

## Full-Length Research Paper

# Effect of Spilled Engine Oil on the kidney of Catfish (*Clarias gariepinus*) and the physico-chemical Parameters of Water

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**ABSTRACT:** Fish is sensitive to devastating consequences of oil contamination. Histological changes in *Clarias gariepinus* fingerlings before death under exposure to different concentrations of spilled engine oil was carried out. The study examined the effect of spilled engine oil, on the physico-chemical parameters in water, determined the effect of spilled engine oil, on the kidney of catfish (*Clarias gariepinus*) and evaluation of the behavioral responses of fish at different concentration of spilled engine oil. Three hundred and sixty *Clarias gariepinus* fingerlings of average weight  $3.50 \pm 0.01$ g and length  $12 \pm 0.000$ m was exposed to different concentrations of (0.01, 0.03 and 0.05 ml/L) of oil with fifteen (15) fishes per tank in two replicates. The median lethal concentration (LC<sub>50</sub>) value of spilled engine oil on *Clarias gariepinus* fingerlings was found to be 0.05ml/L of water. Each treatment was subjected to 96 Hour bioassay test to

determine its acute toxicity in a completely randomized design experiment. Changes in water quality and fish mortality in all treatments were monitored daily in addition, behavioural and histological responses were also investigated. There was a significant difference ( $p < 0.05$ ) in values of temperature, electrical conductivity, salinity, pH and Dissolved Oxygen among all the treatments before and after the experiment. Mortality rate was highest (90%) in fish exposed to 0.05ml/L spilled engine oil water at 96hrs exposure. As concentration and time of exposure increased, the fish showed restlessness, rapid opercula movement, erratic swimming, discoloration and loss of reflex before death.

**Keywords:** *Clarias gariepinus*, kidney, physico-chemical parameters, spilled engine oil

## INTRODUCTION

Fish is highly consumed all over the world, especially for its valuable sources of high protein contents and also serves as source of vitamins. Fish can form very nutritious components of most diet and is rich in most of vitamins consumed, containing a good number of mineral elements as well as essential amino acid obtained from proteins. Fish are excellent sentinels of environmental health as they are sensitive to wide range of xenobiotic chemicals. The presence of metal pollutants in fresh water system posed great danger to the marine organism and fish is known to posed great ability to accumulate heavy metals in their muscles, and since the same is very essential in human diet, it is necessary therefore to carefully examined them for toxic metal contamination before being supplied for human consumption (Adeniyi and Yusuf, 2007;

Ekeanyanwu *et al.*, 2010). Catfish (*Clarias gariepinus*) was selected because it is an economically important group of the tropical inland water and is very important resources of the aquatic system of tropical Africa. To this end, catfish obtained from two ponds and two rivers were deployed for chemical analysis of mineral contents and proximate analysis such as percent of protein contents, moisture contents, ash contents and carbohydrate content.

### Catfish (*Clarias gariepinus*)

Catfish *Clarias gariepinus* of the family *clariidae* is the most common Nigerian fresh water fish species and is prominent in aquaculture practice. They are easily cultured with large economic gains because of their air-

breathing and hardy nature, suitable reproductive strategy, nutritional efficiency and attainment of large size in a short time (Fagbenro, 1993). Furthermore, the sharp tooth catfish (*Clarias gariepinus*) is one of the most important individual's species in traditional freshwater fisheries in Africa. It is widely attributed in Africa, where it occurs in almost any freshwater habitat but floodplains, large sluggish rivers, lakes and dams. The fish is omnivorous, feeding on fishes, birds, frogs, small mammal's reptiles, snails, crabs and other invertebrates. It is also capable of feeding on seeds and fruits (Fagbenro, 1993).

### The Kidney

The kidney is paired organs located in the body cavity either side of the backbone, kidney filters out wastes from the blood and the passes them out of the body. The kidney also plays a major role in regulating the water and salt content of the fish's body. This is partially how some fish are able to survive in saltwater while others survive better in fresh water (Mark, 2010). Seawater contains more dissolved salt than bony fish. This mean that marine and freshwater fishes have different problems with regard to regulating the concentration of salt within their water bodies (2010).

