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EFFECTS OF CRUDE SEED EXTRACTS OF DESERT DATE (*Balanite aegyptiaca)* ON BLOOD AND BIOCHEMICAL PARAMETER OF CAT FISH (*Clarias gariepinus)* JUVENILES

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ABSTRACT
This study investigates the effects of *Balanites aegyptiaca* crude seed extract on juveniles of *Clarias gariepinus*, as it affects the blood and biochemical parameters. The fresh fruits of *Balanites aegyptiaca* were collected from Jos, Nigeria, and processed to obtain a fine powder for analysis. Juvenile *Clarias gariepinus*, averaging 3g in weight and 9cm in length, were acclimatized to laboratory conditions for one week before exposure to varying concentrations (0.05, 0.10, 0.15, 0.20, and 0.25 g/L) of the extract. Hematological results showed initial increases in RBC count and hemoglobin (HGB) at lower concentrations, with RBC peaking at 2.26±0.13 µL (0.05 g/L) and HGB at 189.00±1.00 g/L. Both parameters declined at higher concentrations, showing a non-linear pattern. WBC count exhibited a dose-dependent response, peaking at 97.00±1.30 µL at 0.10 g/L before declining at higher concentrations. Biochemical profiles showed dose-dependent changes in Aspartate Aminotransferase (AST), Alanine Aminotransferase (ALT), Alkaline Phosphatase (ALP), and Lactate Dehydrogenase (LDH). AST levels ranged from 17.50±0.50 µ/L at 0.05 g/L to 36.00±1.00 µ/L at 0.25 g/L, while ALT increased from 11.00±1.00 µ/L at 0.05 g/L to 26.00±1.00 µ/L at 0.25 g/L. ALP values were highest at 84.12±4.69 µ/L (0.25 g/L) and lowest at 54.67±4.46 µ/L (0.10 g/L). LDH increased with concentration, peaking at 418.75±6.85 µ/L (0.25 g/L). Overall, findings indicate that *Balanite aegyptiaca* extract can influence both biochemical parameters and fish health parameters.
Keywords: Biochemical Parameters, Blood indices, *Clarias gariepinus* Juveniles, Toxicant Concentrations, *Balanite aegyptiaca*

INTRODUCTION

Plants extracts have been used by man to collect fish in lentic and lotic waters. Indigenous tribes of South America have long used plants as piscicides (Power, Fuentes, & Harrison, 2008). According to Power *et al*. (2008) macerated material of plant origin is thrown into rivers, streams or shallow ponds, and then the fish which are stupefied float to the surface and can be collected. After analysing the phytochemicals,

the plant material revealed the presence of active compounds such as rotenoids and saponins (Power *et al*., 2008).

Anthropogenic activities have led to an alarming increase in the level of pollutants in aquatic ecosystems. The use of *Balanite egyptiaca* seed extracts by artisanal fishermen has led to a decline in the number of fish catch which causes ripple effects on the economy of the fishermen and the community in general. Like humans and other species of animals, fish have also been found to be polluted with a large number of toxicants such as chemicals, drugs, infections or pollutants/contaminants. The objective of this study is to evaluate the changes in haematological and biochemical parameters of *C. gariepinus* juveniles exposed to crude Seed extracts of *Balanite egyptiaca* compared with the control.

In some parts of North Eastern Nigeria, *B. aegyptiaca* stem is crushed and used by local fishermen to harvest fish. The crude Seed extracts of *Balanite aegyptiaca* is usually indiscriminately used in catching fishes which could be deleterious to aquatic inhabitants including fish. Toxic substances may have a direct effect on the fish immune system by reducing resistance to infectious disease and may also have indirect effects on the environment causing stress in fish or affecting the proliferation or survival of the organisms.

The effect is damaging not only to the individual species and populations, but also to the natural biological communities, and that this occurs when pollutants are discharged directly or indirectly into water bodies without adequate treatment to remove harmful constituents (Agrawal *et al*., 2010).

