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Impact of Different Concentration of Thorn Apple (*Datura stramonium)* Extract on Growth, Histopathology and Blood Profiles Response to Sub- Lethal Exposure of Nile Tilapia (*Oreochromis niloticus)* Juveniles

***[1]OLUSOLA, SE; [1]AYEBIDUN, OV; [2]AMUBIEYA, AV; [1]AMULEJOYE, FD**

**[1]Department of Fisheries and Aquaculture Technology, School of Agriculture, Food and Natural Resources, Olusegun Agagu University of Science and Technology, Okitipupa, Nigeria.*
[2]Department of Biological Sciences (Fisheries and Aquaculture Programme), School of Science, Olusegun Agagu University of Science and Technology, Okitipupa, Nigeria.

**Corresponding Author Email: se.olusola@oaustech.edu.ng, belloolus@yahoo.com; Tel: +2348034110139*
Co-Authors Email: ov.ayebidun@oaustech.edu.ng; adesolavikky7@gmail.com; fd.amulejoye@oaustech.edu.ng

ABSTRACT: This study investigated the toxic effects of *Datura stramonium* leaves extract on growth, histopathology and blood profiles of *Oreochromis niloticus* juveniles. *Datura stramonium* leaves extract of various concentrations were prepared at 0.00 (control), 0.002, 0.02 and 0.2 ml/L for 28 days and 10 days withdrawal in triplicate. Twenty (20) *Oreochromis niloticus* fingerlings (6.05±0.01g) were randomly distributed into each experimental bowl containing 45 L of treatment concentrations. Biological evaluation, blood profiles parameters, and histology of the exposed fish were determined. The result shows an increase within the weight gain and the weight gain was higher in the exposed groups expect TAL 4 than the control. There was no significant difference (P>0.05) among the treatments. The haematological response to *D. stramonium* exposures shows that the values of packed cell volume and haemoglobin were higher in the exposed groups compared to the control while the white blood cell, neutrophils, and lymphocytes decrease as the concentration of the plant increases and were lower than the control. The result revealed a higher value of total protein and albumin in the exposed groups compared to the control. There were decreased in the values of ALT and AST when compared to the control and the values of total protein, albumin, ALT and AST after 10 days of withdrawal show a similar trend in values obtained before the experiment. The result of histopathology shows no observable lesions were observed in control group while there were moderate alterations in gills, liver and the intestine of the exposed groups. This investigation revealed that continuous use of the *D. stramonium* leaves extract in the aquatic environment will affects the growth, blood profiles and histopathology of the exposed fish.

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Nile tilapia *Oreochromis niloticus*, a cichlid fish are found in freshwater and occasionally brackish water environments in Africa, Madagascar, Israel, Syria, India, Sri Lanka, South America, Central America, North America and West Indies (Nelson, 2006), true tilapias are native only to Africa and the Middle East (Barnali *et al*., 2018). It reaches up to 60cm in length and can weigh up to 5kg, the male has better body composition than female, and they are mostly herbivore, feeding on the phytoplankton, algae,

