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EFFECT OF CRUDE OIL EXTRACTS ON EARLY STAGES OF AFRICAN CATFISH HETEROBRANCHUS LONGIFILIS (VAL.) REARED UNDER CONTROLLED CONDITION

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ABSTRACT

Eggs and larvae of *Heterobranchus longifilis* were exposed to extracts of different concentrations of Nigerian Bonny light crude oil and Exxon Mobil Oso off-shore condensate during a comparative toxicity experiment carried out in the Institute of Oceanography Fish Farm, University of Calabar, Nigeria. Petroleum hydrocarbon was extracted from the two oils in separate 30litre glass aquaria and the eggs and young larvae were contaminated by exposing them to three concentrations of both oils, viz: 10^1, 10^3, 10^2ppm for approximately 10 days. Young eggs seemed to be more sensitive from 5 to 30 hours after fertilization. Extract from 10^4 ppm of Bonny Light Crude and Oso condensate caused 40% and 30% mortality respectively, after 100h. Embryos contaminated with Bonny Light crude extract did not recover on transfer to clean water. Delayed development was observed in the two-highest concentrations. *Heterobranchus longifilis* larvae were found to have a "mean critical time" of 4.2 days in the highest concentration of Oso condensate extract when larval integument was damaged. It is concluded that Bonny light crude could be a more dangerous pollutant to juvenile fish than the Oso condensate.

KEYWORDS: *Heterobranchus longifilis,* crude Oil extracts, Mortality, Bonny Light Crude, Exxon Mobil Oso Condensate.

INTRODUCTION

The marine environment adjacent to the Nigerian Gulf of Guinea is impregnated with a galaxy of oil rigs, flow stations and other petroleum based installations. This has resulted in oil entering the marine and adjoining estuarine waters from time to time. Oil drifted ashore and into the swamps could be harmful to littoral organisms as well as estuarine fisheries and fish stock (Kuhnold, 1982, Capuzzo, 1984, Anderson *et al*, 1986, Ewa-Oboho, 2006).

Many commercially important species have planktonic eggs and larvae which are abundant in the uppermost surface layers of oceanic water, and therefore exposed to the influence of petroleum components off-shores and inshore alike. The effect of floating films or undispersed crude oil on fish eggs and larvae has been investigated (Simpson, 1968, Capuzzo, 1984), Ewa-Oboho, 1994, 2006.

Water extracts of different concentrations of crude oil have been reported to show high toxicity on herring eggs when incubated under 10^3 and 2.10^4ppm with a mean survival time of 2.5 to 3.5 days (Kuhnhold, 1982). This high toxicity is mainly caused by low boiling components of petroleum hydrocarbons. The effect of oil condensate on juvenile fish and their early stages have not been seriously investigated even though petroleum associated paraffin hydrocarbons are magnificently exploited in the Gulf of Guinea coast off Akwa Ibom, South East Nigeria. Spills of oil condensate from Oso Exxon Mobil Oil rig readily enter the off-shore marine system and also drift into the adjoining estuaries. The present investigation compares the toxicity of oil condensate from Exxon-Mobil Oso rig and the Bonny Light Crude on the early egg and larval stages of the African catfish (*Heterobranchus longifilis)* which is highly priced in West African countries.

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MATERIALS AND METHODS

Eggs and young larvae of *Heterobranchus longifilis*, which have been fertilized and reared in the Institute of Oceanography (IOC) laboratory, were contaminated with dissolved and dispersed Bonny Light Crude Oil and Oso condensate. Experiments with dissolved petroleum hydrocarbons were carried out by extracting the oil and condensate in separate 30 L-glass aquaria to avoid direct application of oil films in the test containers. Different amounts of oil and condensate (10^4, 10^3, 10^2ppm) were separately poured into the water and left for two days. A calm water circulation in the containers was maintained by means of small pumps to ensure maximum saturation of soluble hydrocarbon compounds. Precaution was taken to reduced the level of evaporation of oil components by not exposing the experiment to direct sun light and strong wing. The oil extract was transferred into the test containers and renewed every two days. The amounts of Bonny light crude oil and Oso condensate used for preparing the extracts, is however not a criterion for the actual amount and types of dissolved hydrocarbons. Preliminary chemical analyses show that under the described test conditions the amount of hydrocarbons dissolved from 10^4ppm of crude oil is in the range of 10ppm. Oil dispersions were obtained by stirring 10^3ppm of Oso condensate for one minute at 10,000 revolutions per minute. Dilutions were then prepared at once. The experiments were started 50 hours later.

