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RESEARCH ARTICLE

ACCUMULATION OF HEAVY METALS AND PESTICIDES IN BLACK CLAM *VILLORITTA CYPRINOIDES* FROM THREE DIFFERENT LOCATIONS

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ABSTRACT

The black clam *Villoritta cyprinoides* are rich source of proteins and are particularly used for human consumption along the west coast of India. The present study deals with the quantization of xenobiotics in water samples and tissue of black clams collected from three different sites namely, Padne, Kuppam and Atholi in the state of Kerala. Among the three heavy metal (Pb, Cd, Hg) screened for their presence, Pb was distributed in water samples from site A: Padne and Cd in the other two sites B: Kuppam and C: Atholi, whereas Hg was absent in all the three sites. However, both Pb and Cd were detectable in the tissue of clams from all the three sites. On analysis of pesticides, none of the water samples in the three sites had any residues of the tested pesticides. On the other hand, BHC (δ form) and endosulphan (α form) were detectable in clams collected from site B and C. These findings strongly suggest that *V. cyprinoides* can be used as a bio- indicator for monitoring heavy metal accumulation and it signals an emergency need to conserve our water bodies harboring aquatic organisms as well as to implement stringent regulations regarding industrial disposal and use of pesticides.

INTRODUCTION

Now-a-days all water sources are getting polluted due to human activities such as agriculture wherein chemical pesticides are used, irrigation, mining and draining of industrial wastes. Due to these continuous activities, the accumulation rate of heavy metals and pesticides in water bodies is increasing continuously, thus affecting the aquatic organisms, and through food chain it reaches human body resulting in serious health hazards. Among the aquatic organism, molluscs are considered as the sensitive indicator of heavy metal pollution in aquatic ecosystem (Sanjay and Jaswant, 2011).

Heavy metals are those which possess density greater than five and are toxic to all organisms when accumulated above a particular level. Trace metals from anthropogenic sources into the water bodies through disposal of domestic sewages, soaps and pesticides from farms are washed into the water bodies and accumulated in aquatic organisms (Goldberg, 1972). Discharge of industrial wastes constitute approximately 62% of the total source of heavy metals such as lead (Pb), zinc (Zn), copper (Cu), nickel (Ni), cadmium (Cd), chromium (Cr) and manganese (Mn) which are responsible for degrading the quality of the water body and also for killing a number of aquatic organisms Abubaker and Garba (2006). These metals are toxic after prolonged accumulation in the body of flora and fauna and later pass on through the food chain from aquatic organisms to man. These prevalence of heavy metals in aquatic animals is becoming a threat, thereby making them unfit for human consumption. Certain heavy metals which are sub hazardous to man, results in the minimal effects on reproduction of the snail *Biomphalaria glabrata* a schistosome vector (Abdallah *et al*, 2002).

The main source of Cd is from phosphate-based fertilizers and pesticides. In west coast of India, people consume these clams regular, unaware of the occurrence of heavy metals in it. Slow accumulation of Cd causes damage to liver and kidneys, and also leads to Osteomalacia and osteoporosis. Similarly accumulation of lead in adults leads to poor muscle co-ordination, increased blood pressure, hearing and vision impairment, reproductive problems etc. It is clear from the findings that endosulphan and BHC are still used despite government order to ban the application of these pesticides. BHC has known to cause oxidative stress and damage the brain's dopaminergic system. Increased concentration of

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endosulphan causes congenital disease in children, physical deformity, skin allergy, cerebral palsy and cancer.

In aquatic system, Cd affects the activity of antioxidant enzymes, metabolic enzymes, as well as reproduction in crabs and shrimps (Xianjiang *et al,* 2012). These heavy metals are also known to reduce the enzyme activity as well as modulate the immune responses in green mussel, *Perna viridis* (Anand *et al,* 2010; Thiagarajan *et al,* 2006). By understanding the levels of xenobiotics in tissues of clams, rate of pollution in collecting sites could be determined. Since the clams *V. cyprinoides* possess open- type circulatory system, they are constantly exposed to changes in environmental factors and pollutants. Based on these observations, *V. cyprinoides* can be used as an ideal sentinel organism to signal the water contaminants.

