



ISSN: 0976-3031

Available Online at <http://www.recentscientific.com>

International Journal of Recent Scientific Research
Vol. 4, Issue, 1, pp.023 - 027, January, 2013

International Journal
of Recent Scientific
Research

RESEARCH ARTICLE

BIOACCUMULATION OF HEAVY METALS IN THE SELECTED TISSUES OF GREY MULLET, (*MUGIL CEPHALUS*) IN THE KADUVAIYAR ESTUARY, NAGAPPATTINAM COASTAL AREA, TAMILNADU, INDIA

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ARTICLE INFO

Article History:

Received 10th December, 2012

Received in revised form 20th, December, 2012

Accepted 5th January, 2013

Published online 17th January, 2013

Key words:

Bioaccumulation of Heavy metals, grey mullet (*Mugil cephalus*), Kaduvaiyar estuary, and Nagapattinam coastal area.

ABSTRACT

The bioaccumulation of heavy metals such as zinc, chromium, copper, cadmium and lead were evaluated in the selected fish tissues of Grey Mullet, (*Mugil cephalus*) The fish organs viz., gill, intestine, liver, kidney and muscle from the selected marine fish was carefully dissected for determination of heavy metals during the year of July 2011 to July 2012 from Kaduvaiyar Estuary, Nagapattinam coastal area, Tamilnadu. The levels of heavy metals were determined using Clicos SI- 176 double beam Atomic Absorption Spectrophotometer (AAS). The highest concentration copper was observed in the intestine and the lowest concentration chromium was observed in muscles. The level of heavy metals accumulated by the freshwater fish species was exceed, the maximum permissible limits values of heavy metals for international agencies. These levels of heavy metals accumulated by the freshwater fish species might be due to increase in agricultural influx waters, domestic waters and some anthropogenic activities.

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INTRODUCTION

The problem of pollution is attracting the attention of people around the world. With increased urbanization and industrialization, there has been a rapid increase in the municipal waste water (sewage water and industrial effluents), which in turn has intensified the environmental pollution. The disposal of industrial effluents and municipal wastes is therefore a major problem for big cities. The major sources of contamination in surface water can be traced to industrial discharges, domestic waste disposal and application of agrochemicals on farm lands (Vutukuru, 2005 and Dirilgen, 2001).

The pollutants like heavy metals after entering into aquatic environment accumulate in tissues and organs of aquatic organisms. The amount of absorption and assembling depends on ecological, physical, chemical and biological condition and the kind of element and physiology of organisms. Also sexuality, weight, age, feeding habits and tissues trace can be effective. These metals after accumulation by the body of aquatic organisms enter into food chain and extremely consumed by human. Reactions of these elements depends on the concentration, physico-chemical properties, chemical bonds and their solution on the absorption, accumulation, distribution in body and physiological effects on metals. Human mediation activities have locally and episodically introduced numerous potentially hazardous metals to the environment since the onset of industrial revolution. (Farombi *et al.*, 2007 and Ashraj, 2005).

Aquatic ecosystems are very vulnerable to water pollution. Notably aquatic ecosystems are often polluted with anomalously high levels of toxicants (organic and inorganic substances), which find their way into the aquatic systems with wastewater and effluents generated from industrial enterprises. Heavy metal accumulation in aquatic ecosystem shows that they are accumulated either in aquatic organisms or in the sediment. In recent years, there has been an increasing interest in the utilization of fishes as bio indicators of the integrity of aquatic environmental system. Several studies have indicated enhanced levels of both non-essential and essential heavy metal load in muscle and liver tissues of fishes (Nayar, 2006).