### Pond water and its physico-chemical characters as pollution indicator,

According to Shaw *et al.* (1991), the quality of water is usually determined by its physico-chemical characteristics and it is a well-established fact that domestic sewage and industrial effluent discharged into natural water result in deterioration of water quality and cultural eutrophication. Tiwana, (1992) cited some important sources of water pollution which includes mass bathing, disposal of dead bodies, rural and urban waste matters, agricultural run-off and solid waste disposal. Furthermore, the physico-chemical characteristics of water bodies are altered due to heavy discharge of effluents and sewages. Correia *et al.* (1994) opined that the chemical pollutants, which are present in the waste water, arise both from the raw materials and as additives used to produce the finished product of pollutant in water. Hence, Kaushik and Saksena (1995) stated that the physico-chemical parameters are useful in detecting effects of pollution on the water quality but changes in the trophic conditions of water are reflected in the biotic community-structure including species pattern, distribution and diversity. Ademoroti (1996) stated that the effluents contain high amounts of dissolved salts from domestic sewage can increase the salinity of the receiving water, which may result in adverse ecological effects on aquatic biota. This according to Peirce *et al.*

(1997) cited that the most important measure of water quality is the dissolved oxygen. The low level of DO recorded could result in the non-maintenance of conditions favorable to the aerobic organisms. This could lead to anaerobic organisms taking over with the resultant creation of conditions making the water body uninhabitable to fish gills-breathing aquatic organisms. Aquatic pollution by natural petroleum products nowadays is one of the most threatened issues for the aquatic organisms particularly to the fish occupying the surface layer (Khan *et al.*, 1999).

According to Karr and Benke (2000) analysis of the physic-chemical characteristics of water samples generally provides only a picture of environmental conditions at the moment samples are taken. It provides chemical quality but do not necessarily reflect the ecological state of the system. This was evidence in Kamal and Khalid (2012) Investigate on "Deltamethrin-induced oxidative stress and biochemical changes in tissues and blood of catfish" deltamethrin is a class of insecticides being used as a substitute for organochlorines and organophosphates in pest-control programs because of their low environmental persistence and toxicity. Their study was aimed to investigate the impact of commonly used pesticides (deltamethrin) on the blood and tissue oxidative stress level in catfish (*Clarias gariepinus*). The catfish were divided into three groups, 1st control group include 20 fishes divided into two tanks each one contain 10 fishes, 2nd deltamethrin group, where Fishes were exposed to deltamethrin in a concentration (0.75µg/l) and 3<sup>rd</sup> Vitamin E group, Fish exposed to deltamethrin and vitamin E at a dose of 12µg/l for successive 4 days (Kamal and Khalid, 2012).

The effect of deltamethrin on renal function tests were observed, urea levels were significantly elevated in deltamethrin exposed group in comparison with control group, this elevation may be due to correlation between urea and increased protein catabolism or from more efficient conversion of ammonia to urea as a result of increased synthesis of enzyme involved in urea production. Urea and creatinine levels increased which may result either from increased breakdown of tissue or dietary or impaired excretion or increased synthesis or decreased urinary clearance by the kidney or decreased degradation of these compounds (Kamal and Khalid, 2012). Their data suggest that polluted fish adept glomerular dysfunction rather than tubular insufficiency as blood levels of urea and creatinine depend largely on glomerular function. In consistent with these explanations of decreased total protein level with deltamethrin administration, urea is the end product of protein catabolism in mammals but in fish ammonia is the end products of protein. So the marked increase in blood urea nitrogen could be attributed to impaired excretion of urea through kidney and this explanation is supported by increasing blood creatinine level which is more sensitive and specific indicator of impaired kidney function

(Kamal and Khalid, 2012).