detritus and aquatic insect larvae. They are maternal mouth brooders and exhibiting parental care (Nico, 2019). Plants are used in aquaculture to stimulate fish catches and these plants have negative effects on the aquatic organism and the aquatic environment because it leads to reduced growth, alteration physiological functions and sudden death. Fishermen use a poisonous plant for capturing fish (Fafioye *et al.,* 2004). The active principles in the plant part used include leaves, seeds, kernel and bark which have varying potencies (Sambasivam *et al*., 2003). The piscicidal plants contain active ingredients known as alkaloids which are toxic to fish and other aquatic organisms at high concentration (Olaifa *et al.,* 1987). Scientific studies have revealed some piscicidal plants used in aquaculture; *Parkia biglobosa* (Fafioye *et al.,* 2004), pawpaw (*Carica papaya*) seed (Ayotunde and Ofem, 2008), *Raphia vinifera* (Fafioye *et al*., 2004), mesocarp of neem plant (*Azadirachta indica)* (Akinwande *et al.,* 2007), *Derris elliptica* (Akinbulumo *et al.,* 2004), *Tephrosia vogelii* (Agbon *et al.,* 2004), *Raphia hookeri* (Adeogun *2004),* Tobacco (*Nicotiana tobaccum*) leaf dust (Omoniyi *et al*., 2002), *Tephrosia candida* (Mohotti and Epa, 2016), *Kigelia Africana* (Ufodiku and Omoregie (1994), *Tetrapleura tetraptera* (Omitoyin *et al*., 1999), Cocoa bean shell (Olaifa *et al*., 2008), *Thevetia peruvina* (Oti and Ukpabi, 2005). Asides their use as traditional piscicidal agents for catching wild fish, these plants are used in aquaculture management for controlling the predatory and wild fishes (Neuwinger, 2004; Fafioye *et al*., 2004). *Datura stramonium* belongs to the family Solanaceae and is known as thorn apple, Jimsom weed or devil snare, the plant species are native to Central America (Preissel and Preissel, 2002). The leaves are soft, smooth, toothed, and irregular undulated with 8 – 20 cm long. The leaves have a bitter and nauseating taste. *Datura* species are rich in alkaloids (e.g. hyoscyamine, hyoscine, atropine, and scopolamine), saponins, flavonoids, phenols, essential oils and cardiac glycosides (Ayuba *et al.,* 2011; Silver, 2005; Singh and Kaushal, 2007). They are used as an aphrodisiac and to induce drowsiness or stupefaction, making strangulation easier (Dash and Mike, 2005). However, there is scanty information on the mode of action and its effects on *O. niloticus* have not been fully documented. Hence this study assessed the toxicity effects of *D. stramonium* leaves extract on weight, histopathology and blood profiles of *O. niloticus* fingerlings.

MATERIALS AND METHODS

Collection and Preparation of D. stramonium Leaves: Datura stramonium leaves (fresh) were collected from a farm in Ilu-titun, Ondo State. The plant was identified at the Department of Biological Sciences (Botany programme), Olusegun Agagu University of Science and Technology, Okitipupa and 200 g of the plant leaves were weighed, washed with sterile distilled water, macerated and squeezed in 10 litres of water to get the aqueous leaves extracts. The aqueous leaves extracts were filtered through a muslin cloth, and the filtrate was stored in a refrigerator until required for bioassay test.

Range Finding Test: Oreochromis niloticus fingerlings (250) with a mean weight of 6.05±0.01g were purchased from a fish farm in Okitipupa, Ondo State, Nigeria. The fish were acclimatized for two weeks and fed twice daily in the pelleted fish feed (Coppens) containing 40% crude protein. Excess feed and faeces were siphoned out to prevent fouling odour that may be resulting from feed residue and faecal. Twenty-four hours before the commencement of the LC_{50} range-finding test, feeding was stopped for the fish. Each of the test solutions was introduced directly into the experimental tank in a single dose and replicate thrice per treatment. The extracts of the *D. stramonium* leaves were dissolved in distilled water and introduced into the experimental bowls containing dechlorinated well-aerated tap water at different concentrations (0.2, 0.4, 0.6, 0.8 and 1.0 ml/L) for range finding test. Fish were randomly selected and stocked at 8 *O. niloticus* fingerlings per experimental bowl for range finding test. The 48 hours LC_{50} value was recorded and the concentration of *D. stramonium* aqueous extract used for the definitive toxicity test was 0.2ml/L

Definitive Test: Twenty (20) fish per treatment was exposed to the sub-lethal concentration for 28 days and were replicated three times. 1/10th value of the resulting 48 hours LC_{50} value (0.2ml/L) was considered as sub-lethal concentration. Four (4) groups were exposed to different concentrations of *D. stramonium*. One group of the fish did not receive *D. stramonium* served as the control group for *O. niloticus*. Other treatments received 0.002, 0.02 and 0.2 ml/L *D. stramonium* with 35 litres of water for *O. niloticus*. The test plant was changed every 72 h and the experiment lasted for 28 days. The fish were fed at 3% body weight with 2 mm Coppens. The diets per day were divided into two; 1.5% in the morning by 8.00 am and evening by 5.00 pm. Measurement of the weight changes was performed every week and the feeding rate adjusted weekly according to the new body weight.

