygen content was monitored using the modified

method of Winkler–Azide [LIND, 1979; APHA, 1985, ROTARU, *et al.*, 2010].

Range Finding Test

After the acclimatization period of 14 days, series of range finding tests was carried out to determine the toxicity level or suitable concentration of the prepared *Piptadeniastrium africanum* aqueous extract of the bark that was used for the acute toxicity test.

The range finding test was carried out for 24 h following the procedure of Solbe [SOLBE;1995].

Three *C. gariepinus* juveniles were stocked in each bowl with the following graded concentrations of aqueous extract of *P. africanum* bark: 0 mg/L, 125 mg/L, 250 mg/L, 1250 mg/L and 2500 mg/L.

Based on the result of the range finding test, acute concentrations of *P. africanum* bark were determined for the 96 h bioassay with a spacing factor of 1.0.

Acute Toxicity Test

A total number of eighteen (18) plastic tanks each with a capacity of 70 L were used for the 96 h bioassay experiment, each containing 30 L of dechlorinated municipal water.

The acute lethal concentrations used were 125 mg/L, 100 mg/L, 75 mg/L, 50 mg/L, 25 mg/L and 0 mg/L (control) in triplicate and a micro–pipette was used to introduce the aqueous extract of *P. africanum bark* extract into the various tanks.

Each of the tanks was stocked with ten (10) healthy juveniles of *C. gariepinus* with mean weight of 18.26±0.04 g.

The fish were selected randomly and used for experiment and were not fed 24 h prior to the start of the experiment and during the exposure period [UNEP;1989, ROTARU, *et al.*, 2011, SELLITTO, *et al.*, 2016].

The Tanks were covered with net to prevent the fish from jumping out.

The whole experiment was aerated continuously using electrically powered and battery powered aerators.

Mortality as well as behavioural changes were observed and recorded 6 h, 12 h, 24 h, 48 h, 72 h and 96 h.

During exposure period, dead fish observed were immediately removed from each test tank and recorded in order to avoid polluting the tanks [FAFIOYE, 2012].

The 96–hour LC_{50} (lethal concentration that cause 50% mortality) was determined using a probit analysis using arithmetic method of percentage mortality data, log conc, graph and slope function, upper and lower confidence limits of the LC_{50} for acute toxicity test recommended by UNEP [UNEP;1989].

Histological assessment

After the acute toxicity test, the histopathological assessments were carried out from each treatment in the following stages:

Collection of samples: the livers and gills of *Clarias gariepinus* exposed to aqueous exract of *Piptadeniastrium africanum*bark after acute toxicity test was obtained.

Fixation: the livers and gills obtained from the fish were immediately fixed in 10 % formaldehyde to prevent spoilage.

Dehydration: this was done by putting the fixed livers and gills in various grades of alcohol over different periods ranging from 70 % alcohol for 3–8 h; 90 % alcohol for 16 h; absolute alcohol 1 for 2–3 h; absolute alcohol 11 for 3 h; to absolute alcohol 111 for 3 h and finally into xylene for 17 h.

This process ensures hardening of the tissue and impregnation with wax 1 and wax 11

Embedding: samples were embedded in wax and embedment mould. After solidifying, the samples were trimmed and mounted on wooden block to fit in microtone.

Sectioning: Microtone was used to section samples into 5µm for all histological samples.

Floatation: the samples were floated with warm water in a floating out bath to unfold the tissue. The tissue was picked, put on a slide and dried on a hot plate stained.

Staining: the samples were stained with hematoxylin and eosin and were cover–slipped by a thin glass plate.

This process facilitates microscopic examination and micrography.

Microscopy: the samples were read under a microscope at × 400 magnifications for various changes in the architectural make up of organs under discussion and then followed by microphotographs.

Data Analysis: Data collected from the various experiments were subjected to analysis of variance using Genstat package Edition 12 and the treatment means was compared using Minitab 14 [MINITAB, 2003, BUTNARIU, et al., 2005].

