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EFFECTS OF CRUDE OIL ON THE GASTROPOD, *Tympanotomus fuscata* IN THE CROSS RIVER ESTUARY, SOUTH-EAST NIGERIA

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ABSTRACT

Edible periwinkle, *Tympanotonus fuscata,* was collected from the Cross River estuary, allowed to acclimate to laboratory conditions for a week and then exposed to Nigerian light crude oil at different concentrations of 50ml, 100ml, and 150ml per litre of water for a period of 96 hours. Mud (80g) was added to all the 4-litre aquaria used for the experiment. The oil was thoroughly mixed with the estuarine water by stirring vigorously. During the 4-days experimental period, most of the specimens suffered suffocation and became moribund and mortality recorded. Within the first 24 hours mortality was absent in the 50ml and 100ml concentrations, although most specimens moved out of the medium by creeping up the aquaria. Specimens in the control seemed lively and unaffected by oiling throughout the duration of the experiment. Mortalities were as high as 117 out of 180 individuals during the entire experimental period, with higher concentrations recording higher mortalities. There was steady increase in mortality from 4 individuals in the first 24 hours to 12 individuals after 72 hours in the 150ml/l concentration. The LC_{50} values were obtained by graphical interpolation. It is concluded that the soluble fraction of the Nigerian light crude oil could be detrimental to shellfish even at a concentration as low as 1000ml/l of the estuarine water.

KEYWORDS: Crude Oil, Water Soluble Fraction, *Tympanotonus fuscata,* Cross River estuary, Nigeria.

INTRODUCTION

Exploration, exploitation, drilling transportation, processing and storage of crude oil have greatly affected the marine environment (Richard *et al*., 1978; Doerter, 1992; Ewa-oboho and Abby Kalio, 1994). The majority of oil pollution occurs in the estuaries and near shore waters (Richard *et al*., 1978; Brown *et al*., 1990). Petroleum hydrocarbons are the major contaminants of the estuarine and coastal environments (Lee *et al*., 1980; Widdows *et al*., 1982). Enin (1997) reports that offshore petroleum production facilities located adjacent to the mouth of the Cross River estuary introduce small-scale oil spills and leakages into the estuary by wave and tides.

When oil spills on the sea surface undergo a number of major physical and chemical processes such as drifting, evaporation, dissolution, dispersion, emulsification, sedimentation, biodegradation and pho-oxidation and these processes, whether physical or chemical, help in the distribution of the spills and partitioning of the oil component into the water, air, ambient atmosphere, organisms and sediments as the case may be (Clark, 1992).

In Nigerian waters, cases of oil spillage have been recorded between 1958 to date releasing about 2.4 million barrels of crude oil into coastal aquatic environment. Of importance are the Exxon Mobil spills, Idoho disaster, 1998, Ogoni oil spills disasters, 1958-2005 (Udo, 2007).

This poses a great risk to aquatic organisms like periwinkle, which is a source of protein for the coastal dwellers in Nigeria. Doertter (1992) reports that oil spills causes substantial mortality among fish and invertebrates. Other effects include changes in species composition, low abundance, loss of species and tainting (Windows *et al*., 1982). To address these effects which are mostly caused

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by Water Soluble Fractions (WSF) of the crude oil by dissolution, attention must be given to the biological effect of WSF and the residual of petroleum (Clark, 1992)

The concentrations of toxicants in the environment vary with duration of exposure, volume of discharge, as well as the nature of hydrocarbon. The acute toxicity of crude oil to aquatic organisms like *T. fuscata,* is assessed by the measurement of LC_{50} value, i.e. the concentration of hydrocarbon mixture or specific hydrocarbon that results in 50% mortality of test organism during the designated exposure period, usually 72 or 96 hours. The aims of this study were to asses the effects of a known quantity of Nigerian light crude oil on the periwinkle, *Tympanotonus fuscata,* of the Cross River estuary, determine the mortality rate of *T. fuscata* at different concentration of the Water Soluble Fraction (WSF), and to examine the tolerance limit of *T. fuscata* exposed to Water Soluble Fraction (WSF) of the (Nigerian light) crude oil.

