ASSESSMENT OF TOXICOLOGICAL IMPACT OF LIGHT CRUDE OIL ON *CLARIAS GARIEPINUS* (Burchell, 1822) FINGERLINGS.

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ABSTRACT

Oil spillage is of critical concern in Nigeria because of the problems of pollution and associated disturbances .It is therefore necessary to study the effects of oil pollution on fish using *Clarias gariepinus,* a widely cultured fish species in the tropics.

A short –term (96-hour), static bioassay was carried out to determine the toxicity of crude oil (Qua Iboe light) on *C .gariepinus* fingerlings. The bioassay involved five treatments viz: 0, 25, 50, 75 and 100µL/L

The water and fish samples were analyzed for total hydrocarbon content (THC) and heavy metals at the end of the experiment. There were significant differences ($P<0.05$) among the five treatments. The LC_{50} (concentration at which 50 percent of the test population died) was 1.58µL/L.Lead, iron, cadmium, and chromium were not detected in the water contaminated with the crude oil sample.

Keywords: toxicity, fingerlings, light crude oil.

INTRODUCTION

Crude oil is toxic to most species of plants and animals and when spilled, contaminates the environment .The formation of a film of oil on a water body disturbs natural aeration, causing death of organisms trapped underneath and fish may imbibe unpalatable or off flavours referred to as 'tainting'.

Crude oil contains compounds toxic to aquatic organisms. The high molecular weight, multi-ring compounds of petroleum like benzo(a)pyrenes and benzanthracenes are carcinogenic and mutagenic (Manahan,1992 , Serrazanetti *et al* ,1995, and Canova, *et al,* 1998). Spilled oil undergoes spreading, emulsification, dissolution, evaporation, oxidation of hydrocarbons, dispersion of water-soluble fractions, sedimentation, formation of tar balls, adsorption and biodegradation.

Bioassays are used to determine the toxicity of chemical substances and to indicate which organisms are the most sensitive to such chemicals .These data are used to rank chemicals, determine water quality criteria and set standards for effluent discharges. Standard procedures for conducting acute toxicity tests have been described (Finney, 1971; Sprague, 1971; Litchfield and Wilcoxon, 1949; Ward and Parrish, 1982; Reish and Oshida, 1987 and American Public Health Association, 1989).

According to Forbes (1994), laboratory tests and analyses form an aspect of the procedures used when the interactions between living organisms and their environments are studied. These tests are conducted to warn against the deleterious effects on target organisms when chemicals or pollutants are released into the environment. These tests are also done to comply with legislative requirements for the approval of new drugs or chemicals and act as the link between the dose (concentration) and the . effects (response).

The objective of this study is to carry out a short- term ,static bioassay using a light crude oil on *Clarias gariepinus* fingerlings .*C. gariepinus* is in high demand in Nigeria and highly tolerant to stress conditions.

MATERIALS AND METHODS

Acute toxicity tests were conducted using a light crude oil as toxicant and *C. gariepinus* (mean weight 10.5 g) as test organism in 13- litre glass aquaria. The experiment was carried out in the Laboratory of the Department of Wildlife and Fisheries Management, University of Ibadan using the method described by Odiete, (1999) .The concentrations of crude oil used are presented on table 1.

There were 15 fish per replicate and no aeration. Dead fish (total lack of opercular and body movements) were promptly removed. The LC_{50} was determined by the logarithm method (Litchfield and Wilcoxon, 1949) and a follow up test using least significant difference (LSD) .The data obtained were also subjected to analysis of variance (table 6). Chemical analyses were conducted to determine the heavy metal (FAO/SIDA, 1983) and total hydrocarbon (Rump and Krist, 1988) content of the fish and water used .The compositions of Nigerian crude oils (including Qua Iboe Light) have been reported by Eco-consultancy Group, (1987).

Table 1: The Concentration Of The Crude Oil Used For The Bioassay

Treatment	Crude oil concentration(µL/L)	Volume of Crude oil (ml)	Volume of dilution water(ml)	volume of crude oil X3(ml)	Volume of dilution water X3 (ml)
5	0	0	1000	0	3000
4	25	2.5	997.5	7.5	2992.5
3	50	5	995	15	2985
2	75	7.5	992.5	22.5	2977.5
1	100	10	990	30	2970

RESULTS

On introducing the toxicant, there was commotion in the aquarium as fish attempted to avoid the oil. The surface of the water in treatment 1 (100µL/L) was totally covered by the crude oil while other treatments had some exposed water surfaces. All fish in the 100µL/L concentration died and all but two fish died within the first three hours of the experiment in treatment 2 (75µL/L). The remaining two fish in treatment two died within the next three hours .The numbers and percentage mortalities during the 96-hour test period are presented on table 2. The survivors in the experiment continued to live for two more weeks after the termination of the experiment.