Kori (2008) also studied "The effects of sub lethal concentrations of potassium on nitrogenous waste products of African catfish". Changes in the nitrogenous waste products of the fish (Kori, 2008) subjected to sub lethal concentrations (2.0, 6.0, and 10.0mg/L) of potassium permanganate over a period of 192hrs were studied in a semi static (renewal) system. The nitrogenous waste metabolism was reflected in the changes in total plasma bilirubin, plasma uric acid, plasma creatinine and plasma urea. Increased creatinine concentration in the blood suggest the inability of the kidney to excrete these products, a manifestation of nephritic damage, decreased plasma urea may be attributable to the depleted protein levels. The effects of potassium on the levels of creatinine were elevated while urea was depleted during their research. Plasma creatinine levels in the treated fish showed a seemingly dose and time-dependent trend with the maximum elevation percentage (40.10) being recorded in 10mg/l KMnO<sub>4</sub> exposed fishes at 192 hrs.

In a research work by Adekunle (2010) on the "Potential nephrotoxicity in African mud catfish following exposure to compost derived humic acid" influence of compost-derived humic acid (HA) on nephrotoxicity in juvenile African mud catfish was evaluated in static water culture. Fish samples were exposed to different HA concentrations (0, 100, 250, 500 and 1000mg L<sup>-1</sup>) for 45 days at 5 samples per aquarium. Renal functions were assessed spectro photo metrically via levels of creatinine by Jaffe method and urea by Nessee realization method. The results revealed that the mean value of urea in the exposed group (I) at each HA concentration was lower than the value found in I relative to II: Significant ( $p < 0.05$ ) variations for I and II were obtained, relative to increasing HA concentrations, decreasing albumin (0.84 to 0.43gdL), urea (5.21 to 1.95mgdL) and increasing creatinine (0.20 to 1.53mgdL) were recorded. The objective of the study was to assess the nephrotoxicity potential of humic acid isolated from composted wastes (organic fertilizer) of Nigeria origin to African mud catfish (*C.gariepinus*) grown in static water culture using the three biomarkers, serum creatinine, albumin, and urea. The creatinine level in the control group was  $0.36 \pm 0.09$  mgdL<sup>-1</sup> and ranged from 0.26 to 1.53mgdL<sup>-1</sup> in the test groups. Positive correlation was obtained for creatinine and HA concentrations ( $+0.704$ ;  $p < 0.10$ ) indicating rising blood levels of creatinine with increasing HA concentration in water. The urea level in the control was  $5.61 \pm 0.07$  mgdL<sup>-1</sup> in the test groups. Correlations gave negative coefficients for urea versus HA concentration for urea versus HA concentration ( $-0.568$ ;  $p > 0.10$ ) indicating a decrease trend with increasing HA concentration in water. Various scholars and researchers like Ogundiran *et al.*, (2009), George *et al.*, (2014), Akinsorotan (2014) and Isyaku and Solomon (2016). All opined that the mortality increased with

increase in concentration of Dizensate glyphosate on catfish fingerlings and time of exposure. The physico-chemical parameters also showed a slight increase as the concentration increased. It could be concluded that Dizensate glyphosate, an aquatic herbicide, has harmful effects on the physiology, biochemistry and histological structure of fish which in turn affects the growth rate.

### Objectives of the study

The objectives of this study are to investigate the following:

Evaluate the effect of oil spillage, on the physico-chemical parameters in water.

Determine the effect of oil spillage, on the kidney of catfish (*Clarias gariepinus*).

Examine the behavioral responses of fish at different concentration of oil spillage.

### Research questions

What is the effect of oil spillage, on the physico-chemical parameters of water?

How does oil spillage affect the kidney of catfish (*Clarias gariepinus*)?

To what extent does oil spillage, influences the behavioral responses of fish at different concentration in water affect fish growth?

## METHODOLOGY

### Study area

Study area is Keffi Local Government in Nasarawa state. It has an area of 138km and a population of 92,665 at 2006 census; resources used for the study were obtained from Nasarawa State University. The toxicological analysis was carried out in the Faculty of Agricultural Sciences, Nasarawa State University Lafia Campus. This study was carried out with the use of 12 plastics tanks in Keffi, Nasarawa state. Resources used for the study were obtained from Nasarawa State University located in Keffi.