MATERIALS AND METHODS

The study Area

The Cross River estuary lies between latitudes $4^0$30' N and $5^0$15' N and longitudes $8^0$00' E and $8^0$40' of the equator (Fig. 1).

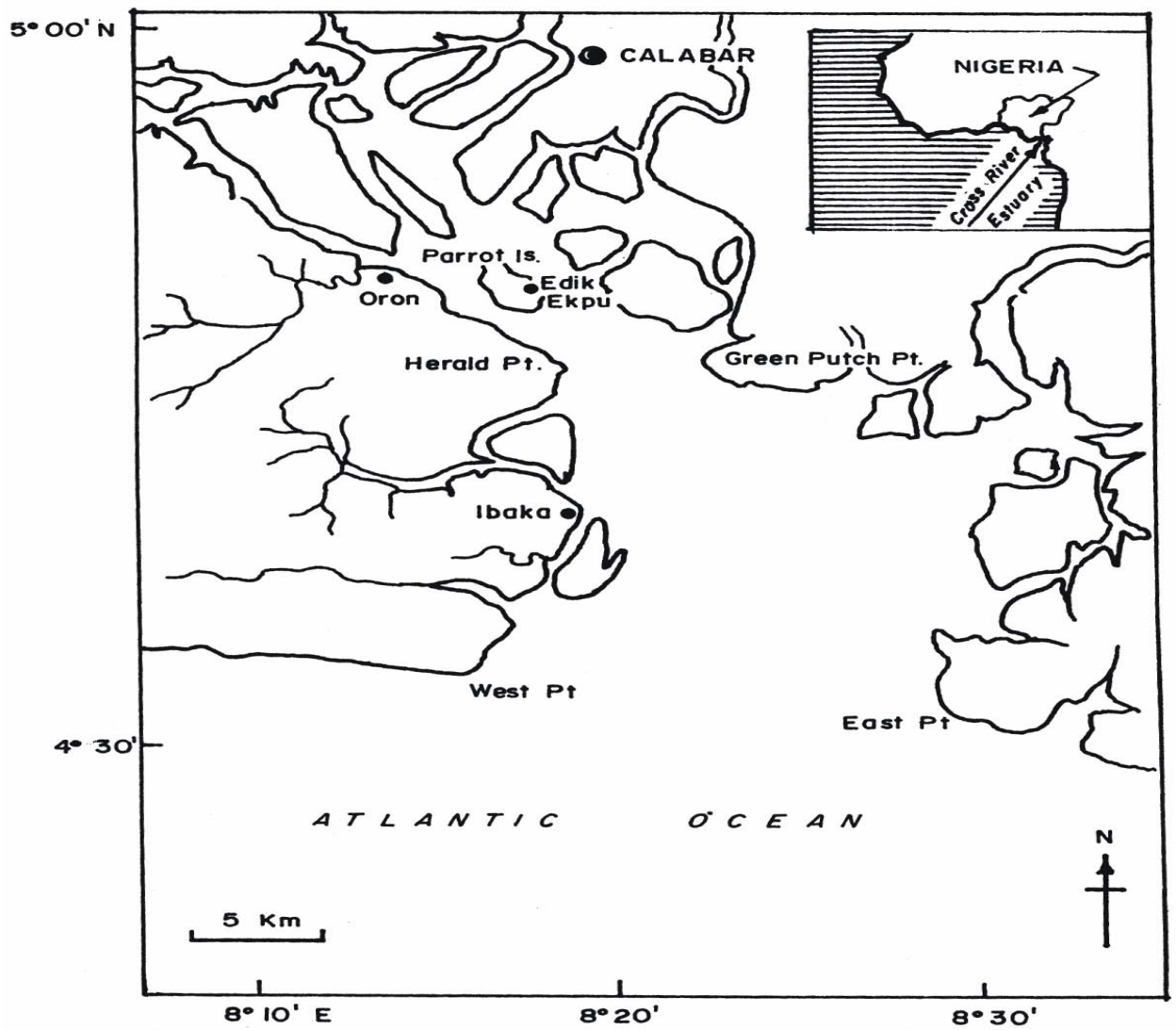

Fig. 1: Cross River Estuary, Nigeria showing Calabar the sampling site.

