Full Length Research Paper

The comparative toxicities of cadmium, copper and lead to *Macrobrachium rosenbergii* and *Penaeus monodon* postlarvae

¹Fafioye, O.O. and ²Ogunsanwo, B.M.

¹Department of Biological Sciences, Olabisi Onabanjo University, Ago-Iwoye, Ogun State, Nigeria. ²Department of Chemical Sciences, Olabisi Onabanjo University, Ago-Iwoye, Ogun State, Nigeria.

Accepted November, 2006

Comparative acute toxicity tests were carried out with three heavy metals viz. cadmium, copper and lead on giant prawn (*Macrobrachium rosenbergii*) and tiger prawn (*Penaeus monodon*) post larvae. Lethal concentrations at which 50% of the shrimp died (LC_{50}) at 96 h were 3.23 mg/l Cd, 3.02 mg/l Cu and 10.0 mg/l Pb for *M. rosenbergii*; 2.91 mg/l Cd, 2.16 mg/l Cu and 7.28 mg/l Pb for *P. monodon*. The metals showed toxicities in the order Cu>Cd>Pb. The LC_{50} values recorded for *M. rosenbergii* were higher than the values recorded for *P. monodon*, hence the specific metals were more toxic to *P. monodon* than *M. rosenbergii*. The safe concentrations for all these metals were also calculated. This study has produced LC_{50} values that may be useful in screening potentially toxic substances in our waters.

Key words: Toxicity, heavy metals, shrimp, post larvae.

INTRODUCTION

Heavy metals such as cadmium, copper and lead are found in most of the industrial, mineral exploration and other miscellaneous anthropogenic effluxents. In Nigeria, these effluents are indiscriminately discharged into natural waters, thereby contaminating aquatic ecosystem (Fafioye et al., 2002). The variation of heavy metal in salt water was low but the mean concentration was high and exceeded most of the levels recorded from a variety of inland surface waters in Africa (Okoye, et al., 1994).

Shrimp farmers in Nigeria do not have access to an adequate supply of healthy, domesticated seed to stock their ponds because post larvae typically are produced from wild-caught brood stock. Wild brood stocks are often polluted with anthropogenic wastes in the lagoon (Moss, 2004). This accounts for decrease in availability and quality of wild brood stock.

The giant prawn, *Macrobrachium rosenbergii* and the tiger prawn, *Penaeus monodon* are commercially

important shrimp for the Nigerian fisheries industry. These crustaceans need low salinity (Saha et al., 1999) and so thrive better in both fresh water and brackish environments, with salinity of between 0.2-15 ppt (ASEAN, 1978; Mohanta, 2000). Asejire lake and Epe lagoon are deep waters with high primary production and this quality has been reported to be favourable to shrimp production (Prasad, 2001).

Many Nigerian farmers have observed poor growth and survival of both P. monodon and M. rosenbergii stocks, resulting in decreased production and profitability. The reason for this is mainly on the condition of shrimp seed being used for stocking their ponds. Although Asejire lake has not been reportedly polluted, but the estuaries around Agbara and Lekki industrial areas of Lagos contain heavy metals in their surficial sediments (Okoye et al., 1994, Otitoloju, 2000). Cadmium, copper and lead are among the recently reported heavy metal contaminants that are abundant in Lagos Lagoon (Ajao, 1996, Oyewo, 1998). These metals are readily seeped by industries into our waters daily, thereby increasing their accumulation level. Therefore, it is necessary to study the toxicity of cadmium, copper and lead with a view to predict their level of toxicity to M. rosenbergii and P. monodon post larvae.

^{*}Corresponding author. E-mail: ofafioye@yahoo.com.

MATERIALS AND METHODS

Shrimp collection, transportation and holding:

Macrobrachium rosenbergii postlarvae (average weight of 0.32 ± 0.07 g) and *Penaeus monodon* postlarvae (average weight of 0.26 ± 0.04 g) of similar state of development were collected from Epe lagoon, Nigeria. The shrimp were transported separately in circular plastic bowls (30 litres capacity) to the laboratory, aerated and held under laboratory conditions (temperature = $27.0 \pm 1.5^{\circ}$ C, dissolved oxygen (DO) = 6.8 ± 1.2 mg/l, pH = 6.9 ± 1.4 and salinity = 2.0 ± 2.2 mg/l) typical of the environment in which they were collected for 168 hours before use. Shrimp were fed supplementary feed containing 40% protein at 2% of the standing biomass.

Metal stock preparation: The metal salts used for this study were $CdCl_2 21/2H_2O$, $CuSO_4 7H_2O$ and $Pb (CH_3COO)_2 3H_2O$. Four different concentrations (1.0, 5.0, 10.0 and 15.0 mg/l) were selected after preliminary screening tests had been conducted. These concentrations were prepared by adding a calculated volume of stock solution in 5 litres of water. A control, which had no toxicant, was also maintained.

