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Phyto-qualitative Evaluation and Effects of Aqueous Extracted *Erythrophleum suaveolens* on sub-adult *Clarias gariepinus* (Burchell, 1822)

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Traditional means of capturing fish dwells on active components in plant which elicit harmful effects. *Erythrophleum suaveolens* is a typical example of such plant, which the study investigated for its stem-back (ASE) and leaf (ALE) aqueous extracts sub-lethal effects on *Clarias gariepinus*. Qualitative phytochemical screening of both extracts was carried out using 300 *C. gariepinus* sub-adults maintained in 10-fish sample per 1000 L circular fibre tanks during the research; the extracts were applied on every other day at 0.23 mg/ L ASE and 0.26 mg/ L ALE, which elicited LC_{50} effects from the range finding tests, for 96 h compared with control set-up. Haematological (during the experiment) and histopathological (at the terminal of the experiment) parameters of the fish were analysed. Results showed significant variations in the treated fish haematological responses compared with the control. More impact was observed in ALE treated fish and linked to three active components (phenols, saponins and steroids) higher than in ASE, with no negative observations on the fish under the control treatments. Fish exposed to ALE showed severely eroded mucosa of the gill secondary lamellae while to ASE revealed signs of severe erosion of the entire gill mucosa. Liver of fish exposed to ALE showed a severe diffuse vacuolation of hepatocytes with several hepatocytes necrotic while to ASE revealed a severe diffuse vacuolation and necrotic hepatocyte with mild to moderate cellular infiltration. The study concluded that both extracts had sub-lethal toxicological effects on the exposed fish as they both elicited polycythemia.

Key words: Active components, Gill mucosa, LC50 effects, Necrosis, Nigeria

The various forms of structural diversity of natural products contribute to the development of new synthetic active substances that are of commercial importance (Sukari *et al.,* 1992). Plants like all other organisms employ enzymes that act as an intermediary in chemical reactions or pathways which are used in synthesizing several molecules required to build up their tissues and ensure optimal functioning (Wink, 1999). This chemical reaction expressed through the metabolic pathways in conjunction to the primary and secondary metabolites of the compounds produced. Plant primary metabolites are compounds responsible to build and maintain cell components such as chlorophyll and adenosine triphosphate (ATP), while secondary metabolites include the flavonoids, alkaloids, saponins, tanins, rotenones (flavonoids) are required for defence mechanisms in plants and therapeutic purposes, as well as repellant or poisons to invader (Kauffman *et al.,* 1999).

Plant poisons are widely applied for hunting and fishing, and poisonous plants termed piscicide or ichthyotoxins are found to be effective on fishes if applied generally rather than being applied against individual targets (Jett, 1991). In order to increase catch yields in traditional fishing, diverse fish poisons of plant origin have been explored (Kamalkishor and Kulkarni, 2009; Agbon *et al.,* 2013). The latter authors also reported that toxic plants can be applied to control diseases in aquaculture. A typical poisonous plant that belongs to the Fabaceae family is *Erythrophleum suaveolens* which is widespread in tropical Africa and confirmed to be extremely toxic. Toxicity mechanisms of *E. suaveolens* were extensively studied without limitation to its anaesthetics potentials on clarrid fish; acute toxicity on albino mice; anti-fungal properties, anti-bacterial properties, phytochemical and toxicological properties, toxicity and mutagenic activity, sub chronic toxicity on rabbits; fungal activities on wood; as well as anti-oxidant and anti-bacteria of its saponin fractions, and wound healing activity (Mgbenka and Ejiofor, 1999; Idyu *et al.,* 2004; Adedotun *et al.,* 2006; Aiyegoro *et al.,* 2007; Abia *et al.,* 2008; Ogunsanwo and Adedeji, 2010; Akanji and Sonibare, 2015). Limited works on the toxicity of *E. suaveolens* on fish due to its widespread use by artisanal fisherfolks had been carried out (Sowunmi and Adeogun, 2002).

Therefore, investigations into the plant toxic effects on commercially available fish species during acute, chronic and sub-lethal exposures are not only necessary but also appropriate (Singh and Singh, 2002). Thus, the study was carried out to evaluate possible sub-lethal exposures of *E. suaveolens* stem-bark and leaf extracts on haematology and histopathology (gill and liver) of *C. gariepinus.* Gills are extremely important in respiration, osmoregulation, acid–base balance and excretion of nitrogenous wastes in fish and thus, represent the greatest area of the animal in contact with external environment (Heath, 1995). On the other hand, fish liver is one of the organs that are most affected by toxicants in the water (Rodriques and Fanta, 1998), with lesions being observed in the liver to indicate the extent of damages (Roche and Boge, 1996). Liver is an organ that is most associated with detoxification and biotransformation process, and its function and position (Van der Oost *et al.,* 2003).