### Acclimatization of fish

The fish were held in 42.5cm by 28cm by 26cm, plastic tank containing non chlorinated water. The fish were allowed to acclimatize for more than one week under laboratory conditions to allow fishes adapt to experimental conditions ( $27 \pm 2^\circ\text{C}$ ). The period of acclimatization was extended beyond one week to ascertain the condition of the fish. Fish were inspected for disease conditions and general fitness. Fish were fed

during the period of acclimatization and the water was changed every three days in order to remove faecal and unconsumed feeds.

### Experimental fishes

Apparently five hundred and forty fishes of *Clarias gariepinus* fingerlings of mean weight 3.50g were obtained from Nasarawa State University Entrepreneur center Keffi and transported in an aerated container to the place where the experiment is going to take place at Dadin Kowa, Keffi, the fishes were acclimatize for at least 48h.

During the period, the water was aerated with air pump (aerator). Fish fed twice daily with aqual feeds (Ingredient composition - fish meal, wheat flour, soybean meal, corn meal, yeast, vitamins and mineral salt; Proximate analysis - crude protein mineral; 40%, crude fat mineral; 4%, crude fibers, maximum; 5% and moisture, maximum; 10%). Unconsumed feed and fecal wastes were removed and water replenished three times weekly as recommended by Oyelese and Faturoti (1995). Water quality of the test and reference solutions fish were monitored throughout the duration of the experiment.

### Range finding test

The range finding test was conducted for accurate LC<sub>50</sub> (Median lethal concentration that causes 90% mortality of exposed fish). Three lethal concentrations of 0.01ml/L, 0.03ml/L and 0.05ml/L were prepared by adding determined amount of toxicant to the tanks, Fifteen fishes were randomly placed in each tank and exposure lasted for 96 hours.

### Experimental set up

Fish were stocked in twelve rectangular tanks of size 45.5 x 28 x 26cm, each had 15 fishes replicated twice in three concentrations (0.01ml/L, 0.03ml/L and 0.05ml/L) including the control. After determining the LC<sub>50</sub>, for the Concentrations of 0.01ml/L, 0.03ml/L and 0.05ml/L toxicant and the control 0ml/L were used for the experiment. Water quality variables such as Temperature, pH, Electrical conductivity, Alkalinity, Hardness, Atmospheric temperature and Dissolved Oxygen (DO) were monitored for each concentration. Behavioural responses and mortality rate after 96 hours of exposure were determined (Table 1).

### Preparation of test medium

Twelve plastic tanks were used as test containers. The water soluble fraction of oil spillage was prepared by mixing 1L of oil obtained from a mechanic work shop and dilute with 4L of water in accordance to Esenowo and Ugwumba, (2010). The mixture was stirred with a stick for

48h. It was made to stand for 12h before it was poured into a separating funnel and allowed to stand for 6h. The lower layer of the water was decanted into the plastic tanks (Afolabi *et al.*, 1985). It was repeated until sufficient quantity of water soluble fraction was obtained.

### Exposure of test organism

The water soluble fraction was made into three concentrations (0.01, 0.03 and 0.05ml, with two replicates each) for oil spillage. Dilution was made with aerated water. Fifteen fingerlings were exposed each to three concentrations of toxicants in 25L capacity plastic containers. Feeding was discontinued for 24 hrs before the start of the experiment (Reish and Oshida, 1987). The volume of water to the weight of fishes was calculated in accordance with Reish and Oshida (1987) standard of 12g of fish to 5L. Catfish fingerlings were observed for 96hrs and any behavioral changes and mortality rate was recorded.

### Physico- chemical parameters

The records of Temperature, pH, Dissolved oxygen, alkalinity, Hardness and Electrical conductivity were taken before and during the exposure period. Temperature, pH and electrical conductivity were measured using a T<sup>o</sup>/pH/ Electrical Conductivity (EC) meter model HI 98129. The Dissolved oxygen was measured using a Dissolved oxygen meter Model 9150.

### Process of preparing stock solution

Fish were fed twice daily with 40% crude protein diet at 1% biomass daily (morning and evening). A range finding text (trial test) was carried out using the toxicant (oil spillage) three concentration were prepared from the stock solution (250ml/L, 1675ml and 40g/ml) respectively in accordance to Grinshaw, (1978) The test trial concentrations were 0.01ml 0.03ml and 0.05ml in two replicate fifteen fishes were exposed to each concentration of toxicant (spilled engine oil).