The river whose catchments area is from Oban hill and basin, covers an estimated area of 54, 000km^2. It is rich in clay materials and is located within the tropical rain forest region. Thus the mineral-rich catchments area in combination with the dense vegetation and frontal rainfall pattern characteristic of this area plays a tremendous role in the biological chemical regulation of organic and inorganic nutrients in the estuary (Akpan and Ofem, 1993; Asuquo *et al*., 1998). These nutrients encourage high primary productivity, species abundance and diversity in the estuary. The estuary is also prone to allochthonously imposed negative changes in the environment, principally due to oil pollution activities adjacent to the mouth of the estuary (Enin, 1997). The Cross River estuary, the largest in Nigeria with tidal amplitude of 3m (Asuquo *et al.*, 1998) is delineated into three aquatic ecological habitats. Ranging from fresh water in the upstream region through brackish water in the middle reaches to the marine environment at the mouth of the estuary (down stream).

The climate is of equatorial type, always hot and wet. During the period of November to April, the region lies in the high-pressure belt from where the dry trade winds start. From May-October, the climate change is one of the challenges the shell and finfish in the estuary face incessantly, which also brings about fluctuation in the number and species composition at different periods of the year.

LABORATORY CONDITION

Nigeria Light Crude Oil was collected and diluted into various concentrations with estuarine water at laboratory temperature, 28-30^o C. The concentrations were prepared by placing the oil over the water and stirred vigorously using hand stirrer and shaking gently manually. The excess floating oil was not removed from the medium after mixing in order to reduce the loss of volatile hydrocarbons through evaporation from the aqueous phase during the dosing period.

The concentrations were grouped. Group I was 50ml of crude oil to 1,000ml of estuarine water; Group II was 100ml of crude oil to 1,000ml of estuarine water; Group III was 150ml of crude oil to 1,000ml of estuarine water and Group IV, the control, was 1,000ml uncontaminated estuarine water.

The test organism, mudflat periwinkles *(Tympanotonus fuscata)* were collected from the Cross River estuary and allowed to acclimate to laboratory condition for one week before exposing them to crude oil. The largest samples measured 25.4mm, 7.1mm, and the smallest 16.9mm, 5.2mm in both height and with respectively.

Three pairs of 4-litre glass aquaria were use as experimental chambers, and one glass aquarium as the control. Each pair had the same concentration. 30 specimens were then placed inside all the aquaria including the control. Some quantity of mud (80g) was introduced into the aquaria to serve as food and also to provide a natural condition for the shellfish. Before stocking, the aquaria were thoroughly washed and rinsed three times.

Both the control and experiments I and II chambers were monitored every 24 hours for four days. Mortality was investigated by careful observations. Experiment II was used to consolidate the result of experiment I. Deaths were ascertained by apparent immobility of periwinkle when left for 24 hours undisturbed and opening of the operculum (lid). The operculum when tapped with pin showed little or no reactions.

RESULTS

The mortality data for periwinkle, *Tympanotonus fuscata* for two replicate experiments (I and II) are shown in Table1. In both trials the acute toxicity after 24 hrs corresponded to the different concentrations of 50ml/l, 100ml/l and 150ml/l. There was no significant difference using homogeneity test (Chi-square, F=0.241, P>0.05) in mortality in both. The test shows that the same procedure was followed in both trials. Mortalities in all the three concentrations were also compared using students' t-test which showed significance of difference (F=12.96, 11.67 and 11.0, p<0.05) for 100ml/50ml/l, 150ml/l/50ml/l and 150ml/l /100ml/l respectively. The values were also transformed using Log transformation (x1 = $Log_{10}(x=^1)$) (Table 2) and converted into percentage (Table3).