MATERIALS AND METHODS

Sourcing, qualitative phytochemical screening and preparation of the plant

Fresh stem-back and leaf of Erythrophleum suaveolens were sourced from Ilewo-Orile, Abeokuta North Local Government, Ogun State, Nigeria and identified by experts at the Department of Forestry and Wildlife Management, Federal University of Agriculture, Abeokuta (FUNAAB). Phytochemical screening for eight constituents (Phenols, Flavonoids, Alkaloids, Oxalates, Tannins, Saponins, Steriods and Cardiac glycosides) were carried out using standard qualitative methods as previously described (Harborne, 1992; Sofowora, 1993). The stem-back and leaf of E. suaveolens were first air-dried for 3 days at room temperature, oven dried for 24 h using GallenKamp oven at 60 ºC (Oshimagye et al., 2014), the stem-back parts were pulverized using mortar and pestle into finely grounded particles while the leaf parts were pulverized with Paulson electric blender. The final pulverized products were kept in black polythene bags; 980 g stem-back was soaked in 8 L of water for 24 h and the concoction was sieved with a muslin cloth (Agbon et al., 2004), and 1.12 kg leaf was prepared the

same way.

Collection and acclamatization of sub-adult Clarias gariepinus

Three hundred (300) C. gariepinus sub-adults with mean weight of 121.95 ± 0.31 g and standard length of 24.47 ± 0.89 cm were collected from the Directorate of University Farms (DUFARMS), FUNAAB and transported in batches in 50 L capacity plastic containers to the FUNAAB hatchery. The fish were first acclimatized in an outdoor concrete tank (2 x 3 x 1.5 m) for two weeks before being transferred into a 1000 L circular fibre tanks inside the hatchery and fed to satiation with 4 mm commercial feed (Skretting) twice daily during the experimental periods. Water was changed every other day following the previously recommended method (Agbon et al., 2002).

Haematological and histopathological examination

About 0.5 – 1 mL of blood of the experimental fish was drawn with 5 mL hypodermic needle and syringe from the region between the pelvic fin weekly for 28 days after exposure to aqueous stem-back (ASE) and leaf extract (ALE) of Erythrophleum suaveolens. The drawn blood samples were emptied into 10 mL sample bottles previously treated with anticoagulant (Ethlylene Diamine Tetracetic Acid, EDTA), and moved to the Veterinary Teaching Hospital Laboratory, FUNAAB, where haematological tests were carried out using standard procedures. The quantifed haematological parameters included PCV and Hb; RBC and WBC; and differential counts of RBC and WBC were carried out as adopted respectively by Wintrobe (1978) and Shaw's diluting fluid (Miale, 1982) using haemacytometer (improved Neubauer, Weber Scientific International Ltd, UK). The erythrocytes indices (MCV, MCH and MCHC) were calculated from the values of RBC, PCV and Hb using the formular proposed by Dacie and Lewis (2001). Histological analyses of the experimental and control fish were carried out at the Department of Anatomy, Faculty of Veterinary Medicine, University of Ibadan, Ibadan, Nigeria. The analyses were performed by removing the gill and liver of the ASE and ALE E. suaveolens exposed fish to know their responsive changes which had occurred organs using Hematoxylin and Eosin techniques (Samuelson, 2007).

Data analysis

Obtained data from phytochemical analysis and haematological parameters were presented using descriptive (Table, Mean ± S.D) and inferential statistics (Analyses of Variance and Duncan Multiple Range Test, at $p < 0.05$) using Microsoft Office 2010 and IBM SPSS Statistics 22.

RESULTS

Qualitative phytochemical analysis of Erythrophleum suaveolens plant

The phytoanalysis of the plant under study reflected higher quantities of the eight organic chemicals (phenols, flavonoids, alkaloids, oxalates, tannins, saponins, steroids and cyanogenic glycosides) considered in the leaf part than the stem-back part. Steroids chemical was however not detected in the stem-back. The levels of higher presence of these chemicals (phenols, saponins and steroids) in the leaf might make its extracts more toxic with deleterious effects when applied.

