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Identification of sensitive and tolerant organisms from a tropical estuarine lagoon to heavy metal intoxication

E. O. Oyewo¹ and K. N. Don-Pedro²

¹Nigerian Institute for Oceanography and Marine Research, P.M.B. 12729, Lagos, Nigeria

Oyewo2000@yahoo.co.uk

²Toxicology Laboratory, Zoology Unit, Department of Zoology, Marine Biology and Fisheries, University of Lagos, Lagos, Nigeria

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ABSTRACT: Sequel to the establishment of the most prominent heavy metals in industrial effluents, drainage channels and the Lagos lagoon environment, the relative susceptibility of six animals from this tropical estuarine complex to some of the identified heavy metal species was investigated using standard semi-static bio-assay techniques.

Against the test metals (Fe, Mn, Cu and Hg), *Mugil sp.*, *T. guineensis* and *Cypris sp.*, had relatively low susceptibility thresholds and were identified as sensitive species based on results of semi-static laboratory tests.

On the other hand, *C. africanus*, *N. senegalensis* and *T. fuscatus* were identified as relatively tolerant animals from the Lagos lagoon system. The results are discussed in terms of their relevance to environmental management, associated standards, guidelines and consumer safety.

Key words: Coastal lagoon; Heavy metal; Sensitivity; Tolerance; Environmental management; Consumer safety.

Introduction

Population pressure and the preferential use of coastal space for industrialization often result in the deterioration in quality of coastal ecosystems and the vitiation of their value as multiple-usage amenities. Anthropogenic activities and influences in general are responsible for the contamination and or pollution of coastal ecosystems by critical pollutants including the class known as heavy metals.

Environmental protective measures and standards can be usefully informed by the results of field and laboratory studies including bio – monitoring and the identification of sensitive and tolerant organisms.

In West Africa, the Lagos lagoon complex is the largest of the four lagoon systems of the Gulf of Guinea coast (Hill and Webb, 1958) and probably typifies similar tropical lagoons world-wide. This lagoon system borders the rain forest belt and receives a number of major rivers and streams including Majidun, Ogun, Ona, Shasha and Oshun which drain about 103,637 km² of vast country. This expanse of water is generally shallow with a depth of between 0.3 and 3.2m in most parts with the exception of some dredged parts, notably in the Lagos harbour, where depths greater than 10m have been recorded. The tidal range is small being only 0.3-1.3m. The tides are the semi-diurnal type. The main body of this lagoon

system lies between longitudes 3° 22' and 3° 40' East and latitudes 6° 17' and 6° 28' North. The Lagos lagoon complex also receives an input of sea water from the Atlantic Ocean into which it opens via the Lagos Harbour. The cosmopolitan city of Lagos which is built around the Lagos lagoon, may have more than 12 million inhabitants while more than 80% of all industries in Nigeria are situated in and around the metropolis. Therefore, the Lagos lagoon complex receives waste waters/effluents from industrial and domestic sources as well as urban storm waters and contaminated stream or river waters.

In order to protect this and similar ecosystems from the known and potential negative impacts of pollution by heavy metals and other critical pollutants, there have been several studies aimed at understanding relevant characteristics of the environment and man-made inputs as benchmarks for judicious management in different parts of the world (Ayodele *et al.*, 1991; Bryan and Langston, 1992; Chen *et al.*, 1991; Clements *et al.*, 1990; Flos *et al.*, 1987; Ortego and Benson, 1992). The focus of most of these studies is the determination of ambient concentrations of critical contaminants and pollutants and sometimes aspects of their biological effects. Such attempts have usually precluded ranking of the investigated animal species in order of susceptibility as a means of identifying sensitive and tolerant ones.

The plants and animals of this system have been the subject of scientific studies by previous workers. For example, Hill and Webb, (1958); Fagade (1969); Olaniyan (1969); Fagade and Olaniyan (1974); Mah-Essiet (1986); Omoniwa (1988); Ajao (1989); Ajao and Fagade (1990); Nwankwo (1996) and a body of knowledge already exists on the biology and ecology of a number of pelagic, benthic and sessile Lagos lagoon animals including *Mugil sp.*, *Tilapia guineensis*, *Cypris spp.*, *Tympanotonus fuscatus*, *Clibanarius africanus* and *Nerita senegalensis* which are the test animals in this study. There is however little or no information on their relative sensitivity or tolerance to contaminants and pollutants. Yet such information will be invaluable in identifying organisms that could be targeted for particular purposes in schemes designed to protect and manage this valuable ecosystem.