The effects of floating oil and dispersions were investigated by the addition of a relatively non-toxic dispersant corexit 7664. 10 and 100ppm of corexit were added when the oil was mixed with water representing 1 and 10 percent of the oil dispersed.

RESULTS

The mortality rates of *H. longifilis* eggs at different ages in the water extracts from Bonny Light Crude and Oso condensate, are represented in Figure 1. Fig. 1 shows that young eggs put into the oil extracts 5 to 30 hours after fertilization were more sensitive. On the other hand, after about 100 hours the extract from 10^4 ppm of Bonny Light Crude had caused a 40% higher mortality than the control, unlike Oso condensate which caused a mortality of about 30% higher than the control.

The relationships between the mortality of *H. longifilis* eggs and duration of pollutant influence is represented in Table 1 which indicates that the mortality was lower when one series of eggs was kept in the extract throughout the test period.

Table 1: Relationships between mortality of *H.longifilis* eggs and duration of pollutant influence

Amount of Bonny Light Crude oil (ppm)	Test for 100 hours in oil extract % dead eggs	Test continued until hatching	
		In oil extract % dead eggs	In clean water % dead eggs
10^4	30	97	43
10^3	23	65	35
10^2	14	38	20
Control	13	-	20

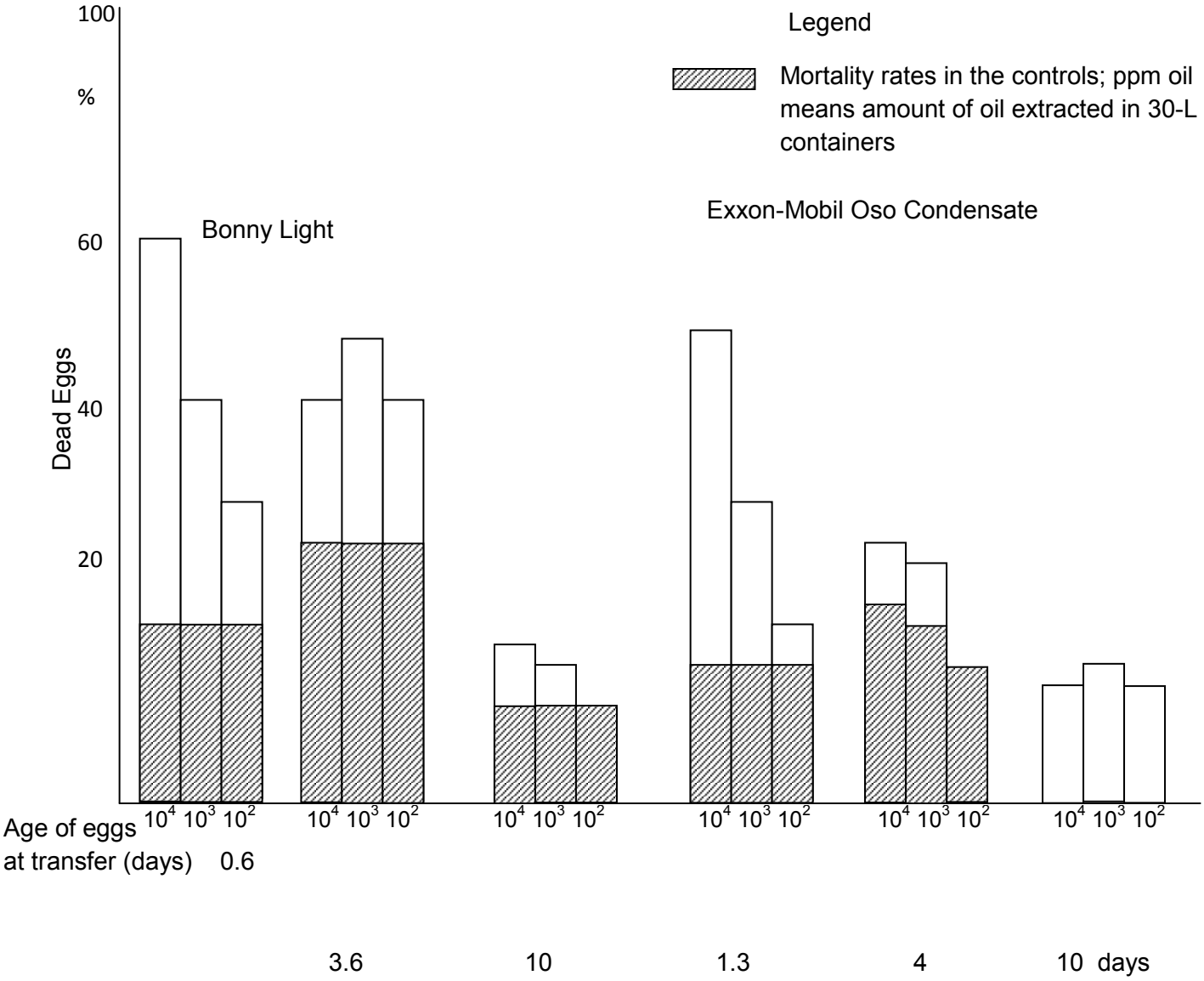

Fig.1: Mortality rates of *H. longifilis* eggs at different ages in water extracts from Bonny Light Crude and Oso condensate.

The hatching rates of *H.longifilis* eggs expressed as percentage of the control are shown in Fig. 2. After 5 to 30 h of fertilization, young eggs of *H. longifilis* exposed to extracts of 10^4 and 10^3 ppm were found to exhibit similar hatching rates (<20%) of Bonny Light Crude as in the Oso condensate. However, higher hatching rates of >80% were obtained