Results and discussion

The phytochemical screening of *P. africanum* bark extract revealed the presence and quantity of certain secondary metabolites in the plant extract as shown in Table 1.

Table 1.

Phytochemical Composition of *P. africanum* bark Obtained

Chemical constituent	**Inference**	**Concentrations** (%)
Flavonoid	+ve	5.63
Saponins	+ve	2.95
Steroid	+ve	2.35
Tannins	+ve	1.95
Cardiac glycoside	+ve	1.05
Anthraquinone	–ve	–
Alkaloid	–ve	–
Terpens	+ve	2.70

KEY: +ve (present), –ve (absent).

The physical and chemical properties of toxicant water exposed to *P. Africanum* bark showed variation in:

- pH,
- Temperature,
- Dissolved oxygen concentration,
- electrical conductivity and
- total dissolved solids. pH,
- electrical conductivity, temperature and
- total dissolved solids

were observed to be increasing with increase in concentration while dissolved oxygen was observed to be decreasing with increase in concentration of *P. africanum* bark extract as shown in the table 2.

Table 2.

Physicochemical parameters of toxicant water during the acute toxicity test of aqueous extract of *P. africanum* bark to *Clarias gariepinus* juveniles.

Treatment (mg/L)	Water Quality Parameters				
	pH	Temperature (^{0}C)	Electrical Conductivity (µS/cm)	Total Dissolved Solids (mg/L)	Dissolved Oxygen (mg/L)
Control	7.90± 0.00^f	26.63± 0.09	841.33±0.33^f	420.33± 0.33^e	5.00± 0.01^a
25	8.14± 0.01^e	26.60± 0.15	870± 0.88^e	434.67±0.33^d	4.64± 0.01^b
50	8.19± 0.01^d	26.77 ± 0.07	912.33± 0.33^d	456.00± 0.58^c	4.55± 0.01^b
75	8.28± 0.02^c	26.57± 0.12	929.67±0.88^c	465.33± 0.33^c	4.42± 0.01^c
100	8.35± 0.00^b	26.57± 0.12	953.33 ±1.66^b	47.67±1.33^b	4.26± 0.01^d
125	8.41± 0.01^a	26.60± 0.15	991.33± 0.67^a	496.67± 0.33^a	4.10± 0.01^e

Mean in the same column with different superscripts differ significantly (n=3, P≤0.05).

There was 83.3 % mortality in the fish exposed to the highest extract concentration of 125 mg/L while the least mortality of 36.7 % was recorded in the fish exposed to the lowest extract concentration of 25 mg/L as shown in table 3.

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Table 3.

Mortality of *Clarias gariepinus* juveniles exposed to acute concentrations of aqueous *Piptadeniasttrium africanum* bark extract for 96 h.

Concentration	No of fish	Total mortality	% mortality	Log conc	Probit
0.0	30	0	0.0	0.00	0.00
25.0	30	11	36.7	1.34	4.67
50.0	30	15	50.0	1.70	5.00
75.0	30	18	60.0	1.88	5.25
100.0	30	21	70.0	2.00	5.52
125.0	30	25	83.3	2.10	5.95

However, no mortality (0 %) was recorded in the control experiment. Mean mortality increased significantly ($p<0.05$) with increase in extract concentrations but decreased with exposure period.

The LC_{50} of the aqueous extract of *P. Africanum* bark in the exposed *C. gariepinus* juveniles over the 96–hour exposure period was calculated to be 42.21 mg/L with the 95 % upper and lower confidence limit of 30.37–58.67 mg/L respectively.

The regression equation of the relationship was calculated to be probit y=1.551x+2.479, log concentration and on R^2=0.899.

The expression R^2 value indicates that, mortality rate of fish increased with increase in concentration of *P. africanum* bark extract as shown in figure 1.

