BIOACCUMULATON, RECOVERY AND TOXICITY OF CADMIUM CHLORIDE IN BARYTELPHUSA GUREINI.

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ABSTRACT

The present study reflects the toxicity, bioaccumulation and recovery of cadmium in some vital organs of Barytelphusa gureini such as hepatopancreas, exoskeleton, muscle, gill, and gonad. The acclimated crabs of equal size were treated with different concentrations of cadmium chloride (2 - 4.5 µg/L). After finding LC50 the crabs were treated with a sub lethal concentration of cadmium chloride (3.5 µg/ L) for 2, 4, 6, 8 and 10 days and the bioaccumulation of cadmium was estimated. The experimental crabs were placed in clean water without toxicant for the same period and the cadmium was again estimated to study the recovery of it from the same organs. The trend of degree of accumulation of cadmium in different organs was as follows Exoskeletion> hepatopancreas> gill> gonad> muscle. The toxicity, bioaccumulation, recovery and its role is correlated with the findings of the earlier researchers.

KEY WORDS: *Barytelphusa gureini,* Cadmium, Bioaccumulation, Recovery

INTRODUCTION

Cadmium is ubiquitous non essential element which possesses high toxicity to human. The main source of non occupational exposure to cadmium includes smoking, air, food and contaminated water by cadmium. Acute or chronic exposure of cadmium causes respiratory distress, lung, breast and endometrial cancers, cardiovascular disorders and endocrine dysfunction (Nagata et al., (2005) and Naithani et al., (2010). It is classified as the second most dangerous metal in our environment. It occurs naturally in the environment and in an

insignificant amount. In the recent past, its concentration in aquatic systems is steadily and considerably increasing due to anthropogenic activities. Its deleterious effects on aquatic fauna by adverse effect on various physiological, biochemical and cellular processes have been reported. Experiments showed that accumulation of heavy metals in a tissue is mainly dependent on water concentrations of metals, exposure period, ecological needs, age of individuals, their life cycle, life history, feeding habits and the season of capture. Although some other environmental factors such as salinity, pH, hardness and temperature play significant roles in metal accumulation Singh et al., (2007), Mansour and Sidky (2002).

Cadmium toxicity has become the focus of intense research globally next to mercury as the most notorious of heavy metal pollutant. After absorption into the gastro-intestinal tract it is transferred to the liver, kidney and finally excreted via urine. It becomes toxic when it is not metabolized by the body and accumulates in soft tissues, liver, kidney mostly as metalloprotein . Cadmium toxicity to aquatic animals depends on complex biochemical interaction and imbalance between rates of absorption, detoxification and excretion. In aquatic animals (e.g. crabs, shrimps, oysters and mussels) heavy metals enter in to various compartments of body through different way such as respiratory tract, digestive tract, surface penetration etc., Wang et al., (2003) and Zhao et al., (1995). They are seriously harmful to the growth of aquatic life and survival, resulting in decline of their population. At the same time, as aquatic food product, these animals exposed to cadmium might threaten human health.

Crustaceans are generally appreciated as one of the major part of the human diet due to high protein contents, vitamins, minerals, low saturated fat and sufficient omega fatty acids which are known to support good health. Therefore, various studies have been taken worldwide on the contamination of different such species by heavy metals. They have been widely use as bio indicators of pollution by metals. Muscle tissue of them is the most frequently used for analysis because it is the major target tissue for metal storage and is the main edible part. Always it is important to determine the bioaccumulation capacity of heavy metals in organisms especially the edible ones in order to assess potential risk to human health Ototoloju, (2002).

Assessment of toxic heavy metals in aquatic animals can serve as bio-indicator of their impacts on these organisms as well as gives an insight to the degree of pollution of the water body in particular Farkas et al., (2000). The aim of present study was to estimate the bioaccumulation and recovery in and from the various organs and tissue as well as the health of the fresh water crab *Barytelphusa gureini* on the basis of above cited researchers.

MATERIALS AND METHODS

Acclimatization:

The adult specimens of fresh water field crab *Barytelphusa gureini*, were collected from the out skirt of paddy fields of Pune district (Maharashtra), and were brought to the laboratory. They were acclimatized in the laboratory for seven days before they were used for experiments. Only healthy crabs weighing between 30-40 gram were selected for experimentation to avoid problems of sex and size. The animals were fed with small pieces of goat flesh and uncooked oats.

