Establishment of the Toxicity Ranking Order of Heavy Metals and Sensitivity Scale of Benthic Animals Inhabiting the Lagos Lagoon

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Abstract

A toxicity ranking order/scale for some pertinent metals (zinc, cadmium, lead, copper and mercury) in the Lagos lagoon was established by testing metallic salts against three typical lagoon animal species. Within limits of slight variations, Hg was found to be most toxic of the test metal compounds followed by Cd, Cu, Zn and Pb in a decreasing order of toxicity against *Tympanotonus fuscatus, Clibanarius africanus* and *Sesarma huzardi.* On the basis of the computed toxicity factors (96 h LC_{50} ratios), Hg was found to be 5-3810 times more toxic than the other test metals against the exposed animals. The establishment of the sensitivity scale for the test animals revealed that, generally, *C. africanus* was the most sensitive animal followed by *T. fuscatus* and *S. huzardi* in a decreasing order of sensitivity. On the other hand, with regards to tests involving the Hg compound, *S. huzardi* was found to be the most sensitive test animal contrary to the sensitivity ranking order established for other metals tested. The importance of the toxicity ranking order and sensitivity scales in choosing heavy metal based raw materials and establishing safe limits of pollutants is discussed.

Introduction

Due to the high risk of biological damage posed by heavy metals from industrial and domestic sources over long periods in aquatic and terrestrial ecosystems, considerable research on metal pollution has taken place in the industrialized countries of Europe, America and Asia. For example, Krishnaja *et al.* (1987) evaluated the acute toxicity of some heavy metal salts against the intertidal crab, *Scylla serrata,* in India. They observed that phenyl mercuric acetate with the LC_{50} value of 0.54 mg/l was the most toxic and lead nitrate with LC_{50} (370 mg/l) the least toxic. Khangarot *et al.* (1982) also demonstrated that Hg^{2+} was the most toxic where different metallic compounds were tested singly against the freshwater pulmonate snail, *Lymnaea acuminata,* in Indonesia.

However, research work in Nigeria on the toxicity of heavy metals is scanty and recent, with considerable information gaps particularly on the biological effects of these pollutants on local plant and animal species (Oyewo, 1998). Therefore, it is necessary to establish dose-response relationships where locally available animals/plants species are exposed to varying concentrations of heavy metals, in order to extrapolate lethal (LC_{50}/LD_{50} values) and sublethal concentrations. These derived toxicity indices are used as a tool in heavy metal pollution identification, control and management, particularly in the establishment of toxicity scales or ranking orders in addition to the establishment of environmental safety limits of the pollutants in the environment (Mason, 1992).

In Nigeria, there is yet no relevant and nationally-generated safe limits of pollutants for the protection of aquatic organisms. What is available is an adopted interim industrial effluent limitation guideline (FEPA, 1991) which most earlier workers have described as an importation from western countries, particularly the USA and United Kingdom (Singh *et al.*, 1995). This observation is quite valid because there is a general lack of information on the toxic levels of pollutants against locally occurring animals and plants specie (FEPA, 1991). These toxicity data are usually required in all schemes used for establishing environmental safe limits or as the basis of standards aimed at protecting aquatic lives. The importance and lack of these toxicity data in Nigeria prompted this present work, the objective of which is to determine the relative toxicity of heavy metals (zinc, cadmium, lead, copper and mercury) found to be very common in the Lagos lagoon (Oyewo, 1998) to three benthic animals, *Tympanotonus fuscatus, Clibanarius africanus* and *Sesarma huzardi* of the Lagos lagoon.

Materials and methods

Test animals

(a) *Tympanotonus fuscatus* var. *radula* L. (Periwinkle) (Mollusca; Gastropoda, Mesogastropoda, Potamididae).

(b) *Clibanarius africanus* (Aurivillus) (Hermit crab) (Arthropoda; Crustacea, Decapoda, Paguridae).

(c) *Sesarma huzardi* (Desmaret) (Mangrove crab) (Arthropoda; Crustacea, Decapoda, Grapsidae).

Laboratory animal cultures, acclimatisation and selection of test animals for bioassays

All the test animals were collected from the Ikoyi Experimental Fish Farm of the Nigerian Institute for Oceanography and Marine Research (NIOMR) because the ponds in the NIOMR fish farm had remained protected over the years, with only limited and controlled connection with the open Lagos lagoon system. Therefore, the animal stocks in the farm were shielded from the full impact of pollutants in the open lagoon and were of known stock history. The test animals were brought to the laboratory and kept in holding tanks with a thin layer of sediment for 5–6 days to allow them acclimatise to laboratory conditions (relative humidity-70 ± 2%; temperature - 26 ± 2 °C; salinity- 16 psu) before using them in bioassays. The test animals were acclimatized gradually to the salinity condition of 16 psu to prevent a sudden osmotic change. This salinity condition was used in all bioassays in order to standardize and simulate a typical brackish water medium.