For fish, gills, skin and digestive tract are potential sites of absorption of water- borne chemicals. The chemicals once absorbed are transported by the blood to either a storage point such as the bone, or to the liver for transportation. If transported by the liver, it may be stored there, excreted in the bile, or passed back into the blood for possible excretion by the kidney or gills or stored in extra hepatic tissues such as fat. Thus, the contamination of fish and the aquatic environment by heavy metals is viewed with serious concern (Malik *et al.*, 2010)

MATERIALS AND METHODS

The study was carried out at in Kaduvaiyar river, Kaduvaiyar estuary and Coastal waters (st-I,II, and st-III) at Nagapattinam during the period July 2011 –June 2011.. Fish samples, specimen of uniform size were collected in order to avoid the

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possible error due to size differences. The different organs *Viz.* (Gills, liver, Intestine and Kidney) were carefully dissected after rinsing with double distilled water and oven dried at 110⁰ C. The heavy metal concentrations in the dried samples were estimated after acid digestion following standard methods as laid down in using Atomic Absorption spectrophotometer. The results were expressed in µg/g⁻¹ metal per dry Weight.

Fish samples were de-scaled and rinsed with ultrapure water before dissection for the isolation of the following internal organs as test samples, Gills, Intestine, Kidney, Liver and flesh Muscles. Cares were taken during dissection of the internal organs to prevent any injuries and metal contaminations of the organ samples by using stainless steel dissecting kits. The isolated organs were manually cut into small pieces with stainless-steel scissor and weighed accurately to 3.00 ± 0.05 g (wet weight basis) into individual sanitised porcelain crucibles and subsequently subjected to oven drying at 180°C for 4hours. The dried samples were later ashed at 500°C for 12 hours inside a muffle furnace (THERMCONCEPT, Germany). The cooled ashes were digested with 1.5mL of concentrated analytical grade 65% HNO₃ (Merck Chemicals, Germany) and subsequently diluted with ultrapure water to 30 mL. Diluted final test solution samples were filtered through Whatman No. 1 filter paper prior to AAS analysis.

All glassware and porcelain crucibles were soaked and sanitized in aqua regia of 1:1 analytical grade 37% HCl and 65% HNO₃ (Merck Chemicals, Germany) solution, subsequently rinsed with ultrapure water, and were air-dried for 12 hours prior to usage. Sample blanks were prepared in the similar way to the test samples for background correction. Standard solutions for Cd and Pb were prepared from stock solutions (100 ppm). The test solution samples were then analysed thrice for Cd and Pb using air acetylene Flame AAS (Perkin Elmer Analyst 100). Detected metals were expressed as mg/kg wet weight (Suhaimi *et al.*, 2005).

RESULTS

Concentration of zinc was minimum 3.04 mg/l during summer season and the maximum 18.81 mg/l during monsoon season at Station 1. Concentration of zinc was minimum 3.61 mg/l during post monsoon season and the maximum 25.89 mg/l during monsoon season at Station 2. Concentration of Zinc was minimum 8.67 mg/l during summer season and the maximum 40.82 mg/l during monsoon season at Station 3. Muscle showed minimum accumulation of zinc and maximum concentration was found in the intestine.

Minimum 1.86 mg/l and maximum 4.81 mg/l values of chromium in fish tissues were recorded during summer and monsoon seasons at Station 1. Minimum 2.66 mg/l and maximum 7.71 mg/l values of chromium if fish tissues were recorded during summer and monsoon seasons at Station2. Minimum 0.18 mg/l and maximum 3.54 mg/l values of chromium in fish tissues were recorded during summer and monsoon seasons at Station 3. Muscle showed minimum accumulation of chromium and maximum concentration was found in the intestine.

Minimum 0.02 mg/l and maximum 6.03 mg/l values of copper in fish tissues were recorded during summer and monsoon seasons at Station 1. Minimum 4.9 mg/l and the maximum 18.98 mg/l values of Cu were recorded during the summer and monsoon seasons at Station2. Minimum 14.42 mg/l and maximum 92.9 mg/l were recorded during the summer and monsoon seasons at Station 3.