Toxicity bioassay:

The aqueous stock solution of cadmium chloride was used to test the toxicity with appropriate dilution by tap water. A group of 10 crabs was exposed to six different concentrations ranging from (2 - 4.5 µg/L) of cadmium chloride. The mortality rate was noted up to 10 days, the test medium and dead crabs were removed with the interval of two days immediately. The LC_{50} was calculated by using probit analysis method Finney, (1971). After finding the $LC_{50,}$ one set of six crabs treated with a sub lethal concentration of cadmium chloride (3.5 µg/L) for 2, 4, 6, 8, and 10 days respectively. The other set of six crabs kept as control under the similar conditions without toxicant. The physicochemical parameters of water were estimated and were as follows: dissolved oxygen: 7.2-7.4 ppm, pH: 7.0-7.2, temperature: 29 ± 2^0c, salinity: 0.4-0.5 µg/ml and the total hardness: 280-288 mg/L.

Cadmium bioaccumulation:

The crabs were exposed to 3.5 µg/L sub lethal concentration of cadmium chloride for the 2, 4, 6, 8, and 10 days to determine cadmium chloride bioaccumulation in different organs like exoskeleton, hepatopancreas, gonad, gill and muscle. The organs were dissected out after the end of each exposure period and dried in an oven at 70^0c for 3 days. The dried samples were powdered using mortar and pestle, 100 mg dried powder was taken in a beaker and 10 ml of concentrated nitric acid was added to it. The mixture was shaken well and kept on hot plate to evaporate the solution. After its complete evaporation 10ml of nitric acid and 2ml of 5N per chloric acid was added. The solution was mixed well and kept on hot plate till it evaporate and become colorless. Third time again 10ml of concentrated nitric acid was added, and thoroughly mixed solution was kept on hot plate for digestion till 5ml remains in the beaker. It was cooled and made up to 25ml with 2M solution of concentrated nitric acid. This 25ml sample solution was used for metal estimation directly following the nitric acid digestion method of APHA (1998) and the PC based Atomic Absorption Spectrophotometer (AA1275BD Varian Techtron, USA) was used at 228.9 nm wavelength. The values are expressed in µg cadmium/ mg weight of sample.

Recovery of bioaccumulation:

For recovery studies cadmium chloride exposed crabs were transferred to clear water without toxicant. During recovery span on 2^{nd}, 4^{th}, 6^{th}, 8^{th}, and 10^{th} day crabs were sacrificed and the above mentioned methodology was followed for the estimation of cadmium in different organs.

Statistical analysis:

Two ways ANNOVA was used to test the differences between two groups. In all statistical analysis significance was assessed at $p<005$ level. Thus the results were found to be not statistically significant.

RESULT AND DISSCUSSION:

The results of bioaccumulation and recovery of cadmium chloride in fresh water field crab *Barytelphusa gureini* was studied at sub lethal concentration (3.5 µg/L) for 2, 4, 6, 8 and 10 days. In the present study the toxicity of cadmium chloride to fresh water field crab *Barytelphusa gureini* is determined along with the accumulation and recovery of cadmium in and from the gill, hepatopancreas, exoskeleton, gonad and muscle. Results were presented in table 1&2 and the values expressed in µg cadmium/mg weight of sample. The maximum accumulation of cadmium found in exoskeleton whereas minimum in muscle as compared to control. Values of accumulation were found intermediate in hepatopancreas, gonad and gill as compared to exoskeleton and muscle. On the whole the degree of cadmium accumulation trend in different organs was as follows exoskeleton > hepatopancreas> gill > gonad> muscle. During recovery span except exoskeletion other organs recorded a decrease in accumulation and showed recovery trend.

Bioaccumulation:

Bioaccumulation is the ability of an organism to concentrate an element or a compound from food chain and water to level higher than that of its environment. Bioaccumulation is the resultant process of much interaction within the compartments of the organisms. Metals uptake and their toxicity in aquatic fauna are influenced by many factors such as pH, hardness of water, alkalinity, temperature etc. metals exist in a variety of states and their toxicity depends on its nature and chemical forms whether it is in ionic form or in an oxidized or reduced state in combination with other organic substances and other metals. Bryan (1966) reported that the absorption of heavy metals in the alimentary canal of aquatic animals depends upon their precise chemical form. Cheroff and Dooley (1989) reviewed the literature on metal concentration in fish tissue and concluded the relationship between metal concentration, fish length, age and weight appears to be specific for metal and species of fish.