Test chemicals

The heavy metal salts used in this study were analar grades of Fisons laboratory reagents and consist of the following:

(a) Copper as $CuS0_4.5H_20$
(b) Zinc as $ZnC0_3.3H_20$
(c) Lead as $Pb(N0_3)_2$
(d) Cadmium as $3CdS0_4.8H_20$
(e) Mercury as $HgCl_2$

The choice of heavy metals for this study was based on the available, common and abundant metals from the results of a chemical survey of industrial effluents that are discharged into the Lagos lagoon carried out by Oyewo (1998).

Preparation of test media including application of toxicant

A pre-determined amount of each heavy metal compound was weighed (using an Oertling 30TD top loading balance) out and made up to a given volume to obtain a stock solution of known strength. The resultant stock solution was serially diluted to obtain solutions of required concentrations. The ratio of the sediment to volume of test medium was 100 g to 500 ml for the tests involving *T. fuscatus* and *C. africanus*, and 160 g to 2000 ml for test involving *S. huzardi.*

Assessment of quantal response (mortality)

(a) *T. fuscatus* var. *radula*: This animal was taken to be dead when it was observed to be totally retracted into its shell and failed to emerge or protrude its muscular foot during an observation period of 15 min in untreated dilution water (salinity - 16 psu) in a petri dish; or if the foot was retracted at the start of the observation and the animal failed to respond by withdrawing the foot into the shell on prodding with a glass rod.

(b) *C. africanus* or *S. huzardi*: An animal was taken to be dead if it showed no movement of the body and or appendage, even on prodding with a blunt object or failed to emerge or protrude its legs/body during an observation period of 3 – 4 min in untreated brackish water at salinity of 16 psu placed in an observatory petri dish.

Bioassays

(a) Bioassays with periwinkles *T. fuscatus* var. *radula* and hermit crab *C. africanus*. These were carried out in plastic bowls (bottom diameter – 14.5 cm, top diameter – 21cm and height – 9 cm).

(b) Bioassays with mangrove crab (*S. huzardi*). These were carried out in plastic tanks (35.5 cm × 21 cm × 22 cm).

The ratio of the sediment to volume of test medium was 100 g to 500 ml for the tests involving *T. fuscatus* and *C. africanus*; and 160 g to 2000 ml for test involving *S. huzardi.*

Differential acute toxicity of heavy metals against benthic animal species

Following the establishment of the range of activity of the test compounds in the preliminary range of finding experiments, the test animals were exposed to several concentrations of each heavy metal salt and untreated control as follows:

A) *T. fuscatus*

(a) Cadmium ($3CdS0_4.8H_20$) against *T. fuscatus* at 4, 6, 8, 10, 50, 100, 120, 140, 160 mg/l.

(b) Mercury ($HgCl_2$) against *T. fuscatus* at 1.0, 2.5, 5, 7.5, 10, 12.5, 15, 17.5, 20.0, 22.5, 25.0 mg/l.

(c) Zinc ($ZnC0_3.3H_20$) against *T. fuscatus* at 5, 10, 20, 50, 100, 200, 500, 800, 1000, 1200, 1400, 1600, 1800, 2000, 2200 mg/l.

(d) Lead ($Pb(NO_3)_2$) against *T. fuscatus* at 50, 100, 200, 400, 600, 800, 1000, 1400, 1800, 2000, 2200, 2600, 2800, 3000 mg/l.

(e) Copper ($CuS0_4.5H_20$) against *T. fuscatus* at 10, 15, 30, 40, 50, 60, 70, 80, 90, 100, 120, 140, 160, 180 mg/l.

B) *C. africanus*

(a) Cadmium ($3CdS0_4.8H_20$) at 4, 6, 8, 10,20,40,60,80,100,120,140,160 mg/l.

(b) Mercury ($HgCl_2$) at 0.4, 0.6, 0.8, 1.0, 4.0, 8.0, 10, 20, 40, 60 80, 100, 120 mg/l.

(c) Zinc ($ZnC0_3.3H_20$) at 8, 10, 20, 50, 80, 110, 140, 170, 200, 240 mg/l.