Mortality was absent during the first 24 hours in 50ml/l and 100ml/l concentrations, while each recorded in 4 dead in 150ml/l concentration. At 48 and 72 hrs, higher mortality values were recorded with 150ml/l concentration having the highest values of 12 dead. There was a sudden drop at 96 hrs recording as low as 1 dead in 50ml/l concentration, indicating that the toxic effect of WSF reduces with duration of exposure due to evaporation of the volatile component and

coagulation of the residue (Clark,1992) (Fig.2a and 3a)

Out of 180 stocked samples, a total of 117 individuals were dead with the following break down: first 24 hrs, 8; 48hrs, 34;72 hrs, 54; and 96 hrs,21 dead. The experimental mortalities were contrary to the control, which recorded no dead throughout the period. However, the crude oil toxicity increased with longer exposure up to the third day (72hrs) with a sudden fall on the fourth day (96hrs) (Figs. 2b and 3b). After 24-hrs exposure, mortality was observed only at the 150ml concentration with less than 14%. This implies that the 24-hrs LC_{50} is outside the range of concentration utilized for the experiments (Figs. 2c and 3c). There were increases in mortality after 48-hrs at about 14%, 20% and 47% corresponding to concentrations for 50ml, 100ml, and 150ml respectively. Exposures over a 72-hrs period resulted in mortalities of about 34%, 47%, and 80% respectively. At 96-hrs, about the following percentages were also recorded: 40%, 60% and 100% (Tables 3a and 3b) (Fig. 2c and 3c).

Apart from mortality caused by the toxic effects of WSF, a layer of oil in the water column would result in a drop in respiration rate of test organisms leading to death.

DISCUSSION

In Figs. 2 and 3, the mortality rate of test organisms is presented. Figs. 2 and 3 represent Experiments I and II respectively, which showed no significant difference using homogeneity test (x^2, F=0.241, p>0.05). This implies that the results were the same without any noticeable variation, and that the same produce was followed in each experiment. While Figs. 2a and 3a represent a normal count in mortality, Figs. 2b and 2c represent the transformed values using (x^1 = Log10 (x+1)). Fig. b of both experiments I and II depicts each day's morality while Fig. c determines the LC_{50} of the test organisms.

The rate of mortality of *T. fuscata* was attributed to the concentration of the spilled oil, the period of exposure, ability of the shellfish to bio-accumulate and the nature of the oil (Clark, 1992; Ewa-oboho and Asuquo, 2006; Daka *et al*., 2003).

Mortality in 50ml concentration fell below 50% (median%) indicating that LC_{50} is outside the range of concentration throughout the experimental period, while 100ml and 150ml were within and above the range that causes LC_{50} mortality (Figs. 2c and 3c). These results agree with those of Widows, *et al*., 1982 and Daka, *et al*., 2003. Certain invertebrates have the capability to retain high concentration of hydrocarbon in their tissues, hence high concentrations are necessary to elicit a lethal response (White, 1981).

The delay in mortality in the concentrations of 50ml and 100ml during the first 24hrs could be attributed to the time taken for WSF to dissolve and residue to emulsify (Clark, 1992). As deposit feeders, *T. fuscata* take up fine droplets of emulsified oil in the course of normal feeding process, which is subsequently incorporated into their tissues. The sudden drop after 72 hrs could be as a result of evaporation after dissolution. According to Clark (1992) the dissolved component is still susceptible to evaporation. The waxy excess oil on the surface could hinder evaporation during the first few hours till the oil coagulates into chocolate mouse. Coagulation takes place between 10 to 70 hrs after oil is spilled (Clark, 1992).

It was observed that the size of the test organisms collected from the Cross River estuary was smaller than the reported size in the literature. Edmunds (1978) reports that *T. fuscata* range from 45 to 70mm in height and about 1/3 of the height in width, but the observed height and width in the estuary were between 16.9mm to 25.4mm and 5.2mm to 7.1mm respectively. This difference could be attributed to favourable environmental conditions prevailing at the coastal area.