Haematological responses of *C. gariepinus* exposed to *E. suaveolens* aqueous extracts

In comparison to the control treatments, increase in the levels of quantified packed cell volume (PCV), haemoglobin (Hb) and red blood cell (WBC) was observed with possible implications that the test fish in either aqueous stem-bark extract (ASE) or aqueous leaf extract (ALE) responded to abate the toxicological effects being imposed on from day7 to day28. The quantified levels were higher in the ALE than in the ASE. The residual parameters could have influenced the observation as the renewed quantities increased across the experimental periods and higher in the ALE treatments due to higher presence of phenols, saponins and steroids. Increase in the levels of Hb for survival of the test fish under study would influence carriage of more oxygen across the fish system to adapt to the toxicological threats. For higher quantity/ level of oxygen to be circulated for the test fish survival, more of PCV and RBC would in turn or directly be produced (Fig. 1). Influences of the various treatments were not significantly ($p > 0.05$) different on the RBC unlike ($p < 0.05$) on both PCV and Hb (Table 2).

White blood cell (WBC) in the fish occurred inherently to subdue xenobiotics (ASE and ALE) in the blood stream; the WBC was quantified highest in the control fish blood compared to the ASE and ALE, where there were reduced quantities from the efforts of the test fish to abate the threats posed by xenobiotics (ASE and ALE) introduced as treatments. In turn, the quantified accompanied indices (mean corpuscular volume; MCV, mean corpuscular haemoglobin; MCH, and mean corpuscular haemoglobin concentration; MCHC) were higher in the experimental fish under ASE and ALE treatments compared to the control fish. The WBC from the influence of ALE treatments became least quantified in days 21 and 28, when the three indices (MCV, MCH and MCHC) became highest. Residues of ALE triggered by the presence of higher quantities of phenols, saponins and steroids could have been more threatening and toxic than ASE's in days 21 and 28 (Fig. 2). Influence of ASE treatments was not significantly ($p > 0.05$) different on WBC and MCH, while there was no significant ($p > 0.05$) different on the levels of MCH and MCHC under the control (Table 2).

Responses of ASE and the control were inversely proportional to both heteroplphils (HET) and eosiniphils (EOS) while ALE treatment influences were moderate. Percentage levels of HET were highest in the ASE treatments, where EOS was least, these occurred in 10-manifold. It might be inferred that ALE influenced (traceable to higher presence of phenols, saponins and steroids) more toxicological effects than ASE (Fig. 3). There were significant ($p < 0.05$) differences from the treatments on the both levels of HET and EOS (Table 2). Also, ALE treatments were observed to increasingly influence highest levels of lymphocytes (LYM) and monocytes (MON), which were reducing in the control fish from day 7 to 28. There was no significant ($p > 0.05$) difference in the levels LYM as well as MON in the control treatment. Their (LYM and MON) levels were significantly ($p < 0.05$) different in the ASE and ALE (Table 2).

Histo-pathological responses of sub-adult *C. gariepinus* to ASE and ALE

There was no lesion on the gills in the control unlike the various distortions observed under either the ASE (aqueous stem-back extract) or ALE (aqueous leaf extract), the former showed that there was a very severe erosion of the entire gill mucosa while the latter induced severe eroded mucosa of the secondary lamellae (Fig. 5). Observation indicated normal condition of the fish liver having normal hepatocytes and a slightly congested portal vessel, while the fish liver under ASE treatment in one hand had a severe diffuse vacuolation and necrosis of the hepatocyte associated with mild to moderate cellular infiltration, the fish liver under ALE treatments on the other hand was not without a severe diffused vacuolation of hepatocytes accompanied several hepatocytes necrotic.