in 10^2 ppm extracts of both pollutants after 10 days. The relationship between larval sensitivity, pollutant concentration and exposure duration expressed as mean critical times, is shown in Table 2. *H. longifilis* larvae showed a mean critical time of 4.2 days when brought into the highest concentration of Oso condensate at the ages of 1 and 0.5 days when 10 days old. (Table 2). In the lowest concentration values were 14 days and 5-5 days respectively. Resistances to oil extracts seem to

decrease with advancing resorption of the yolk sac. Embryos contaminated with Bonny Light Crude extract did not recover although they were transferred to clean water after 48h.

The biological effects revealed more severe damage, because most of the larvae that hatched had deformed bodies or abnormal flexures of the tail which hindered normal swimming and most died within one day. The deformation was observed from gastrulation onward. A delay of development was observed in the two highest concentrations beginning from 3.5 to 4 days after putting the eggs into the test milieu. In some cases, hatching was delayed or did not occur though the embryo looked normally developed.

The larvae on the other hand showed behavioural symptoms in oil extracts for example, increase in activity (especially in higher concentrations) was followed by a reduction of swimming activity, which finally stopped. The larvae then showed signs of narcosis, which gradually deepened until the critical point when no responses of larvae were obtained even by touching. These observations are similar to those reported by Wilson (1970).

Table 2: Mean Critical Time (days) of *H. longifilis* larval in dissolved petroleum hydrocarbons.

Age of larvae at transfer	Amount of Oil extracted (ppm)		
	10^2ppm	10^3ppm	10^4ppm
1	14	8.4	4.0
3	10	7.4	3.6
5	8.1	5.8	2.4
10	5.5	4.3	0.6