(d) Lead ($Pb(N0_3)_2$) against *C. africanus* at 50, 100, 200, 300, 400, 500, 600, 700, 800, 900, 1000 mg/l.

(e) Copper ($CuS0_4.5H_20$) against *C. africanus* at 1, 4, 8, 10, 20, 40, 60, 80, 100 mg/l

C) *S. huzardi*

(a) Cadmium ($3CdS0_4.8H_20$) against *S. huzardi* at 20, 50, 200, 300, 400 mg/l.

(b) Mercury ($HgCl_2$) against *S. huzardi* at 0.1, 0.5, 1.0, 1.5, 2.0, 2.5 mg/l.

(c) Zinc ($ZnC0_3.3H_20$) against *S. huzardi* at 300, 400, 500, 600, 700, 800 mg/l.

(d) Lead ($Pb\ (N0_3)_2$) against *S. huzardi* at 2000, 2200, 2400, 2600, 2800, 3000 mg/l.

(e) Copper ($CuS0_4.5H_20$) against *S. huzardi* at 100, 200, 300, 400, 500, 600 mg/l.

Mortality assessments were carried out once every 24 h over a 96 h period.

Dose-response data analysis

Toxicological dose-response data involving quantal response (mortality) were analysed by probit analysis (Finney, 1971) based on a computer programme written by Ge Le Pattourel, Imperial College, London, as adopted by Don-Pedro (1989):

Toxicity factor (T.F.) for relative potency measurements, e.g. ratio of 96 h LC_{50} of a compound LC_{50} values at equivalent time intervals =

$$\frac{\text{96 h } LC_{50} \text{ of a compound tested against a species}}{\text{96 h } LC_{50} \text{ of another compound tested against same species}}$$

Results

Physico-chemical conditions in bioassays during toxicity testing

In all bioassays, except where stated otherwise in particular tests, the dissolved oxygen content of test media remained greater than 5.5 mg/l over the 96 h test period during the acute toxicity evaluations. The measured *p*H, temperature and salinity values also remained fairly constant at 7.8±0.2; 26±2 °C and 16.5±1psu, respectively, from the beginning to the end of the bioassays in 96 h. The sediment type used for all bioassays involving *T. fuscatus* was composed of 72.6% clay, 1.1% sand and 26.3% silt. The organic matter content of the soil was about 6.34%. The sediment substrate for all bioassays involving *C. africanus* and *S. huzardi* was found to be composed of 0.6% clay, 94.6% sand and 4.8% clay. The organic matter content was about 0.5%.

Relative acute toxicity tests of heavy metals tested against test animals species

Tympanotonous fuscatus *var.* radula. On the basis of 96 h LC_{50} values, Hg with a 96 h LC_{50} value of 3.685 mg/l was the most toxic metal tested against *T. fuscatus* followed by Cd, Cu, Zn and Pb (96 h LC_{50} = 609.78 mg/l) in descending order of toxicity (Table 1). Mercury was significantly (no overlaps in 95% C.L. of 96 h LC_{50} values) more toxic than each of the other metallic compounds tested against *T. fuscatus*. Computed toxicity factors (96 h LC_{50} ratios) showed that Hg was about 166×, 23×, 11× and 8× more toxic than Pb, Zn, Cu and Cd, respectively, when tested against *T. fuscatus* (Table 1).

Clibanarius africanus. On the basis of 96 h LC_{50} values, Hg with a toxicity value of 0.9 mg/l was the most toxic metal compound tested against *C. africanus* followed by Cu, Cd, Zn and Pb (96 h LC_{50} = 370.76 mg/l) least toxic in descending order of toxicity (Table 2). The Hg compound was significantly (no overlap in 95% C.L. of 96 h LC_{50} value) more toxic than each of the other metallic compounds tested separately against *C. africanus*. Copper was also found to be significantly more toxic than Cd and each of the other metallic compounds. Computed toxicity factors (96 h LC_{50} ratio) showed that the mercury compound was 464×, 37×, 17× and 5× more toxic than Pb, Zn, Cd and Cu, respectively (Table 2).