Kidney showed minimum accumulation of copper and maximum concentration was found in the intestine. Concentration of cadmium was minimum 0.02 mg/l during summer season and the maximum 0.04 mg/l during monsoon season at Station1. Concentration of cadmium was minimum 0.3 mg/l during summer season and the maximum 0.71 mg/l

Table 1 Bioaccumulation of selected Heavy Metals in different Seasons and Organs at Station - I during the Present Study (July 2011 - June 2012)

Heavy Metal	Season	Organs					Annual Average
		Gill	Intestine	Liver	Kidney	Muscle	
Zn	Premonsoon	14.36	22.18	6.02	5.28	3.42	10.25
	Monsoon	416.81	68.26	14.62	7.21	5.64	28.50
	Postmonsoon	3.86	10.21	5.03	3.14	1.92	4.83
	Summer	3.92	14.62	4.06	2.46	1.18	5.24
	Total	17.23	18.81	7.43	4.52	3.04	
Cr	Premonsoon	5.33	8.46	2.97	2.16	1.63	4.10
	Monsoon	11.36	7.48	5.21	3.16	2.13	5.86
	Postmonsoon	1.01	1.8	3.31	2.41	1.16	1.79
	Summer	1.86	1.26	3.92	3.16	2.91	2.74
	Total	4.81	4.57	3.85	1.86	1.95	
Cu	Premonsoon	6.14	9.18	2.14	1.85	1.92	4.24
	Monsoon	8.26	9.45	4.22	2.13	4.92	4.19
	Postmonsoon	2.01	2.96	0.92	0.81	3.62	2.06
	Summer	2.16	2.54	0.82	0.60	4.81	2.19
	Total	2.64	6.03	0.02	1.34	3.81	
Cd	Premonsoon	0.082	0.03	0.062	0.068	0.086	0.06
	Monsoon	0.074	0.032	0.07	0.056	0.062	0.05
	Postmonsoon	0.016	0.039	0.020	0.03	0.051	0.03
	Summer	0.011	0.023	0.016	0.037	0.066	0.02
	Total	0.04	0.02	0.04	0.04	0.03	
Pb	Premonsoon	0.016	0.012	0.036	0.091	0.066	0.04
	Monsoon	0.019	0.034	0.068	0.087	0.059	0.03
	Postmonsoon	0.026	0.038	0.051	0.066	0.072	0.04
	Summer	0.042	0.047	0.063	0.078	0.069	0.05
	Total	0.02	0.02	0.05	0.06	0.060	

Table 2 Bioaccumulation of selected Heavy Metals in different seasons and organs at Station - I during the present study (July 2011 - June 2012)

Heavy Metal	Season	Organs					Annual Average
		Gill	Intestine	Liver	Kidney	Muscle	
Zn	Premonsoon	26.18	57.08	11.36	10.26	10.67	17.95
	Monsoon	43.80	82.28	17.46	14.32	8.12	33.19
	Postmonsoon	2.90	8.63	8.06	7.21	13.11	7.98
	Summer	11.82	15.30	7.66	5.47	3.39	8.72
	Total	21.17	40.82	11.33	9.06	8.67	
Cr	Premonsoon	6.92	9.8	2.57	2.62	3.82	5.16
	Monsoon	9.52	13.66	4.73	4.33	4.99	7.34
	Postmonsoon	0.91	2.81	2.89	3.01	0.92	2.10
	Summer	2.48	4.56	1.62	0.98	1.21	2.17
	Total	4.15	7.71	2.97	2.66	2.73	
Cu	Premonsoon	15.28	20.46	3.92	5.24	5.38	10.01
	Monsoon	18.90	30.82	5.87	7.74	8.92	14.45
	Postmonsoon	13.01	9.36	4.32	4.90	2.87	6.84
	Summer	8.64	15.36	4.28	1.92	2.89	6.61
	Total	13.95	18.98	4.57	4.9	5.01	
Cd	Premonsoon	0.82	0.56	0.88	0.67	0.42	0.67
	Monsoon	0.96	0.89	0.93	0.72	0.63	0.82
	Postmonsoon	0.65	0.72	0.46	0.33	0.10	0.45
	Summer	0.08	0.09	0.58	0.44	0.08	0.25
	Total	0.62	0.56	0.71	0.53	0.30	
Pb	Premonsoon	0.036	0.048	0.056	0.066	0.094	0.05
	Monsoon	0.048	0.056	0.069	0.011	0.096	0.06
	Postmonsoon	0.103	0.098	0.074	0.082	0.069	0.08
	Summer	0.132	0.101	0.092	0.090	0.079	0.09
	Total	0.07	0.07	0.06	0.07	0.07	

during premonsoon season at Station 2. Concentration of cadmium was minimum 0.79 mg/l during post monsoon season and the maximum 1.63 mg/l during monsoon season at Station 3. Intestine showed minimum accumulation of cadmium maximum concentration was found in the gill.