MATERIALS AND METHODS

V. cyprinoides: collection and maintenance

V. cyprinoides were collected with water samples from three different districts of Kerala, (Fig.1) (A) Padne, Kasaragod (latitude-12.18175, longitude-75.15054), (B) Kuppam, Kannur (latitude-12.05041, longitude-75.30880) and (C) Atholi, Kozhikode (latitude-11.3802, longitude-75.7527). The clams were maintained under laboratory conditions (26±2° C) with continuous aeration and the water was changed every day.

Water analysis

The pH and temperature of water samples were recorded immediately at the collection site. Heavy metals such as lead, cadmium, mercury and dissolved pesticides like dichloro diphenyltrichloroethane (DDT), benzene hexachloride (BHC) and endosulphan concentrations were determined using standard operating procedure for testing heavy metals in packaged drinking water/natural mineral water.

Extraction and analysis of heavy metals and pesticides in tissues

The whole tissues from *V. cyprinoides* collected from each location were carefully removed by shelling the bivalves with a plastic knife were dried and powdered. This tissue powder was weighed and digested with 15-20 mL concentrated nitric acid (HNO_3) at 100° C until the sample became colorless. To this, 1 mL of perchloric acid was added and brought to near dryness. This solution was cooled, made up to 25 mL using double distilled water and subsequently analyzed using Atomic Absorption Spectroscopy (AAS).

Preparation of standard

From stock solution of 1000 ppm, standard solution (nist traceable, 0.01, 0.02, 0.03, 0.04, & 0.05 mg) of heavy metals Cd, Pb and Hg were prepared using concentrated HNO_3. From this solution 100 mL of sample was transferred to a beaker and digested with 0.5 mL of concentrated HNO_3 and 5ml of HCL till the volume reduced to three fourth; then cooled and made up to 100 mL using distilled water. The AAS was set and the absorbance of Cd, Pb and Hg in standard and samples was measured at 228.8 nm, 283.3 nm and 253.7 nm respectively. A standard calibration curve was plotted with absorbance versus standard concentration. The samples were quality checked with standard at limit of detection 0.05 mg/L or limit of quantization 0.01 mg/L.

Analysis of pesticides using gas chromatograph

In 1 L of the water sample collected from each location, 100 g of sodium chloride was dissolved by addition of 60 mL of dichloro methane. This mixture was extracted by vigorous shaking for 30 seconds with periodic venting to release the excess pressure and left undisturbed for 2 minutes thus allowing the organic layer to separate from water phase. The dichloromethane layer was collected in a round bottom flask after passing through anhydrous sodium sulphate. This procedure was repeated and re-extracted the pesticide residue with dichloromethane and collected in round bottom flask by passing through the anhydrous sodium sulphate. The samples were concentrated by evaporating the dichloromethane at 40° C to 1 mL. Two portions of 10 mL ethyl acetate added and further this solution was evaporated to a residue at 40° C using nitrogen concentrator. This residue was dissolved in 1 mL ethyl acetate from which 1 mL was injected to Gas Chromatograph – Electron Capture Detector (GC-ECD) for analysis of organochlorine pesticide residues.

RESULTS AND DISCUSSION

The pH and temperature of water samples recorded from the collection locations of *V. cyprinoides* are given in Table 1. The heavy metals Cd, Pb and Hg levels in the water samples from these locations (Table 2) revealed that Pb is present in site A whereas it is absent in sites B and C. On the other hand, Cd was present in sites B and C but absent in Site A. Interestingly Hg was not detectable in the samples from all the three sites.