Sesarma huzardii. On the basis of 96 hLC_{50} values, Hg compound with a toxicity value of 0.69 mg/l was found to be the most toxic metal compound tested against *S. huzardi* followed by Cd, Cu, Zn and Pb (96 hLC_{50} = 2320.13 mg/l) least toxic in a descending order of toxicity (Table 3). Hg

TABLE 1

Relative acute toxicity of selected heavy metal compounds against T. fuscatus

Metals	*LC_{50} $^{+}$C.L. (mg l^{1})*	*LC_{95} $^{+}$C.L. (mg l^{1})*	*Slope±SE**	*DF$^{□}$*	*Probit line equation*	*TF^{6}*
24 h						
Hg	11.764(13.011-10.635)	28.711(36.04-22.81)	4.258±2.06	4	Y=0.442+4.258X	1
Cd	424.33(566.44-317.62)	7968.71(15179-4155)	1.295±0.14	5	Y=1.596+1.295X	36.07
Cu	84.51(89.02-80.24)	139.92(154.8-126.1)	7.54±0.69	5	Y=-9.52+7.54X	7.18
Pb	1144.51(1307.66-1001.61)	4394.51(5664.25-3384.3)	2.824±0.27	6	Y=-3.64+2.824X	97.29
Zn	459.75(581.01-363.70)	5031.73(7839-3202)	1.588±0.13	6	Y=0.773+1.588X	39.08
48 h						
Hg	6.699(8.09-5.52)	29.74(39.34-22.53)	2.55±1.597	4	Y=2.895+2.549X	1
Cd	136.71(194.04-96.11)	5527.02(10753-2852)	1.03±0.09	6	Y=2.807+1.027X	20.41
Cu	69.03(75.21-63.36)	161.62(196.60-132.26)	4.47±0.51	5	Y=-3.213+4.47X	10.3
Pb	922.58(1061.31-801.88)	3331.85(4293.37-2567.8)	2.958±0.29	5	Y=-3.772+2.96X	137.7
Zn	273.79(346.48-216.22)	3141.19(4837.7-2023.36)	1.557±0.13	6	Y=1.205+1.56X	40.87
72 h						
Hg	4.917(5.992-4.018)	30.464(42.09-21.85)	2.08±0.19	5	Y=3.559+2.08X	1
Cd	57.57(82.64-4012)	2770.23(5848.7-1345.1)	0.98±0.08	6	Y=3.274+0.981X	11.71
Cu	53.51(61.00-46.94)	161.62(196.60-132.26)	2.86±0.32	5	Y=0.064+2.856X	10.88
Pb	687.34(820.15-575.91)	3331.85(4293.37-2567.8)	2.355±0.27	4	Y=-1.682+2.355X	139.8
Zn	155.47(200.85-120.33)	3141.19(4837.68-2023.4)	1.432±0.12	6	Y=1.862+1.432X	31.62
96 h						
Hg	3.685(4.55-2.99)	24.684(38.73-16.47)	1.998±0.2	4	Y = 3.68+1.998X	1
Cd	28.246(41.24-19.37)	1353.38(3216-599.90)	0.982±0.1	5	Y = 3.575+0.982X	7.7
Cu	39.254(44.68-34.48)	141.992(183.14-109.52)	2.955±0.3	5	Y = 0.29+2.955X	10.7
Pb	609.779(731.50-508.16)	3106.18(4403.9-2173.6)	2.333±0.3	4	Y =-1.499+2.333X	165.5
Zn	83.167(110.51-62.72)	1229.396(2256.56-674.6)	1.410±0.1	5	Y = 2.292+1.410X	22.6

$^{+}$CL = 95% Confidence limit
SE^{s} = Standard error
DF$^{□}$ = Degrees of freedom
TF^{6} = Toxicity factor = $\frac{\text{24/48/72/96 h } LC_{50} \text{ value of other metals}}{\text{24/48/72/96 h } LC_{50} \text{ value of most toxic (Hg) metal}}$