Table 1. Qualitative phytochemical screening of Erythrophleum suaveolens plant

Parameters	Stem-back	Leaf
Phenols	+	++
Flavonoids	+	+
Alkaloids	++	++
Oxalates	+	+
Tannins	++	++
Saponins	+	++
Steroids	-	+
Cyanogenic glycosides	+	+

(-): Not present (+): Mild (++): High

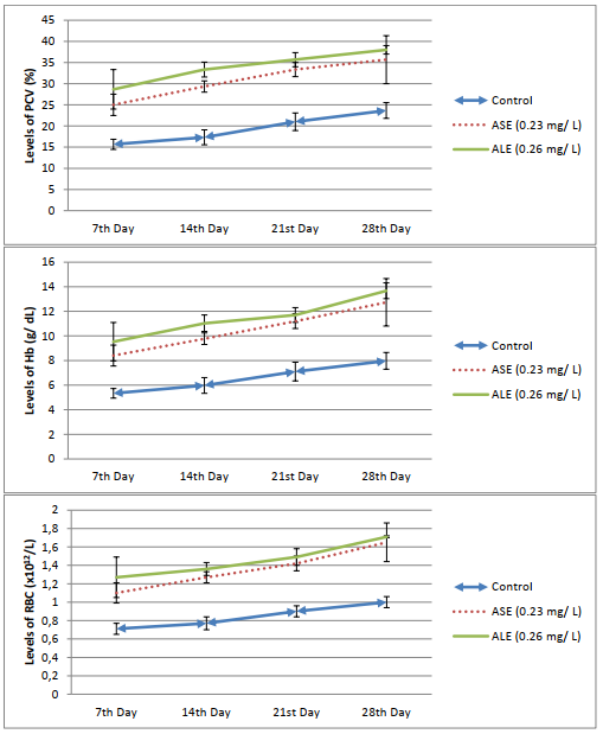

Figure 1. Determined levels of haematological parameters (PCV, Hb and RBC) under the control, ASE and ALE treatments; ASE is aqueous stem-root extract, ALE is aqueous leaf extract, PCV is pack cell volume, Hb is haemoglobin, RBC is red blood cell.

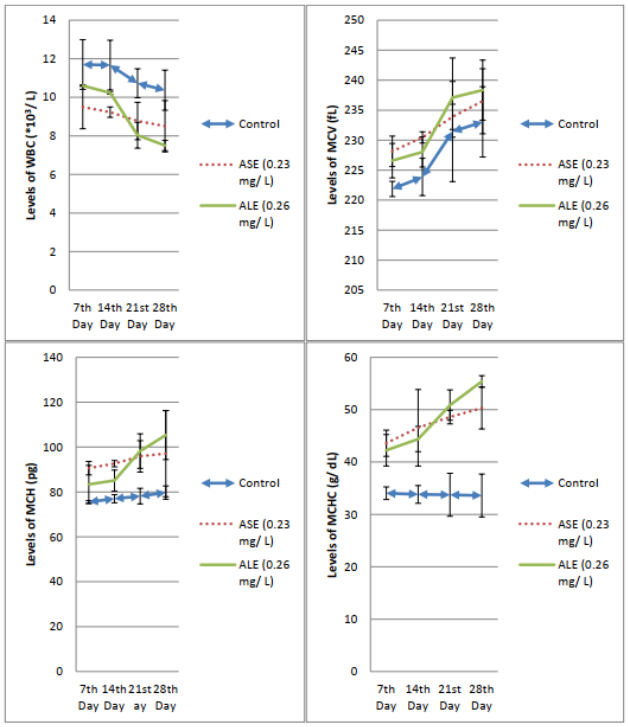

Figure 2. Determined levels of haematological parameters (WBC, MCV, MCH and MCHC) under the control, ASE and ALE treatments; ASE is aqueous stem-root extract, ALE is aqueous leaf extract, WBC is white blood cell, MVC is mean corpuscular volume, MCH is mean corpuscular haemoglobin, MCHC is mean corpuscular haemoglobin concentration.

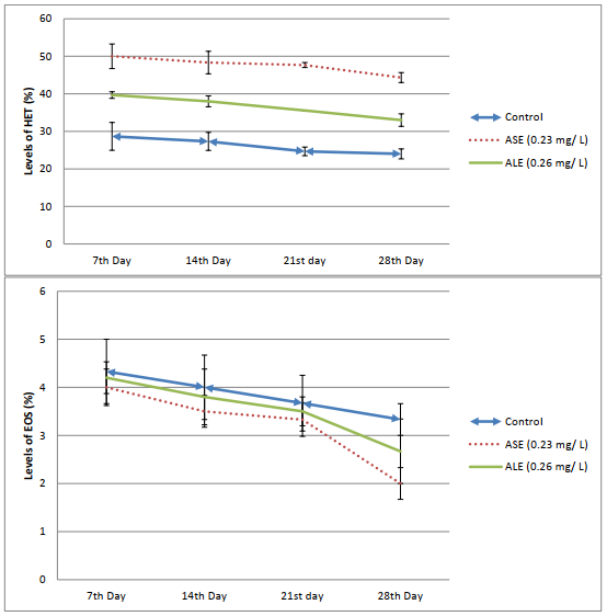

Figure 3. Determined levels of haematological parameters (HET and EOS) under control, ASE and ALE treatments; ASE is aqueous stem-root extract, ALE is aqueous leaf extract, HET is Heterophils, EOS is Eosinophils.