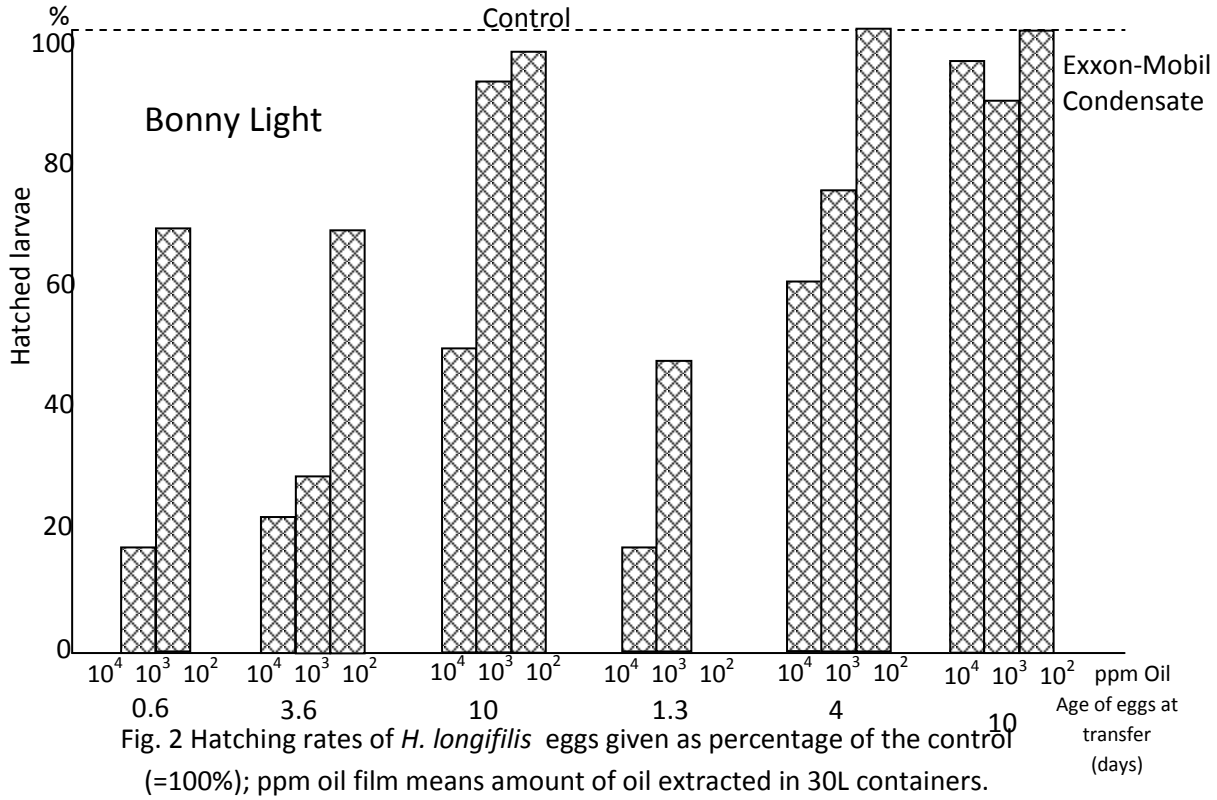

Fig. 2 Hatching rates of *H. longifilis* eggs given as percentage of the control (=100%); ppm oil film means amount of oil extracted in 30L containers.

DISCUSSION

The acute toxicity of petroleum hydrocarbons to juvenile *H. longifilis* fish is assessed in the present studies by measurement of LC_{50} value … ie. the concentration of a hydrocarbon mixture that results in 50% mortality of the test fish during a designated exposure period. It is difficult to estimate the $50LC_{50}$ or $100CL_{50}$ of the eggs as mortality rate in the controls was 0-30% after 100hrs and highly dependent on the spawn quality. Young eggs put into the oil extracts 5 to 30h after fertilization seemed to be most sensitive because of their delicate physiological stage which is usually very sensitive to pollutant.

Pure corexit solution of 10 and 100ppm concentrations were found to be non-toxic to larvae. Oil dispersions with and without corexit caused "mean critical times" of 3 to 6h at 10^3ppm of Oso condensate and 60 to 100h at 20ppm. After two days, the pure oil-water dispersion containing 100ppm of dispersant had kept or even slightly increased its toxicity at all dilution tested. The larval integument was damaged especially in higher concentrations. Typical row of blisters are formed on the primordial fins and tail fins. Larvae did not seem to avoid milky clouds of even higher concentration of dispersed oil, probably because their chemoreceptor were blocked very quickly at the first contact with oil component. It has been shown that toxic compounds are extracted from oil films in natural conditions in the sea during pollution, injuring larvae and young stages of floating eggs and yolk sac.

Several investigators have demonstrated that early embryonic and larval stages of fish are more sensitive to petroleum hydrocarbons than later larval stages (Ceas, 1974, Wells and Sprague, 1976, Donahue *et al* 1977, Linden, 1978, Cucci and Epifanio, 1979; Capuzzo and Lancaster, 1981, Ewa-oboho, 1988, Ewa-oboho and Abby-kalio, 2006, Asuquo *et al*, 2007). This is consistent with the results of the present study. The early embryonic stages were more sensitive than later embryonic and larval stages, possibly as a result of reduced membrane permeability to hydrocarbons (Sharp *et al*., 1979) or increased capability for detoxication (Binder and Stegeman, 1980) among later stages. The Bonny light crude possibly had higher concentrations of toxic aromatic components than Oso condensate which may probably be less dense and evaporates slowly than the Bonny light crude, hence, making the Bonny light crude more toxic than Oso.

To conclude, Bonny Light Crude seems to have a greater impact on early stags of *H. longifilis*. Dispersions from both types of oils can have a ten to hundred fold higher toxicity to both eggs and larvae, delaying growth or in most cases destroying the organisms.

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