Introduction of the toxicant (spilled engine oil) was done with the aid of pipette and suckers. The water was changed every three days and the plastic tank was washed with a piece of foam. The test solution was renewed immediately as in the acclimation period of ninety-six (96) hours. Purpose of test was to determine the range of concentration to be used for the definitive (main) test. The sub-lethal concentration for the definitive test was done based on the Range finding test. The concentration were prepared by taken 1%, 2% and 5% using serial dilution from stock solution of toxicant (oil spillage) three concentration were prepared from the stock solution (250ml/L, 1675ml and 40g/ml) and making

**Table 1:** Effect of oil spillage on growth of catfish (*C. gariepinus*).

	CONC(ML)WI	CONTROL		Oil Spillage	
		W(g)	L(cm)	W(g)	L(cm)
NOV	0.01	3.5	12	3.5	12
	0.03	3.5	12	3.5	12
	0.05	3.5	12	3.5	12
DEC.	0.01	10.5	25	5.5	23
	0.03	12.03	25.05	8.55	22.01
	0.05	10.33	25.05	5.33	18.06
JAN.	0.01	14.55	29	10.33	18.66
	0.03	16.55	27	10.2	20.22
	0.05	16.55	25.05	9.59	16
FEB.	0.01	19.66	28.56	12.05	24.61
	0.03	20	29.05	10.56	20.01
	0.05	20	29	10.01	20
MAR	0.01	23	30	11.59	23.66
	0.03	21	34.5	11	21.8
	0.05	22	34.56	10.45	20.05
APR	0.01	25.55	33.98	11.72	23.01
	0.03	25.77	34.98	11.05	22.07
	0.05	26.71	35.45	10.55	20.02

it up to 25 litre with well water in the plastic tank to make 0.01ml 0.03ml and 0.05ml .

## Histopathology

### Kidney

Histopathological section through the Control kidney transverse section through the kidney shows normal tissues where the blood cells are created, normal tissues and cells. The organ (kidney) was removed and prepared for histopathological examination. The kidneys were fixed in bouin's fluid for 24 hours, washed with 70 percent ethanol and dehydrated through a graded series of ethanol (Kelly, 1979). They were embedded in paraffin, sectioned at 4-5um thickness stained with haematoxylin and eosin and examined using light microscope and photomicrography (Keneko, 1989).

### Statistical analysis

The statistical analysis used in this research are; method of least significant difference (LSD) using the SPSS statistical package to analyzed the variables of the physico-chemical parameters. Duncan Multiple Range Test was used to assess the toxicity effect at three levels of concentrations (0.01, 0.03 and 0.05ml/L) of the toxicant (oil spillage) on weight and length of experimental fish. T-test was used to determine if a water toxicity level has significant difference on the weight and length of the fish (*C.gariepinus*). Scientific package for social sciences (SPSS) Inc. Chicago, USA, was

employed to calculate the significance of the differences between control and experimental means and within various treatments P values of 0.05 or less was considered statistically significant.

## RESULTS AND DISCUSSION

### Effect of oil spillage on growth of catfish (*C. gariepinus*)

Table 1 shows effect of oil spillage on the growth of catfish from the month of November to April, were the weight and length of fish is taken at different concentration level.

### Behavioral pattern and morphological changes of *Clarias gariepinus* fingerlings exposed to toxicants

Table 2 shows behavioural pattern and morphological changes of fish exposed to different concentration of toxicant and this behavioural pattern and morphological includes, Erratic swimming, Discolouration, Loss reflex Moulting, Air gapping and Haemorrhage

### Mean values of physico-chemical parameter by oil spillage

Temperature of the control tank ranged between  $26.03 \pm 0.02$  to  $32.6 \pm 0.5(^{\circ}\text{C})$ , Temperature was high in March and April ranging from  $31 \pm 0.04$  to  $32 \pm 0.0(^{\circ}\text{C})$ ,  $p^{\text{H}}$  of  $7.5 \pm 0.00$  and  $7.6 \pm 0.01$ , while Dissolved oxygen was  $4.50 \pm$