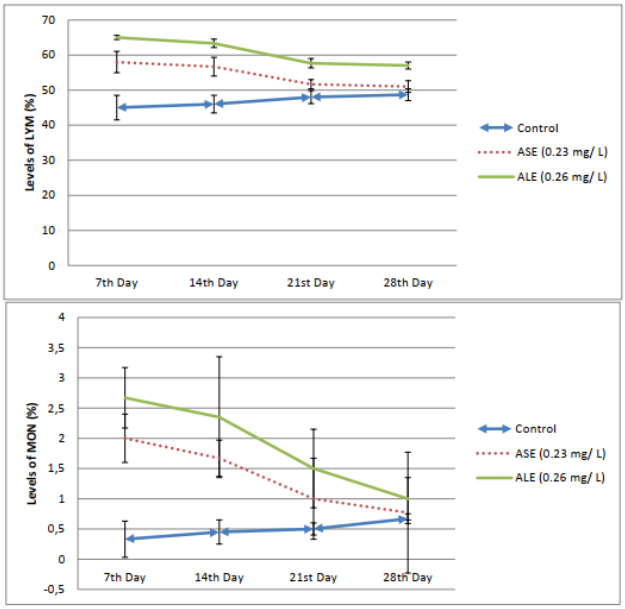

Figure 4. Determined levels of haematological parameters (LYM and MON) under control, ASE and ALE treatments; ASE is aqueous stem-root extract, ALE is aqueous leaf extract, LYM is Lymphocytes, MON is Monocytes.

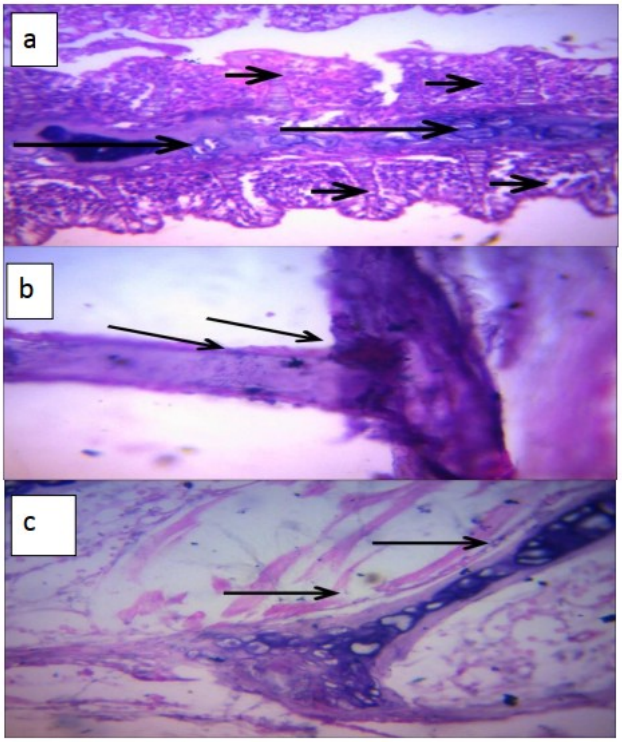

Figure 5. Sub-adult *C. gariepinus* gill exposed to sub-lethal (LC_{50}) concentration of *E. suaveolens* at 28th day; a: Normal cartilaginous core of the primary lamellae (long arrows) and the mucosa of the secondary lamellae (short arrows) under control, b: A very severe erosion of the entire mucosa (arrows) under ASE, c: Severely eroded mucosa of the secondary lamellae (arrows) under ALE (H & E, X400).