MATERIALS AND METHODS

Collection and Preparation of Experimental Toxicant (Desert Dates) and Collection Area

The fresh part (fruit) of the experimental plant was collected from Jos, Jos North Local Government Area of Plateau State North-central Nigeria. Plateau gets its name from the Jos Plateau, a mountainous Area in the North of the State with captivating rock formations. Bare rocks are scattered across the grasslands, which cover the Plateau. The Jos Plateau makes it the source of many River in northern Nigeria including Kaduna, Gongola, Hadejia and Yobe rivers. The State shares borders with Kaduna State to the North, Kaduna and Nasarawa States to the East, Benue to the South and Taraba State to the East. Plateau State is located between latitude $80^0$24’N and longitude $80^0$32’ and $100^0$38’ East. (Diala, 2022).

Preparation of Toxicant

The preparation of the toxicant was done according to the method of Audu *et al.* (2014): The *Balanites aegyptiaca* fruits was transported to Hydrobiology and Fisheries research Laboratory of University of Jos, Nigeria where the seed was removed and dried under shed and pound using pestle and mortar and

sieved with 0.5mm sieve into fine powder before taking the sample for phytochemicals and proximate analysis in the department of biochemistry University of Jos.

Collection and Acclimation of Experimental Fish (*Clarias gariepinus*) Juveniles

A total of 300 Live and healthy, mixed sex Juveniles of *C. gariepinus* (Plate 2) with mean weight of 3g and total length of 9cm were purchased for both acute and sub-acute from a Private Fish Farm in Zarmaganda, Jos South, Plateau State and transported to Hydrobiology and Fisheries Research laboratory of University of Jos, Nigeria. Fish were transferred into six (6) plastic tanks each with 10L dechlorinated tap water and were allowed to acclimatize to laboratory conditions for a period of one week. Fish were fed with commercial feed (Blue crown) twice daily. Three quarters of the water in the tank were siphoned out on daily basis to remove left over feed and faecal matter and replaced with fresh water. Mortality observed during acclimation period were replaced and allowed to stabilize to zero. Feeding stopped 24 hours prior to exposure to the bioassay media.

Haematological parameters of C. *gariepinus* Juveniles exposed to crude Seed extract of *Balanite aegyptiaca.*

Total count of RBC

Total red blood cells (TRBCs) were counted using an improved Neubaur haemocytometer. Blood was diluted at a ratio of 1:200 with Hayem's fluid. Erythrocytes were counted in the loaded haemocytometer chamber and total numbers were reported as 106 mm^{-3} (Maheswaran *et al.*, 2008).

Total count of WBC

Total white blood cells (WBC) were counted using an improved Neubaur haemocytometer. Blood was diluted at a ratio of 1:20 with Turk's diluting fluid and placed in haemocytometer. Four (4) large (1 sq mm) corner squares of the haemocytometer were counted under the microscope (Olympus) at 640 X. The total number of WBC was calculated in mm^3 $x10^3$(Maheswaran *et al.*,, 2008).

Estimation of Haemoglobin

The hemoglobin contents of blood were analyzed using Cyanmet hemoglobin referential methods of Darbkins Fluid as described by Sharma (2013). Briefly, twenty micro litre (20 µl) of blood was mixed with 5ml Darbkin's working solution. The absorbance was measured using a spectrophotometer at wavelength of a 540 nm. Hemoglobin contents were expressed as g/dL

Total erythrocyte count

The total erythrocyte count was also estimated using the approach of Sharma (2013). Briefly, blood was drawn from the caudal vein into EDTA bottle to prevent the blood cells from lysis and clotting. The blood

was diluted to 1:200, with RBC counting pipette. The mixture was shaken well to suspend the cells uniformly in the solution. Then the cells were counted using a haemocytometer as follows:

Number of RBC/mm^3 = N x 10000 (where, N= total number of red blood cells counted in 5 squares of the haemocytometer slide and 10,000 is the dilution factor) (Sharma, 2013).

Total leukocyte count

The total leukocuyte count was determined using the method reported by Sharma (2013). Briefly, the blood collected in the EDTA bottle was diluted up to 1:20 ratio with WBC diluting fluids using WBC counting pipette. The mixture was shaken well to suspend the cells uniformly in the solution. Then the cells were counted using a haemocytometer as follows:

Number of WBC/mm^3 = N x 50 (where, N = total number of white blood cells counted in 4 squares of the hemocytometer slide and 50 is dilution factor) Sharma (2013).