during premonsoon season at Station 2. Concentration of Lead was minimum 0.54 mg/l during post monsoon season and the maximum 1.58 mg/l during monsoon season at Station 3. Kidney showed minimum accumulation of lead and the maximum concentration was found in the muscle.

Table 3 Bioaccumulation of selected heavy metals in different seasons and organs at Station - III during the present study (July 2011 – June 2012)

Heavy Metal	Season	Organs					Annual Average
		Gill	Intestine	Liver	Kidney	Muscle	
Zn	Premonsoon	39.26	19.81	9.32	4.39	3.82	15.32
	Monsoon	45.81	32.2	14.62	5.83	2.69	20.23
	Postmonsoon	7.13	4.46	7.09	6.12	6.93	6.34
	Summer	11.36	9.81	2.58	2.91	1.03	5.53
	Total	25.89	16.57	8.40	4.81	3.61	
Cr	Premonsoon	3.45	4.06	2.23	1.07	0.12	2.194
	Monsoon	6.73	5.82	0.66	0.83	0.049	2.906
	Postmonsoon	3.86	2.82	0.91	0.16	0.02	1.554
	Summer	0.090	0.05	0.045	0.012	0.010	6.06
	Total	3.54	3.19	0.96	0.51	0.18	
Cu	Premonsoon	92.39	96.64	34.78	15.06	12.32	44.838
	Monsoon	122.26	116.17	28.62	16.31	22.17	61.106
	Postmonsoon	71.26	69.43	16.22	14.33	18.60	37.924
	Summer	82.91	78.31	12.18	12.01	10.31	39.144
	Total	92.90	90.08	22.95	14.42	15.85	
Cd	Premonsoon	0.92	0.86	0.63	0.32	0.18	0.59
	Monsoon	2.36	2.61	1.32	1.26	1.12	1.75
	Postmonsoon	0.39	0.30	0.59	0.66	0.92	0.67
	Summer	2.38	1.26	1.83	0.91	0.90	1.46
	Total	1.63	1.25	1.09	0.79	0.77	
Pb	Premonsoon	1.34	1.86	0.91	0.86	0.32	0.99
	Monsoon	1.66	0.81	0.29	0.33	0.87	0.79
	Postmonsoon	1.99	1.17	0.66	0.37	2.01	1.24
	Summer	1.65	1.87	0.89	0.63	0.59	1.12
	Total	1.58	1.42	0.68	0.54	0.94	

Concentration of Lead was minimum 0.02 mg/l during summer season and the maximum 0.06 mg/l during monsoon season at Station I. Concentration of Lead was minimum 0.06 mg/l during summer season and the maximum 0.07 mg/l

DISCUSSION

Osman *et al.*, (2007) who reported that the monitoring of heavy metals in fish and other food items is concerned with human health protection. The linkage between predictions

from fate models and monitoring heavy metals in environmental samples shows major promise as an early warning tool for heavy metal pollution. In terms of preference, as fishes are the prime sea food to Indians, marine fishes play a significant role in determining the heavy metal levels of body tissues of Indians, who are consuming sea foods.

Heavy metal concentration in the biota is a function of the element, its source, time of sampling, total metal load, and the organism tested. Therefore, varying heavy metals concentrations and different distribution pattern at different environments may be of common occurrence. Bioaccumulation of heavy metals in tissue of marine organisms has been identified as an indirect measure of the abundance and availability of metals in the environment (Farombi *et al.*, 2007).