Histopathological Examination: The fish from both the exposed and control group was dissected, fish tissues such as liver, intestine and gills from each

treatment were carefully removed and washed in 0.9% saline and fixed in 10% formalin in heparinized bottles (without EDTA) and taken for histopathological examination at Histopathology Unit of Veterinary Pathology Department, University of Ibadan, Nigeria. These organs were sections and staining as described by the modified methods of Rodrigues, (2007) and Omitoyin *et al.,* (2006). A light photomicroscope attached to a 35 mm camera was used to examine the organs sections.

Haematological and Biochemical Assessment: Blood samples (5 ml) was collected before the experiment and at the end of the experiment (day 28) and 10 days after withdraw (to assess physiology recovery of fish) from the caudal peduncle of both the exposed and control fish into heparin bottles (with EDTA) and transported in ice packed to the Microbiological Laboratory unit of Ondo State Specialist Hospital, Okitipupa for the analysis of complete haematology using standard methods as described by Dacie and Lewis, (1991).

Plasma and Serum Biochemistry: Blood samples were centrifuged at 3000 rpm for 15 minutes to obtain serum biochemical parameters. Serum from the centrifuged blood was carefully siphoned out and the concentration of Glucose, total proteins, albumin and globulin were estimated as described by Dacie and Lewis, (1991).

Biological Evaluation: The weight of the test animals in exposed and an unexposed (control) group was recorded at the commencement and after the experiment (28 days) of the sub-lethal test. Weight changes in the fishes were carried out every week with the aid of a battery-operated weighing scale and the following biological evaluation was measured as Weight gain = Final Body Weight – Initial Body Weight, Percentage weight gain = (Final Body Weight – Initial Body Weight) / Initial Body Weight × 100**,** Specific Growth Rate (SGR) = 100 (loge FBW – loge IBW) / Time**,** Feed Conversion Ratio (FCR) = Dry weight of the feed / Fish weight gain**,** Nitrogen Metabolism (NM) = 100 (number of fish stocked - Mortality) / Number of Fish Stocked**,** Survival Rate (SR) = Initial number of fish stoked – Mentality/Initial number of fish stoked x100**,** Nitrogen metabolism = (0.549)(a+b)h/ 2**,** where, a = Initial body weight, b = Final body weight**,** h = Duration of the experiment (Olusola and Olawoye, 2019).

Statistical Analysis: Data obtained were subjected to one-way analysis of variance (ANOVA) while the means were compared for significant differences using the Duncan test.

RESULTS AND DISCUSSION

Behavioural Responses to Toxicity of D. stramonium: During the range-finding test the following observations were observed in the exposed fish; loss of appetite, erratic movement, restlessness, frequent opercula movements, state of motionlessness, mucus secretion, adoption of different postures, swirling movements, loss of reflex and loss of balance.

Growth response and nutrient utilization of O. niloticus exposed to different concentration of D. stramonium for 28 days: The result of the present study revealed an increased weight among the treatments, but there was no significantly different (P > 0.05) in initial body weight, final body weight, weight gain, percentage weight gain specific growth rate, feed conversion ratio and nitrogen metabolism among the dietary groups (Table 1)

Table 1: Growth response and nutrient utilization of *O. niloticus* fingerlings on commercial feed for 28 days.

Parameters	Control (0 ml/L)	TAL2 (0.002 ml/L)	TAL3 (0.02 ml/L)	TAL4 (0.2 ml/L)
Initial Body Weight (g)	6.05±0.01^{a}	6.05±0.00^{a}	6.05±0.01^{a}	6.05±0.00^{a}
Final Body Weight (g)	7.32±1.21^{a}	7.31±0.41^{a}	7.51±0.48^{a}	7.07±0.20^{a}
Weight gain (g)	1.27±1.20a	1.36±0.28^{a}	1.50±0.49^{a}	1.25±0.57^{a}
Weight Gain (%)	21.02±1.88^{a}	20.78±6.82^{a}	24.70±8.09^{a}	16.89±3.36^{a}
Specific Growth Rate	0.29±0.26^{a}	0.29±0.08^{a}	0.34±0.09^{a}	0.24±0.04^{a}
Feed Conversion Rate	7.19±1.48^{a}	4.39±0.89^{a}	3.76±1.22^{a}	5.21±1.04^{a}
Survival Rate (%)	90.00±0.00^{ab}	75.00±0.07^{a}	95.00±7.07^{b}	85.00±0.01^{ab}
Nitrogen Metabolism	102.69±9.34^{a}	102.69±3.15^{a}	103.30±2.18^{a}	100.84±1.53^{a}