Packed Cell Volume (PCV) or Haemotocrit

The packed cell volume was estimated by adopting the method of Bull, *et al*. (2000) In brief, a pair of scissors was used to cut off the fin and the caudal peduncle and heparinized capillary tubes were placed to obtain blood by capillary action, up to ¾ filled. One end of the capillary tube was sealed with plaster seal and placed in the micro-haematocrit centrifuge (model (No)-Hawksley and sons Ltd., Country) and spinned for 5 minutes at 12,000 revolutions per (rpm). The result was read using the micro-haematocrit reader and expressed in percentage directly from the reader.

Biochemical analysis

The excised gills, and liver were rinsed in distilled water to remove traces of blood and the organs were macerated in normal saline, crushed with laboratory pestle and mortar (Devi *et al.* 2013), then centrifuged for 5 minutes at 1000 rpm to obtain supernatants which were transferred into non–heparinised tubes before storage in a refrigerator (0^oC) for biochemical analysis (Audu, *et al.,* 2020).

The activities of various enzymes; alkaline phosphatase (ALP), alanine aminotransferase (ALT), and aspartate aminotransferase (AST) in the liver and gills of C. *gariepinus* juveniles exposed to aqueous fruit extract of *B. aegyptiaca* were determined using colorimetric procedure described by Sanghavin and Jivani (2017). Briefly, one milliliter (1 ml) of the desired substrate is pipetted into a test tube, and placed in a water-bath at constant temperature (40^0C) for 10 min. Upon the addition of 0.2 ml. of serum, the contents were mixed and incubation for 60 minutes. The tube was removed from the water-bath. One milliliter of the 2,4-dinitrophenylhydrazine reagent was added immediately, thereby stopping the reaction. After the tube was permitted to stand at room temperature for a minimum of 20 min., 10 ml. of 0.4N sodium hydroxide was added. Then a rubber stopper was inserted and the contents were mixed by

inversion. At the end of 30 min., the optical density of the solution was measured at 505 m/L using water as the blank reference. Any filter in the range of 490 to 530 m/u may be used satisfactorily. While the specimens were incubating, a control for each serum was prepared. One ml. of the substrate, 0.2 ml. of serum, and 1 ml. of 2,4-dinitrophenylhydrazine reagent were mixed in a test tube. After a minimum of 20 min., 10 ml. of 0.4N sodium hydroxide was added, and the optical density of the solution was measured as described in the preceding paragraph. For convenience, the addition of the alkali may be performed along with the incubated samples (Sanghavin and Jivani, 2017).

Experimental Design

A total of 12 transparent circular plastic tanks of 16 liters capacity each with 10 liters of de-chlorinated tap water were used in a complete randomized design. Each test tank was replicated and designated as A1-C2, E1-B2, C1-A2, D1-F2, B1-E2 and F1-D2. Each tank contained ten (10) mixed sex juveniles of *C. gariepinus*. Fish were sorted into cohort and distributed in batches of ten fish/per experimental tank with the control.

Statistical analyses

Statistical analyses were performed using IBM SPSS (version 23) software. Data were analysed by One-Way Analysis of Variance (ANOVA). Treatment means were separated using Turkey's multiple comparisons test. The level of significance was determined at a $p=0.05$ level of probability and a $p<0.05$ value was considered statistically significant. Data were presented as Mean± SE (standard error).



Experimental set-up for the study.

RESULTS

Table 1 presents the mean haematological indices of *Clarias gariepinus* juveniles exposed to varying concentrations (0.00–0.25 g/L) of aqueous crude seed extract of *Balanites aegyptiaca*. White blood cell (WBC) count was significantly ($P < 0.05$) affected by the extract concentration, whereas lymphocyte (LYM), red blood cell (RBC), haemoglobin (HGB), haematocrit (HCT), mean corpuscular volume (MCV), mean corpuscular haemoglobin (MCH), mean corpuscular haemoglobin concentration (MCHC), platelet (PLT), mean platelet volume (MPV), and platelet distribution width (PDW) did not differ significantly ($P > 0.05$) among treatments. The WBC count increased markedly from 25.30 ± 1.20 µL in the control group to 89.00 ± 2.30 µL, 97.00 ± 1.30 µL, 90.34 ± 1.54 µL, and 88.90 ± 1.10 µL in fish exposed to 0.05, 0.10, 0.15, and 0.20 g/L, respectively, before declining to 66.10 ± 31.20 µL at 0.25 g/L.