In the present study higher levels of bioaccumulation of all the organs was found to be high during monsoon season and low values during summer season such a concentration may be explained as follows. The concentration of a pollutant alone is not a reliable criterion in the accumulation other physical and chemical factors are also very important (Alinnor and Obiji, 2010). In the present study temperature has a minimum impact on the bioaccumulation of heavy metals in fish. Any change in the process of absorption and accumulation of heavy metals in fish can be effective due to various factors such as aquatic type, tissue, sex, weight and age of aquatic food habits fish characteristics, ecological characteristics and environmental conditions and the physical and chemical properties such as hardness of water environmental, pH, temperature, nutrients and fish growth. The probable reason for this would be that in tropical regions like India, temperature ranges at different seasons are not great and the animal was not found to show much correlation with temperature fluctuation (Mohammadi *et al.*, 2011).

In the present study heavy metals in water (Zn, Cu, Cr, Cd & Pb) were more during monsoon season. Hence, the occurrence of high bioaccumulation levels of metal in fishes during monsoon season can be attributed to the background levels under the favourable physico-chemical conditions. These findings are in agreement with the findings of (Karadede *et al.*, 2004 and Chandrasekar *et al.*, 2003).

Faromi *et al.*, (2007) who reported that the other possible reason for high levels of heavy metals in fish tissues during monsoon season may be attributed to the physiology of the fish, as these fishes shows higher feeding rates between the months October to December as the breeding season or prespawning period occurs between these months.

In general, accumulation of copper and zinc is more in the present study probable as they are the essential elements, as they have biological importance as a constituent of metal enzymes (Rojas de Astudiuo, 2005).

Olaifa *et al.*, (2004) have reported that the studies on the distribution of heavy metals in different organs of an organism are important to understand not only the extent of environmental pollution, but also the role of these metal ions in metabolic processes. During the process of intake, some of the metals actually get accumulated in the body of the organs because of the affinity between certain metals and certain

organ systems. The presence of metal binding protein, metalothionein, has favoured the accumulation of heavy metals in the kidney as it was shown to have the higher level of some of the heavy metals analysed in the investigation in the fish species. The variations in the accumulation at different stations could be attributed to the point sources of heavy metals to the ambient medium at that station

The order of accumulation of metals at different stations in different fish tissues are

Station 1: Intestine > Gill > Liver > Muscle > Kidney

Station 2: Intestine > Gill > Liver > Kidney > Muscle

Station 3: Gill > Intestine > Liver > Muscle > Kidney

In the present study, metals were accumulated at higher level in gills and intestine. The possible reason for the high levels of metals in gills may be due to the route of entry through active or passive diffusion between the gill membrane and actual medium as gill are active sites of respiration.

The higher levels of metals in intestine can be attributed to the feeding habits of fish. Since the fish *Mugil cephalus* is detritus feeder and feed either by sucking up the surface layer of the mud or by grazing on the rock surfaces leaving to the transfer of mineral particles into the system along with food. The sediments are usually enriched with trace metals and as a result, detritus feeders are exposed to more quantity of metals than fishes that are pelagic in nature (Osman *et al.*, 2010; Rejomon *et al.*, 2010 and Anim *et al.*, 2010). The elevated level of metals also can be attributed to the feeding habits. Since fishes largely prey on invertebrates – like crustaceans and molluscs – which also accumulate metals. Similar observations were also made by (Nayar, 2006). (Mukherjee and Kaviraj, 2011) have studied the higher levels of metals in kidney can be explained as follows: Kidney, as an excretory organ, is a major route of accumulation. It showed a particularly high affinity for metals, relative to other organs, thus suggesting a prominent role in the metal metabolism. The decrease in concentrations could be due to either decreased rate of accumulation or due to the fact that once the accumulation had reached a limit, proportionally and totally depending on physiological environmental conditions, the fish tissues would try maintaining their level by rejecting the excess metals by regulatory processes (Alinnor and Obiji, 2010).

Acknowledgement

The authors are deeply indebted to Professor and Head, Department of Zoology, Annamalai University, Annamalai Nagar, Tamil Nadu, India for their inspiring help, constant support and for providing adequate laboratory facilities in the department to carry out the research work.

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