TOXICITY OF CRUDE OIL PRODUCTS AND DETERGENT ON SERUM ALKALINE PHOSPHATASE CONCENTRATION OF *Clarias gariepinus* JUVENILES

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ABSTRACT

The comparative effect of exposing Clarias gariepinus juveniles (100.20 ± 0.8g) to different concentrations of crude oil products and detergent were studied. Bonny Light Crude oil (BLCO), Premium motor spirit (PMS), Dual purpose kerosene (DPK) and Ariel Enzymax Detergent (AED) were respectively applied at three concentrations, 2.00, 4.00and 6.00mi/L as well as a control experiment 0.00ml/L with no toxicant for two hours (2 hours) toxicity period. Increased serum alkaline phosphatase and low degree of toxicity were recorded in the fish samples exposed to the various pollutants than in the fish control. The comparatively low toxicity noticed within the sample fish was attributed to the reduced length of exposure to the pollutants this probably lowered the pressure of the toxicant on organs of the fish.

Keywords: *Clarias gariepinus,* Serum alkaline phosphatase, Bonny light crude oil, Premium motor spirit, Dual purpose kerosene, Ariel enzymax detergent

INTRODUCTION

The degree of hazardous effect of crude oil products as well as detergent is dependent on the degree of their concentration, chemical components and solubility in water. These products have been recognized as a potential environmental contaminant shortly after the beginning of twentieth century (Albers, 1995). Researches carried out across the globe on the toxicity of Bonny Light crude oil (BLCO), Premium motor spirit (PMS) and Dual purpose kerosene (DPK) on aquatic organisms have revealed their lethal, acute, short and long term effect (Brain, 1971). It has been found that as little as 0.1ppm of oil can seriously affect fish, crustaceans and plankton. "Oils" float and coats things and has the potential to kill quickly by coating aquatic lives, interfering with gas exchange necessary for life, when it sediment at the bottom, the contaminating impact has longer effect and benthic organisms become particularly susceptible (Baker, 1971).

Detergents are common household and industrial products and their role as polluting agent of natural water cannot be ignored. The Validity of a Detergent as a toxicant depends on three factors response of the test animal, the substances mode of action and toxicity of the substance in relation to its chemical and physical structure (Leader and Devaminck, 1952; Goldacre, 1968). The effect of this product is not only on the aquatic lives but also on terrestrial organisms of which man is not an exception. Detergents (`Ariel Enzymax) contains chemicals (water softener, processing acids, cleaning agents, perfume etc) that exerts a devastating effect on exposure to aquatic lives, causing severe damages and disruptions of activities of the mucus secreting glands indicating poisoning effects on organs, glands and tissues (Omoregie *et al.*, 1990). They are also known to damage olfactory epithelium in the nasal capsule, inhibit enzyme activity and cause oxygen stress by affecting the gills (Okwuosa and Omoregie, 1995).

The primary reason for analyzing ALP enzymes are to determine whether an increased level of ALP is due to bone or liver enzyme; however Ayalogu *et al.* (2001) reported that the elevation of ALP activity appears to reflect some inflammatory diseases or injuries to the liver or stress. Other studies also indicated increase in the activities of the enzyme following liver damage in fish and albino

mouse exposed to petroleum products (Dheer *et al*., 1987).

There are many more evidences that different organic and inorganic pollutants elevate the serum alkaline phosphatase in fish. This study thus, presents the results of the study of the exposure of Dutch *Clarias gariepinus* juveniles to different concentrations of crude oil products and Detergents and their effect on the alkaline phosphatase. The essence of this was to ascertain the impact of the various concentrations of this pollutant on energy metabolism of this Nigerian highly priced food fish.

MATERIALS AND METHODS

One hundred and fifty (150) juveniles of *Clarias gariepinus* (100.20 ± 0.08g) were transported from a private fish hatchery at Ugwuomu, Emene in Enugu State, Nigeria to Heldin Fisheries, Unit, New Haven, Enugu. At the fishery unit, the fish were acclimatized for 14 days and fed 38 % crude protein diet at 3 % body weight daily (Table 1).

Twenty six (26) juveniles fish of Dutch *Clarias gariepinus* were subjected to different concentrations (0.00mlL^{-1}, 2.00mlL^{-1}, 4.00mlL^{-1}, and 6.00mlL^{-1}) of Bonny-Light Crude Oil (BLCO), Premium Motor Spirit (PMS), Dual Purpose Kerosene (DPK) and 30g/l, Ariel Enzymax detergent (AED). Each of these four pollutants was introduced to 24 plastic containers (90 L) at the above stated concentrations. The control experiment (0.00mlL^{-1}) consisted of two (2) plastic containers without any oil treatment. The fish juveniles were randomly stocked in 26 plastic containers (90L) at four fish per container. Each container was filled to 10 litres mark with rain water and labeled according to treatment. The fish in each set up were exposed to 2 hours in the different concentrations of the pollutants. The application of the pollutants marked the beginning of the toxicity phase.

Blood samples from the fish juveniles exposed to the different concentrations of the pollutants were collected from razor cut in the musculature, behind the opercula region on the dorsal surface. The collected blood samples were allowed to cloth in EDTA bottles in order to get blood serum. Blood serum was collected from prepared blood samples for analysis of alkaline phosphatase using Kind and King (1954) method, and thus serum alkaline phosphatase concentrations were determined. All enzymes assays were conducted spectophotometrically at appropriate wave length.