Although not statistically significant ($P > 0.05$), lymphocyte percentage ranged from 17.56 ± 1.86% to 47.66 ± 24.65%, while RBC counts varied between 0.92 ± 0.44 µL and 2.26 ± 0.13 µL. Haemoglobin concentration ranged from 64.50 ± 0.05 g/L to 189.00 ± 1.00 g/L, whereas haematocrit values ranged from 13.14 ± 2.62% to 29.39 ± 1.66% across treatments. Similarly, erythrocyte indices (MCV, MCH, and MCHC) and platelet indices (PLT, MPV, and PDW) were not significantly influenced by increasing concentrations of the extract.

Table 1: Mean Haematological Indices of *C. gariepinus* Juveniles Exposed to different Concentrations of Aqueous Crude Seed Extract of *B. aegyptiaca*

Parameters	0.00(g/L)	0.05(g/L)	0.10(g/L)	0.15(g/L)	0.20(g/L)	0.25(g/L)	P-values
WBC (µL)	25.30±1.20	89.00±2.30	97.00±1.30	90.34±1.54	88.90±1.10	66.10±31.20	0.04
LYM (%)	23.52±1.27	17.56±1.86	47.66±24.65	19.31±0.10	20.81±0.74	23.67±5.30	0.42
RBC ((µL)	1.44±0.33	2.26±0.13	2.22±0.80	1.98±0.95	0.92±0.44	1.93±0.15	0.37
HGB (g/L)	86.00±8.00	189.00±1.0	160.50±12.5	148.00±99.0	64.50±0.05	119.00±32.00	0.41
HCT (%)	20.09±4.71	28.76±1.70	29.39±1.66	23.16±10.66	13.14±2.62	24.54±0.64	0.32
MCV (fL)	138.85±1.35	127.30±0.50	131.95±2.75	118.35±2.95	120.35±4.45	127.55±6.55	0.06
MCH (pg)	64.05±19.96	84.15±5.25	72.70±8.30	66.33±18.01	62.86±15.15	63.46±21.46	0.91
MCHC	462.5±148.	661.0±44.0	552.0±166.0	564.0±166.0	518.0±107.0	490.0±143.00	0.88

(g/L)	5	0					
PLT (µL)	36.50±3.50	141.50±27.5	90.00±7.00	170.50±27.5	169.50±39.5	221.50±139.5	0.43
MPV (fL)	12.33±0.09	11.93±0.30	12.13±0.36	11.38±1.67	10.37±0.05	11.00±1.14	0.61
PDW (%)	16.05±0.45	16.20±1.60	17.10v1.10	15.90±1.30	14.80±0.20	11.00±0.00	0.57

[a, b] Means in the same row not sharing superscript are significantly different at P<0.05.

SEM = Standard error mean; WBC=White blood cells; LYM= Lymphocytes; HGB= Haemaglobin; MCV= Mean corpuscular volume; MCH= Mean corpuscular haemoglobin; MCHC= Mean corpuscular haemaglobin concentrations; PLT= platelets; MPV= mean platelets volume; PDW= platelets distribution width