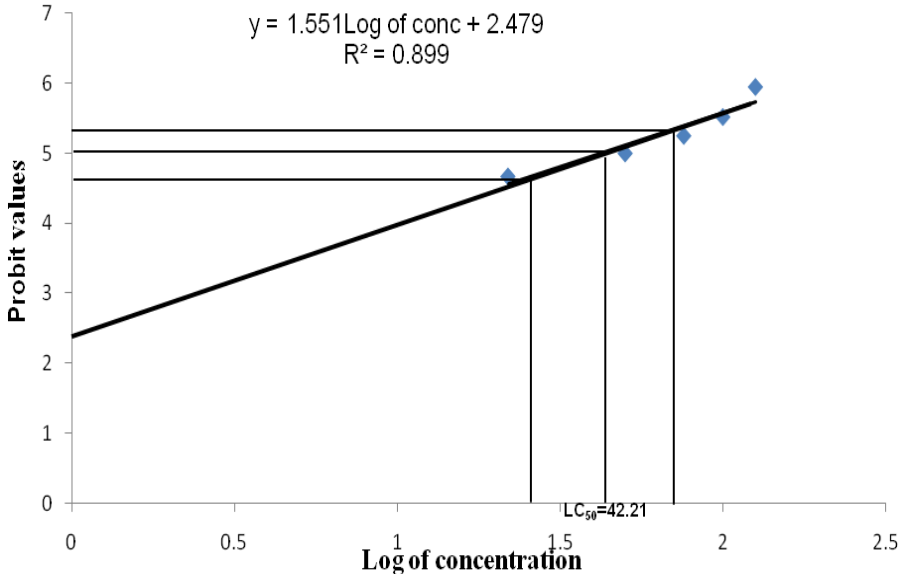

Figure1. Linear relationship between mean probit mortality and log concentration of *clarias gariepinus* juveniles exposed to acute concentrations of aqueous extract of *piptadeniasttrium africanum* bark for 96 h.

Several behavioural changes were observed ranging from excessive mucus secretion, erratic swimming, discoloration, air gulping, restlessness and settling at the bottom motionless just before death.

These behavioural changes increased with increasing extract concentration and with exposure period. The histopathological alterations observed in the gill and liver of *C. gariepinus* juveniles as presented in plates 1–12 is an indication of the toxic effect of *P. africanum* bark extracts.

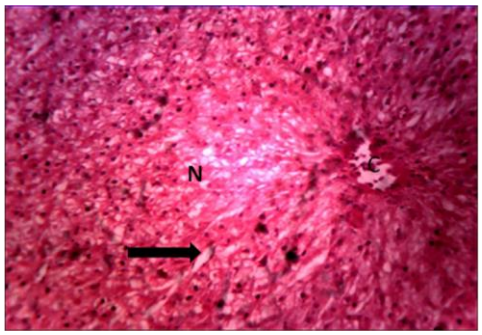

Plate 1: Microphotograph of liver cells of *Clarias gariepinus* juveniles exposed to aqueous extracts of *P. africanum* bark at control (0.0 mg/L) showed normal hepatic tissue (black arrow), hepatocytes with granular cytoplasm (C) and central and round nucleus (N). (x400).

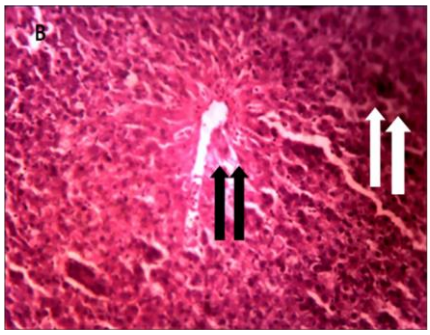

Plate 2: Microphotograph of liver cells of *Clarias gariepinus* juveniles exposed to aqueous extracts of *P. africanum* bark at 25 mg/L showed focal necrosis (black arrow), bile stagnation (white arrows), and nuclear degeneration (B) (x400)