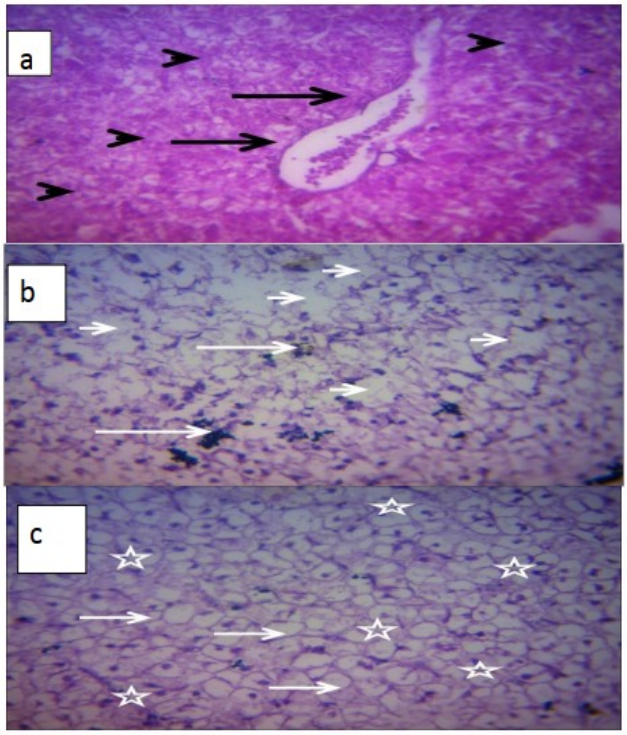

Figure 6. Sub-adult *C. gariepinus* gill exposed to sub-lethal (LC_{50}) concentration of *E. suaveolens* at 28th day; a: Normal hepatocytes (arrowheads) and slightly congested portal vessels (long arrows) under control, b: A severe diffuse vacuolation and necrosis of the liver cells (short arrows), with mild to moderate cellular infiltration (long arrows) under ASE, c: A severe diffuse vacuolation of hepatocytes (stars) with several hepatocytes necrotic (arrows) under ALE (H & E, X400).

Table 2. Levels of significance of various haematological parameters

Parameters	Control				ASE (0.23 mg/ L)				ALE (0.26 mg/ L)			
	7th	14th	21st	28th	7th	14th	21st	28th	7th	14th	21st	28th
PCV (%)	a	ab	b	c	a	b	c	d	a	b	c	d
Hb (g/dl)	a	ab	b	c	a	b	c	d	a	b	bc	c
RBC ($x10^{12}$/L)	a	a	a	a	a	a	a	a	a	a	a	a
WBC ($x10^{3}$/L)	a	a	b	b	a	a	a	a	a	a	b	c
MCV (fL)	a	ab	c	c	a	b	c	d	a	ab	c	cd
MCH (pg)	a	a	a	a	a	a	a	a	a	a	b	c
MCHC (g/dl)	a	a	a	a	a	b	c	d	a	ab	c	d
HET (%)	a	ab	c	cd	a	ab	b	c	a	b	c	d
EOS (%)	a	b	c	d	a	b	bc	d	a	b	c	d
LYM (%)	a	a	a	a	a	a	b	bc	a	a	b	bc
MON (%)	a	a	a	a	a	b	c	d	a	b	c	d

Same alphabets across the rows under each treatment were not significantly ($p > 0.05$)

DISCUSSION

Different bioactive constituents were determined in qualitative screening through liquid infusion of *E. suaveolens* stem-back (ASE) and leaf (ALE). Phytochemicals could either be harbinger or elixir depending on the usefulness. Protective mechaanisms of Glutathione S-transferase (GST) when minimizing the toxic potential of oxygen intermediate is prone to be affected by rotenone (flavonoids) in plant (Ibiam *et al.,*

2015). The elicited harbinger effect on the fish under study could be linked to alkaloids being one of the detected components which may not only pose Na^+ - K^+ ATPase, DNA polymerase, cytochrome P-450 system inhibition (Ibiam *et al.,* 2015) but also impairing respiration and osmo-regulation (Annune *et al.,* 1991). Presence of saponin is deleterious to fish for having respiratory epithelia damaging effect and could result in asphyxiation because of its haemolytic in nature (Prance and Balick, 1990). The cyanogenic glycoside has been recorded to have negative effect on central nervous system and nerve mechanisms of the heart (Prance and Balick, 1990). The compound was determined in the ALE of *E. suaveolens* in this study but was not in the past study (Hassan *et al.,* 2007) using leaf of *E. africanum*.