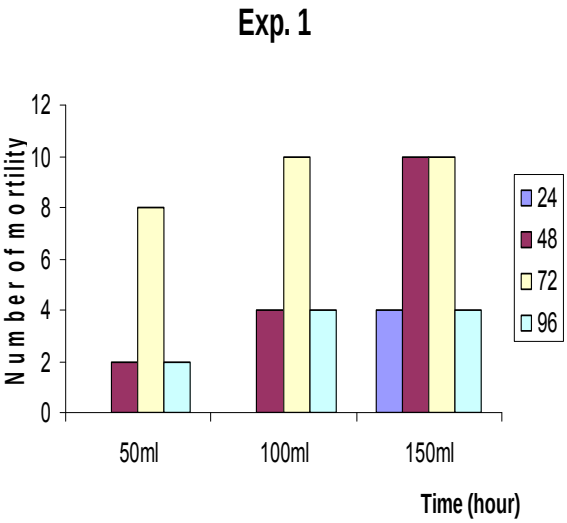

Fig. 2: Mortalities of test organisms during the 96 hours experimental period

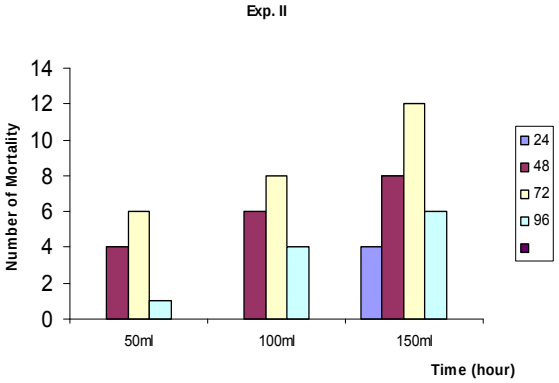

Fig. 3: Mortalities of test organisms during the 96 hours experimental period

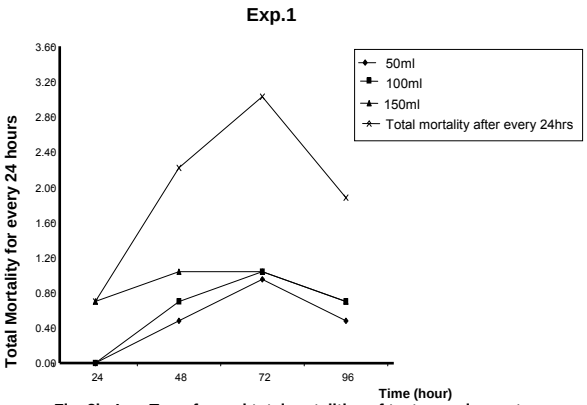

Fig. 2b: Log Transformed total motalities of test organisms at every 24 hours.

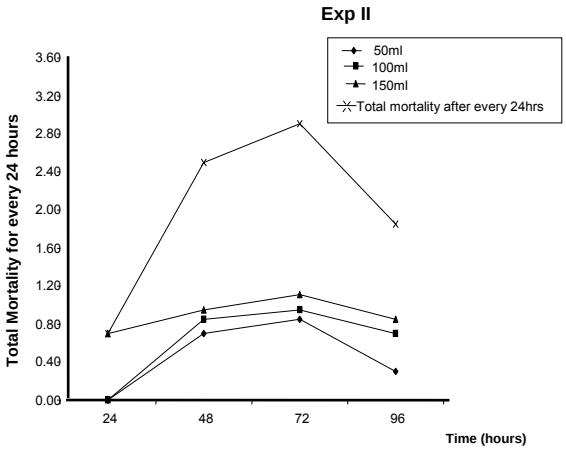

Fig.3b: Log Transformed total mortalities of test organisms at every 24 hours.