Test Procedure

The toxicity tests (i.e. two tests) were conducted according to standard procedure of FAO (1985). Four concentrations and a control were set up in triplicate using fifteen rectangular glass tanks ($15x60x14 \text{ cm}^3$) per experiment. Twenty hardy shrimp of the same size ($10.2 \pm 0.6 \text{ mm}$ for *M. rosenbergii*; $8.6 \pm 0.4 \text{ mm}$ for *P. monodon*) were separately transferred from the holding tanks into the control and experimental tanks. The whole set was aerated continuously, while test solution in each tank was changed with requisite fresh solution every 24 hours. Observations for mortality were made twice (10.00 am and 6.00 pm) daily, while dead shrimp

(i.e. shrimp that failed to respond to mechanical stimulation) were promptly removed.

Analysis: The 96 h LC50 values were calculated using probit analysis of Finney (1971), while the safe concentration of the metals was determined by the methods of Kameswara-Rao (1974) and Miller & Miller (1986) following the application factor, 1/100th of the 96 h LC₅₀ value. Analysis of variance (ANOVA) was employed to validate the significance and insignificance values of the results. Duncan's multiple range test (DMRT) was used to separate means and correlation coefficient analysis was done to determine the relationships.

RESULTS

During the toxicity tests, the temperature, salinity and ph of the test media remained fairly constant at $26.5\pm1.5^{\circ}$ C, 3.1 ± 2.6 mg/l and 7.2 ± 0.4 respectively, while dissolved oxygen was higher than 5.6 mg/l. *Macrobrachium rosenbergii* and *Penaeus monodon* postlarvae swimming behaviour were weakened progressively with time, while all the shrimp moulted at a faster rate than control (Table 1). There was 100% survival at initial exposure in the different concentrations, but the survival rate started declining with an increase in concentrations and time of exposure (Table 2). Significant differences (P<0.05) in the toxicities were observed in the different concentrations and between the different heavy metals used.

When exposed to cadmium, *M. rosenbergii* recorded 80% and 70% mortality in 15 mg/l and 10 mg/l respectively at 96 h duration. In *P. monodon*, 90% and 75% mortalities were recorded at 96 h exposure to 15 mg/l and 10 mg/l of Cd, respectively. The lowest concentration (1.0 mg/l) produced 30% and 35% mortality at 96 h in *M. rosenbergii* and *P. monodon*, respectively.

In copper, *M. rosenbergii* percentage mortality at 96 h was 90% in 15 mg/l and 75% in 10 mg/l of Cu, while 30% mortality occurred in 1.0 mg/l at 96 h. *P. monodon* had 100% mortality in 15 mg/l and 80% mortality in 10 mg/l with 40% mortality in 1.0 mg/l at 96 h.

M. rosenbergii exposed to lead concentration recorded 70% mortality in 15 mg/l and 50% in 10.0 mg/l with 20% mortality in 1.0 mg/l at 96 h. *P. monodon* mortality was 75% in 15 mg/l and 55% in 10.0 mg/l at 96-h, while percentage mortality was 25% in 1.0 mg/l of Pb.

Table 3 shows the 96 h LC_{50} values, safe concentration, slope and intercept of the shrimps exposed to the heavy metals. In *M. rosenbergii*, the LC_{50} values for 96 h were 3.23 mg/l Cd, 3.02 mg/l Cu and 10.0 mg/l Pb. *P. monodon* recorded 96 h LC_{50} values of 2.91 mg/l, 2.16 mg/l and 7.28 mg/l in Cd, Cu and Pb respectively.

DISCUSSION

In the present study, it was observed that exposed *M. rosenbergii* and *P. monodon* postlarvae to various concentrations of cadmium, copper and lead were weakened progressively with time prior to mortality. This behaviour could not have arisen from the influence of the physico-chemical parameters of the test water since results showed little or no fluctuation. However, the rate at which the swimming behaviour of these shrimps slowed down depended on the amount of potency of each heavy metal. Similarly, the toxic effect of the metals produced moulting in the shrimp at a faster rate than control. These facts, therefore, affirm that heavy metals can cause physiological stress and dysfunction in crustaceans (Gao and Zou, 1995).

Shrimp survival rate started declining from 24 h exposure in the different concentrations of heavy metals and this produced varied percentage mortalities and LC_{50s} at 96 h exposure. The 100% mortality of *P. monodon* in 15 mg/l of Cu at 96 h and LC_{50} of 2.16 mg/l of Cu showed that Cu is more toxic than either Cd with LC_{50} of 2.91 mg/l or Pb with LC_{50} of 7.28 mg/l which had lower percentage mortality. Similarly exposure of *M. rosenbergii* to heavy metals that produced mortality of 90%, 80% and 70% in 15 mg/l Cu, Cd and Pb had different LC_{50} values of 3.02, 3.23 and 10.0 mg/l, respectively, which also confirmed Cu as most toxic to the shrimp than Cd and Pb.