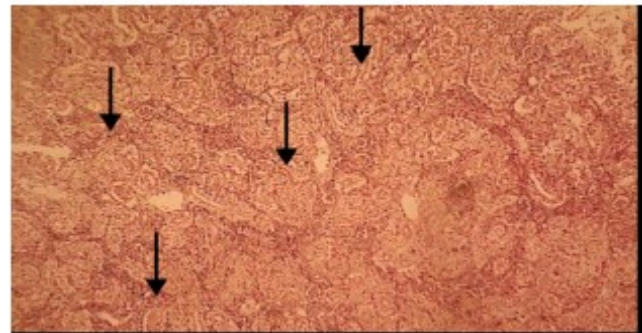
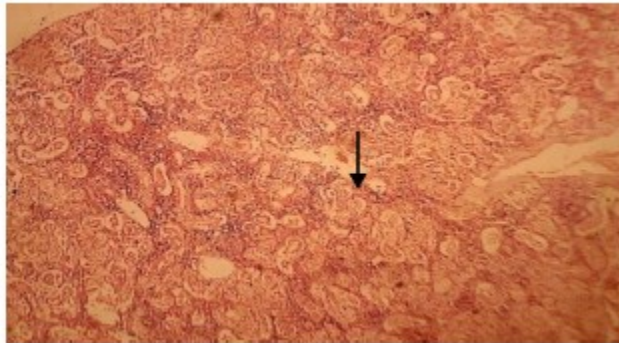
**Table 2:** Behavioral pattern and morphological changes of *Clarias gariepinus* fingerlings exposed to toxicant (oil spillage).

Behavioural patterns	12hrs	24hrs	48hrs	72hrs	96hrs
Erratic swimming	-xxx	-x--	-xxx	-xxx	-xxx
Discolouration	-xx-	-xxx	---x	-xxx	-xx-
Loss reflex	-x--	-xx-	-xxx	-xxx	-x--
Moulting	-xxx	-xx-	-xxx	-xxx	-xxx
Air gapping	-xx-	--xx	-xxx	-xx-	--xx
Haemorrhage	-xxx	-xxx	-xx-	-xx-	-xx-

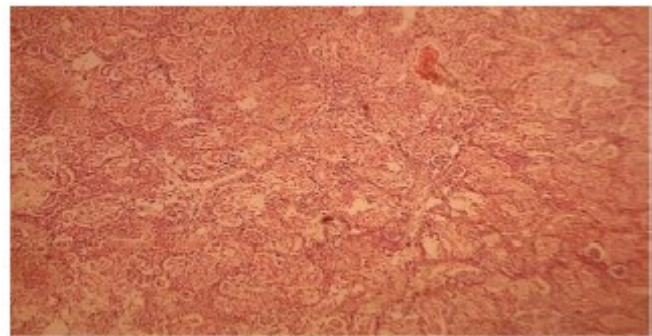
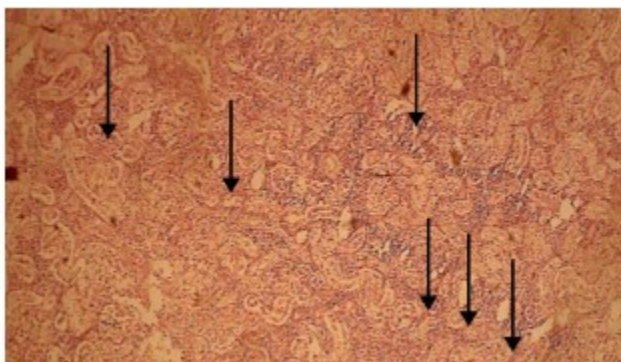
Key; (x) present; (-) not present