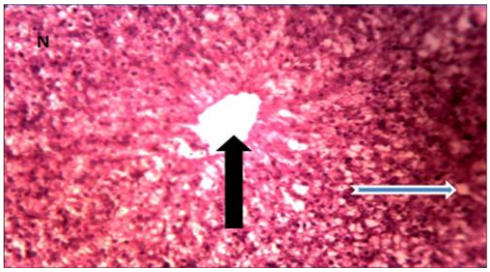

Plate 3: Microphotograph of liver cells of *Clarias gariepinus* juveniles exposed to aqueous extracts of *P. africanum* bark at 50 mg/L showed hepatocytes with irregular shaped nucleus (black arrows), eosinophilic granules in the cytoplasm (arrowheads) and nuclear hypertrophy

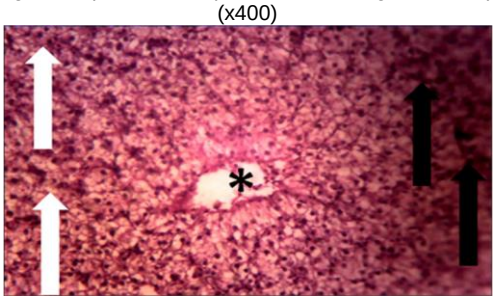

Plate 4: Microphotograph of liver cells of *Clarias gariepinus* juveniles exposed to aqueous extracts of *P. africanum* bark at 75 mg/L showed vascular congestion (black arrow) fibrosis (white arrow) and vacoulation of cytoplasm (*)(x400)

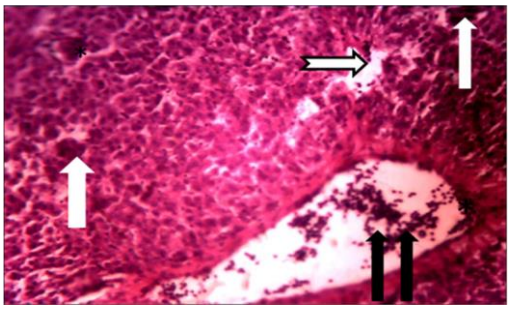

Plate 5: Microphotograph of liver cells of *Clarias gariepinus* juveniles exposed to aqueous extracts of *P. africanum* bark at 100 mg/L showed liver with hepatic tissue revealing focal necrosis (black arrow), bile stagnation (white arrows), nuclear degeneration (arrowhead) and cytoplasmic degeneration (*) (x400)

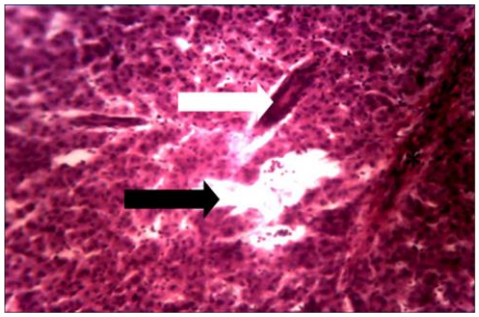

Plate 6: Microphotograph of liver cells of *Clarias gariepinus* juveniles exposed to aqueous extracts of *P. africanum* bark at 125 mg/L showed cytoplasmic vacuolation (black arrows), bile pigment disintegration (white arrows) and eosinophilic granules in the cytoplasm (*)(x400)

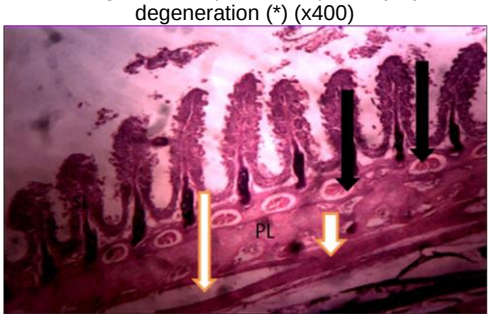

Plate 7: Photomicrograph of gill filament of *Clarias gariepinus* juveniles exposed to aqueous extracts of *P. africanum* bark at control (0.0 mg/L) Showed normal aspect of the gill filaments (black arrow), a pillar cell (white arrow), epithelial lining (arrowhead), primary lamellae (PL) and secondary lamellae (SL) (x400)

