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Evaluation of the Effect of Short-Term Cadmium Exposure on Brackish Water Shrimp-*Palaemonetes africanus*

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ABSTRACT: This study evaluated the effect of short-term cadmium exposure on brackish water shrimp-*Palaemonetes africanus*. Tests were carried out by exposing the shrimps to the test solutions containing various concentrations of the cadmium (0, 0.1, 1.0, 4.0, 6.0, 8.0, 10.0, and 100.0) mg/l using the semi-static agitation test procedure. Mortality was recorded at 2, 4, 6, 8, 10, 12, 14 and 16 hours exposure periods. Test results indicated that the brackish water juvenile shrimp, *Palaemonetes africanus* were sensitive to the cadmium solution especially at concentration above 4.0mg/l. Though no death was recorded after 16hrs for 0.1mg/l, 1.0mg/l and 4.0mg/l respectively, however, for 6mg/l, 60% mortality was recorded after 4hrs and 100% after 8hrs, for 8mg/l, 20% mortality was recorded after 2hrs, 60% after 4hrs, 80% after 6hrs and 100% mortality after 8hrs respectively, for 10mg/l, 40% mortality was recorded after 2hrs and 100% after 4hrs and 100% mortality was recorded for 100mg/l after 2hrs. The LC_{50} value calculated using Arithmetic Method of Karber was 5.0mg/l. It is therefore evident that the effects of acute toxicity of cadmium are concentration-related; the greater the concentration, the greater the effect. @ JASEM

Cadmium is highly toxic to wildlife; it is cancer-causing and teratogenic and potentially mutation-causing, with severe sub-lethal and lethal effects at low environmental concentrations (Eisler 1985a). It is associated with increased mortality, and it affects respiratory functions, enzyme levels, muscle contractions, growth reduction, and reproduction. It bio-accumulates at all trophic levels, accumulating in the livers and kidneys of fish (Sindayigaya, *et al*. 1994; Sadiq, 1992). Crustaceans appear to be more sensitive to cadmium than fish and mollusks (Sadiq 1992). Metal accumulation in tissues of fish is dependent upon exposure dose and time as well as other factors such as temperature, age of fish, interaction with other metals, water chemistry and metabolic activity of the fish (Pagenkopf 1983, Heath 1987, Goyer 1991).

Studies revealed effects of the combination of cadmium and zinc in an aquatic environments (Kargin 1996). Zinc has an antagonistic and protective action in the uptake and toxic effects of cadmium, probably because of Zn-induced synthesis of metallothionein that detoxifies cadmium by firmly binding this metal (Hemelmad, *et al*., 1987). Similar effects have been observed in the interaction of heavy metals such as selenium and mercury (Lucu and Skreblin, 1981).

Generally, the effect of heavy metals on aquatic organisms ranges from slight reduction in growth rate to death. Severe imbalances in concentration can lead to death, while marginal imbalances may cause poor health and retarded growth. Concentrations determined to be lethal in the laboratory tests occur commonly in nature (Abowei and Sikoki, 2005). Research has shown that metal accumulation is more rapid than metal elimination, probably due to the presence of metal binding proteins in tissues and this has contributed to their deleterious effect. This is because several factors influence the elimination of metals from the tissues of aquatic animals. These include time, temperature, interacting agents, age of fish, metabolic activity of animals and biological half lives of metals (Larson *et al.,* 1985; Heath, 1987, Rao *et al.*, 1988, Douben, 1989, Woo *et al.*, 1993, Kargm 1996, Nielsen and Andersen 1996). Elimination routes of metals from fish are generally through gill, bile, urine, skin, and via mucus (Varanasi and Markey 1978, Heath 1987).

The objective of this work is to determine the potential acute toxicological effect of different cadmium concentrations on brackish water shrimp-*Palaemonetes africanus.* This will help to determine the concentration-time effect and LC_{50} value as applicable.

MATERIALS AND METHODS

Semi- static Bioassay Techniques:

Acute toxicity tests were carried out with aquatic organisms by exposing them (test organisms) to test solutions containing various concentrations of the test sample, using the semi-static agitation test procedure as recommended by Department of Petroleum Resources (DPR, 2002).

Sampling Test Organisms: Brackish water juvenile shrimp, *Palaemonetes africanus* were collected from the brackish water at Eagle Island Waterside, Port Harcourt, Nigeria. Juvenile shrimps were collected

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with the aid of sieves of appropriate mesh size during spring tide. They were transferred into 10L coolers containing the habitat water (APHA, 1998).