Key: Mean of triplicate data, mean value in each row with similar superscripts are not significantly different

Mean Haematological Parameter of O. niloticus on Sub-lethal Concentration of D. stramonium Leaves Extracts: This study shows the variation in the value of some haematological parameters among the treatments after 28 days exposure of *D. stramonium*. Pack cell volume, haemoglobin, neutrophils, lymphocytes, and monocytes were not significant (P >0.05) among the treatments. The withdrawal recorded relative similar values in some haematological parameters among the treatments and when compared to the values of the exposed groups (Table 2).

Table 2: Mean haematological parameter of *O. niloticus* fingerlings before, after exposure to different concentrations of *D. stramonium* for 28 days and 10 days withdrawal period

		28 days of exposure				10 days withdrawal period			
Parameter	**Before**	**Control (0 ml/L)**	**TAL2 (0.002 ml/L)**	**TAL3 (0.02 ml/L)**	**TAL4 (0.2 ml/L)**	**Control (0 ml/L)**	**TAL2 (0.002 ml/L)**	**TAL3 (0.02 ml/L)**	**TAL4 (0.2 ml/L)**
PVC%	40.00±1.82^{a}	40.00±0.02^{a}	41.00±0.82^{a}	43.00±0.02^{a}	44.00±0.02^{a}	40.00±0.03^{a}	41.00±1.00^{a}	41.00±0.83^{a}	41.00±0.90^{a}
HB(g/dl)	13.30±0.28^{a}	13.30±0.00^{a}	14.30±0.28^{bc}	13.70±0.08^{c}	13.70±0.10^{ab}	13.30±0.30^{a}	14.00±0.30^{b}	14.00±0.20^{b}	14.10±0.10^{b}
RBCx10$^{12/l}$	4.70±0.20^{a}	4.80±0.20^{a}	4.50±0.20^{a}	5.10±0.60^{a}	4.70±0.06^{a}	4.10±0.08^{a}	4.40±0.00^{a}	4.30±0.01^{a}	4.60±0.28^{a}
WBCx10a/l	11.20±0.30^{ab}	11.70±0.03^{b}	11.40±0.30^{ab}	10.90±0.02^{a}	10.80±0.01^{a}	11.10±0.30^{a}	11.40±0.25^{a}	11.80±0.40^{b}	12.10±0.80^{b}
Platelet(m/µl)	29.10±0.30^{a}	30.00±2.83^{a}	28.70±0.30^{a}	31.50±0.20^{a}	30.50±0.30^{a}	30.10±0.28^{a}	31.40±0.08^{b}	30.40±0.05^{a}	30.50±0.00^{a}
MCV (F1)	85.30±0.30^{a}	83.30±0.30^{b}	95.60±0.60^{c}	86.30±0.08^{c}	87.30±0.30^{a}	97.600±0.30^{ab}	97.70±0.30^{ab}	100.00±2.83^{b}	95.70±0.28^{a}
MCH(pg/cal)	28.30±0.30^{ab}	27.70±0.02^{a}	31.80±0.30^{d}	28.84±0.01^{bc}	29.20±0.07^{c}	32.40±0.30^{a}	32.50±0.10^{a}	32.30±0.02^{b}	32.00±0.10^{a}
MCHC (g/dl)	33.30±0.20^{a}	33.30±0.06^{a}	38.30±0.70^{b}	33.40±0.20^{a}	33.40±0.10^{a}	33.30±0.01^{a}	33.30±0.03^{a}	33.30±0.04^{a}	33.40±0.08^{a}
NEU (%)	66.00±2.83^{a}	69.00±2.83^{a}	68.00±1.83^{a}	67.00±2.38^{a}	67.00±2.05^{a}	69.00±2.0.04^{a}	69.00±0.02^{a}	68.00±0.03^{a}	69.00±0.05^{a}
LYMP (%)	32.00±0.20^{a}	32.00±0.09^{a}	31.00±0.20^{a}	29.00±0.00^{a}	27.00±0.01^{a}	28.00±0.02^{a}	30.00±2.83^{a}	30.00±2.53^{a}	28.00±0.22^{a}
MONO (%)	1.00±0.30^{a}	1.00±0.01^{a}	2.00±2.02^{a}	1.00±0.00^{a}	2.00±0.20^{a}	1.00±0.00^{a}	1.00±0.28^{a}	2.00±1.83^{a}	3.00±2.83^{a}
EOS (%)	1.00±0.30^{b}	1.00±0.30^{b}	0.00±0.00^{a}	0.00±0.00^{a}	1.00±0.16^{b}	1.00±0.30^{b}	0.00±0.00^{a}	1.00±0.30^{b}	0.00±0.00^{a}