TABLE 2

Relative acute toxicity of selected heavy metal compounds against C. africanus

Metals	LC_{50}, $^{+}$*C.L. [mg* l^{-1}*]*	LC_{95}, $^{+}$*C.L. [mg* l^{-1}*]*	*Slope±SE*^{s}	*DF*$^{\square}$	*Probit line equation*	*TF*^{6}
24 h						
Hg	13.466(20.25-9.001)	321.27(771.55-147.05)	1.198±0.017	5	Y=3.64+1.198X	1
Cd	72.75(84.46-62.65)	231.95(323.83-165.40)	3.276±0.5	4	Y=-1.10+3.276X	5.4
Cu	33.996(41.78-27.68)	123.93(184.34-83.45)	2.937±0.365	3	Y=2.268+2.422X	2.5
Pb	380.75(132.24-105.87)	1011.79(1275.25-798.6)	3.887±0.47	5	Y=-5.031+3.887X	28.3
Zn	118.324(132.24-105.87)	251.604(316.0-199.6)	5.036±0.64	3	Y=-5.439+5.04X	8.8
48 h						
Hg	4.014(5.376-3.008)	31.466(57.54-18.85)	1.845±0.21	4	Y=3.886+1.845X	1
Cd	36.51(45.20-29.49)	176.88(304.12-103.01)	2.407±0.39	3	Y=1.239+2.407X	9.1
Cu	14.332(19.455-10.559)	150.898(311.35-75.998)	1.614±0.22	4	Y=3.134+1.614X	3.6
Pb	292.11(338.82-251.81)	810.92(1097-596.5)	3.721±0.5	3	Y=-4.174+3.721X	72.8
Zn	79.885(92.68-68.84)	227.62(299.16-172.4)	3.628±0.48	4	Y=-1.902+3.628X	19.7
72 h						
Hg	1.668(2.24-1.27)	10.324(21.37-6.34)	2.08±0.27	3	Y=4.537+2.084X	1
Cd	21.605(27.64-16.93)	113.296(197.57-65.86)	2.293±0.31	3	Y=1.94+2.293X	12.95
Cu	8.068(11.330-5.734)	93.67(219.67-43.30)	1.549±0.24	3	Y=3.595+1.549X	4.8
Pb	241.83(279.3-209.32)	607.16(779.4-470.81)	4.127±0.6	3	Y=-4.836+4.127X	14.98
Zn	55.35(67.50-45.37)	249.72(379.50-163.87)	2.521±0.34	4	Y=0.605+2.521X	33.2
96 h						
Hg	0.8(0.97-0.67)	2.75(5.45-1.80)	3.077±0.6	3	Y=5.298+3.077X	1
Cd	13.423(16.87-10.71)	64.422(112.04-37.68)	2.422±0.4	3	Y=2.268+2.422X	16.8
Cu	4.342(6.82-3.20)	33.326(63.70-18.48)	1.864±0.3	3	Y=3.811+1.864X	5.4
Pb	370.76(447.572-307.31)	1823.69(3164.9-1044.2)	2.385±0.4	4	Y=-1.127+2.385X	463.5
Zn	29.801(36.47-24.35)	127.931(186.85-87.64)	2.608±0.3	4	Y=1.156+2.608X	37.3

$^{+}$CL = 95% Confidence limit

SE^{s} = Standard error

DF$^{\square}$ = degrees of freedom

TF^{6} = Ttoxicity factor = $\frac{\text{24/48/72/96 h LC}_{50}\text{ value of other metals}}{\text{24/48/72/96 h LC}_{50}\text{ value of most toxic(Hg) metal}}$

was significantly (no overlap in 95% C.L. of 96 h LC_{50} values) more toxic than each of the other metallic compounds tested separately against *S. huzardi* (Table 3). Computed toxicity factors (96 h LC_{50} ratio) showed that Hg was 3810×, 864×, 398× and 202× more toxic than Pb, Zn, Cu and Cd respectively (Table 3).

Relative susceptibility/sensitivity of test animal species to heavy metals

Cadmium (Cd). Clibanarius africanus was the most susceptible/sensitive test animal to Cd salt, with a response level based on 96 h LC_{50} value of 13.423 mg/l followed by *T. fuscatus* and *S. huzardi* (the most tolerant species, 96 h LC_{50} = 122.998 mg/l) in a descending order of susceptibility (Table 4). On the basis of the computed susceptibility factors, *C. africanus* was found to be about two times and nine times more susceptible to the toxic effect of Cd than *T. fuscatus* and *S. huzardi*, respectively (Table 4).

Copper (Cu). Clibanarius africanus was the most susceptible/sensitive test animal to Cu salt, with a response level based on 96 h LC_{50} value of 4.342 mg/l