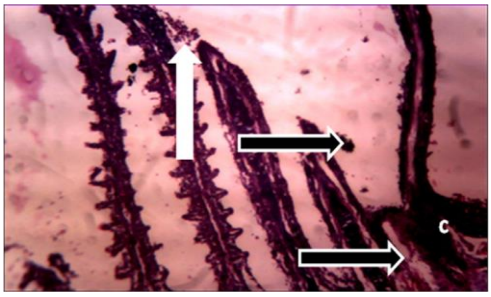

Plate 8: Microphotograph of gill filament of *Clarias gariepinus* juveniles exposed to aqueous extracts of *P. africanum* bark at 25 mg/L showed degeneration of primary and secondary lamellae (black arrows), cellular axis vacuolation(C)and epithelial erosion (white arrow)(x400)

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Banat's Journal of Biotechnology
2017, VIII(15)

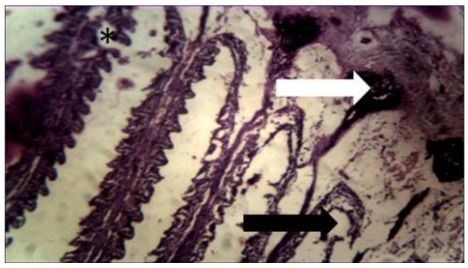

Plate 9: Microphotograph of gill filament of *Clarias gariepinus* juveniles exposed to aqueous extracts of *P. africanum* bark at 50 mg/L showed lamellae with the marginal channel dilated (black arrow), hyperplasia of the epithelial cells (white arrow) and epithelial lifting (*)(x400).

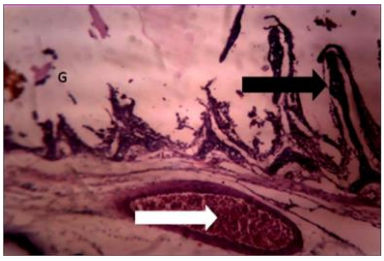

Plate 10: Microphotograph of gill filament of *Clarias gariepinus* juveniles exposed to aqueous extracts of *P. africanum* bark at 75 mg/L showed blanketing of the lamellae with distortion of pillar cells (black arrow) and hyperplasia of epithelial cells (white arrow), epithelial lifting and oedema (G)(x400)

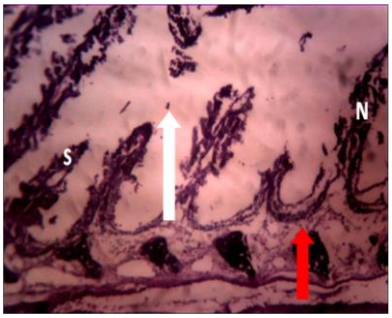

Plate 11: Microphotograph of gill filament of *Clarias gariepinus* juveniles exposed to aqueous extracts of *P. africanum* bark at 100 mg/L showed epithelial erosion with haemorrhage (white arrow), hyperplasia of lamellar epithelium (red arrow), necrosis (N) and malformed secondary lamellae (S) (x4oo)

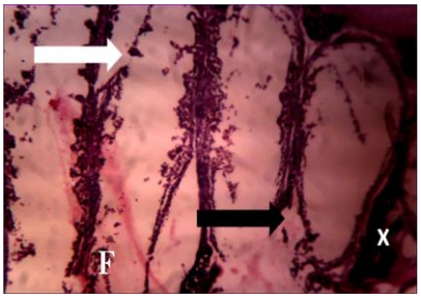

Plate 12: Microphotograph of gill filament of *Clarias gariepinus* juveniles exposed to aqueous extracts of *P. africanum* bark at 125 mg/L showed epithelial rupture with haemorrhage (black arrow) hyperplasia of lamellar (X) blanketing (white arrow) and epithelial and diffusion of secondary lamellae (F) (x400)

The results obtained from the phytochemical screening of *P. africanum* is similar with that reported by Akinlami, Obomanu, Absalom, and Akpa [AKINLAMI *et al;* 2013; OBOMANU *et al*; 2005; ABSALOM *et al;* 2013 and AKPA *et al* ;2010].