Exoskeleton:

Experiments have confirmed that cadmium absorption, accumulation and recovery by crabs obviously showed differences among the various body organs. Accumulated cadmium

distributed to all organs with the highest proportions of body content being found in the exoskeleton, hepatopancreas, gills, and so on. Exoskeleton was found to accumulate more amount of cadmium when compared to other organs. Cadmium has similar chemical properties to calcium the main component of the exoskeleton, such as the same charge number, the similar ion diameter and electronic number. Jennings & Rainbow (1979) worked on the uptake of cadmium by the crab *Carcinus maenas* and found the highest accumulation in exoskeleton. Therefore, the cadmium in water can replace the calcium entering the body via exoskeletons. Bryan (1996) found that exoskeleton and gill have higher cadmium than other organs in many aquatic organisms. The probable reasons for their death were attributed to unequal accumulation of metallic ions in various tissues and their translocation within the body from one tissue to the other.

Hepatopancreas:

Next to the exoskeleton, hepatopancreas found to accumulate considerable amount of cadmium in the present study. Hepatopancreas is the main metabolizing organ, and it is also contains all the detoxifying enzyme machinery. The toxicant in the hepatopancreas may be changed into less toxic form or may get detoxified resulting in less amount of cadmium accumulation Thus the cadmium concentration is higher in the hepatopancreas next to exoskeleton. Saltes and Bailey (1984) have reported accumulation of cadmium in hepatopancreas as a site of high bioaccumulation in aquatic organisms.

Gill:

The gill is a respiratory organ for crabs. It plays an important role in the absorption and transport of cadmium and is the target organ of cadmium in water Silvestre et al., (2004). The accumulation of cadmium in the gill may be due to adsorption to gill surfaces and dependent on the availability of proteins to which the cadmium may bind. The low concentration may be due to development of some defensive mechanism such as excessive mucous secretion and clogging of gills. The slow penetration of cadmium across the gill may be the reason for low toxicity of

this metal in the experimental animal. Huges and Floss (1978) also reported the low accumulation of zinc and cadmium in the gill of rainbow trout.

Gonad &Muscle:

Gonad and muscle were found to accumulate small amount of cadmium and might have received it through circulation via exoskeleton. It is suggested that the low accumulation of cadmium in gonad and muscle may be due to lack of binding affinity of cadmium with the proteins of gonad and muscle. During the exposure period the accumulation of cadmium in different organs of *Barytelphusa gureini* was less, but it was noticed that concentration increased with increased exposure period. It is suggested that on chronic exposure to cadmium the crab may accumulate more cadmium or may be acclimatized to toxic environment.

Recovery span:

When experimental crabs were placed in clean water after exposure to cadmium, the recovery of cadmium in almost all organs was rapid and significant. The concentration of cadmium in the organs depends on level of exposure, nature of metal and ability of the experimental crabs to metabolize or excrete the compound. Metals might be excreted by one or more of the routes proposed by Bryan (1996) across the body surface, gill, gut wall, exoskeleton and fecal matter.

It is concluded that the experimental crab from polluted area when transferred to clean water developed the capacity to metabolize or degrade the pollutant and recover from pollutant influence. In the present study muscle and gonad showed almost complete recovery as compare to other organs. Hence the crab as a food to human being is not affected by cadmium toxicity on short term of exposure.

CONCLUSION:

The cell and its molecular contents of living organisms always prone to toxicants, which cause alteration at its physiological, pathological, biochemical and immunological levels. The role of cadmium among the living organisms is very significant but its excessive discharge to aquatic environment can have an adverse effect on aquatic animals and its consumers. The present study reveals the magnitude of cadmium accumulation and recovery in the tissue and organs of *Barytelphusa gureini.* The maximum accumulation of cadmium in exoskeleton where as minimum accumulation was found in muscle, as compare to control. Values of accumulation were found intermediate in gill, hepatopancreas and gonad. During recovery span except skeleton and gill other organs recorded a decrease in accumulation and showed recovery trend. It is suggested that the studies are highly required to monitor the ecosystem and assess the toxic effect on aquatic organisms particularly crustaceans and fishes. Thus the present experimental organism becomes an agent for the evaluation of potency of cadmium as toxic substance. Further it is concluded that the bioaccumulation in the experimental crab did not exceed the permissible limits set for cadmium by FAO, (1983) and WHO, (1992a) therefore the crabs were fit for consumption.