Table 1: Average Weight of Test Organisms

S/N	Name of Organism	Type of Habitat	Weight of Organism
1.	*Palaemonetes africanus*	Brackish water	90 ± 5mg

Table 2: Average Length of Test Organisms

S/N	Name of Organism	Type of Habitat	Weight of Organism
1.	*Palaemonetes africanus*	Brackish water	2.5 ± 0.5cm

Acclimatization Procedure: All test organisms were first acclimatized for ten days at room temperature 28 ± 2^{o}C. They were acclimatized in dark glass tanks in which air (oxygen) was continuously bubbled into, through an aerator. They were also fed with fish feed obtained from the Institute of Fisheries at Aluu near University of Port Harcourt during the period of acclimatization. The water in the acclimatization units was replaced with the fresh water from the organism's habitat daily. There was controlled lighting system, as 12hours of light and 12hours of darkness was employed (DPR, 2002).

Selection of Test Organism: Twenty test organisms of fairly equal size were randomly caught with a hand net from acclimatization tanks and carefully transferred into the test vessel. The organisms were not touched with hand during the selection so as to avoid stress due to handling. Only healthy and active test organisms were selected.

Choice of Bioassay Tanks: The bioassay containers were made of dark glass of 30cm × 21cm × 25cm dimension. These were constructed with removable stands for easy cleaning and movement. The tanks were washed with detergent, rinsed with distilled water and dried overnight to avoid contamination.

Range Finding Test: Range Finding Test was carried out to establish a preliminary working range by obtaining the least concentration that gives no effect and the minimum concentration that gives 100% death. Test design incorporated multiple, widely spaced concentrations with single replicates. Exposure times were 4hr., 8hr., 24hr., and 48hr.

Source of Cadmium Standard: Cadmium standard was gotten from Buck Scientific, PURO-GRAPHIC^{tm} Standards, East Norwalk, CT 06855, Canada.

Test Medium: Four different concentrations of the test sample (0.1mg/l, 1.0mg/l, 4.0mg/l, 6.0mg/l, 8.0mg/l, 10.0mg/l, and 100.0mg/l) were prepared using habitat water of the particular organism as diluent. This followed after a preliminary range finding test. The corresponding aliquots were added to the test vessels constructed of glass. The aliquots were first stirred for 5mins and subsequently at 4 hourly intervals.

Serial Dilutions Used: For 100mg/l, about 100ml aliquot of the cadmium standard concentration was measured and diluted to 1litre with the brackish water (diluent). For 10mg/l, 8mg/l, 6mg/l, 4mg/l and 1mg/l, 0.1mg/l) concentrations, about 10ml, 8ml, 6ml, 4ml, 1ml and 0.1ml aliquots of the cadmium standard concentrations respectively were diluted to 1litre with the diluent.

Twenty test organisms were used in each concentration. Healthy, active test organisms were carefully introduced into bioassay vessels representing different concentrations. Three hundred and sixty test organisms (*P. africanus*) consisting of twenty organisms per concentration made up of six concentration levels. Controls containing dilution water, and twenty test organisms were prepared without the toxicant and this served as the control. Each of the test concentrations was labeled appropriately. After each day, the media were replaced with fresh one. Dead organisms were also removed at the end of each exposure period. This was done to avoid contamination of live organisms by bacteria from dead decaying organisms. Mortality was recorded at 2, 4, 6, 8, 10, 12, 14 and 16 hour exposure periods (Finney, 1978 and Sprague, 1973).

LC_{50} Determination: The number of dead fishes per group was recorded against the time of their death in a tabular form as specified by Sprague (1972). The data obtained was used to calculate the median lethal concentration (LC_{50}) of the Cadmium solution on *P. africanus* using Arithmetic Method of Karber

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PHYSICO-CHEMICAL ANALYSIS:

- **pH, Temperature, Conductivity, TDS and Dissolved Oxygen**:

These chemical properties were determined electrometrically with a multi- parameter data logger (Hanna model HI991300).

- **Salinity**:

Salinity was determined titrimetrically in accordance with APHA 2520A.

- **Alkalinity**:

Alkalinity was determined in accordance with ASTM D 1067B. This method involved titration of 100ml of sample against 0.01M HCl with the addition of 2-3 drops of phenolphthalein indicator.