TABLE 3

Relative acute toxity of selected heavy metal compounds against S. huzardi

Metals	*LC_{50} $^{+}$C.L. (mg l^{-1})*	*LC_{95}, $^{+}$C.L. (mg l^{-1})*	*Slope±SE$^{\leq}$*	*DF$^{\square}$*	*Probit line equation*	*TF^{6}*
24 h						
Hg	1.707(2.084-1.402)	4.424(9.289-2.553)	3.991±1.2	2	Y=4.073+3.991X	
Cd	352.24(408.36-303.86)	606.89(899.81-408.27)	6.98±2.1	1	Y=-12.79+6.98X	
Cu	463.41(593.54-361.98)	1443.60(3118.6-666.25)	3.34±0.99	2	Y=-3.913+3.343X	
Pb	2518.89(2615.1-2426.3)	2965.4(3216.8-2731.07)	23.28±4.99	2	Y=-74.17+23.28X	
Zn	794.31(880.53-716.56)	1323.29(1752.3-716.56)	7.44±1.7	3	Y=-16.59+7.44X	
48 h						
Hg	1.456(1.907-1.118)	4.648(10.973-2.878)	3.273±0.75	2	Y=4.466+3.273X	
Cd	275.57(348.72-217.83)	836.02(1627.7-428.30)	3.423±0.9	2	Y=-3.35+3.423X	
Cu	348.10(427.02-283.79)	982.07(1573.8-610.56)	3.663±0.75	3	Y=-4.31+3.663X	
Pb	2371.56(2463.8-2282.8)	2876.9(3112-2656)	19.67±3.7	3	Y=-61.38+19.67X	
Zn	689.21(761.18-624.04)	1189.90(1548.8-911.39)	6.96±1.6	3	Y=-14.75+6.96X	
72 h						
Hg	0.934(1.330-0.618)	5.284(18.590-3.005)	2.192±0.5	3	Y=5.065+2.192X	
Cd	167.94(223.61-126.09)	776.51(1533-393)	2.481±0.53	3	Y=-0.52+2.48X	
Cu	293.09(355.07-241.90)	748.27(1087.6-512.9)	4.053±0.77	3	Y=-4.99+4.053X	
Pb	2345.92(2439.9-2255.5)	2864.2(3103.9-2639.9)	19.03±3.66	3	Y=-59.14+19.03X	
Zn	593.60(657.66-535.78)	1042.72(1371.6-790.21)	6.743±1.56	3	Y=-13.702+6.74X	
96 h						
Hg	0.609(0.871-0.382)	3.370(8.681-2.052)	2.221±0.4	3	Y=5.478+2.221X	1
Cd	122.998(171.65-88.21)	683.304(1532.8-306.67)	2.215±0.5	3	Y=0.370+2.215X	202
Cu	242.102(297.73-196.80)	651.509(945.06-447.43)	3.838±0.7	3	Y=-4.149+3.838X	397.5
Pb	2320.13(2412.9-2230.9)	2823.31(3050.7-2609.8)	19.355±3.7	3	Y=-60.139+19.355X	3809.7
Zn	526.119(585.7-472.668)	887.22(1095.9-716.218)	7.270±1.3	3	Y=-14.782+7.270X	863.9

$^{+}$CL = 95% Confidence limit
SE$^{\leq}$ = Standard error
DF$^{\square}$ = Degrees of freedom
TF^{6} = Toxicity factor =

$$\frac{\text{24/48/72/96 h } LC_{50} \text{ value of other metals}}{\text{24/48/72/96 h } LC_{50} \text{ value of most toxic(Hg) metal}}$$

followed by *T. africanus* and *S. huzardi* (the most tolerant with 96 h LC_{50} = 242.102 mg/l) in descending order of susceptibility (Table 4). On the basis of the computed susceptibility/sensitive factor, *C. africanus* (most sensitive) was found to be about nine times and 56 times most susceptible to the toxic effect of Cu than *T. fuscatus* and *S. huzardi*, respectively (Table 4).

Lead (Pb). Clibanarius africanus was also found to be the most susceptible/ sensitive animal to the test lead compound with a 96 h LC_{50} value of 370.767 mg/l followed by *T. fuscatus* and *S. huzardi* (the most tolerant, 96 h LC_{50} = 2320.13 mg/l) in a descending order of susceptibility (Table 4). On the basis of the computed susceptibility factors, *C. africanus* was found to be about two times and six times more susceptible to the toxic effect of Pb than *T. fuscatus* and *S. huzardi*, respectively (Table 4).