In an earlier study, the most prominent heavy metal species discharged by industries around the Lagos lagoon complex were identified while the magnitude of their occurrence in the environment was determined (Oyewo, 1998). The present study describes the susceptibility ranking of *Mugil spp.*, *Ti guineensis*, *Cypris spp.*, *T. fuscatus*, *Clibanarius africanus* and *N. senegalensis* to some of these heavy metal species (Fe, Mn, Cu and Hg) in an attempt to identify sensitive and tolerant ones to heavy metal intoxication.

Materials and Methods

Evaluation of sensitivity/tolerance was based on laboratory bio-assays involving the test animals and metals.

Semi-Static Bioassay Technique

All bio-assays followed standard semi-static procedures with the observance of all necessary traditions and precautions (Sprague, 1973, Ward and Parish, 1982). Appropriate modifications were made where necessary (Oyewo, 1998; Tokolo, 1988, Alegbeleye *et al.*, 1989).

Test Animals

All the test animals were collected from the Ikoyi Experimental Fish Farm of the Nigerian Institute for Oceanography and Marine research which was protected, with only limited and controlled connection with the open Lagos Lagoon System. The animals were mainly from cultured stocks of known history which are usually preferred for toxicological bioassays rather than animals collected from the wild which may have acquired increased tolerance to pollutants over years of exposure (Callahan and Weiss, 1983; Ward and parish, 1982; Le blanc, 1982; Salami, 1990).

The test animals employed in this study included fingerlings of the mullet *Mugil sp.* (Pisces, Mugilidae) (TL = 70 ± 5mm), *Tilapia guineensis* (Pisces: Cichlidae) (Total Length (TL) = 65 ± 4mm), *Cypris sp.* (Ostracoda) (0.25 – 0.30mm diameter), *Tympanotonus fuscatus* (Mollusca; Gastropoda) (shell length 43 ± 4mm); *Clibanarius africanus* (Arthropoda, Crustacea) (weight of shucked animal = 0.48 ± 0.05g), and

Nerita senegalensis (Mollusca, Gastropoda) (Length = 7.5 ± 2 mm). They were subsequently transported to the laboratory and acclimatized before use in bioassays.

Test Temperature and Salinity

All test animals were acclimatized to a temperature regime of 22 – 24.5°C and salinity of 15 psu. The only exception was *Cypris*. It was tested under the same temperature regime but at 5 psu based on results of earlier salinity tolerance tests (Oyewo, 1998).

Test Chemicals

The heavy metals and salts used in this study were: Iron as FeCl_2 ; manganese as MnSO_4 ; Copper as $\text{CuSO}_4 \cdot 5\text{H}_2\text{O}$ and Mercury as HgCl_2 . All were of analar grade quality. With the exception of Mercury, the choice of heavy metals for this study was based on the commonest metals from the results of the chemical survey of industrial effluents, lagoon waters and sediments for heavy metals in an earlier related study (Oyewo, 1998).

Assessment of Quantal Response

The quantal responses adopted in this study included death, cessation of opercular or other body movement, failure to respond to prodding, failure to protrude foot during a defined observation period in untreated dilution water and loss of hold-fast capacity depending on the test animal (Oyewo, 1998).

Mortality and other responses were assessed every 24 hours.

Statistics

Toxicological dose-response data involving quantal responses 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 (1996b).

Results

Iron (Fe)

Mugil sp. was the most susceptible test animal to Fe with a response level based on 96-hr LC_{50} value of 50.32 mg L^{-1} followed by *Cypris sp.*, *Ti. Guineensis*, *N. senegalensis* and *T. fuscatus* (the most tolerant species, 96-hr $\text{LC}_{50} = 17409 \text{ mg L}^{-1}$) in a descending order of susceptibility (Tables 1 – 6). The susceptibility thresholds of the three most sensitive test animals, *Mugil*, *Tilapia* and *Cypris sp.*, to Fe were significantly lower than those of the more tolerant animals, *N. senegalensis* and *T. fuscatus* by several orders of magnitude. *C. africanus* was not tested against Fe.