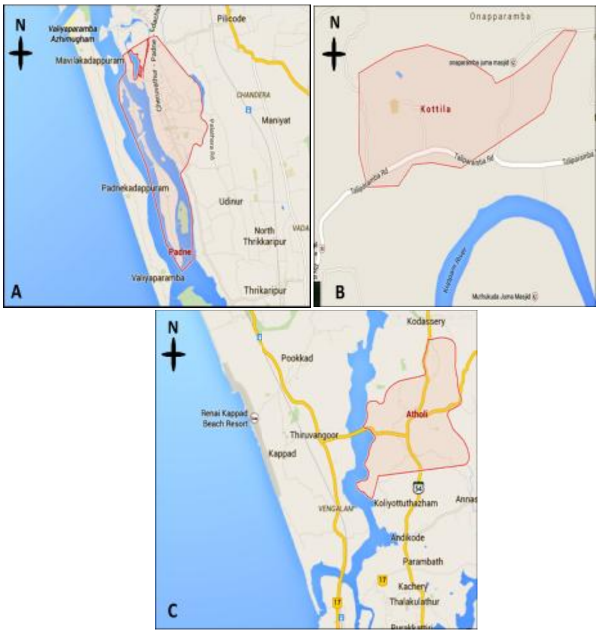

Figure 1 Map indicating sample collection site. A: Padne, Kasaragod, B: Kuppam, Kannur, C: Atholi, Kozhikode

On analysis of tissue sample in AAS data the presence of Pb and Cd in all the three sites with an overall higher concentration of Pb compared to Cd in the tissue. The greater accumulation of Pb (8.54 mg/kg) was notable in site B compared to other two sites, but its concentration was very low at site A and undetectable in water from site B and C.

Table 1 pH and Temperature of water samples from three sites

Site	pH	Temperature (°C)
A	7.4 ± 0.1	25 ± 2
B	7.2 ± 0.1	30 ± 2
C	7.6 ± 0.1	30 ± 2

A: Padne, Kasaragod, B: Kuppam, Kannur, C: Atholi, Kozhikode

organisms possess intracellular regulatory mechanism. Hg, a non-essential heavy metal is reported absent in the tissues of the clam *Galatea paradoxa*. Similarly, our observations indicate the absence of Hg in both water samples and test organism.

No trace of pesticides could be detected in any of the water samples from all the three sites. On the other hand, pesticides such as BHC and endosulphan were detectable in tissues of the clams as observed in fresh water snails (Margelette and Eric, 2007). δ BHC and α endosulphan are detectable in the tissue of samples collected from sites B and C in our study.

Table 2 Concentration of heavy metals and pesticides in the water samples and tissue of *V. cyprinoides* from three different sites

Heavy metals & Pesticides (mg/ kg)		Water collected at Site A	Water collected at Site B	Water collected at Site C	Tissue of clams collected from Site A	Tissue of clams collected from Site B	Tissue of clams collected from Site C
Heavy metals	Lead	0.1	-	-	5.05	8.54	4.14
	Cadmium	-	0.27	0.020	0.174	1.09	0.532
	Mercury	-	-	-	-	-	-
DDT	O,P DDD	-	-	-	-	-	-
	O,P DDE	-	-	-	-	-	-
	P,P DDT	-	-	-	-	-	-
	P,P DDE	-	-	-	-	-	-
	O,P DDT	-	-	-	-	-	-
	P,P DDE	-	-	-	-	-	-
BHC	α BHC	-	-	-	-	-	-
	β BHC	-	-	-	-	-	-
	γ BHC	-	-	-	-	-	-
	δ BHC	-	-	-	-	0.06	0.1
Endosulphan	α	-	-	-	-	0.16	0.08
	β	-	-	-	-	-	-
	Endosulphan sulphate	-	-	-	-	-	-
	Lindane	-	-	-	-	-	-

DDT: Dichlorodiphenyl trychloroethane, O,P DDT, P,P DDT are the isomers of DDT
DDE: Dichlorodiphenyl dichloroethylene, P,P and O,P DDE are the isomers of DDE
DDD: Dichlorodiphenyl dichloroethane, O,P DDE is the isomer of DDE
BHC: Benzene hexachloride, α, β, γ and δ are the various forms of BHC tested.
α and β endosulphan are the configurational isomers of endosulphan.