They reported on the presence of these bioactive compounds in several piscicidal plants including: *Piptadeniastrium africanum, Lepidagathis alopecuroides, Balanite egyptica and Tephrosia vogelii* respectively used in obnoxious fishing practices.

The observed signs of toxicity including histological alterations as well as the ultimate mortality in some of the exposed fish might be due to the presence of these bioactive substances [OLOGE and SOGBESAN., 2007, BUTNARIU, *et al.,* 2015a].

Tannins are reported to be cytotoxic, antineoplastic,nephrotoxic and hepatotoxic [RIEVERE *et al*; 2009; BAGEPALLIS *et al,* 1992, BUTNARIU, *et al.,* 2016] due to their ability to precipitate exogenous and endogenous proteins.

Steroids are known to inhibit oxidative phosphorylation with subsequent impairment of oxygen consumption in exposed fish [TIWARI and SINGH; 2003, BUTNARIU and CORADINI, 2012] and therefore, might have contributed to the observed signs of respiratory distress in the exposed fish.

Although, the physicochemical parameters of the test water fluctuated slightly during the bioassay but were not thought to have affected fish mortality since they were within tolerance range [FAFIOYE., 2012, BUTNARIU, *et al.,* 2015b].

This is in line with the findings of Ayuba and Ofojekwu., Absalom,Audu,and orji [AYUBA and OFOJRKWU;2002, ABSALOM *et al*., 2013, AUDU *et al*., 2014 and ORJI *et al*., 2014].

The observed behavioural changes in this study are similar with that reported by other studies [AYOTUNDE *et al*; 2010; FAFIOYE *et al*;2004; ABALAKA and AUTA 2010] who exposed *C. gariepinus* to various levels of piscicides.

The exhibited clinical signs and eventual deaths of exposed fish may be due to direct poisoning leading to pathological alterations in their tissues and organs [ABLAKA and AUTA; 2010].

The agitated behaviours correspond to both the contact and exertion phases of fish's response to toxicants exposures

[BESCH; 1975] and it is their natural response in trying to escape from absorbing these offending extracts. The presence of excessive mucus secretion as reported by other studies [ANUNNE and AJIKE; 1999, FAFIOYE *et al*; 2000, ABALAKA and AUTA, 2010, ORJI *et al.,* 2014] as observed in this study was because of the increase in the activities of mucus cells subsequent to exposure to pollutants [ANUNNE *et al*., 1991; 1994, BUTNARIU, 2012].

However, such excessive mucus secretions are reported to reduce respiratory activity in fishes [ADEOGUN *et al.,* 2012, BUTU, *et al.,* 2014b] which together with decreasing oxygen content of reconstituted extracts results into hypoxic states in exposed fishes [USMAN *et al*; 2005. BUTNARIU, 2006, IANCULOV, *et al.,* 2004] leading to subsequent respiratory distress and deaths in exposed fishes [OMITOYIN *et al*; 2006].

The findings of this study revealed that the 96 hour LC_{50} for African catfish exposed to *Piptadeniastrium africanum* bark was 42.21 mg/L.

The 96 hour LC_{50} had been reported for *C. gariepinus to be* 0.32 mg/L by Dahunsi and Oranusi (2013) in effluents from rubber processing plant, 0.35 mg/L by Orji [ORJI *et al;* 2014] in *Psychotria mychrophilla leaves*, 7.35 mg/L by Abalaka [ABALAKA *et al*; 2014] in*Adeniumobesum*stem bark and 12.9 mg/L by Ayotunde [AYOTUNDE;2010] in *Carica papaya* seeds.

Comparatively, the 96 h LC_{50} value of 42.21 mg/L obtained from this study was much lower than 430 mg/L, 105.83 mg/L; and 204.17 mg/L reported by other reports [AYOTUNDE; 2006; ABALAKA and AUTA ;2010 and AYUBA and OFOJEKWU 2002] respectively who worked with different plant poisons.