Zinc (Zn). Clibanarius africanus was the most susceptible/sensitive test animal to Zn compound with a 96 h LC_{50} value of 29.801mg/l followed by *T. fuscatus* and *S. huzardi* (most tolerant species, 96 h LC_{50} value = 526.119 mg/l) in a descending order

TABLE 4

Relative susceptibility of test benthic animals against heavy metal compounds, based on 96-hour mortality data

Animals	*LC^{50} $^{+}$C.L. (mg l^{-1})*	*Slope±SE$^{\leq}$*	*DF$^{\square}$*	*Probit line equation*	*SF$^{\leq\leq}$*
Hg					
T. fuscatus	3.685(4.551-2.993)	1.998±0.2	4	Y=3.68+1.998X	6.1
C. africanus	0.813(0.970-0.670)	3.077±0.6	3	Y=5.298+3.077X	1.3
S. huzardi	0.609(0.870-0.380)	2.221±0.4	3	Y=5.478+2.21X	1
Cd					
T. fuscatus	28.246(41.240-19.37)	0.982±0.1	5	Y=3.575+0.982X	2.1
C. africanus	13.423(16.870-10.710)	2.422±0.4	3	Y=2.268+2.422X	1
S. huzardi	122.998(171.650-88.210)	2.215±0.5	3	Y=0.370+2.215X	9.2
Cu					
T. fuscatus	39.254(44.680-34.480)	2.955±0.3	5	Y=0.29+2.955X	9.04
C. africanus	4.342(6.820-3.200)	1.864±0.3	3	Y=3.811+1.864X	1
S. huzardi	242.102(297.73-196.80)	3.838±0.7	3	Y=4.149+3.838X	55.8
Pb					
T. fuscatus	609.779(731.50-508.16)	2.333±0.3	4	Y=-1.499+2.333X	1.7
C. africanus	370.767(447.57-307.31)	2.385±0.4	4	Y=-1.127+2.385X	1
S. huzardi	2320.13(2412.9-2230.96)	19.355±3.7	3	Y=-60.139+19.355X	6.3
Zn					
T. fuscatus	83.167(110.51-62.72)	1.410±0.1	5	Y=2.292+1.410X	2.8
C. africanus	29.801(36.47-24.35)	2.608±0.3	4	Y=1.156+2.608X	1
S. huzardi	526.119(585.615-472.67)	7.270±1.3	3	Y=-14.782+7.270X	17.7

$^{+}$CL = 95% confidence limit

SE$^{\leq}$ = Standard error

D.F$^{\square}$ = Degrees of freedom

S.F$^{\leq\leq}$ = Susceptibility factor =

$$\frac{\text{96 h } LC_{50} \text{ value of other test animals}}{\text{96 h } LC_{50} \text{ value of the most sensitive test animal}}$$

of susceptibility (Table 4). On the basis of the computed susceptibility factors, *C. africanus* was found to be about three times and 18 times more susceptible to the toxic effect of Zn compound than *T. fuscatus* and *S. huzardi*, respectively (Table 4).

Mercury (Hg). Unlike in the other tests above, *Sesarma huzardi* was the most susceptible/sensitive test animal to Hg compound with a 96 h LC_{50} value of 0.60 mg/l followed by *C. africanus* and *T. fuscatus* (most tolerant species, 96h LC_{50} = 3.685 mg/l) in descending order of susceptibility (Table 4). On the basis of the computed susceptibility factors (96 h LC_{50} ratios), *S. huzardi* was found to be about 1.3 times and six times more susceptible to the toxic effect of Hg than *C. africanus* and *T. fuscatus* (most tolerant), respectively (Table 4).

Discussion

In this study, the general toxicity ranking order of the investigated heavy metals against the three benthic animals of the Lagos lagoon was as follows:

$$Hg > Cd > Cu > Zn > Pb$$

The ranking order was, however, different in *C. africanus*, in that Cu was found to have a higher toxicity than Cd.

Many authors in different parts of the world including Nigeria (Khangarot & Ray, 1987; Mackie, 1989; Oyewo, 1998) have observed the differential toxicity of heavy metal compounds against different test organisms. The observed differential toxicity of heavy metals can be attributed to several factors such as metal compound tested, solubility of salts, predominant ions in test solution, physico-chemical characteristics of the test solution and the mechanism(s) of action of the different metals. All of these factors determine the availability of the metals to the test animals hence their toxicity. Other factors which may affect metal's toxicity or susceptibility of test animals include the formation of complexes with protein, e.g. metallothionien complexes, formation of encapsulated metal granules, metabolism and excretability; all of which differ considerably between test organisms and heavy metals types.

Mercury (Hg) compound was observed to be consistently the most toxic of the metals compounds tested against the test benthic animals while lead (Pb) compound was the least toxic. According to Cross *et. al.* (1973), the high toxic nature of Hg is due to its high electronegativity and the resultant affinity to sulphydryl groups. Bryan & Langston (1993), however, attributed the high toxic nature of Hg to its methylated form, methymercury, which was found to have high lipid solubility which inadvertently increases its penetrability or transfers across membranes.