Shrimp	Heavy metal	Concentration (mg/l)	No. of shrimp	Moulting/hour
P. monodon	Cadmium	0.0	20	0
		1.0	20	1
		5.0	20	3
		10.0	20	5
		15.0	20	8
	Copper	0.0	20	1
		1.0	20	2
		5.0	20	4
		10.0	20	7
		15.0	20	9
	Lead	0.0	20	0
		1.0	20	1
		5.0	20	2
		10.0	20	4
		15.0	20	6
M. rosenbergii	Cadmium	0.0	20	0
		1.0	20	1
		5.0	20	2
		10.0	20	3
		15.0	20	5
	Copper	0.0	20	1
		1.0	20	2
		5.0	20	4
		10.0	20	6
		15.0	20	9
	Lead	0.0	20	1
		1.0	20	1
		5.0	20	1
		10.0	20	3
		15.0	20	5

 Table 1. Moulting of Macrobrachium rosenbergii and Penaeus monodon postlarvae exposed to cadmium (Cd) copper (Cu) and lead (Pb) for 24-h duration

The 96 h LC₅₀ values of 2.91 and 3.23 mg/l observed in P. monodon and M. rosenbergii exposed to Cd respectively showed that P. monodon was more susceptible to Cd than *M. rosenbergii*. However, the 96 h LC₅₀ values of 2.88, 3.02 and 3.11 mg/l of Cd reported to be toxic to P. monodon (Diaz, 1995), P. pencillatus (Gao and Zou, 1995) and P. indicus (Chinni and Yallapragda, 2000), respectively, further confirmed P.monodon to be more sensitive to cadmium than any other shrimps, while the documented LC₅₀ values of these authors were in the same range of the values reported for this study. The observed 96 h LC₅₀ value of Cu in *P. monodon* was 2.16 mg/l and in *M. rosenbergii* was 3.02 mg/l. Again P. monodon was more vulnerable to Cu than *M. rosenbergii.* These results conform to the earlier

results of 2.05 mg/l Cu documented for *P. japonicus* (Bombang et al., 1995) and 2.535 mg/l Cu for *P. indicus* (Chinni and Yallapragda, 2000). This shows that Copper was most toxic to either of the shrimp.

Penaeus monodon exposed to lead concentrations produced 96 h LC₅₀ of 7.28 mg/l, while for *M. rosenbergii* it was 10.0 mg/l of Pb. These values compared well with the values of 7.223 mg/l reported by Chinni and Yallapragda (2000) for *P. indicus*.

The present observed lethal concentrations in the toxicity of Cd, Cu and Pb on *M. rosenbergii* and *P. monodon* are very much comparable with other results as discussed above, while lead was most tolerated by the shrimp, Cu was most potent and the toxicities are in the order of Cu>Cd>Pb. The specific metals were more toxic

Shrimp	Heavy	Concentration (mg/l)	No of	Percentage mortality/hour				
	metal		the test shrimp	Initial	24	48	72	96
P. monodon	Cd	0.0 control	20	0	0	0	0 ^d	0 ^d
		1.0	20	0	05	20	30 ^c	35 [°]
		5.0	20	0	10	25	40 ^c	55 [°]
		10.0	20	0	15	30	50 ^b	75 ^b
		15.0	20	0	25	40	65 ^a	90 ^a
	Cu	0.0 Control	20	0	0	0	0 ^d	0 ^d
		1.0	20	0	10	15	30 ^c	40 ^c
		5.0	20	0	20	30	45 ^b	65 ^c
		10.0	20	0	25	40	60 ^b	80 ^b
		15.0	20	0	30	55	80 ^a	100 ^a
	Pb.	0.0 Control	20	0	0	0	0 ^c	0 ^d
		1.0	20	0	0.5	10	20 ^b	25 ^b
		5.0	20	0	10	15	30 ^b	45 ^b
		10.0	20	0	15	25	40 ^a	55 ^b
		15.0	20	0	20	30	55 ^a	75 ^a
M. rosenbergii	Cd	1.0 control	20	0	0	0	0 ^c	0 ^d
		1.0	20	0	0.5	15	25 ^b	30 ^c
		5.0	20	0	10	20	35 ^b	55 ^b
		10.0	20	0	15	25	45 ^a	70 ^a
		15.0	20	0	20	35	55 ^a	80 ^a
	Cu	0.0 Control	20	0	0	0	0 ^c	0 ^d
		1.0	20	0	10	10	20 ^b	30 ^c
		5.0	20	0	15	25	40 ^b	60 ^b
		10.0	20	0	20	35	55 ^a	75 ^a
		15.0	20	0	25	50	70 ^a	90 ^a
	Pb.	0.0 Control	20	0	0	0	0 ^d	0 ^c
		1.0	20	0	05	05	15 [°]	20 ^b
		5.0	20	0	10	15	25 ^b	30 ^b
		10.0	20	0	15	20	35 ^b	50 ^a
		15.0	20	0	20	30	55 ^a	70 ^a

Table 2. Mean percentage mortality of Macrobrachium rosenbergii and Penaeus monodon postlarvae exposed to cadmium (Cd).

