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Lethal effect of water soluble fractions of Forcados crude oil on African catfish, *Clarias gariepinus*

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ABSTRACT: Juvenile and fry *Clarias gariepinus* (mean weight $13.5 \pm 0.48\text{g}$ and $2.13 \pm 0.64\text{g}$ respectively) were exposed for 96 hours to different lethal concentrations (1.0 to 15.0mg/L) of water soluble fractions (WSF) of the Forcados crude Oil. Each static tank contained ten fish randomly selected in triplicates. The fry was more sensitive while the juvenile was more resilient. The 96-h LC50 value obtained for *C. gariepinus* fry was 4.0mg/L and for juvenile was 7.4mg/L. The pulsation counts and opercular beats per minute fluctuated with time and concentration; they increased from initial to a certain level and then decreased to a lethal condition. The lethal pulsation count at 48th hour in 15.0mg/L (CGJ) was 38, and in 12.0mg/L (CGF) was 33; sublethal pulsation counts in both CFG and CGJ ranged between 33 and 39 while non-lethal pulsation ranged from 40 counts and above.

Key Words: African catfish; *Clarias gariepinus*; Forcados crude oil; Environmental pollution.

Introduction

Importance of petroleum to man cannot be over-emphasised, while only very few countries of the world including Nigeria produce it in large quantity.

Oil spillage is a common global phenomenon, though it is most common in the producing nations. The effect is pollution of the world aquatic ecosystem (Omoriegbe *et al.*, 1997). Petroleum and its products contain complex mixture of hydro-carbons which cause cellular and tissue damages in living organisms (Capuzzo, 1987). Several lethal studies document that petroleum hydrocarbons cause various stressful conditions in fish. Ghatak and Konar (1991) reported that the feeding rate of tilapia was reduced when exposed to petroleum hydrocarbon at various concentrations. The respiratory response of juvenile Chinook salmon and striped bass reduced when exposed to crude oil (Brockson and Bailey, 1973). There was reduction in opercular beating rates of pink salmon fry exposed to toluene, naphthalene, and water soluble fractions of Cook Inlet Crude oil (Thomas and Rice, 1979). There is therefore the need for lethal effect of water soluble fraction of Forcados oil on African catfish. Forcados blend (SHELL) contains low molecular weight aromatic hydrocarbons which readily dissolve in water (Laws, 1981).

Clarias gariepinus is a mud fish in water (Laws, 1981). The fish can tolerate low oxygen tension and this enables it to occupy adverse environmental conditions (Tsadu and Adebisi, 1997). It is widely distributed throughout Africa. *C. gariepinus* (Teugels, 1986) is named after its common locality, the Garipep river (Orange river) in South Africa. It is economically important and highly valued fish.

Materials and Methods

Juvenile (mean weight $13.5 \pm 0.48\text{g}$) and fry ($2.13 \pm 0.64\text{g}$) *C. gariepinus* were procured from Biya Fish Farm, Ibadan and brought to Fisheries research Laboratory, Department of Zoology, University of Ibadan in an open circular bowl. These fish were held in glass chambers (40l capacity) and fed daily with compounded feed of 30% crude protein at 2% of their body weight for two weeks acclimation. Only 2% fish mortality was recorded during acclimation.

Preparation of the WSF of Forcados crude oil was carried out using the method of Reish and Oshida (1986). The following WSF concentrations (1.0, 2.0, 4.0, 8.0 and 12.0mg/L for fry and 1.5, 3.0, 6.0, 10.0 and 15.0mg/L for juveniles) were used for the toxicity test. Ten (10) six sets of chambers (five different concentrations and a control). The fish were starved throughout the experiment.

Physico-chemical characteristics of the test concentrations were maintained at a dissolved oxygen of $6.1 \pm 0.32\text{mg/L}$, pH at 6.9 ± 0.11 and water temperature at $26.5 \pm 0.81^\circ\text{C}$ during the experiment.

Fish mortality was recorded at 24-h intervals for 96-h duration. Opercular beat and pulsation level per minute and the behaviour of experimental fish per concentration were recorded. The LC₅₀, percentage mortality and 95% confidence limited probit kill were determined.

Result

The little fluctuation in the physicochemical characteristics of the test media during the toxicity test produced mild effect on fish mortality. Both fish stages exhibited similar stressful behaviour like spiral swimming, spontaneous air gulping, loss of balance and jumping, but at different concentrations and duration of exposure.