Manganese (Mn)

Cypris sp. was the most susceptible to Mn with a response level, based on 96-hr LC_{50} value of 90.14 mg L^{-1} followed by *Mugil sp.*, *T. guineensis*, *N. senegalensis*, *C. africanus* and *T. fuscatus* (the most tolerant species, 96-hr $\text{LC}_{50} = 8409 \text{ mg L}^{-1}$) in a descending order of susceptibility (Tables 1 – 6). The susceptibility threshold of the three most sensitive test animals; *Mugil sp.*, *Tilapia* and *Cypris* were significantly (no overlaps in 95% CL of 96-hr LC_{50} values) lower than those of the more tolerant animals – *N. senegalensis*, *C. africanus* and *T. fuscatus* (Table 1-6).

TABLE 1: THE RELATIVE SUSCEPTIBILITY OF *Mugil* sp TO SELECTED HEAVY METALS.

Time (h)	(LC ₅₀ mg/L} 95% Confidence Limits	LC ₉₅ (mg/L} 95% Confidence Limits	LC ₅ (mg/L} 95% Confidence Limits	Chi sqd	Df	P	Slope ± SE	Probit line Equation
IRON (Fecl ₂)								
24	137.20(104.49-179.94)	655.73(408.62-1049.37)	28.71(15.92-51.78)	5.518	4	0.238	2.43+0.36	Y = -0.191 + 2.43x
48	71.71(54.24-94.68)	275.52(177.83-426.93)	18.67(10.98-31.65)	1.201	3	0.753	2.82+0.43	Y = -0.237+ 2.82x
72	66.82(50.94-87.62)	227.79(142.65-364.35)	19.60(11.92-32.16)	1.424	2	0.491	3.10+0.51	Y = -0.653 + 3.10x
96	50.32(37.92-66.71)	202.04(113.56-360.52)	12.54(6.37-24.55)	1.255	2	0.534	2.73+0.56	Y = 0.349 + 2.73x
MANGANESE (MnSo4)								
24	209.70(157.71-278.51)	1096.04(665.6-1799.3)	40.12(21.86-73-70)	2.284	4	0.684	2.30+0.34	Y = -0.333 + 2.30x
48	129.99(99.41-169.78)	530.58(335.93-835.71)	31.85(17.71-57.29)	3.298	3	0.348	2.70+0.44	Y = -0.710 + 2.70x
72	121.03(95.61-153.12)	451.81(283.51-717.65)	32.42(18.23-57.77)	0.384	3	0.943	2.88+0.52	Y = -1.008 + 2.88x
96	121.03(95.61-153.12)	451.81(283.51-717.65)	32.42(18.23-57.77)	0.384	3	0.943	2.88+0.52	Y = -1.008 + 2.88x
COPPER (CuS ₀₄ .5H ₂₀)								
24	0.37(0.30-0.45)	1.48(1.09-2.14)	0.09(0.06-0.14)	8.623	4	0.071	2.72+0.33	Y = 6.187 + 2.72x
48	0.20(0.16-0.25)	0.72(0.49-1.09)	0.05(0.03-0.09)	2.696	2	0.260	2.95+0.44	Y = 7.078 + 2.95x
72	0.19(0.15-0.23)	0.59(0.41-0.86)	0.06(0.04-0.09)	1.989	2	0.370	3.33+0.50	Y = 7.423 + 3.33x
96	0.16(0.12-0.19)	0.55(0.36-0.83)	0.04(0.03-0.08)	2.902	2	0.234	3.02+0.54	Y = 7.448 + 3.02x
MERCURY (Hgcl2)								
24	0.053(0.042-0.067)	0.25(0.17-0.37)	0.011(0.007-0.018)	6.34	4	0.175	2.42+0.28	Y = 8.094 +2.42x
48	0.031(0.025-0.039)	0.12(0.09-0.174)	0.008(0.005-0.013)	5.18	3	0.159	2.80+0.36	Y = 9.201 + 2.80x
72	0.020(0.016-0.024)	0.07(0.05-0.103)	0.006(0.003-0.009)	2.74	3	0.433	2.97+0.44	Y = 10.040 + 2.97x
96	0.018(0.015-0.023)	0.07(0.05-0.102)	0.005(0.003-0.008)	2.97	3	0.396	2.81+0.43	Y = 9.889 + 2.81x

TABLE 2: THE RELATIVE SUSCEPTIBILITY OF *Tilapia guineensis* TO SELECTED HEAVY METALS.