The accumulation of Pb in the tissue samples from site B and C but its absence in water from the same site clearly indicates that these organisms are capable of the removal of heavy metals from the surrounding water by their filter feeding. These observations on the detectable amount of Cd in the tissue sample of clams from site A but absent in the water sample of the same site and higher concentrations noticeable from tissue samples of clams from site B and C compared to their presence in the water sample from the same site strongly suggest that these clams can serve as bio indicator.

Similarly, Cd was highest in site B which is one- and five- fold increase compared to site C and A respectively. Based on the biological factors, there could be differences in bioaccumulation between bivalve species. Within a single species accumulation is reported as a function of age, sex, size, genotype, phenotype, feeding activity and reproductive state (Margelette and Eric, 2007). Thus, Cd in water seems to be absorbed by these clams through respiratory and digestive systems as well as body surface without significant excretion (Margelette and Eric, 2007; Ravera, 2001).

Similar to the observations by Amisah *et al*, (2009), essential heavy metals are usually kept at equilibrium, since the

In addition, the concentration of endosulphan is one- fold greater in the site B compared to site C. These observations suggest that the clams can recruit pesticides like BHC in delta form and endosulphan in alpha form from the environment during filter feeding.

CONCLUSION

In the current study, detection of heavy metals and pesticides such as BHC and endosulphan in the tissue of black clams but their absence in the surrounding water strongly indicates that *V. cyprinoides* can serve as sentinel organism for the back waters of the Indian west coast.

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References

Aanand, S., C.S. Purushothaman, A. K. Pal and K.V. Rajendran. (2010).Toxicological studies on the effect of

Copper, Lead and Zinc on selected enzyme in the adductor muscle and intestinal diverticula of the green mussel *Perna viridis. Indian journal of Marine Sciences.* 39: 299-302.

Abdallah, A.T., M.Q.A. Wanas and S.N. Thompson. (2002). Dissolved heavy metals, lead, cadmium and mercury, accumulate in the body of the schistosome vector, *Biomphalaria glabrata.* (Gastropoda; Pulmonata). *Journal of Molluscan Studies*. 69: 35-41.

Abubakar, M. and S. Garba. (2006) Level of heavy metals in terrestrial crabs. *Best Journal.* 4: 47- 80.

Amish, S., D. Adjei-Boateng, K. Obirikarang and K. K. Quagrainie. (2009). Effects of clam size on heavy metal accumulation in whole tissues of *Galatea paradoxa* (Born, 1778) from the Volta estuary, Ghana. *International Journal of Fisheries and Aquaculture*. 1: 014-021.

Goldberg, E.D., (1972). The Changing Chemistry of the Oceans. Wiley Interscience Division, New York. 250-255.

Margelette, E.T and J. Eric. (2007). Acute toxicity of Endosulphan to three fresh water snails in Jamaica. *Caribbean Journal of Science.* 43: 277-299.

Ravera, O., (2001). Monitoring of the aquatic environment by species accumulator of pollutant. A review, *Journal of Limnology*. 60: 63-78.

Sanjay kumar and Jaswant, S. (2011). Evaluation of mollusks as sensitiveindicator of heavy metal of pollution in aquatic system review. *IIOAB Journal*. 2: 49-57.

Thiagarajan, R., S. Gopalakrishnan and H. Thilagam. (2006). Immunomodulation in the marine green mussel *Perna viridis* exposed to Sublethal concentrations of Cu and Hg. *Archives of Environmental Contamination and Toxicology*. 51: 392-399.

Xianjiang, K., M. Shumei and L. Wenyan. (2012). Toxic effect of cadmium on crabs and shrimps. *Toxicity and Drug Testing*. 221-236.

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