Table 2 presents the differential white blood cell counts of *Clarias gariepinus* juveniles exposed to graded sub-lethal concentrations of *Balanites aegyptiaca* seed extract. Significant differences ($P < 0.05$) were observed in lymphocyte, neutrophil, eosinophil, and monocyte percentages among the treatment groups, whereas basophil percentage was not significantly affected ($P > 0.05$). Lymphocyte percentage was highest ($P < 0.05$) in fish exposed to 0.05 g/L (73.00%) and progressively decreased with declining extract concentration, reaching the lowest value of 62.00% in the control group. Fish exposed to 0.00625 g/L and 0.00325 g/L recorded intermediate values of 71.00% and 70.00%, respectively. Neutrophil percentage exhibited an opposite trend, with the highest values (28.00%) recorded in fish exposed to 0.00325 g/L and the control treatment, while the lowest values (19.00%) occurred at 0.025 g/L. Fish exposed to 0.0125 g/L recorded an intermediate value of 22.00%. Eosinophil percentage varied significantly among treatments, with the highest value (10.00%) recorded at 0.0125 g/L, whereas fish exposed to 0.00325 g/L had no detectable eosinophils (0.00%). The remaining treatments ranged between 2.00% and 5.00%. Basophil percentage showed no significant differences ($P = 0.136$) among treatments. Basophils were absent in most treatment groups but reached 2.00% at 0.0125 g/L and 1.00% in the control group. Monocyte percentage differed significantly ($P < 0.05$), with the highest values (10.00% and 9.00%) recorded at 0.025 g/L and 0.0125 g/L, respectively. The lowest value (2.00%) was observed at 0.00325 g/L, while intermediate values were recorded in the remaining treatments.

Table 2: Differential white blood cell (WBC) counts of *C. gariepinus* exposed to sub-lethal concentration of *B. aegyptiaca*

Parameters	A	B	C	D	E	Control	SEM	P-value

	0.05	0.025	0.0125	0.00625	0.00325	0.00		
Lymphocyte %	73.00[a]	69.00[c]	66.00[d]	71.00[b]	70.00[bc]	62.00[e]	0.89	0.000
Neutrophil %	20.00[c]	19.00[c]	22.00[b]	20.00[c]	28.00[a]	28.00[a]	0.93	0.000
Eosinophil %)	2.00[c]	2.00[c]	10.00[a]	3.00[c]	0.00[d]	5.00[b]	0.8	0.000
Basophil %	0.00[b]	0.00[b]	2.00[a]	0.00[b]	0.00[b]	1.00[ab]	0.27	0.136
Monocyte%	5.00[c]	10.00[a]	9.00[a]	7.00[b]	2.00[d]	4.00[c]	0.71	0.000

[a, b] Means in the same row not sharing superscript are significantly different at P<0.05.

SEM = Standard error mean

The mean serum biochemical profiles of *Clarias gariepinus* juveniles exposed to acute concentrations of aqueous crude seed extract of *Balanites aegyptiaca* for 96 hours are presented in Table 3. The activities of aspartate aminotransferase (AST), alanine aminotransferase (ALT), alkaline phosphatase (ALP), and lactate dehydrogenase (LDH) differed significantly ($P < 0.05$) among the treatment groups. AST activity decreased slightly from 19.00 ± 2.00 U/L in the control to 17.50 ± 0.50 U/L at both 0.05 and 0.10 g/L before increasing progressively with increasing extract concentration to 21.50 ± 0.50 U/L, 28.00 ± 1.00 U/L, and 36.00 ± 1.00 U/L at 0.15, 0.20, and 0.25 g/L, respectively. Similarly, ALT activity showed a slight reduction at 0.05 g/L (11.00 ± 1.00 U/L) compared with the control (13.50 ± 0.50 U/L) but increased steadily with increasing extract concentration, reaching the highest value (26.00 ± 1.00 U/L) at 0.25 g/L. ALP activity declined from 67.06 ± 0.78 U/L in the control to 54.67 ± 4.46 U/L at 0.10 g/L, after which it increased progressively to 84.12 ± 4.69 U/L at the highest concentration (0.25 g/L). Lactate dehydrogenase (LDH) activity followed a similar pattern, decreasing from 270.05 ± 3.85 U/L in the control to 244.35 ± 0.85 U/L at 0.10 g/L before increasing markedly to 365.60 ± 15.30 U/L and 418.75 ± 6.85 U/L at 0.20 and 0.25 g/L, respectively.

Table 3: Mean Biochemical profiles of *C. gariepinus* Juveniles Exposed to Acute Concentrations of Aqueous Crude Seed Extract of *B. aegyptiaca* for 96 Hours

Conc (g/L)	AST (µ/L)	ALT (µ/L)	ALP (µ/L)	LDH (µ/L)
0.00	19.00±2.00	13.50±0.50	67.06±0.78	270.05±3.85
0.05	17.50±0.50	11.00±1.00	62.22±3.79	248.90±2.90
0.10	17.50±0.50	13.50±0.50	54.67±4.46	244.35±0.85

0.15	21.50±0.50	15.00±1.00	61.39±2.07	257.50±2.90
0.20	28.00±1.00	16.50±1.50	67.36±1.02	365.60±15.30
0.25	36.00±1.00	26.00±1.00	84.12±4.69	418.75±6.85
P-values	0.00	0.00	0.00	0.00

[a, b] Means in the same row not sharing superscript are significantly different at P<0.05.