Time (h)	(LC ₅₀ mg/L} 95% Confidence Limits	LC ₉₅ (mg/L} 95% Confidence Limits	LC ₅ (mg/L} 95% Confidence Limits	Chi sqd	df	P	Slope ± SE	Probit line Equation
IRON (Fecl ₂)								
24	476.90(393.28-578.07)	2064.24(1475.5-2867.7)	110.18(71.02-171.98)	7.001	5	0.221.	2.59±0.31	Y = -1..945+ 2.59x
48	314.20(254.14-388.21)	1322.93(946.77-1836.9)	74.62(47.72-117.28)	7.342	4	0.119	2.64±0.31	Y = -1.599 + 2.64x
72	314.15(259.43-380.30)	1320.99(902.21-1922.4))	74.71(48.42-115.91)	5.133	4	0.274	2.65±0.34	Y = -1.605 + 2.65x
96	239.65(195.99-292.88)	992.33(695.75-1406.69)	57.88(36.28-92.79)	3.895	4	0.420	2.67±0.35	Y = - 1.362 + 2.67x
MANGANESE (MnSo4)								
24	976.17(768.12-1241.12)	5133.3(2780-9423)	185.6(112.1-309.4)	1.742	3	0.767	2.29±0.36	Y = -1.842 + 2.29x
48	470.98(384.83-576.13)	1844.4(1333-2533)	120.27(78.15-186.09)	3.601	4	0.463	2.78±0.34	Y = -2.439 + 2.78x
72	292.88(232.54-368.65)	1117.1(819.57-1670.9)	73.12(47.39-113.22)	8.231	3	0.041	2.74±0.33	Y = -1.753+ 2.74x
96	231.39(185.14-289.01)	920.22(641.4-1313.4)	58.19(36.60-92.85)	3.250	3	0.355	2.75±0.36	Y = -1.507+ 2.75x
COPPER (CuS _O 4.5H ₂₀)								
24	0.44(0.33-0.56)	1.70(1.13-2.93)	0.11(0.06-0.18)	7.206	3	0.066	2.79±0.43	Y = 6.008 + 2.79x
48	0.29(0.21-0.39)	1.53(0.92-2.89)	0.06(0.03-0.10)	4.407	3	0.221	2.28±0.37	Y = 6.231 + 2.28x
72	0.19(0.14-0.27)	0.97(0.60-1.74)	0.04(0.02-0.08)	1.746	3	0.627	2.35±0.41	Y = 6.677 + 2.35x
96	0.16(0.11-0.22)	0.71(0.41-1.34)	0.03(0.01-0.08)	2.257	2	0.323	2.50±0.52	Y = 7.016 + 2.50x
MERCURY (Hgcl ₂)								
24	0.10(0.08-0.13)	0.74(0.47-1.19)	0.014(0.008-0.024)	10.49	4	0.062	1.91±0.22	Y = 6.904 + 1.91x
48	0.07(0.05-0.09)	0.49(0.30-0.82)	0.010(0.005-0.018)	7,90	4	0.096	1.94±0.24	Y = 7.246 + 1.94x
72	0.03(0.02-0.04)	0.14(0.088-0.212)	0.006(0.003-0.010)	2.40	3	0.504	2.40±0.32	Y = 8.724 + 2.40x
96	0.02(0.017-0.03)	0.095(0.061-0.146)	0.005(0.003-0.008)	3.20	3	0.362	2.51±0.38	Y = 9.224 + 2.51x

TABLE 3: THE RELATIVE SUSCEPTIBILITY OF *Cypris* sp. TO SELECTED HEAVY METALS.