Keys: TAL= Thorn Apple Leaves, PVC= Packed Cell Volume, HB= Haemoglobin, RBC= Red Blood Cell, WBC= White Blood Cell, MCV= Mean Cell Volume, MCH= Mean Cell Haemoglobin, MCHC= Mean Cell Haemoglobin Concentration, LYMP= Lymphocytes, MONO= Monocytes, EOS= Eosinophil. Mean of triplicate data, the mean value in each row with similar superscripts are not significantly different (p>0,05)

Table 3: Mean Plasma Biochemistry parameter of *O. niloticus* fingerlings before and after exposure to different concentration of *D. stramonium*

Parameter	28 DAYS EXPOSURE					10 DAYS WITHDRAWAL PERIOD			
	Before	**Control (0)**	**TAL2 (0.002 ml/L)**	**TAL3 (0.02 ml/L)**	**TAL4 (0.2 ml/L)**	**Control (0)**	**TAL2 (0.002 ml/L)**	**TAL3 (0.02 ml/L)**	**TAL4 (0.2 ml/L)**
TP (g/dl)	74.00±0.50^{a}	69.00±0.30^{a}	74.00±2.30^{a}	70.00±0.02^{a}	71.00±0.30^{a}	72.00±1.80^{a}	72.00±1.53a	72.00±1.83^{a}	73.00±2.83^{a}
ALB (g/dl)	36.00±0.70^{a}	33.00±0.20^{a}	39.00±0.05^{a}	40.00±0.20^{a}	40.00±0.01^{a}	35.00±2.80^{a}	35.00±1.60^{a}	35.00±1.40^{a}	34.00±0.80^{a}
GLO (g/dl)	38.00±1.30^{b}	36.00±0.02^{ab}	35.00±0.80^{ab}	30.00±0.30^{a}	31.00±0.02^{ab}	38.00±2.83^{ab}	35.00±0.03^{a}	38.00±2.83^{ab}	36.00±0.93^{a}
ALB/GLO Ratio	1.00±0.20^{a}	0.90±0.01^{a}	1.10±0.50^{a}	1.30±0.08^{a}	1.30±0.10^{a}	1.00±0.20^{a}	1.10±0.30^{a}	1.00±0.10^{a}	0.90±0.01^{a}

KEYS: TAL=Thorn Apple Leaves, TP= Total Protein, ALB= Albumin, GLO= Globulin, ALB/GLO= Albumin-Globulin ratio. Mean of triplicate data, the mean value in each row with similar superscripts are not significantly different (P >0.05).

Table 4: Mean blood serum biochemistry parameter of *O. niloticus* fingerlings before and after exposure to different concentration of *D. stramonium*

Parameter	28 DAYS EXPOSURE					10 DAYS WITHDRAWAL PERIOD			
	Before	**Control (0)**	**TAL2 (0.002ml/L)**	**TAL3 (0.02 ml/L)**	**TAL4 (0.2 ml/L)**	**Control (0)**	**TAL2 (0.002ml/L)**	**TAL3 (0.02ml/L)**	**TAL4 (0.2 ml/L)**
ALT (iu/i)	11.00±1.80^{a}	10.00±0.90^{a}	10.00±0.80^{a}	11.00±1.80^{a}	10.00±0.40^{a}	12.00±1.83^{a}	11.00±0.33^{a}	10.00±0.01^{a}	10.00±0.03^{a}
AST (iu/i)	13.00±0.01^{a}	13.00±0.10^{a}	10.00±0.20^{a}	12.00±0.10^{a}	10.00±0.00^{a}	14.00±1.85^{a}	13.00±0.65^{a}	13.00±0.80^{a}	15.00±2.82^{a}
GLU (mol/l)	5.10±0.28^{as}	4.40±0.50^{ab}	5.40±0.70^{c}	3.90±0.30^{a}	4.00±0.80^{a}	4.30±0.23^{a}	4.70±0.70^{a}	4.70±0.80^{a}	4.00±0.00^{a}