SEM = Standard error mean; AST= Aspartate Aminotransferase; ALT= Alanine Aminotransferase; ALP= Alkaline Phosphatase; LDH= Lactate Dehydrogenase

The biochemical parameters of *Clarias gariepinus* juveniles exposed to sub-lethal concentrations of aqueous crude seed extract of *Balanites aegyptiaca* are presented in **Table 4**. Alanine aminotransferase (ALT), aspartate aminotransferase (AST), alkaline phosphatase (ALP), total protein, creatinine, urea, and calcium (Ca^{2+}) differed significantly ($P < 0.05$) among the treatment groups. ALT activity was highest ($P < 0.05$) in fish exposed to **0.00625 g/L** (83.00 IU/L), followed by **0.00325 g/L** (56.00 IU/L) and **0.025 g/L** (55.00 IU/L), whereas the control recorded the lowest value (33.00 IU/L). Fish exposed to **0.05 g/L** and **0.0125 g/L** had intermediate ALT values of **45.00 IU/L** and **47.00 IU/L**, respectively. AST activity also differed significantly among treatments, with the highest value (**202.00 IU/L**) recorded at **0.00625 g/L**, followed closely by **199.00 IU/L** at **0.025 g/L**. The lowest AST activity (**19.00 IU/L**) occurred at **0.00325 g/L**, while the control recorded **61.00 IU/L**. Alkaline phosphatase (ALP) activity was significantly highest at **0.0125 g/L** (77.00 IU/L), followed by **0.00625 g/L** (74.90 IU/L), whereas the lowest value (**58.90 IU/L**) was recorded in the control group. Total protein concentration ranged from **54.30 g/dL** in the control group to **85.40 g/dL** at **0.00625 g/L,** with all treatment groups exhibiting significantly higher values than the control. Creatinine concentration varied significantly among treatments. The highest value (**45.20 µmol/L**) was observed at **0.025 g/L**, while the lowest values (**15.10–15.60 µmol/L**) occurred at **0.00325 g/L** and **0.00625 g/L**, respectively. Urea concentration increased markedly with treatment, reaching the highest value (**45.40 mmol/L**) at **0.00625 g/L,** whereas the lowest value (**5.70 mmol/L**) occurred at **0.00325 g/L**. Calcium concentration also differed significantly ($P = 0.002$), with fish exposed to **0.025 g/L** recording the highest value (**6.70 mmol/L**) and those exposed to **0.00625 g/L** recording the lowest (**2.00 mmol/L**).

Table 4: Biochemical Parameters of *C. gariepinus* exposed to sub-lethal concentration of *B. aegyptiaca*

Parameters	A	B	C	D	E	Control	SEM	P-value

	0.05	0.025	0.0125	0.00625	0.00325	0.00		
Alt IU/L	45.00^{d}	55.00^{b}	47.00^{c}	83.00^{a}	56.00^{b}	33.00^{e}	3.73	0.000
Ast IU/L	89.00^{d}	199.00^{b}	132.00^{c}	202.00^{a}	19.00^{f}	61.00^{e}	16.48	0.000
Alp IU/L	65.30^{d}	68.00^{c}	77.00^{a}	74.90^{b}	65.00^{d}	58.9.00e	1.51	0.000
Total protein g/dl	67.40^{e}	78.00^{c}	80.50^{b}	85.40^{a}	72.00^{d}	54.30^{f}	2.47	0.000
Creatinine umol/L	19.90^{d}	45.20^{a}	23.00^{c}	15.60^{e}	15.10^{e}	32.70^{b}	2.6	0.000
Urea mmol/L	8.70^{c}	7.90^{c}	21.20^{b}	45.40^{a}	5.70^{d}	6.90c^{d}	3.43	0.000
Ca^{2}+	4.60^{b}	6.70^{a}	3.30^{bc}	2.00^{c}	3.00^{bc}	4.10^{b}	0.41	0.002

[a, b] Means in the same row not sharing superscript are significantly different at P<0.05.