Time (h)	(LC ₅₀ mg/L} 95% Confidence Limits	LC ₉₅ (mg/L} 95% Confidence Limits	LC ₅ (mg/L} 95% Confidence Limits	Chi sqd	df	P	Slope $\pm$ SE	Probit line Equation
IRON (Fec/2)								
24	210..64(177.99-249.20)	786.23(574.69-1069.10)	56.43(38.28-83.65)	10.523	5	0.062	2.88 $\pm$ 0.35	Y = -1.702 + 2.88x
48	201.88(170.18-239.39)	758.78(556.28-1028.69)	53.71(36.31-79.88)	8.252	5	0.143	2.87 $\pm$ 0.34	Y = -1.614 + 2.87x
72	175.80(138.61-222.98)	1379.11(763.27-2481.80)	22.41(12.47-40.45)	7.194	5	0.207	1.84 $\pm$ 0.25	Y = 0.859 + 1.84x
96	90.08(68.62-118.23)	684.76(344.72-1361.86)	11.85(5.76-24.33)	4.168	3	0.244	1.87 $\pm$ 0.30	Y = 1.339 + 1.87x
MANGANESE (MnSo4)								
24	293.33(249.90-344.22)	908.27(687.52-1193.11)	94.73(65.53-137.66)	10.282	4	0.036	3.36 $\pm$ 1.43	Y = -3.293 + 3.36x
48	249.06(211.10-293.78)	939.01(669.73-1308.63)	66.06(45.12-97.25)	9.695	5	0.084	2.86 $\pm$ 0.35	Y = -1.860 + 2.86x
72	154.48(121.13-196.85)	1112.01(693.11-1778.27)	21.44(12.35-37.36)	16.521	5	0.006	1.92 $\pm$ 0.23	Y = 0.789 + 1.92x
96	90.14(68.02-119.24)	709.97(409.60-1228.18)	11.45(5.70-22.94)	10.412	4	0.034	1.84 $\pm$ 0.25	Y = 1.401 + 1.84x
COPPER (CuSo ₄ .5H ₂₀)								
24	0.32(0.28-0.38)	1.47(1.11-1.99)	0.07(0.05-0.09)	12.504	6	0.052	2.51 $\pm$ 0.23	Y = 6.232 + 3.36x
48	0.25(0.22-0.29)	1.14(0.88-1.52)	0.06(0.04-0.07)	8.120	7	0.322	2.52 $\pm$ 0.20	Y = 6.503 + 2.52x
72	0.22(0.19-0.26)	1.09(0.79-1.55)	0.04(0.03-0.06)	10.885	6	0.092	2.36 $\pm$ 0.21	Y = 6.558 + 2.36x
96	0.16(0.14-0.19)	0.77(0.55-1.08)	0.04(0.02-0.05)	3.215	5	0.667	2.46 $\pm$ 0.24	Y = 6.935 + 2.46x
MERCURY (Hgcl ₂)								
24	0.30(0.22-0.38)	1.60(1.102-2.496)	0.054(0.030-0.088)	6.453	4	0.168	2.25 $\pm$ 0.26	Y = 6.193 + 2.25x
48	0.16(0.12-0.21)	0.86(0.55-1.39)	0.029(0.017-0.047)	2.925	3	0.403	2.24 $\pm$ 0.26	Y = 6.801 + 2.24x
72	0.13(0.10-0.16)	0.84(0.53-1.39)	0.019(0.010-0.033)	3.958	4	0.412	2.00 $\pm$ 0.25	Y = 6.803 + 2.00x
96	0.12(0.10-0.15)	0.61(0.39-0.99)	0.022(0.013-0.037)	5.905	3	0.116	2.30 $\pm$ 0.31	Y = 7.143 + 2.30x

Copper (Cu)

T. guineensis, *Mugil sp.* and *Cypris sp.* had similar magnitude of responses to Cu, (with overlaps in 95% CI of 96-hr LC₅₀ values) and therefore constituted the three most susceptible animal species to Cu with response levels of approximately 0.16 mg L⁻¹ in each case, followed by *C. africanus*, *N. senegalensis* and *T. fuscatus* (the most tolerant sp., 96-hr LC₅₀ = 8.84 mg L⁻¹) in a descending order of susceptibility (tables 1-6). The response level of any of the three most sensitive animal species was significantly (no overlaps in 95% CI of 96-hr LC₅₀ values) higher than those recorded for the most tolerant animals – *C. africanus*, *N. senegalensis* and *T. fuscatus*.

Mercury (Hg)

Mugil sp. was the most susceptible to Hg with a response level, based on 96-hr LC₅₀ value of 0.018 mg L⁻¹ followed by *T. guineensis*, *Cypris sp.*, *C. africanus*, *N. senegalensis* and *T. fuscatus* (the most tolerant sp. 96-hr LC₅₀ = 2.47mg L⁻¹) in a descending order of susceptibility (Tables 1-6). The susceptibility threshold of the three most sensitive test animals; *Mugil sp.*, *T. guineensis* and *Cypris* were significantly lower than those recorded for the more tolerant test animals – *C. africanus*, *N. senegalensis* and *T. fuscatus*.