Haematological changes are possibly reflecting in the physiology of fish, and have been used as an index of health status of fish species. So, alterations elicited by ASE and ALE treatmets compared to the control on the treated fish haematological parameters in this study was similar to the work of Alwan *et al.* (2009) observed on *Coptodon zillii* exposed to aluminium. Different possible impacts on most of the haematological parameters by both treatments compared to the control might have resulted from stimulation of growth and differentiation factors in the bone marrow influenced formation or proliferation of blood cells or improved erythropoietin production as reported by Elgerwi *et al.* (2013).

The effect of both *E. suaveolens* ASE and ALE treatments on the hematological profile of fish is polycythemia elicited by increase in RBC, Hb and PCV as observed and reported previously (Mckim *et al.,* 1970; Taylor *et al.,* 1985). Responses of PCV, Hb, and RBC could be as a result of erythropoiesis increase in amount of circulating RBC, which is a first mechanism through which the fish might compensate for poor oxygen uptake in prevailing hypoxic conditions and also the release of erythroblasts, resulting from an increased rate of RBC catabolism (Wepener *et al.,* 1992). Annune *et al.* (1994**)** reported a significant increase in RBC count of *C. gariepinus*, unlike in this study, when subjected to Zn treatment. They attributed the RBC elevation to blood cell reserve as well as cell shrinkage resulting from osmotic alterations of blood by the toxicant effects. The increased values of RBC and associated parameters (Hb, PCV) were indication of polycythemia (ADA, 2000). The increase and difference in RBC till the 28th day of exposure might be due to stimulation of erythropoietin by the elevated demands for O_2, CO_2 transport as a result of increased metabolic activity or destruction of gill membranes causing faulty gaseous exchange. The increase in Hb content could be explained as a process where the treated fish tried to replace the oxidized denatured Hb (Cyria *et al.,* 1989). There release of high quantity of mature RBC in the general circulations might also trigger by stimulation of the β-adrenergic action on the hemopoietic tissues to contract and release stored mature RBC and compensate for short-term oxygen concentration variations in blood or water (Nespolo and Rosenmann, 2002). The significant increase observed on increased RBC volume could be as a result of either osmotic change due to ion losses from the blood plasma or adrenergic-splinic contraction in hypoxic conditions (Witters *et al.,* 1990, 1991). Increases in PCV and the RBC indices (MCV, MCH and MCHC) quantities were attributed to swelling of RBC due to increased CO_2 in blood, hypoxia or stressful procedures (Nemesok and Boross, 1999). This corroborated the work of Zaki *et al.* (2011) when investigated haematological responses in *Clarias lazera* to phenol toxicity.

There was a decrease in the WBC count of sub-adult *C. gariepinus* throughout the 28-day sub-lethal exposure to both ASE and ALE of *E. suaveolens* which Oyewole *et al.* (2009) had associated to immune-suppressant ability of a toxicant all through the exposure period. The WBC reduction might also be from redistribution into tissues to abate the harbinger influences of the both ASE and ALE (Guyton and Hall, 1996), or as a result of bone marrow depression and toxins competition for folic acid utilization (Jain, 1986). The WBC count reduction of the exposed fish may also be due to release of epinephrine during stress posed by the extracts, capable of initiating contraction of spleen and decrease of leucocytes count resulting possibly in the waning of immune system (Svoboda, 2001; Witeska, 2003). However, increasing or decreasing in the volume of WBC is a normal reaction to a toxicant (Kori-Siakpere *et al.,* 2006), to signify effects of the test fish immune

system under either ASE and ALE threats as similar to the work of Ogbonnaya and Uadia (2016) when evaluated haematological responses associated with sub-acute exposure of rats to *Telfaira occidentalis* root, pod and stem extracts.

The decreased WBC from the 21st day (prominent in ALE, which had high levels of phenols, saponins and steroids) might have been caused by hemolysis and hemorrhaging damage to hematopoietic tissues, enzyme dysfunction, faulty gaseous exchange leading to hypoxia and stress mediated hormonal imbalance (Thangnan *et al.,* 2016). This is in agreement with the findings of Sampath *et al.* (1993) when they exposed Nile tilapia (*O. niloticus*) to a toxic environment. The WBC decrease could also be the result of bio-concentration of the plant extract organic constituents in the fish organs: kidney and liver. This had been associated to hindering of granulopoiesis or lymphpoiesis induced by primary or secondary changes in haematopoietic organs (Tomaszewski, 1997), similar to the work of Ololade and Oginni (2010) on toxic stress and haematological responses on *C. gariepinus*. There was progressive reduction in the quantified percentage lymphocytes (LYM) and monocytes (MON) under the ASE and ALE treatments compared to the control. This was said to be owing to cortisol secreted during stress reaction to not only shorten their (LYM and MON) life spans and promote their apoptosis (Verburg van Kemenade *et al.,* 1999) but also reduce their proliferation (Wyets *et al.,* 1998). Decrease in LYM and MON counts and their activity was being observed as effects of stress irrespective of the toxicant. Shah and Altindag (2004) reported that impairments in haematological parameters in diverse fish species on exposure to stressors could be attributed to haemodilution, haemoconcentration, haemolysis and haemorraging, damage to hematopoietic tissues, accelerated erythroclasia, increased mechanical fragility, impaired osmoregulation, enzyme dysfunction, faulty gaseous exchange at gills leading to hypoxia and stress mediated hormonal imbalance.