- **Hardness**:

Total hardness in water was determined in accordance with APHA 2340C. This method involved the addition of a standard EDTA solution to 50ml of the sample in 250ml conical flask. The mixture was stirred continuously until a reddish tinge disappeared and there was a color change to sky blue.

RESULTS AND DISCUSSION

Table 3: Some Physicochemical Characteristics of the Brackish Water

Parameter	Test Result
pH	6.61
Temperature (°C)	26.5
Electrical Conductivity (μScm^{-1})	1670
Total Dissolved Solids (mg/l)	843
Dissolved Oxygen (mg/l)	7.97
Total Hardness (mg/l)	740
Alkalinity (mg/l)	60

Table 4: Concentration-Time Effect of Cadmium on Brackish water Shrimp (*Palaemonetes africanus*)

	% Mortality						
Concentrations (mg/l)	0.1	1.0	4.0	6.0	8.0	10.0	100.0
2Hours	0	0	0	0	20	40	100
4Hours	0	0	0	60	60	100	100
6Hours	0	0	0	60	80	100	100
8Hours	0	0	0	100	100	100	100
10Hours	0	0	0	100	100	100	100
12Hours	0	0	0	100	100	100	100
14Hours	0	0	0	100	100	100	100
16Hours	0	0	0	100	100	100	100

Table 5: LC_{50} Determination Based on Arithmetic Method of Karber.

Concentration (mg/l)	Concentration Difference	Number Alive	Number Dead	Mean Death	Mean Death Dose Difference
0 (Control)	-	20	0	-	0
0.1	0.1	20	0	0	0
1.0	0.9	20	0	0	0
4.0	3.0	0	0	0	0
6.0	2.0	0	20	10	20
8.0	2.0	0	20	20	40
10.0	2.0	0	20	20	40
100.0	90.0	0	20	20	1800
					∑ = 1900

$$LC_{50} = LC_{100} - \frac{\sum \text{Conc. Diff} \times \text{Mean Death}}{\text{No. of org. per group}}$$

$$= 100 - \frac{1900}{20}$$

$$LC_{50} = 100 - 95 = 5.0 \text{ (mg/l)}.$$

Table-3 presents some physico-chemical characteristics of the brackish water. As indicated the pH was 6.61, Temperature (26.5°C), Electrical conductivity (1670μScm^{-1}), Total dissolved solids (843mg/l), Dissolved oxygen (7.97mg/l), Total hardness (740mg/l) and Alkalinity (60.0mg/l).

The dose - time effect of the different concentrations of Cadmium on the brackish water shrimp - *Palaemonetes africanus* is presented in table 4. Test results indicated that at 0.1mg/l and 1.0mg/l and 4.0mg/l concentrations respectively, there was no death of the shrimps that is, mortality rate was 0%. For 6mg/l after 2hrs, mortality rate was 0%, after 4hrs and 6hrs respectively; mortality rate was 100% after 8hrs. For 8mg/l, 20% mortality rate was noticed after 2hrs, 60% mortality rate after 4hrs and 80%

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after 6hrs and 100% mortality after 8hrs. It was observed that while 40% mortality rate was recorded after 2hrs and 100% after 4hrs for 10mg/l concentration, 100% was observed after 2hrs for 100mg/l. Research has shown that chemicals at a given concentration which would have been harmless as a stand alone may cause deleterious effects by interacting in the general milieu of contaminated waters (Joel, *et al.,* 2009). Therefore, the lower concentrations as seen in this test that resulted to no death of the test organisms may have toxic effect if there are other contaminants in the aquatic habitat.

As effluent from many sources enters natural waters, the negative impact on the aquatic ecosystem is due to a mixture of metals, rather than individual component metals. When metal mixtures are discharged into the environment they may show a number of effects, which are synergistic, antagonistic or additive in nature (Grobler *et al.,* 1989).

The LC_{50} value calculated using Arithmetic Method of Karber was 5.0mg/l (Table 5). From the standard toxicity rating of chemicals by Sprague, the test solution was slightly toxic (Sprague, 1973).

Conclusion: The semi – static bioassay revealed that the higher the concentration of the cadmium solution, the higher the % mortality rate. It is therefore evident that the effects of acute toxicity of cadmium are concentration-related; the greater the concentration, the greater the effect.

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