This showed that *P. africanum* bark extract was more toxic to *C. gariepinus* than other reported plant extract as higher LC_{50} values signify less toxicity [HELFRICH *et al*.,2009, RODINO, *et al.,* 2014] and vice versa.

The variations observed in these studies can be attributed to the type of plants and part of the plants used, size of fish, environmental factors, food, or water parameters and selective action of toxicants [BUTU, *et al.,* 2014c, CAUNII, *et al.,* 2015].

The toxic nature of the *P. africanum* bark might be the reason behind the global use of the plant to poison fish [OBOT; 2006, BUTNARIU and GIUCHICI, 2011, FERENCZ, *et al.,* 2012].

As earlier reported by Absalom [ABSALOM *et al.,* 2014] in [TREASE and EVANS, 1989] that LC_{50} is the convenient reference point for expressing the acute lethal toxicity of a given pollutant to fish.

Gills and liver pathological alterations have been reported in fishes exposed to toxicants. [ANUNNE and AJIKE., 1999., AYOTUNDE 2006., and OLUSEGUN and ADEDAYO; 2014].

The role of the gill in diffusion of oxygen and excretion predisposes it in such a way that slight structural damage can render a fish very vulnerable to osmoregulation as well as respiratory difficulties [ADEDAYO and OLUSEGUN., 2014].

The lesion observed in gills of *C. gariepinus* juveniles exposed to *P. africanum* bark extract in this investigation may cause a drastic difficulty in respiration.

The gills from the control experiment had a normal architectural makeup of gill morphology of primary and secondary lamellae arising from these parallel with them and perpendicular to the filament axis.

The treated gills showed histo–pathological changes. These alterations could lead to several physiological stresses in fish and difficulty in overcoming oxygen diffusion in test fish resulting to hypoxic respiratory responses leading to eventual suffocation and death of fish [ADEOGUN; 2004, BUTU, *et al.,* 2014a].

The liver is considered the most important target organ from a toxicological point of view because of its role in detoxification, biotransformation and excretion of xenobiotics [OLUSEGUN and ADEDAYO; 2014, BUTNARIU, 2014, BARBAT, *et al.,* 2013, BUTU, *et al.,* 2015, DIMITRIU, *et al.,* 2016].

After enteric uptake of injurious chemicals, it is the first organ to be exposed to hazards via portal circulation and many xenobiotics tend to accumulate in the liver at high concentrations [OLUSEGUN and ADEDAYO; 2014, BUTNARIU, *et al.,* 2012, PETRACHE, *et al.,* 2014, HAMBURDA, *et al.,* 2016].

The liver of C. gariepinus juveniles at the control showed normal architectural make-up of the organ while the liver of fish exposed to the aqueous extract of *P.*

africanum showed different histological damage.

The resultant damage of liver could be due to the sensitive nature of liver to toxicants. This agrees with works of Rahman and Ayotunde where livers of freshwater fishes were exposed to acute–lethal and chronic–sub lethal concentrations of different toxicants [RAHMAN *et al*., 2002; AYOTUNDE; 2006, BUTNARIU, and BOSTAN, 2011, MIHALACHE, *et al.,* 2016].

The pathological changes in the gills and liver were very prominent and pronounced and strongly implicate *P. africanum* as a toxicant because these organs are important for diffusion of oxygen and detoxification of xenobiotics.

Conclusions

The essence of this research work was to determine the toxicological effects of *P. africanum* on *Clarias gariepinus* juveniles. The study has shown that, the aqueous bark extract of *Piptadeniastrium africanum* was toxic to African catfish (*Clarias gariepinus)* with 96–hr LC_{50} value of 42.21 mg/L.

The toxic effects were reflected in the death of fish during the acute toxicity test as well as gill and liver pathological alterations in the test fish.

Generally, one could deduce from this research work that the indiscriminate introduction of *P. africanum* into water bodies is toxic and would threaten the life and existence of fish and other aquatic organisms thereby reducing fish production and aquatic fauna.

This study may be useful in providing information for possible further use of *Piptadeniastrium africanum* in aquaculture practices.

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