The results of the heavy metals and total hydrocarbons in water and experimental fish are presented on tables 3 to 7. Lead, iron copper, cadmium and vanadium were not detected in both the water used and fish muscle and higher concentrations of metals were detected in fish muscles than in the water. Lead was not detected in the survivors of the experiment at the end of recovery period of two weeks. The highest concentrations of THC in water was observed in the 100ul/L and least in the control .The highest observed THC in fish muscle (21.76 mg/kg) was recorded for the 75µL/L concentration.

DISCUSSION

The activity of crude oil on *C. gariepinus* fingerlings was assessed in an acute study using crude oil-in-water bioassay. More mortalities ($p<0.05$) were reported for treatments with higher concentrations of crude oil (75-100µL/L).The toxicity of crude oil is directly correlated to its contents of soluble aromatic derivatives: benzene, naphthalene, phenanthrene and their alkyl groups and organism – specific but lighter crude oils are more volatile, soluble and toxic than the heavy or medium crude oils (Eco-Consultancy Group, 1987). Fish may increase or decrease the concentration of oil through bioaccumulation and / or direct degradation (Palmer *et al*, 1997).It has been observed that fish have the ability to induce detoxifying enzymes though the enzymes do not offer resistance to acute exposures (Payne and Penrose, 1975).

CYPIA is an enzyme induced in oil-affected animals, usually measured by the activity of 7-ethoxyresorufin O-deethylase (EROD) activities in the liver of affected animals. EROD is the activity of the P450 monoxygenase, a family of biochemical catalysts that allow detoxification and excretion of toxic substances like polycyclic aromatic hydrocarbons (PAHS) by transforming them into polar, water-soluble compounds (Marine Pollution Monitoring Group, 1998). The changes observed in the fish could have been due to the induction of these enzymes in response to the oil. This observation is supported by D'Adamo *et al.* (1997).

The fish in the 100µL/L concentration died within the first three hours of the experiment and were removed while a longer period passed (6 hours) in the 75µL/L concentration .This time allowed greater exposure to the oil and time for more absorption which may account for the higher THC of tissues. The concentrations of THC in all treatments except in the control were greater than 1ppm. The LC 50 obtained using the crude oil in water was 1.58uL/L. based on the LC_{50} (figure 1). The permissible level of the concentration is usually set at a fraction (1-10%) of the LC50 value.

The heavy metal contents recorded in the fish muscles during the 96-hour period were generally below the World Health Organizations'(W.H.O.) limits of 2.0, 2.0. 30.0 and 1000µL/L for Cd, Pb Cu, and Zn respectively. The concentrations of iron were also within WHO limits. However, levels of iron differed among treatments with 75µL/L having the lowest and 25µL/L the highest. The differences observed in the metal concentrations during the 96-hour and recovery periods may be due to reactions by fish to the presence of the pollutant.

CONCLUSION

From this study, it may be concluded that immediately an oil spill occurs, the fish that are exposed to high concentrations die while those that survive attempt to eliminate the excess oil through physiological processes leading to the adaptation to the presence of THC in their

tissues. It has been observed that the liver, brain and other visceral organs accumulate more THC than the muscles. It is therefore recommended that the processes which reduce the entrance of crude oil into the environment should be encouraged.

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Table 2: Number and percentage mortality of fish in 96 hours

Concentration µL/L	No. of Test organisms	No of deaths at 96 hours			Total deaths out of 45	Percentage Mortality of fish		
		Replicates				Replicates		
		1	2	3		1	2	3
0	45	0	0	0	0	0	0	0
25	45	4	3	2	9	26.67	20	13.33
50	45	3	3	2	8	20	20	13.33
75	45	15	15	15	45	100	100	100
100	45	15	15	15	45	100	100	100

Table 3: Heavy Metal concentration in water after 96 hours

Concentration (µL/L)	Pb	Fe	Mn	Cr	Cd	Zn	Cu	V
0	Nd	Nd	Nd	0.375	Nd	0.375	Nd	Nd
25	Nd	Nd	Nd	0.375	Nd	0.5	Nd	0.1
50	Nd	Nd	1.625	0.375	Nd	0.375	Nd	0.2
75	Nd	Nd	Nd	0.375	Nd	0.375	Nd	0.39
100	Nd	Nd	Nd	0.375	Nd	0.375	Nd	0.625
Tap water	Nd	Nd	Nd	0.25	Nd	0.25	Nd	Nd

Nd = not detected

Table 4: Heavy Metal Concentration in Fish Muscle after 96-Hour Bioassay

Conc. (µL/L)	Pb	Fe	Mn	Cr	Cd	Zn	Cu	V
0	Nd	24.375	9.25	1.125	0.25	10.875	1.125	Nd
25	Nd	43.75	10.125	0.625	0.25	9.5	1.625	0.125
50	Nd	30.35	11.625	1.125	0.375	17.136	1.125	0.92
75	Nd	8.625	8.375	0.625	1.125	7.25	0.375	0.875
100	Nd	24.25	13.625	1.000	0.125	10.500	1.375	1.000

Table 5: Total hydrocarbon content (thc) in water and fish muscle after 96 hours

Concentration µL/L	THC in water µL/L	THC in fish Muscle mg/kg
0	0.0	0.16
25	5.119	0.72
50	7.429	4.76
75	7.361	21.76
100	14.21	18.32

Table 6: ANOVA Table for Oil –In- Water Bioassay

SV	df	SS	MS	F	P-LEVEL
Treatment	4		158.7667		0.000000*
10		2.667*	.266667*	593.3750*	

Table 7: Heavy Metal Content of Surviving Fish after Two Weeks of Recovery.