SEM = Standard error mean

DISCUSSION

Haematological parameters are used in medical analysis of fish body function which is determined by the effect of the surrounding water (Ojutiku *et al.,* 2013), thus reveal the level of damage in fish. Fish exposed to toxicants can exhibit a variety of physiological responses, including blood disturbances (Osman *et al.,* 2010; Thangam, *et al.,* 2014). The anisocytosis of red blood cells in this study indicates stress mediated release of blood cells by haematopoietic tissues (Al-Zaidan, 2017) and poikilocytosis implies destruction of blood cells due to the effects of the toxicant on the exposed fish (Ajima *et al.,* 2014) while normocytic and normochromic cells indicate that concentrations of the plant extract had no effect on the blood cells. Decrease in WBC and LYM recorded in this study opposed the findings of Adewoye (2010), which reported an increase in WBC and LYM due to the fish's attempt to fight against the toxicant by producing more WBC and LYM to recover their health condition. The decrease in RBC as concentrations of the test material increase in this study is similar to the findings of Ajima *et al.* (2017) that RBC decreased after chronically treating *C. gariepinus* with urea fertilizer. It also agrees with the findings of Suely *et al.* (2015) after exposing *Heteropneustes fossilis* to concentrations of *Terminalia arjuna*. A significant decrease in RBC and HGB level after exposure to pesticides and other toxicants were reported by several investigators (Adewoye, 2010). The present study also corroborates the findings of Adedeyi and Odo (2017) who reported decline in HCT following elevation in concentration of selenium on *C. gariepinus*. Similar reduction had been reported by Thangam, *et al.* (2014) after exposing *C. carpio* to copper. In contrast with results of the present study, Riaz-ul-Haq *et al.* (2018) have previously observed an increase in HCT (or PCV) of freshwater fish *Channa punctatus* exposed to endosulphan pesticide.

AST, ALT, and LDH are major stress indicators providing specific information about organ dysfunction (Ishikawa *et al.,* 2007). High AST levels indicate greater energy demand associated with cell synthesis activities (Svačina *et al.,* 2016), while increased LDH levels, influenced by exercise, may contribute to mortality during fish capture and transport (Okey *et al.,* 2022). Aminotransferases like AST and ALT play a crucial role in monitoring fish health status and diagnosing damages caused by xenobiotics to various tissues, they also function in amino acid utilization for oxidation activities and gluconeogenesis, serving as links between carbohydrate and protein metabolism under different physiological, pathological, and stress-induced environmental conditions (Daniel, 2009). Increased transaminase activities, including ALT and AST, observed in all acute toxicity scenarios in the present study, suggest amplified transamination processes, aiding amino acid input into the tricarboxylic acid (TCA) cycle to cope with energy crises during stress. Alanine aminotransferase is also known for mobilizing L-amino acids for gluconeogenesis and functioning as a link between carbohydrate and protein metabolism under varied physiological, pathological, and induced environmental conditions (Adewale *et al.,* 2017). Elevated ALT levels indicate efficient amino acid utilization for metabolic purposes (Adewale *et al.,* 2017). Stress-induced elevation of the transamination pathway likely contributed to increased transamination activities in this study (Congleton, 2006), as observed in rainbow trout (*Oncorhynchus mykiss*) treated with MS-222.

CONCLUSION AND RECOMMENDATION

Conclusion

The findings of this study indicated that the crude seed extract of *Balanite aegyptiaca* contains a variety of phytochemical components with distinct effects on fish physiology. These phytochemicals adversely impact water quality and fish organs, leading to changes in behavior and increased mortality among fish populations. Consequently, the crude seed extract of *B. aegyptiaca* could be considered an aquatic pollutant. Moreover, the extract has to be cautiously applied in fish water environment. The observed physiological alterations in haematological and biochemical parameters in *C. gariepinus* juveniles exposed to the crude seed extract of *B. aegyptiaca* likely contributed to a decline in overall fish health and subsequent mortality.

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