Discussion

The series of acute bioassays carried out permitted the establishment of a susceptibility scale for six resident Lagos lagoon animal species, against each test metal. Overall, *Mugil sp.*, *T. guineensis* and *Cypris sp.* were the most susceptible and were significantly less tolerant to the heavy metals than the three other test species namely; *N. senegalensis*, *C. africanus* and *T. fuscatus* – the least susceptible sp. in all cases. Although it generally recognized that results of laboratory tests can not be directly extrapolated to predict field toxicity situations accurately, there is a regular attempt to correlate results of laboratory toxicity tests with field toxicity in order to explain observations in the wild (baron, 1995). In spite of their limitations, result of laboratory test are known to serve as fair indications of field situations and have been variously used to understand complex field interactions or complement field studies. Thus, the test animals can be broadly classified as sensitive and tolerant species based on the results obtained. It is however desirable to carry out similar screening on other organisms and against other critical pollutants to enrich the data pool for appropriate and informed decisions.

The differential susceptibility shown by the test animals to heavy metals serves as an extension of scientific knowledge that has been demonstrated elsewhere (Torres *et al.*, 1987; Mackie, 1989; Chen *et al.*, 1991). In general, differential responses amongst organisms have been attributed to factors such as; nature of the cuticle or body covering with respect to penetrability, metabolic transformation capacities of the organisms; for example, availability of the right type of enzymes and optimal physico-chemical conditions, excretory capacity and the rate of elimination of the by-products of metabolism, availability and sensitivity of site of action, body size, age and life cycle stage as well as ecology with particular reference to location and activity coefficient and possibly, behavioural attributes (Don-Pedro, 1987; Don-Pedro, 1996b). The observed differential susceptibilities are thus expected.

The susceptibility or sensitivity scale established in this work represents useful information which in conjunction with similar ones will be invaluable for the management of the Lagos lagoon and similar ecotypes with respect to heavy metal pollution in a number of ways.

The most sensitive species can serve as sensitive indicators that can be employed in the early detection of heavy metal pollution since they respond at low contamination levels thereby serving as early warning signals.

More efficient and wider environmental protection of living organisms can be achieved by employing the response levels of the more sensitive species as a basis or starting point from which no effect levels and consequently safe limits can be extrapolated and employed in fixing realistic standards and or allowable levels in effluents and the recipient natural ecosystems such as the Lagos lagoon.

TABLE 4: THE RELATIVE SUSCEPTIBILITY OF *Tympanotonus fuscatus* TO SELECTED HEAVY METALS.