Samples were assayed in duplicates and data collected were analyzed using analysis of variance (ANOVA) and LSD to determine statistical differences ($P<0.05$) among the treatment means.

Table 1: Gross and proximate composition of the experimental diet fed to *Clarias gariepinus* juveniles

Feed ingredients	% composition
Yellow maize	9.81
Soya bean meal	54.76
Fish meal	16.43
Blood meal	10.95
Palm oil	0.45
Salt	0.25
Vitamin mix[1]	0
Mineral mix[2]	1.80
Total	100.00%
Nutrients	
Crude protein	37.58
Ether extracts	5.18
Ash	10.48
Dry matter	11.80
Nitrogen-free extract	34.46
Total	100.00

1)Vitamin mix provided the following constituents diluted in cellulose (mg/kg of diet); thiamine, 10; riboflavin, 20; pyridoxine, 10; folacin, 5; pantothenic acid, 40; chlorine chloride, 3,000; niacin, 150; vitamin Biz, 0.06, retinyl acetase (500,000 IU/g), 6; menadione – Na – bisulphate, 80; inositle, 400; biotin, 2 vitamin C, 200 alphatocopherol, 200; cholecalcipherol, 000,000 IU/g.

2) Contained as g/kg of premix: $Feso_4.7H_20$, 5; $MgSo_4.7H_20$; K_2So4, 329.90; KI, 0.15; NaCl, 45; Na_2So_4, 88; $AICL_3$, 0.15; $Cocl_2.6H_20$, 0.05; $Cuso_4.5H_20$, 0.50; $NaSeO_3$, 0.11, $MnSo_4.H_20$, 0.70 and cellulose 390.97(Faturoti et al., 1986).

RESULTS

Table 1 shows gross and proximate compositions of the experimental diet fed to *Clarias gariepinus* juveniles during the study periods. Table 2 showed the concentrations of alkaline phosphatase in C*larias gariepinus* juveniles for different concentrations of crude oil products and detergents.

The control fish samples recorded significantly ($P < 0.05$) higher values of alkaline phosphatase concentration in the blood than those polluted or treated with the different concentrations of BLCO, DPK, PMS and AED except for 6 ml BLCO and 2 ml AED which recorded values that were significantly higher than that of the control experiment. Generally, their degree of toxicity on the fish was high when compared with the control, Ariel enzymax detergent gave low concentration, fish exposed to Bonny light crude oil had lower concentrations while kerosene and petrol had higher concentrations of the enzyme.

Table 2: Serum alkaline phosphatase content of *Clarias gariepinus* exposed juveniles to different concentrations of crude oil products and Ariel detergent

Toxicants	Concentration (ml^{-1})	Enzyme Concentration In i.u/l		Total Serum Alkaline Phosphatase Conc. (i.u/l)	Mean Alkaline Phosphatase Conc.
		R1	R2		
Kerosene (DPK)	2ml	62.4	62.4	124.9	62.45±0.04^{b}
	4ml	62.5	62.3	124.8	62.40±0.01^{b}
	6ml	60.4	60.4	120.4	60.40±0.03^{a}
	Control	62.5	62.5	125.00	62.5±0.001^{b}
Crude Oil (BLCO)	2ml	61.1	61.5	122.10	61.05±0.04^{a}
	4ml	60.4	60.2	120.6	60.30±0.01^{a}
	6ml	66.8	66.7	133.50	66.50±0.01^{c}
	Control	62.5	62.5	125.00	62.5±0.00^{b}
Petrol (PMS)	2ml	61.8	61.6	123.4	61.7±0.01^{a}
	4ml	62.5	61.2	123.7	62.32±0.03^{b}
	6ml	61.8	62.05	123.3	61.65±0.03^{a}
	Control	62.5	62.5	125.00	62.5±0.00^{b}
Ariel Detergent (AED)	2ml	66.7	66.6	132.1	66.10±0.01^{c}
	4ml	61.10	61.8	122.9	61.45±0.02^{a}
	6ml	62.4	62.04	124.4	61.20±0.05^{a}
	Control	62.5	62.5	125.00	62.5±0.00^{b}

Fish exposed to Ariel detergent and Bonny light crude oil showed a significant increase in alkaline phosphatase concentration when compared with the control while petrol and kerosene recorded decreased serum alkaline phosphatase concentration. There was generally significant difference ($P < 0.05$) among alkaline phosphatase concentrations in fish exposed to the different products. No death was however recorded among the tested samples. There were comparatively significant differences ($P < 0.05$) among alkaline phosphatase concentrations in fish exposed to different pollutants.

DISCUSSION

The increase noticed in the treated samples were believed to be as a result of the toxic effects of the pollutants on the fish though not all the concentrations elevated serum alkaline phosphatase concentration. The toxic effect of the crude oil products and Ariel Detergent varied and fluctuated among pollutants and their concentrations. Petrol had faster dissolution, this probably accounted for its higher toxic effect on fish than BLCO and DPK. This is consistent with the report of Baker (1971) on the potential ability of petrol to dissolve, coat and kill quickly anything it comes in contact with. The elevation of serum alkaline phosphatase may be as a result of the injury sustained by the organs of the fish, this report concurs with the report of Ayalogu *et al*. (2001) on the elevation of ALP activity which they reported appears to reflect some inflammatory diseases or injury on the liver or stress. The rupture of blood vessels around the gills and disruption of the skin of the fish exposed to Ariel detergent, could cause severe dermal, respiratory and physiological stress, this report is in consonance with Omoregie *et al*., 1990 that reported that detergent has devastating effects on aquatic lives.

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