The control fish showed normal cartilaginous core of the primary lamellae and mucosa of the secondary lamellae whereas the exposure of test fish to ASE and ALE of *E. suaveolens* led to a very severe erosion of the entire gill mucosa and a severely eroded mucosa of the secondary lamellae respectively. Mucus cells contain mucins, polyanions composed of glycoproteins that can be effective in trapping toxicants and promote prevention of toxicant entry into the gill epithelium (Perry and Laurent, 1993). Gill alterations such as epithelial hyperplasia and separation of the epithelial layer from supportive tissues are usually directly related to gill dysfunction, resultantly affecting physiology or eliciting death of the fish (Smart, 1976). Partial fusions of some secondary lamellae are examples of defense mechanisms, resulting in the increase of the distance between the external environment and the blood to serve as a barrier to contaminants entry (Mallatt, 1985). The lesions found on the exposed gills may elicit reduced oxygen-uptake capacity of the gill, making the test fish less able to get adequate oxygen for its total metabolic activity. Winkaler *et al.* (2007) reported that this may in turn lead to internal hypoxia, to cause an increase in hematocrit due to swelling of the erythrocytes that occurs whenever fish blood cells are exposed to hypoxia. Toxic substances (ASE and ALE of *E. suaveolens* in this study) can be injurious to gills, thereby reducing O_2 consumption and disrupting the osmo-regulatory function of the test fish (*C. gariepinus*) (Saravana and Geraldine, 2000). Similar responses had been reportedly observed in *C. gariepinus* exposed to stressors (Gabriel *et al.,* 2007). Tissue hypoxia caused by gill epithelia damage in the exposed fish could also have caused the observed hepatic lesions (Mohammed, 2001). This corroborated the work of Abalaka *et al.* (2015) when exposed *C. gariepinus* to ASE of *Adenium obesum*.

The control fish also showed normal hepatocyte and slightly decongested portal vessels on the liver while the *E. suaveolens* exposed fish liver had severe diffuse vacuolation and necrosis of the hepatocytes with mild to moderate cellular infiltration to ASE and severe diffuse vacuolation of hepatocytes with several hepatocytes necrotic to ALE. The observed necrosis may be due to inability of the fish under study to regenerate new liver cells, with resultant changes of metabolic problems on the liver characterized by the remains of the bile in the form of droplets in the cytoplasm of the hepatocyte as suggested by the submission of Pacheco and Santos

(2002) that this was an indication of possible damage to the hepatic metabolic functions of the liver, leaving the liver of the exposed fish with severe vacuolated cells for evidence of fatty degeneration. Necrosis of some portions of the liver was observed and might probably result from the excessive stress on the test fish to get rid of the organic constituents of the extracts from its body during the process of detoxification by the liver. Increased vacuolations of the hepatocytes could be an indication of fatty change, which are pathological responses to the threats (Pacheco and Santos, 2002; Wolf and Wolfe, 2005) of ASE and ALE exposures. Such a liver fatty change might be due to either excessive fat mobilisation to the liver exceeding its capacity to metabolise it or liver damage that cannot adequately metabolise the fat (Mohan and Mohan, 2011).

CONCLUSIONS

Organic chemical constituents were determined in the extracts (ASE and ALE) of the plant (*E. suaveolens*) under study. The sub-lethal dose of both ASE and ALE of *E. suaveolens* elicited haematological responses of the exposed fish leading to polycythemia and mortality during the study. The treatments also influenced severe damages to both gill and liver of the exposed fish over the period of time.

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CONFLICT OF INTEREST

The authors declare that they have no conflict of interest.

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