Key O = means no mortality.

Values with the same superscripts vertically in a column

Table 3. The 96-h LC₅₀ values, safe concentration, slope and intercept of Macrobrachium rosenbergii and Penaeus monodon post larvae exposed to cadmium (Cd), copper (Cu) and lead (Pb).

Shrimp	Heavy metal	96-h LC ₅₀ (mg/L) mean standard ±error	Safe concentration (mg/l)	Slope	Intercept
M. rosenbergii	Cadmium	3.23±0.92	0.0323	3.45	3.09
	Copper	3.02±0.67	0.0302	4.24	1.56
	Lead	10.00± 0.58	0.1000	6.37	1.72
P. monodon	Cadmium	2.91±0.64	0.0291	3.11	2.75
	Copper	2.16±0.72	0.0216	3.47	1.52
	Lead	7.28±1.23	0.0728	4.13	0.96

REFERENCES

- Ajao EA (1996). Review of the state of pollution of the Lagos lagoon. NIOMR Tech. Paper 106. ISSBN 978-2345-112. p.20.
- ASEAN (Association of Southeast Asian Nations) (1978). Manual on pond culture of penaeid shrimp. ASEAN National coordinating agency of Philippines (ASEAN 77/SHR/CUL 3). iv p.114.
- Bombang Y, Thuet P, Charmantier D, Trilles M, Charmantier JP (1995). Effect of copper on survival and osmoregulation of various developmental stages of the shrimp *Penaeus japonicus* Bate (Crustacea, Decapoda). Aqua. Toxicol., 33: 125-139.
- Chinni S, Yallapragda PR (2000). Toxicity of copper, cadmium, zinc and lead to *Penaeus indicus* postlarvae: Effects of individual metals. J. of Environ. Biol. 21 (3): 255-258.
- Diaz VR (1995). Acute toxicity of cadmium (as CdCl₂2H₂0) to postlarvae stage of tiger prawn (*Penaeus monodon* Fabricius). Third- National- Symposium-in- Mari-Sci of the Philippine Association of Marine Science Pams.
- Fafioye OO, Adeogun OA, Olayinka EA, Ayoade AA (2002). Effects of sublethal concentrations of lead on growth of *Clarias gariepinus* J. of Nigerian Society for Exp. Biol. (NISEB) 2: 11-15.
- FAO (1985). Manual of methods in aquatic environment research. Part 4: Bases for selecting biological tests to evaluate marine pollution. FAO Fish Tech Paper 164
- Finney DJ (1971). Probit analysis. Third edition, Cambridge University Press, London pp. 13-28.

- Gao S, Zou D (1995). Acute toxicity of Cd, Zn and Mn to larvae of Penaeus pencillatus. Mar. Sci- Bul Haiyang Tongbao 14: 83-86.
- Kamewara Rao, K. (1974). The comparative toxicities of organophosphorous and carbamate pesticides. *Mahasagar*, 7 (1-2) 79-82.
- Miller, J.C. and Miller, J.N. (1986). Statistics for Analytical Chemistry, 2nd Edition, Ellis Horwood, Chichester, England. p.50
- Mohanta, K.N. (2000) . Development of Giant Freshwater Prawn Broodstock. Aquabyte. ICLARM, 23(3): 18-20.
- Moss SM (2004). White Shrimp in Asia- Fad or the future! International File. Fish Farmer 18 (4): 20-21.
- Okoye BBC, Oladapo AA, Ajao EA (1994). Heavy metals in the Lagoon sediments. Int. J. Environ. Stud. 37: 35-41.
- Otitoloju AA (2000). Joint action toxicity of heavy metals and their bioaccumulation by benthic animals of the Lagos lagoon. Ph.D. Thesis, University of Lagos. p. 231.
- Oyewo EO (1998). Industrial sources and distribution of heavy metals in Lagos lagoon and their biological effects on estuarine animals. Ph.D. Thesis, University of Lagos, Nigeria. p. 274.
- Prasad G (2001). Length-weight relationships of *Penaeus monodon* reared in semi-intensive culturew systems of Kerala, India. Aquabyte ICLARM 24 (1&2) pp16 17.
- Saha, SB, Bhatacharyya SB, Choudhury A, (1999). Preliminary observation on culture of *Penaeus monodon* in low-saline waters. ICLARM QUARTERLY 22 (1): 30-33