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Bioaccumulation of cadmium in exposed *Barytelphusa gureini*
Table: 1

Organs	Values	Control	Recovery span (In days)				
			2	4	6	8	10
Gonad	Exptl Value	1.123	1.346	1.521	1.637	2.124	2.216
	SEM	±0.022	±0.035	±0.034	±0.045	±0.050	±0.057
	%V		+18.85	+20.60	+36.65	+55.96	+60.59
Exo-skeleton	Exptl Value	2.621	2.689	3.110	3.240	4.200	5.500
	SEM	±0.032	±0.025	±0.045	±0.065	±0.074	±0.085
	%V		+20.28	+31.46	+42.42	+66.45	+72.66
Gill	Exptl Value	1.405	1.436	1.581	1.638	1.841	2.162
	SEM	±0.035	±0.038	±0.055	±0.061	±0.044	±0.038
	%V		+16.28	+19.73	+24.85	+49.80	+60.95
Hepato-pancreas	Exptl Value	1.672	1.854	1.927	2.148	2.259	2.305
	SEM	±0.035	±0.030	±0.029	±0.025	±0.035	±0.044
	%V		+16.34	+20.46	+26.87	+34.62	+36.40
Muscle	Exptl Value	0.587	0.623	0.746	0.885	0.920	0.950
	SEM	±0.025	±0.028	±0.032	±0.034	±0.036	±0.040
	%V		+8.37	+11.65	+18.95	+23.48	+26.67

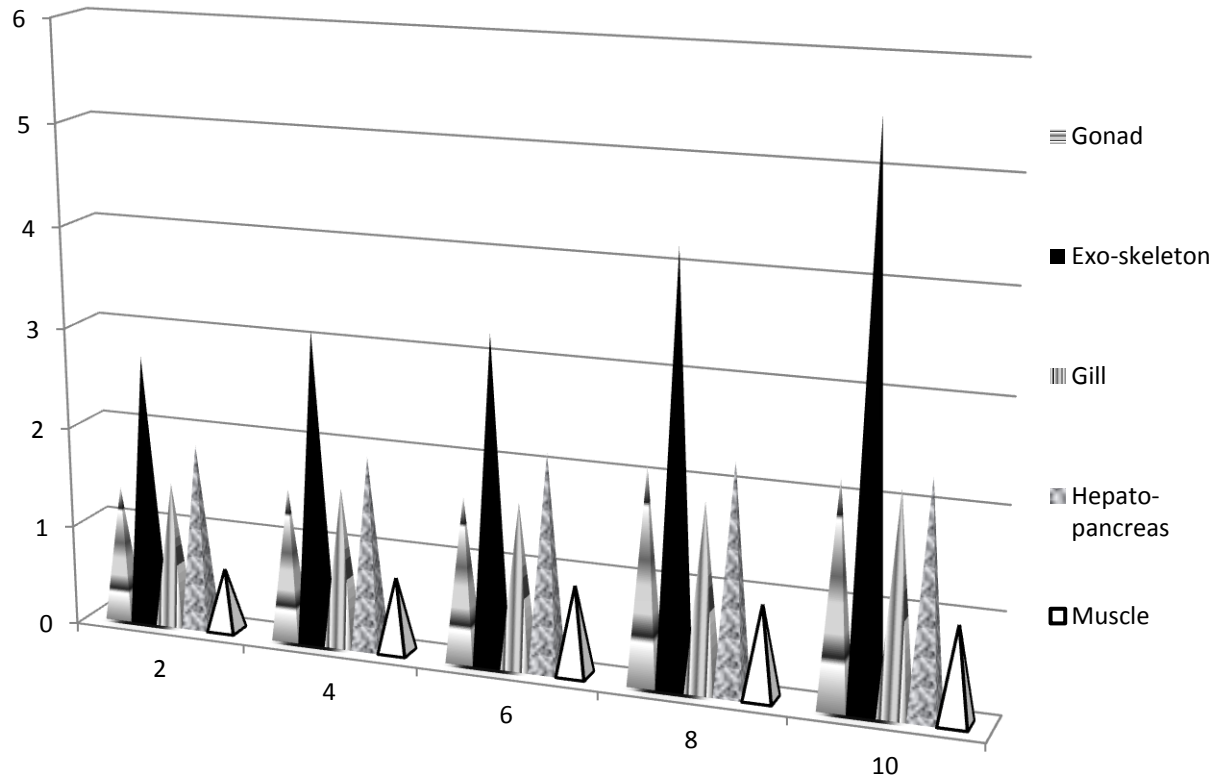
1. Experimental values expressed as µ gram of cadmium /milligram weight of sample.
2. Each value is the mean of six observations.
3. %v is coefficient of variation.
4. Experimental values are statistically different from control with statistical significance at $p < 0.005$ (not significant).