The low toxic nature of Pb has also been reported widely in the literature (Qureshi *et al.*, 1980 Krishnaja *et al.*, 1987, Bryan & Langston, 1993). For example, Krishnaja *et al.* (1987) was unable to establish a 96 hLC_{50} value for lead nitrate against *Scylla serrata* because the compound did not cause any mortality of the exposed animals even at the highest test concentration. The low toxic nature of Pb compound may be because the free inorganic ion Pb^{2+} which is usually dominant in Pb solution is not lipid soluble, hence transfers across membranes may be inhibited.

Furthermore, the toxicity scale/ranking order generated in this study revealed that Hg, Cu and Cd are the most hazardous of the test heavy metals that occur in the Lagos lagoon, hence their introduction into the environment *via* industrial effluents must be minimized through effective control/ management strategies. Additionally, the toxicity scale should also enable industrialists to choose those chemicals/raw materials whose metallic components are amongst metals, which have been found to have relatively lower toxicity.

The single acute toxicity tests also established sensitivity/susceptibility scale for the test benthic animals, which are conspicuous inhabitants of the Lagos lagoon against the test metal compounds. With regards to Cd, Cu, Zn and Pb compounds tested in this study, *C. africanus* was the most sensitive followed by *T. fuscatus* and S. *huzardi* {most tolerant} in decreasing order of sensitivity. With regards to Hg, *S. huzardi,* which was observed to be the most tolerant against other test metal compounds, was found to be most susceptible to the toxic effects of Hg. This clearly indicates that the mechanism(s) of action of Hg against *S. huzardi* is quite different from that of the other metals tested against the test organism.

The differential response of organisms to metal compounds and other chemicals can be attributed to several factors such as the permeability of body membrane or cuticles, metabolism, excretory capacity, sex, age, body size, site of action and behaviour (Don-Pedro, 1996). Although the specific underlying reasons for the observed differential responses of the test animals was not evaluated in this study, explanations based on available literature and observations on their ecology and morphology suggest that the hardy and highly sclerotised nature of *Sersama huzardi* limits the penetratability of metal compounds into the body tissues of the test organism, thus conferring a high degree of tolerance of the animal to the toxic effect of the metal compounds.

In respect to *T. fuscatus*, Oyewo (1998) had observed that the relatively high tolerance level of the animal to some metal compounds was due to behavioural mechanisms, whereby the animal withdraws into its shell and seals off the shell aperture with its operculum upon its detection of noxious compounds in its surroundings. The relatively lower tolerance or higher sensitivity of *C. africanus* to the toxic effect of the metal compounds may probably be due to higher level of exposure arising from the following: (i) it possesses a thinner and less sclerotised membrane of its abdominal region, (ii) the shell it inhabits is an acquired shell and, therefore, can only provide limited protection since its aperture cannot be sealed up, and (iii) observations in this study have shown that the crabs tend to withdraw out of their acquired shells when stressed, thus making them more exposed to the toxicants. The results of bioaccummulation studies of heavy metals by *T. fuscatus* (Otitoloju 2001) and *C. africanus* (Oyewo, 1998) revealed that the animals also accumulated most of the heavy metals in the body tissues to such an extent that the tissue concentration were several folds higher than the ambient concentrations. This implies that metal tolerance in these organisms is not only related to limitation of exposure to the toxicants but may also include other metabolic and physiological processes such as: 1. sequestration of the toxicants in metal-binding proteins, 2. formation of encapsulated metal granules, and 3. other detoxication processes which can be responsible for the observed level of tolerance. The possible metabolic and physiological processes that may be responsible for the high tolerance of the test animals will, therefore, merit future research efforts.

Furthermore, the establishment of the sensitivity scale of the test benthic animals found in the Lagos lagoon should serve as a useful management tool of this very important aquatic resource. For instance, in the derivation of water quality criteria meant for the protection of aquatic life, the toxicity indices, e.g. LC_{50} values of the test compounds against the most sensitive test organism, are utilized in the determination of a minimum adverse concentration or a no-effect level, from which safe limits are extrapolated (Mason, 1992). These safe limits or criteria are then employed in fixing realistic industrial effluent limitation guidelines.

Although this study confined the toxicity testings to adult stages of the test animals, there is also the need to test the heavy metals against the expectedly more sensitive larval stages and other organisms such as *Daphnia* sp. and *Palaemonestes* sp. which may be more sensitive. This is necessary to ensure that the safety limits prescribed would indeed be able to protect all organisms in the aquatic ecosystem.

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