Mortality of juvenile and fry *C. gariepinus* at varying concentration is shown in Table 1. The mortality of CGJ after 96 hours varies between 100% in 15.0mg/l and 10% in 30mg/l. There was not mortality in the lowest concentration (1.5mg/l) and control (0.0mg/l). The lethal time for 50% mortality (LT₅₀) varies between 30 and 48 hours in concentrations 15.0 and 10.0mg/l respectively, while the LC₅₀ is 7.4mg/l (Fig. 1). The 96-h mortality of GCF varies between 100% in 12.0mg/l and 20% in 2.0mg/l (table 1). Zero mortality was recorded in the lowest concentration (1.0mg/l) and control. The time for 50% mortality (LT₅₀) varies between 26, 40 and 50 hours in concentrations 12.0, 8.0 and 4.0mg/l respectively. The 96-h LC₅₀ is 4.0mg/l (Fig. 1).

Table 2 shows the effect of different concentrations of WSF of Forcado oil on the pulsation of both *C. gariepinus* juvenile (CGJ) and fry (CGF).

Pulsation was 48 and 50 counts at the initial and control hours for the juveniles and fries respectively. At the lowest concentration, pulsation counts increased in the CGJ from 48 counts at the initial hour to 56 at 72nd hour, but reduced to 52 at 96th hour. At 6.0mg/l concentration, pulsation was 66 counts at 48th hour, but decreased to 47 at 96th hour. At the highest concentration, pulsation was highest (48 counts) only at the initial hour and decreased to the least count (38) at the 48th hour. Similarly, pulsation in CGF also increased at the lowest concentration from 50 counts at initial hour to 57 at 72nd hour, but decreased to 44 at 96th hour. The highest pulsation of 67 counts occurred in 2.0mg/l at 72nd hour and dropped to 41 at 96th hour, while the least pulsation (33 counts) was recorded in 12.0mg/l at 48th hour.

Figure 2 shows the effects of different concentrations of the oil on the opercular beats of the test fish (CGJ and CGF). No significant difference ($P > 0.05$) in the opercular beats at the initial time of the experiment in all the concentration. However, opercular beats increased with concentrations and duration of exposure. The values recorded for CGJ varied between 74 beats in 15.0mg/l and 50 in 1.5mg/l at 24th hour, between 66 in 10.0mg/l and 30 in 15.0mg/l at 48th hour, between 58 in 10.0mg/l and 55 in 1.5mg/l at 72nd hour and finally between 70 beats in 3.0mg/l and 50 beats in 10.0mg/l at 96th hour. The control opercular beats was 45 from the initial to the 96th hour. The values recorded for CGF varied between 68 beats in 12.0mg/l and 48 in 1.0mg/l at 24th hour, between 62 in 8.0mg/L and 25 in 12.0mg/L at 48th hour between 52 in 1.0mg/L and 53 in 8.0mg/L at 72nd hour and finally between 60 beats in 1.0mg/L and 48 beats in 8.0mg/L at 96th hour. The control opercular beats was 46 from the initial to the 96th hour.

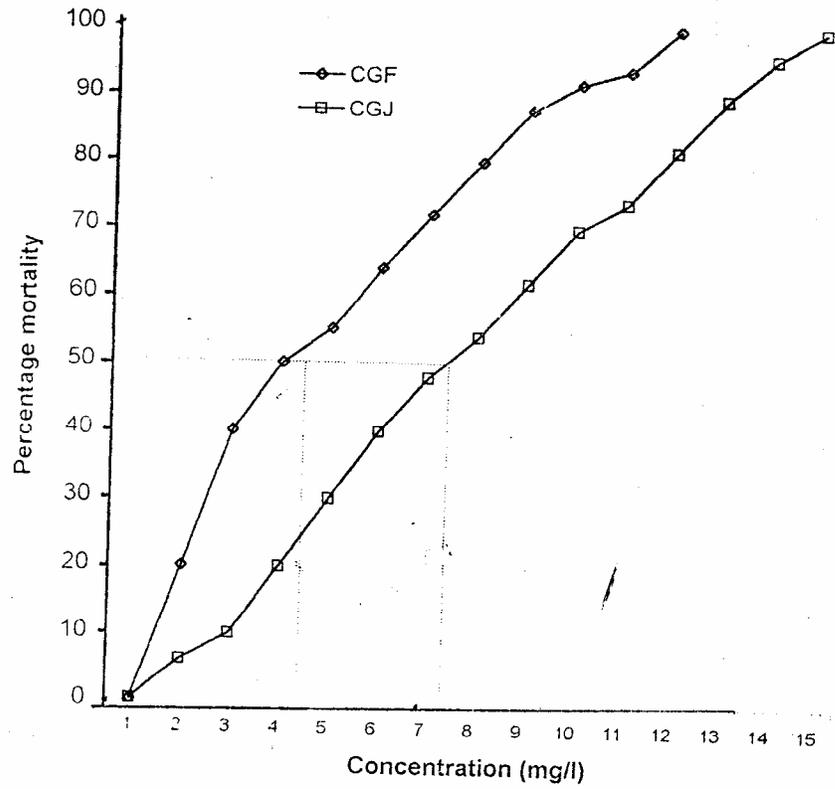


Fig. 1: 96-h percentage mortality curve of *Clarias gariepinus* juvenile (CGJ) and fry (CGF) exposed to lethal concentrations of water soluble fractions (WSF) of Forcados crude oil with LC50s calculated graphically.