KEYS: TAL= Thorn Apple Leaves, ALT= Amino alanine transferase, AST= Aspartate aminotransferase, GLU= Glucose. Mean of triplicate data, the mean value in each row with similar superscripts are not significantly different (p>0.05).

Mean plasma biochemistry parameter of O. niloticus on the sub-lethal concentration of D. stramonium leaves extracts: The result revealed a higher value of total protein and albumin in the exposed groups compared to the control and there was no significant difference (p>0.05) among the treatments. However, the value obtained after 10 days of withdrawal show a similar trend in total protein and albumin. There was no significant difference (p>0.05) among the treatments (Table 3).

Mean blood serum parameter of O. niloticus on the sub-lethal concentration of D. stramonium leaves extracts: The result shows that there was no significant difference (p>0.05) in values of AST, ALT and Glucose among the treatments. They decreased in the values of ALT, AST when compared to the control after 10 days of withdrawal of exposure to different concentration of *D. stramonium* (Table 4).

Histological parameter of O. niloticus fingerlings on the sub-lethal concentration of D. stramonium leaves extract: There is no observable lesion in the experimental organs (gills, liver and intestine) in the control and moderate observable changes at TAL2, but there are severe changes at TAL3 and TAL4 (Plate 1A- C). *Oreochromis niloticus* fingerlings exposed to *D. stramonium* leaves aqueous extract revealed different behavioural changes such as abnormal swimming, turning movement, restlessness, frequent opercula movements, setting at the bottom motionless, mucus secretion, adoption of different postures, swirling movements, loss of reflex and loss of balance. This may be as a result of the active component (atropine) in *D. stramonium* which impaired the vital organ from the transportation of oxygen. This study agreed with the report of Olusola *et al.*, (2021) who recorded symptoms such as abnormal swimming, restlessness and uncoordinated behaviour before death in *C. gariepinus* fingerlings exposed to *D. stramonium* extract. Also, Akinbulumo *et al.*, (2004) and Mohotti and Epa, (2016) reported that similar behavioural changes were observed on *O. niloticus* fingerlings exposed to *Derris* powder extracts and *Tephrosia candida* aqueous extract respectively.

The result of the experiment revealed that there is a general increase in the weight of the fish among the treatments. The weight gain was higher in the exposed groups expect TAL 4 than the control and there were no significant difference (P>0.05) among the treatments. However, the study revealed that feed conversion ratio, specific growth rate, percentage weight gain was not significantly different (P >0.05) among the treatments. The fish exposed to *D. stramonium* recorded a slight increase in weight.

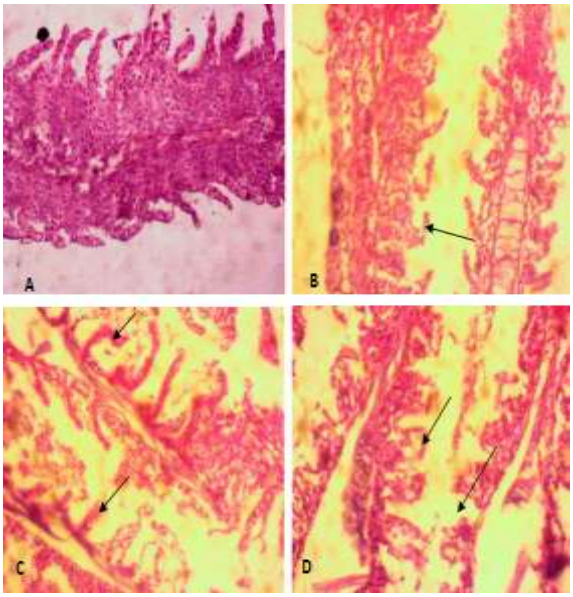

Plate 1A, (A – D): Photomicrograph showing a transverse section of gill filaments of *O. niloticus* fingerlings. (**A**) Control showing there is no observable lesion (**B**) Gill of exposed at 0.002ml/L indicating there is moderate diffuse lamellar atrophy (arrow) (**C**) Gill of exposed at 0.02ml/L indicating there is severe lamellae atrophy (arrows) (**D**) Gill of exposed at 0. 2ml/L showing there is severe lamellae atrophy (arrows) HE x400