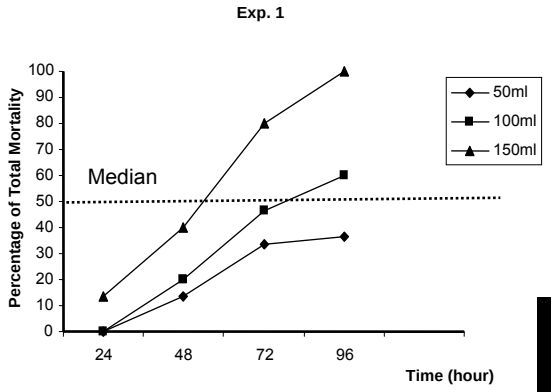

Fig. 2c: Percentage of total mortalities of test organisms (LC_{50} values)

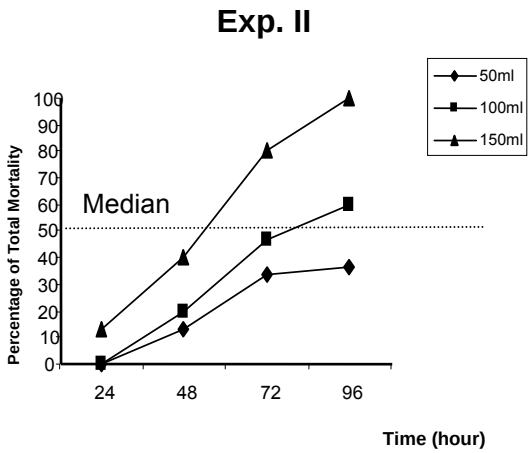

Fig. 3c: Percentage of total mortalities of test organisms (LC_{50} values)

Table 1: Number of Mortality after 24 hours of exposure to Water Soluble Fraction (WSF)

Groups of Experiment						
Conc.(ml) / Time(hrs)	50ml		100ml		150ml	
	Number of deaths		Number of deaths		Number of deaths	
	EXP.I	EXP.II	EXP.I	EXP.II	EXP.I	EXP.II
24	No death	No death	No death	No death	4	4
48	2	4	4	6	10	8
72	8	6	10	8	10	12
96	2	1	4	4	4	6
Total mortality	12	11	18	18	28	30

Table 2a: Log Transformed number of total mortality of periwinkle exposed to Water Soluble Fraction (WSF)

Conc.(ml) / Time(hrs)	Experiment I			
	50ml	100ml	150ml	Total mortality after every 24hrs
24	0.00	0.00	0.70	0.70
48	0.48	0.70	1.04	2.22
72	0.95	1.04	1.04	3.03
96	0.48	0.70	0.70	1.88

Table 2b: Log Transformed number of total mortality of exposed periwinkle to Water Soluble Fraction (WSF)

Conc.(ml) / Time(hrs)	Experiment II			
	50ml	100ml	150ml	Total mortality after every 24hrs
24	0	0	0.70	0.70
48	0.70	0.85	0.95	2.50
72	0.85	0.95	1.11	2.91
96	0.30	0.70	0.85	1.85

Table 3a: Percentage Transformed number of total mortality of periwinkle exposed to Oil Water Soluble Fraction (WSF)

Experiment I			
percentage of Mortality			
Conc.(ml) / Time(hrs)	50ml	100ml	150ml
	Cumm %	Cumm %	Cumm %
24	-	-	13.33
48	6.67	13.33	46.66
72	33.34	46.66	79.99
96	40.00	60.00	93.32

Table 3b: Percentage Transformed number of total mortality of periwinkle exposed to Water Soluble Fraction (WSF)

Experiment II			
percentage			
Conc.(ml) / Time(hrs)	50ml	100ml	150ml
	Cumm %	Cumm %	Cumm %
24	-	-	13.33
48	13.33	20.00	40.00
72	33.33	46.67	80.00
96	36.66	60.00	100.00

CONCLUSION

These experiments showed that the Water Soluble Fractions (WSF) of the Nigeria light crude oil could be detrimental to *Tympanotomus fuscata* even at a concentration as low as 100ml per litre of estuarine water. This benthic fauna is commonly found in the mangrove of Cross River estuary where they serve as food for the coastal dwellers. In the Nigerian coast and estuaries, there are known minor and major oil discharges such as oil well blowout and tanker accident (Enin, 1997). While the edible periwinkle may withstand minor discharge, they are certainly affected by the major oil spills. Because these organisms serve as food as well as a link in the food web, attention should be given to prevent their high degree of mortality through oil spillage in the Nigerian coastal waters.

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