Table 1: Mortality of *Clarias gariepinus* juvenile (CGJ) and fry (CGF) in different concentrations of water soluble fractions (WSF) of Forcados crude oil over a 96-h period.

Concentration (mg/L)	No. of fish	Mortality after 96-h		Percentage Mortality		Cumulative Frequency (C.F.)		Percentage C.F.		Time for 50% Mortality	
		CGJ	CGF	CGJ	CGF	CGJ	CGF	CGJ	CGF	CGJ	CGF
0.0	10	0	0	0	0	0	0	0	0	0	0
1.0	10	-	0	-	0	-	0	-	0	-	0
1.5	10	0	-	0	-	0	-	0	-	0	-
2.0	10	-	2	-	20	-	2	-	8.0	-	0
3.0	10	1	-	10	-	1	-	4.5	-	0	-
4.0	10	-	5	-	50	-	7	-	28	-	52
6.0	10	4	-	40	-	5	-	22.7	-	0	-
8.0	10	-	8	-	80	-	15	-	60	-	40
10.0	10	7	-	70	-	12	-	54.5	-	48	-
12.0	10	-	10	-	100	-	25	-	100	-	26
15.0	10	10	-	100	-	22	-	100.0	-	30	-

Table 2: Effect of different concentrations of WSF Forcados crude oil on pulsation counts of *Clarias gariepinus* juvenile (CGJ) and fry (CGF) per minute.

Exposure Period	CONCENTRATIONS (mg/L)											
	CGJ						CGF					
	0.0	1.5	3.0	6.0	10.0	15.0	0.0	1.0	2.0	4.0	8.0	12.0
Initial	48	48	48	48	48	48	50	50	50	50	50	50
24-h	48	48	51	52	64	46	50	51	52	53	55	42
48-h	48	51	66	56	51	38	50	51	58	51	46	33
72-h	48	56	68	60	44	-	50	57	67	48	39	-
96-h	48	52	51	47	41	-	50	44	41	37	35	-