Conc. Ul/L	Pb	Fe	Mn	Cr	Cd	Zn	Cu	V
0	Nd	24.375	9.25	1.125	0.25	10.875	1.125	Nd
25	Nd	23.125	9.125	2.375	0.375	14.75	1.5	1.0
50	Nd	24.5	16.125	1.25	0.375	14.75	1.75	1.25

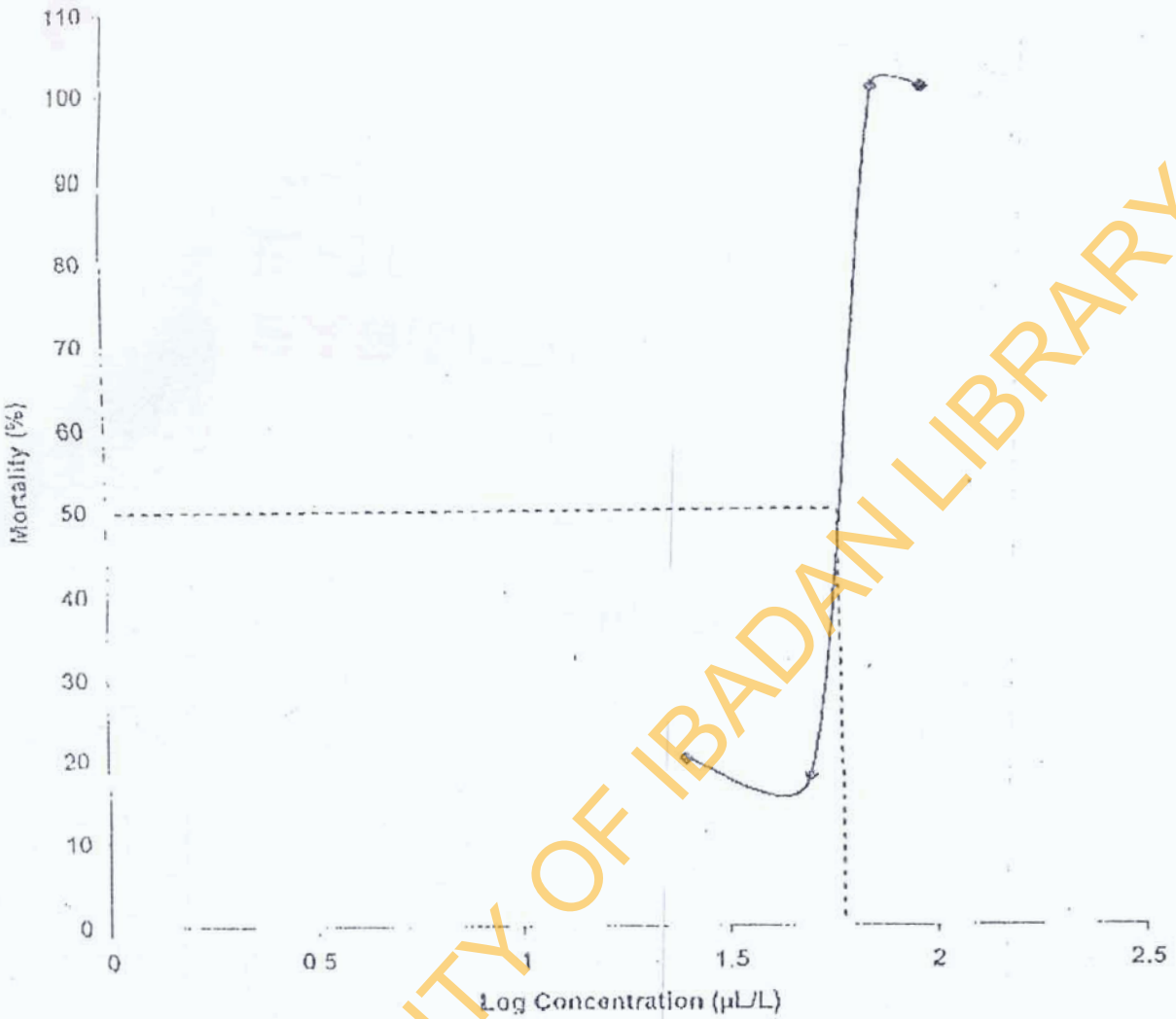

Figure 1: Percentage Mortality versus Log. Concentration for Oil –In –Water Bioassay Showing LC50