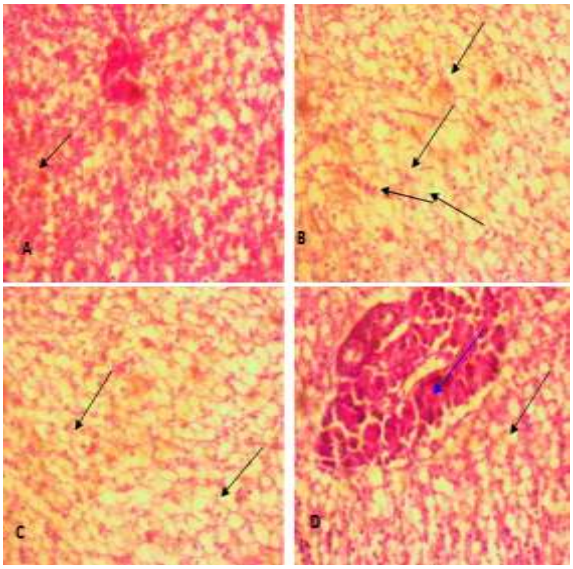

Plate 1B, (A – D): Photomicrograph showing a transverse section of the liver of *O. niloticus* fingerlings (**A**) Control showing there is no observable lesion (**B**) Liver of exposed at 0.002ml/L indicating there is centrilobular hepatocellular vacuolar degeneration (arrows). (**C**) Liver of exposed at 0.02ml/L indicating there is multiple foci of hepatocellular degeneration (arrows) (**D**) Liver of exposed at 0. 2ml/L showing there is random hepatocellular degeneration (black arrow) and melano-macrophages (blue arrow) in hepato-pancreas. HE x400

The result of the present study agreed with the report of Onusiriuka and Ufodike, (1998) on *Clarias gariepinus* exposed to *Blighia sapida* and *Kigelia africana* and Omoregie *et al.*, (1994) on *O. niloticus*

exposed to formalin. This study shows variations (increase and decrease) in the values of some haematological parameters. The increase and decrease in these haematological parameters may be an indication of hyperglycaemia, and hypoproteinemia caused by exposure of the fish to *D. stramonium* aqueous extract. This study was in agreement with the report of Omoregie *et al.,* (1994) who reported similar observations when *O. niloticus* was exposed to different concentrations of formalin. Packed cell volume and haemoglobin recorded an increased value in the exposed groups, the value was higher than the control. This study agrees with the report of Ojikutu *et al*., (2013) who reported an increase in packed cell volume and haemoglobin concentration of juveniles *C. gariepinus* on exposure to different concentration of cypermethrin.

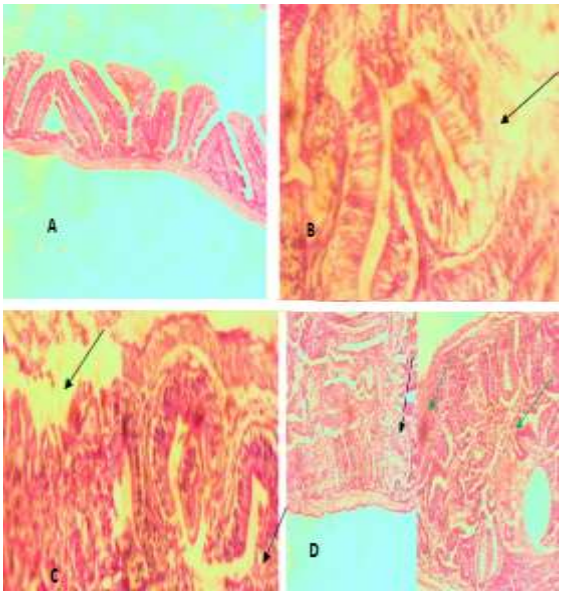

Plate 1C, (A – D): Photomicrograph showing a transverse section of the intestine of *O. niloticus* fingerlings. (**A**) Control showing there is no observable lesion (**B**) Intestine of exposed at 0.002ml/L indicating there is enterocyte degeneration and necrosis with a few inflammatory cells (arrow) (**C**) Intestine of exposed at 0.02ml/L indicating there is a fusion of villi (arrows) and loss of surface enterocytes (**D**) Intestine of exposed at 0. 2ml/L showing there is necrotizing enteritis characterized by clubbed to fused villi (black arrow) and cellular infiltrates (green arrows). HE x400