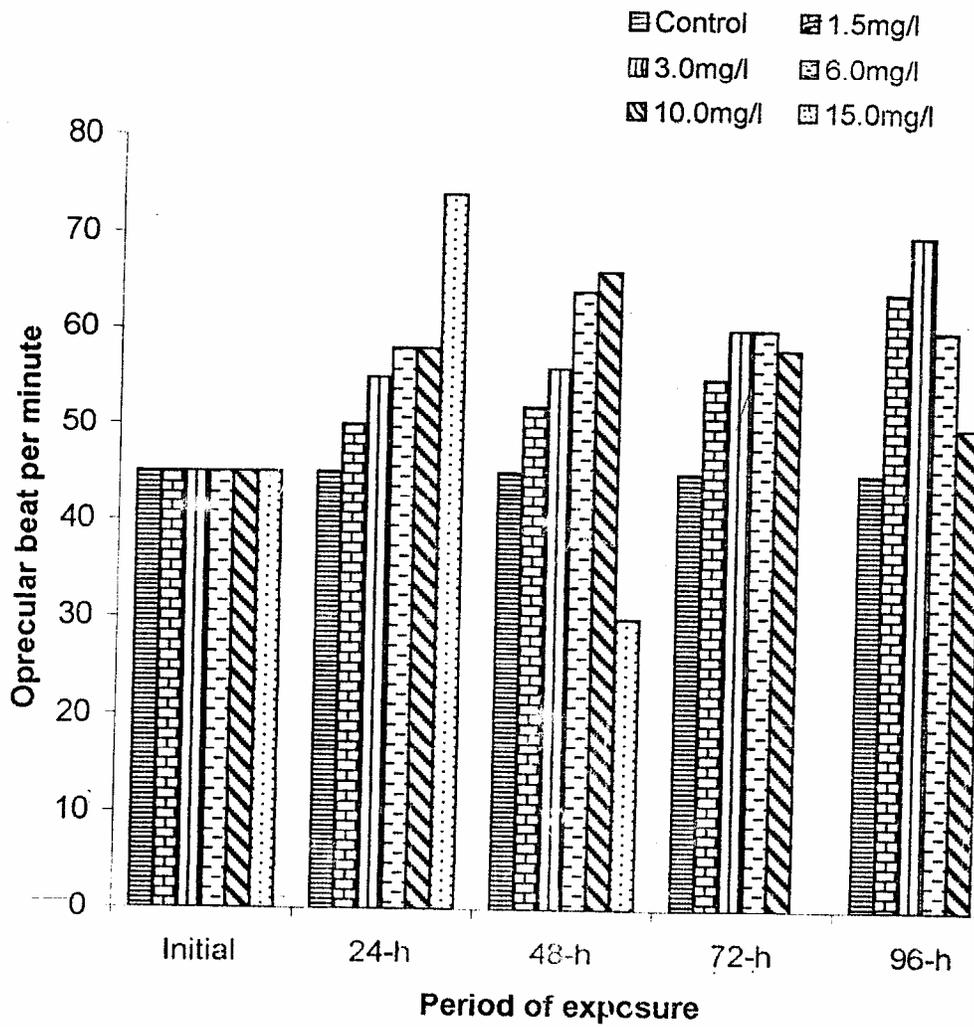


Fig. 2: Effect of different concentrations of water soluble fractions (WSF) of Forcados crude oil on opercular beats of *Clarias gariepinus* juvenile (CGJ).

Discussion

Small increases in temperature reduced the oxygen carrying capacity of the water and was shown to increase the sensitivity to oil of *C. gariepinus*. The stressful behaviours of the CGJ and CGF confirm their increased sensitivity to Forcados oil and which may result to respiratory impairment in the fish. Mitchell and Bennett (1972), Brungs *et al.*, (1978), Steels (1983) and Fafioye and Jeje (In Press) observed similar stressful behaviours in their experimental fishes subjected to varying toxicants. Also, oil on the surface of the test chambers may prevent gaseous diffusion, leading to a build-up of carbon dioxide in the water, causing oxygen stress in *C. gariepinus*.

The high mortality rates of fish at the highest concentrations of the W.S.F. of Forcado oil show that the higher the concentration, the higher the mortality at a given time. Mortality rate of CGF is higher than CGJ in similar concentrations to suggest that CGJ is more tolerant (resilient) to the various concentrations. Also, the 96-h LC₅₀ in CGF is lower (4.0mg/L) than CGJ (7.4mg/L). Similarly the LT₅₀ in CGF is lower (26 hours) in the highest concentration (12.0mg/L) than CGH (30 hours in the highest concentration (15.0mg/L). These findings agree with the results obtained by Abdelmoneim (1995) that the most sensitive fish stage is the fry. However the toxic effect of Forcados oil may be attributed to its active ingredient aromatic hydrocarbons. Edward (1981), Green and Trett (1981), broman *et al.*, (1991) and Fafioye and Owa (In press) reported crude oil containing aromatic hydrocarbons as most toxic and most soluble.

The changes in pulsation at different concentrations signify that pulsation is affected by concentration. The lowest pulsation count at which 100% mortality was recorded in the highest concentrations may imply that lethal pulsation level for CGJ and CGF is any count below 38 and 33 respectively. This lethal pulsation count is realistic by the fact that other concentrations whose pulsation counts never fall below 40 did not obtain total mortality. Similarly concentrations whose pulsation counts never fall below 50 never received any death. based on pulsation counts, the toxicity of WSF of Forcados oil may be grouped into lethal concentration level with pulsation lying between 33 and 49, and non-lethal with pulsation lying above 50.

The observed high level of opercular beats of CGJ in 15.0mg/L and 3.0mg/L and CGF in 12.0mg/L and 2.0mg/L at 24th and 96th hour respectively show that opercular beats are affected by both concentration and duration of the WSF of Forcados oil on the fish. This may also account for respiratory impairment and actions leading to lethal condition. While very low opercular beats at 48th hour in 15.0mg/L (CGJ) and 12.0mg/L (CGF) further confirms the stressful and lethal conditions.

The tolerance of both fish stages of development after 120th hour may be suggested that the toxicant lost its potency due to adsorption and/or volatility of the active ingredient and that the organisms's exudes detoxified the toxicant. Mitchell and Bennett (1972), Thomas and Rice (1979), Hedtke and Puglisi (1982) and Omoregie *et al* (1997) obtained the same result in their experimental fish. As a whole, the toxicity of WSF of Forcados oil becomes lethal only when the concentration is high to compensate for its active ingredients loss and detoxification. However the accumulated effect of this toxicant is not investigated here, but it is suggested that incessant oil spillages into the aquatic environment should be stopped to avert any hazardous bio-magnification effect in fish and other aquatic organisms.

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