**Table 3:** Effect of oil spillage on physico-chemical parameters.

Physico-chemical parameters	NOV.	DEC.	JAN.	FEB.	MAR.	APR.
	Mean/SD	Mean/SD	Mean/SD	Mean/SD	Mean/SD	Mean/SD
TEMP.(°C)	26.22±0.09	23.41±0.09	25.22±0.17	26.22±0.15	31.55±0.03	29.50±0.04
P <sup>H</sup>	6.78±0.26	6.09±0.15	6.71±0.09	6.62±0.09	6.49±0.06	6.69±0.02
DO(mg/l)	4.53±0.09	5.77±0.08	3.33±0.18	4.37±0.22	4.34±0.12	4.91±0.02
EC(µs/cm)	320±0.00	319.66±0.06	321.60±0.00	409.12±0.02	405.28±0.01	411.15±0.02
ALK(PPM)	12.26±0.02	14.73±0.15	14.60±0.06	21.81±0.33	52.89±0.11	35.72±0.47
HD(ppm)	25.00±0.20	37.00±0.07	48.33±0.16	43.67±0.47	57.00±0.04	57.00±0.05
AT(0c)	28.33±0.05	27.33±0.08	27.00±0.07	28.00±0.07	30.33±0.07	29.33±0.08



**Plata 1:** The photomicrograph of *clarias gariepinus* fingerlings, kidney in control tank (0ml/L) showing no pathological lesion. (x4x6.7). **Plata 2:** The photomicrograph of *Clarias gariepinus* fingerlings, kidney exposed to 0.01ml/L of oil spillage, showing surface of Hypertropy but no visible changes.



**Plata 3:** The photomicrograph of *Clarias gariepinus* fingerling kidney exposed to 0.03ml/L of oil spillage, showing degeneration in the renal cells. **Plata 4:** The photomicrograph of *Clarias gariepinus* fingerlings, Kidney exposed to 0.05ml/L of oil spillage, showing hydropic infiltration and degeneration in the renal cell.

0.04 to 5.00 ± 0.09 which led to high mortality of the fishes.

Temperature for oil spillage range between 23.41±0.13 to 31.7±0.03 this shows that as toxicity increases,

temperature and other physico-chemical parameter decreases, the effect of change in temperature on the organism causes reduction in feed intake of catfish. Therefore these led to effect on growth of catfish.

Dissolved oxygen was between  $3.54 \pm 0.10$  to  $5.77 \pm 0.03$  which leads to gasping for air, Oil does not allow easy penetration of air which leads to respiratory distress and this led to death of some organism that could not withstand the stress.  $P^H$  for oil spillage range between  $6.05 \pm 0.17$  to  $10 \pm 0.03$ . It was noticed that the organisms were stressed (Table 3). The observed increasing state of inactivity with both increasing concentrations and exposure period agree with the report of Ayoola (2008). Water quality parameters had little variation, physico-chemical parameter measured seemed to be within optimum range for fish culture as reported.

### Histological examinations

Results of histological examinations are shown in Plates 1 to 4. Kidney histology of *Clarias gariepinus* exposed to different concentrations of oil spillage in (Plates 1-4). Control treatment (0ml/L) has no pathological lesion (Plate 1). Kidney of fish in (0.01ml/L) also showed no visible changes (plate 2). There was degeneration in renal cells of fish in (0.03ml/L), while hydropic degeneration in renal cell was observed in fish treated with 0.05ml/L of water containing oil spillage.

### Conclusion

This study has revealed that, exposure of *C. gariepinus* fingerlings to concentrations of oil spillage effluents is enough to induce various toxicological effects in terms of histological degradations in gill, liver, kidney and other organ structures. The damage in the kidney of test fish was found to be concentration and exposure period dependent. The results of the present study have contributed knowledge on the toxicity effect of chemicals to fish organs. The morphological and physical changes in the kidney recorded in this study were erratic swimming, stagnant growth, anorexia, loss of reflex and discoloration. It was observed in *Clarias gariepinus* fingerlings after exposure to oil spillage shows brief exposure to toxicants for about 96 hours produced irreversible changes in the organ this was also notes in Ferdandez and Mazon (2003) investigation on toxicants effect on catfish. This suggests both inland and coastal waters are vulnerable to oil-based industrial pollution. Some fish exposed to toxicants survived to the end of the exposure period, indicating that these toxicants impact the internal metabolism of the fish and, because they are non-degradable, accumulate in the tissues of the fish, eventually reaching humans through the food chain. As a result, human ingestion of infected fish poses a risk of inherent toxicity.

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