Recovery of cadmium from exposed *Barytelphusa gureini*
Table: 2

Organs	Values	Control	Recovery span (In days)				
			2	4	6	8	10

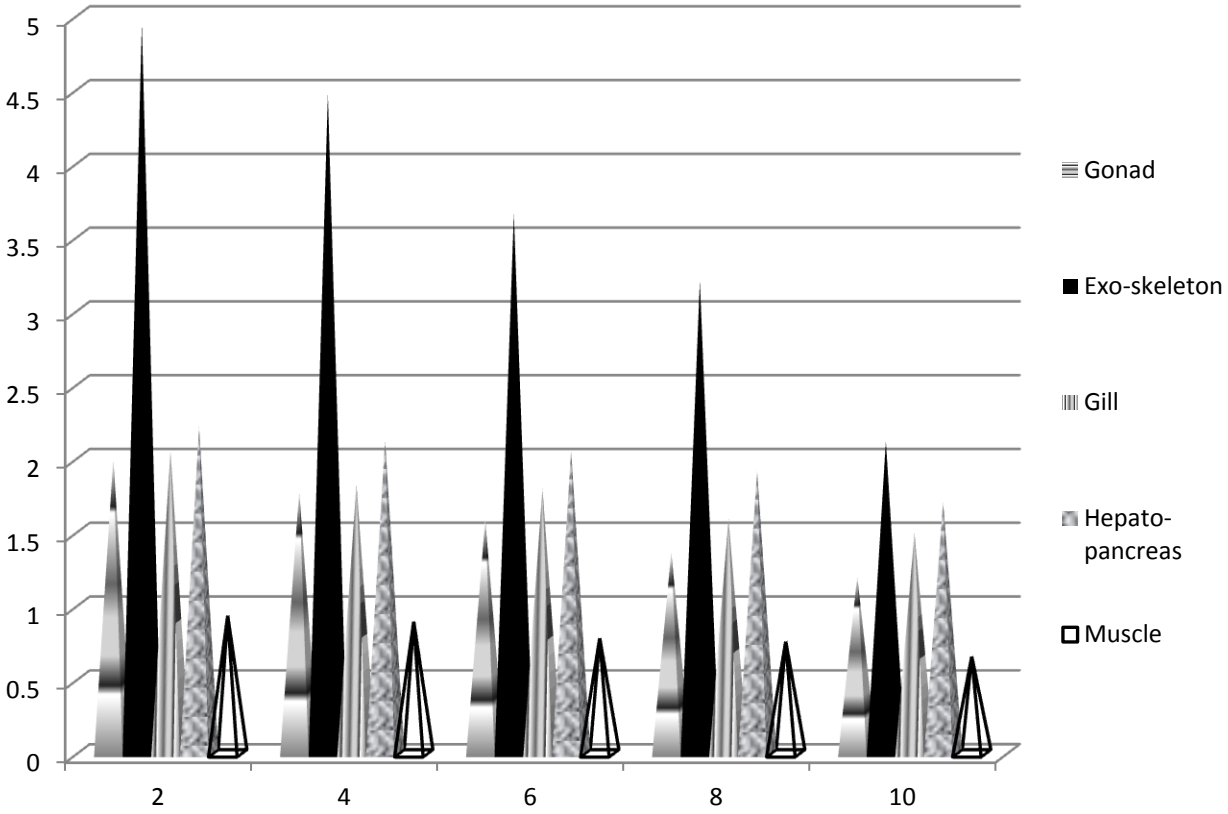
Gonad	Exptl Value	1.123	1.995	1.780	1.585	1.365	1.202
	SEM	±0.022	±0.031	±0.024	±0.023	±0.021	±0.015
	%V		+60.75	+48.14	+36.25	+26.31	+20.28
Exo-skeleton	Exptl Value	2.621	4.948	4.472	3.658	3.205	2.118
	SEM	±0.032	±0.044	±0.036	±0.021	±0.051	±0.035
	%V		+76.52	+72.36	+69.14	+66.24	+59.85
Gill	Exptl Value	1.405	2.050	1.825	1.795	1.593	1.496
	SEM	±0.035	±0.026	±0.030	±0.032	±0.038	±0.044
	%V		+42.75	+38.82	+29.35	+18.18	+16.70
Hepato-pancreas	Exptl Value	1.672	2.215	2.124	2.054	1.917	1.705
	SEM	±0.035	±0.021	±0.023	±0.032	±0.042	±0.051
	%V		+34.51	+26.92	+24.45	+20.65	+18.86
Muscle	Exptl Value	0.587	0.935	0.895	0.782	0.760	0.658
	SEM	±0.025	±0.024	±0.023	±0.022	±0.024	±0.019
	%V		+16.75	+14.10	+13.58	+12.22	+09.10

1. **Experimental values expressed as µ gram of cadmium /milligram weight of sample.**
2. **Each value is the mean of six observations.**
3. **%v is coefficient of variation.**
4. **Experimental values are statistically different from control with statistical Significance at p< 0.005 (not significant).**



Exposure span (days) vs. Exptl values

Bioaccumulation



Exposure span (days) vs. Exptl values

Recovery