Time (h)	(LC ₅₀ mg/L} 95% Confidence Limits	LC ₉₅ (mg/L} 95% Confidence Limits	LC ₅ (mg/L} 95% Confidence Limits	Chi sqd	Df	P	Slope + SE	Probit line Equation
IRON (Fecl ₂)								
24	28788(18795--44104)	59335(22631-154967)	13967(11413-17166)	0.047	1	0.829	5.25±0.04	Y = -1.8.424+5.25x
48	35930(19120-67584)	140691(31728-620193)	9176(6419.1-13219.94)	0.320	2	0.852	2.783±0.92	Y = -7.679 + 2.78x
72	20989(16898-26075)	58153(31556-106560)	7575(5777-9993)	0.223	3	0.974	3.73±0.78	Y = -11.112+ 3.73x
96	17409(15040-20153)	42353(27990-63765)	7156(5544-9284)	2.567	2	0.277	4.27±0.76	Y = -13.122+4.27x
MANGANESE (MnSo4)								
24	28915(11823-70786)	145167(6310.01-3318989)	5759.7(1365.4-24494..6)	0.593	1	0.441	2.87±1.69	Y = -5.504 + 2.355x
48	19154(15483-23699)	51087(27822-93312)	7181.5(5169.4-10032.8)	0.734	2	0.693	3.87±0.89	Y = -11.583 +3.87x
72	14011(12612-15566)	29763(22061-39980)	6596.2(5173.11-8447.9)	1.224	3	0.747	5.04±0.88	Y = -15.909+ 5.04x
96	8409(7273-9722)	24659(18471-32722)	2867.6(2073.7-3989.0)	4.294	4	0.368	3.53±0.45	Y =-8.859 + 3.53x
COPPER (CuSO4.5H20)								
24	19.24(15.04-24.60)	133.36(83.54-214.9)	2.78(1.68-4.53)	4.876	5	0.431	1.96±0.21	Y = 2.480 + 1.96x
48	13.26(10.76-16.33)	64.52(44.31-94.46)	2.73(1.77-4.17)	2.138	5	0.830	2.40±0.27	Y = 2.305 + 2.40x
72	11.66(9.53-14.26)	48.93(33.46-72.04)	2.78(1.84-4.17)	3.788	4	0.435	2.65±0.32	Y = 2.174 + 2.65x
96	8.84(7.05-11.09)	43.84(27.03-72.41)	1.78(1.05-2.97)	7.722	3	0.052	2.37±0.34	Y = 2.754 + 2.37x
MERCURY (Hgcl2)								
24	5.09(4.42-5.87)	17.16(12.98-22.84)	1.51(1.12-2.04)	3.129	6	0.762	3.13±0.33	Y = 2.788 +3.13x
48	3.49(3.16-3.87)	7.79(6.20-9.85)	1.57(1.25-1.95)	4.336	5	0.502	4.74±0.61	Y = 2.426 + 4.74x
72	2.68(2.50-2.87)	4.57(3.95-5.29)	1.57(1.34-1.84)	3.107	5	0.684	7.11±0.91	Y = 1.956 + 7.11x
96	2.47(2.31-2.65)	4.01(3.55-4.55)	1.52(1.31-1.76)	5.211	4	0.266	7.84±0.97	Y = 1.918 + 7.84x

TABLE 5: THE RELATIVE SUSCEPTIBILITY OF *Clibanarius africanus* TO SELECTED HEAVY METALS.

Time (h)	(LC ₅₀ mg/L} 95% Confidence Limits	LC ₉₅ (mg/L} 95% Confidence Limits	LC ₅ (mg/L} 95% Confidence Limits	Chi sqd	df	P	Slope + SE	Probit line Equation
MANGANESE (MnSO ₄)								
24	36109(14701-89390)	1338548(65970-27452909)	974.1(191.12-4988)	4.672	2	0.097	1.05±0.33	Y = 0.207+1.05x
48	18344(10710-31504)	405685(59988-2740839)	829.6(211.3-3277.9)	11.065	2	0.004	1.233±0.32	Y = -0.231+ 1.23x
72	12563(8467-18658)	217397(54913-854786)	726.0(236.2-2251.6)	10.521	3	0.015	1.33±0.28	Y = -0.462+1.33x
96	5057(3515-7270)	83155(36883-185914)	307.6(124.5-765.7)	9.870	4	0.043	1.36±0.19	Y = -0.026+1.36x
COPPER (CuSO ₄ .5H ₂ O)								
24	36.96(29.83-45.76)	160.34(105.94-242.5)	8.52(5.32-13.62)	8.26	3	0.041	2.59±0.32	Y = 0.942 +2.59x
48	0.59(0.52-0.66)	1.42(1.13-1.86)	0.24(0.19-0.30)	1.97	5	0.854	4.31±0.52	Y = 5.991 +4.31x
72	0.40(0.36-0.45)	0.89(0.73-1.11)	0.18(0.14-0.23)	6.02	4	0.198	4.73±0.58	Y = 6.889+ 4.73x
96	0.38(0.34-0.43)	0.83(0.68-1.01)	0.18(0.14-0.22)	1.16	4	0.884	4.97±0.61	Y =-7.065 + 4.97x
MERCURY(HgCl ₂)								
24	43.42(36.08-52.22)	148.37(102.46-214.14)	12.71(8.003-20.22)	8.91	3	0.031	3.09±0.48	Y = 0.064 + 3.09x
48	2.40(1.93-2.98)	10.22(7.16-15.49)	0.57(0.36-0.83)	2.19	4	0.701	2.62±0.31	Y = 4.001 +2.62x
72	0.40(0.36-0.45)	0.89(0.73-1.11)	0.18(0.14-0.23)	6.02	4	0.198	4.73±0.58	Y = 6.889 + 4.73x
96	0.38(0.34-0.43)	0.89(0.71-1.13)	0.17(0.12-0.22)	9.28	3	0.026	4.56±0.63	Y = 6.891 + 4.56x

TABLE 6: THE RELATIVE SUSCEPTIBILITY OF *Nerita senegalensis* TO SELECTED HEAVY METALS.