Also, the value of white blood cell, neutrophils, and lymphocytes decrease as the concentration of the *D. stramonium* increases, this revealed that the concentration of *D. stramonium* altered the immune response of the fish (i.e the higher the concentration of the plant the lesser the immune response of the fish to *D. stramonium* aqueous extract). This could be due to immunological reaction leading to a reduction in antibody production to cope with the stress induced by the toxicant. However, the value obtained after 10 days withdrawal shows relative similar values to the value obtained before the experiment and during the exposure, the value of packed cell volume reduces and shows relative similar value to the control (P>0.05). The value of white blood cell was higher in the exposed groups after withdrawal when compared to the control. The value of neutrophils and lymphocytes were similar in the exposed groups when control after 10 days of withdrawal. The value of MCV, MCH and MCHC shows an increase in the exposed groups and were higher than the control (P < 0.05). This investigation was aligned with the report of Adeyemo *et al*., (2008) and Ojikutu *et al*., (2013) who reported that MCV, MCH and MCHC values were higher in exposed groups compared to the control group of *C. gariepinus* exposed to different concentration of lead nitrate and Cypermethrin respectively. This study shows that total protein and albumin were higher in the exposed groups compared to the control and there was a general reduction in the value of total protein, globulin and albumin of the exposed groups after 10 days of withdrawal and these values were similar to control. This could be correlated with the increase in antibody production which help in the survival and recovery in fish exposed to sub--lethal concentration of *D. stramonium*. This result was in accord with the reported by Musa and Omoregie, (1999) who reported an increase in total protein and albumin of *Clarias gariepinus* that was exposed to sub-lethal doses of malachite green and with the report of Cimanga *et al*., (2018) who reported an increase in TP, ALB and GLO of *C. gariepinus* exposed to sub-acute toxicity of aqueous extract of *C. sanguinolenta* root bark. The value of amino alanine transferase and aspartate aminotransferase were not significantly different (P> 0.05) among the treatments, the values of aspartate aminotransferase was higher in the control compared to the exposed groups and the values of amino alanine transferase were similar in the exposed groups expect TAL3 compared to the control. However, the values obtained after 10 days withdrawal revealed that there were no significantly different (P>0.05) in amino alanine transferase, and aspartate aminotransferase and glucose among the treatments. These values have a similar trend with the value obtained before the experiment. This report was in agreement with the report of Cimanga *et al*., (2018) who reported in lower values in ALT and glucose of fish exposed to different concentration of *C. sanguinolenta.* There were no observable lesions in the control in the gills, liver and intestine while there was moderate diffuse laminar atrophy in the gills and centrilobular hepatocellular vascular generation in the liver at 0.002 ml/L concentration of *D. stramonium* as the concentration increases, there is an increase in severe changes in gills, liver and intestine among the treatments. At 0.2 ml/L concentration there recorded the severe lamilae

atrophy in the gills, random hepatocellular degeneration and melono – macrophages in the hepatopancreas in the liver and necrotizing enteritis characterized by cubbed to fused villi and cellular infilterate in the intestine. This study was in agreement with the report of Olurin *et al.,* (2016) who observed no lesion in gill, liver and kidney of the fish in the control group while there were alterations in the cytoarchitecture of the gills, liver and kidney of *C. gariepinus* juveniles exposed to sub-lethal concentration of phostoxin.

Conclusion: The result of this investigation indicated that sub-lethal levels of *D. stramonium* leaves extract impacted on growth performance and blood indices of *O. niloticus* fingerlings. However, unchecked exposure of fish to *D. stramonium* leaves extract may affect the growth, survival, and availability of the fish as a source of protein to sustain livelihoods.

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