Time (h)	(LC ₅₀ mg/L} 95% Confidence Limits	LC ₉₅ (mg/L} 95% Confidence Limits	LC ₅ (mg/L} 95% Confidence Limits	Chi sqd	Df	P	Slope ± SE	Probit line Equation
IRON (FeCl ₂)								
24	2369.88(2154.9-2606.2)	5547.9(4495.6-6813.1)	1012.3(789.16-1304.97)	32.082	5	0.00	4.47±0.56	Y = 10.073+4.47x
48	1352.08(1117.8-1551.9)	3962.5(3180.3-4907.5)	461.3(339.9-629.69)	10.762	4	0.029	3.53±0.38	Y = 6.062+3.53x
72	1200.79(1042.7-1382.6)	3341.9(2652.5-4186.9)	431.46(318.9-586.82)	11.070	3	0.001	3.71±0.43	Y = 6.430+3.71x
96	1051.14(894.99-1234.50)	3488.02(2565.1-4714.3)	316.77(231.67-435.73)	6.648	3	0.084	3.17±0.36	Y = 4.571+3.17x
MANGANESE (MnSO ₄)								
24	751.64(723.99-780.34)	1021.99(949.73-1097.72)	552.81(512.38-597.52)	5.614	4	0.230	12.37±1.33	Y = -30.561 +12.37x
48	612.73(573.05-655.16)	1123.05(983.68-1277.6)	334.31(282.57-396.92)	18.718	5	0.002	6.27±0.72	Y = -12.477+6.27x
72	509.49(465.98-557.04)	1105.08(930.91-1305.98)	234.90(187.74-295.20)	15.156	5	0.010	4.91±0.58	Y = -8.283+ 4.90x
96	445.87(401.71-494.84)	1087.53(906.06-1298.73)	182.80(141.9-236.57)	19.859	5	0.001	4.26±0.48	Y = -6.287 + 4.261x
COPPER (CuSO ₄ .5H ₂ O)								
24	3.98(3.39-4.66)	15.98(12.08-21.40)	0.99(0.70-1.37)	14.72	5	0.012	2.73±0.27	Y = 3.363 + 2.73x
48	2.87(2.40-3.42)	15.98(11.59-22.52)	0.52(0.36-0.73)	11.54	6	0.073	2.22±0.19	Y = 3.985 + 2.22x
72	2.11(1.73-2.56)	12.99(9.06-19.98)	0.34(0.22-0.50)	6.50	5	0.265	2.09±0.20	Y = 4.323 + 2.09x
96	1.65(1.38-1.98)	7.82(5.68-11.80)	0.35(0.24-0.47)	5.29	5	0.382	2.44±0.23	Y = 4.469 + 2.44x
MERCURY (HgCl ₂)								
24	1.87(1.69-2.07)	4.84(4.06-5.85)	0.72(0.58-0.89)	19.064	6	0.004	4.00±0.37	Y = 3.912 + 4.00x
48	1.36(1.23-1.50)	3.41(2.85-4.25)	0.54(0.43-0.66)	6.866	5	0.231	4.13±0.41	Y = 4.448 + 4.13x
72	1.03(0.92-1.15)	2.68(2.26-3.35)	0.40(0.300-0.49)	2.758	5	0.737	3.98±0.40	Y = 4.944 + 3.98x
96	0.99(0.88-1.09)	2.37(1.99-2.99)	0.41(0.31-0.50)	4.084	4	0.395	4.34±0.47	Y = 5.027 + 4.34x

The more tolerant species identified in this work namely: *N. senegalensis*, *C. africanus* and *T. fuscatus* could also serve as indicator species by providing information on the state of levels of metal contamination in the environment over a period of time. Thus, by measuring the concentration of metals accumulated in the bodies of these relatively tolerant species at regular intervals the state of pollution can be extrapolated and used to further control or regulate the pollutant being monitored.

The tolerance of *T. fuscatus* in particular against heavy metal intoxication has public health implications because it is a delicacy in several Nigerian communities. Even when apparently healthy, their contaminant burden can be transferred to animals like man at higher trophic levels.

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