



RESEARCH ARTICLE

HEAVY METAL INDUCED BIOCHEMICAL ALTERATIONS IN DIFFERENT TISSUES OF THREE FRESHWATER BIVALVE SPECIES FROM GANGAPUR RESERVOIR IN NASIK (MS)

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ABSTRACT

The present study deals with the effect of heavy metals Zn, Cu, Pb and Cd on the total protein, DNA and RNA content from different soft body tissues like mantle, gills, digestive glands and whole soft body tissue of bivalves species, *Lamellidens corrianus*, *Lamellidens marginalis* and *Parreysia cylindrical* collected seasonally (summer, monsoon and winter seasons) from Gangapur reservoir during November 2010 to October 2011. In the present study results revealed highest concentrations of heavy metals Zn, Cu, Pb and Cd during summer season in surface water and three bivalve species sampled from gangapur reservoir. The results also revealed the lowest protein, DNA and RNA concentrations in different soft body tissues in three species of bivalves sampled during summer season, might be due to bivalves were exposed to higher concentration of heavy metals in summer than winter and monsoon seasons.

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INTRODUCTION

In aquatic ecosystem, heavy metals are considered as the most important pollutants due to their highly bio accumulative nature, persistent behavior and potential of higher toxicity (Niyogi and Wood, 2004; Censi *et al.*, 2006). The biochemical composition varies according to the situation like seasonal changes, environmental factors (temperature, salinity), starvation and toxicants in the water (Verlecar *et al.*, 2007; Nandurkar and Zambare, 2010; Salaskar and Nayak 2011). In fact the changes in biological functions, structures and proteins in response to metal pollution may be used to assess the health of aquatic animals as early warning signals of various environmental risks (Venier and Zampieron, 2005) and to assess the fitness of the animal. Mahajan (2007) studied the changes in the biochemical composition of the gill, foot, digestive gland and whole soft body tissues of freshwater bivalve, *Lamellidens marginalis* exposed to chronic concentration of arsenic trioxide, cadmium chloride and lead chloride. Injal and Raut, (2009) reported lead induced alterations in protein levels of gills and mantle of freshwater bivalve, *Lamellidens marginalis*. The seasonal variation in RNA content in *Austrovenus stutchburyi* at different sites was studied by Norkko and Thrush (2006). Singh *et al.* (2010) studied the effect of sublethal treatment of deltamethrin on

protein, amino acid and nucleic acids levels in gonadal, nervous and foot tissue of *Lymnaea acuminata*. Under certain unfavourable conditions, the organism develops certain adaptive methods such as mobilization of energy from reserves to tide over the crisis and to protect themselves (Bryan *et al.*, 1986). Hence, the study of biochemical components would be much meaningful to estimate the nutritive value of the organism, and its further analysis with the metal effect would provide an intricate relation between the metal pollutants and the metabolism of the basic biochemical constituents. Therefore, it is an attempt to investigate the effect of metal stress on the biochemical components.

MATERIALS AND METHODS

The samples were collected in summer, monsoon and winter seasons during November 2010 to October 2011 from gangapur reservoir of Nashik. The heavy metals Zn, Cu, Pb and Cd concentrations were determined in surface water samples and obtained results were summarized in Table 1. The heavy metals Zn, Cu, Pb and Cd concentrations were determined from whole soft body tissues of freshwater bivalve, *Lamellidens corrianus*, *Lamellidens marginalis* and *Parreysia cylindrical* and obtained results were presented in Table 2. The total protein, DNA and RNA contents were determined from soft body tissues like mantle, gills, digestive glands and whole soft body tissues of *Lamellidens corrianus*, *Lamellidens marginalis* and *Parreysia cylindrical*. Mantle, gills,

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Table 1. Seasonal variations of heavy metal concentrations (mg/l) in surface water of gangapur reservoir of Nasik

Name of reservoir	Seasons	Zn	Cu	Pb	Cd
Gangapur reservoir	Summer	0.1008±0.0005	0.0193±0.0005	0.0250±0.0002	0.0070±0.0003
	Monsoon	0.0573±0.0004	0.0128±0.0003	0.0207±0.0003	0.0054±0.0001
	Winter	0.0812±0.0006	0.0145±0.0004	0.0228±0.0005	0.0068±0.0002
WHO standard, 1993 mg/L		03	02	0.01	0.003

± indicate Standard Deviation

Table 2. Seasonal variations in heavy metal concentrations (µg/g dry tissue weight) in whole soft body tissues of different species of freshwater bivalves at gangapur reservoir of Nasik

Species	Seasons	Zn	Cu	Pb	Cd
<i>Lamellidenscorriamus</i>	Summer	353.34±4.93	91.19±1.83	97.63±2.33	11.78±1.24
	Monsoon	213.24±3.64	64.56±1.65	64.56±2.01	05.92±0.87
	Winter	248.61±3.92	78.71±1.24	82.35±1.92	07.88±0.95
<i>Lamellidensmarginalis</i>	Summer	359.15±5.72	98.26±2.14	95.37±2.42	12.51±0.82
	Monsoon	225.09±5.27	74.42±1.45	62.53±1.86	06.37±0.65
	Winter	254.70±4.75	85.11±1.63	79.23±1.92	08.48±0.74
<i>Parreysiacylindrical</i>	Summer	367.81±5.93	91.63±2.08	93.62±2.07	13.67±0.92
	Monsoon	230.91±4.46	69.82±1.15	61.27±1.34	6.45±0.72
	Winter	255.70±4.74	82.35±1.95	77.47±1.92	9.03±0.69

± indicate Standard Deviation

Table 3. Profile of Total protein, DNA and RNA in different body tissues of three freshwater bivalve species from gangapur reservoir of Nashik (Values are in mg/100mg dry tissue weight)

Species	Parameter	Mantle			Gills			Digestive glands			Whole soft body tissue		
		Sum	Mon	Win	Sum	Mon	Win	Sum	Mon	Win	Sum	Mon	Win
<i>L. corrianus</i>	Total	43.51±0.71	53.98±1.68	51.38±1.76	53.81±1.39	64.89±1.92	61.00±2.08	51.72±1.84	63.91±1.86	60.13±2.28	49.47±1.47	62.01±1.96	58.05±1.77
<i>L. marginalis</i>	Protein	44.05±1.28	54.73±1.38	52.48±1.18	53.18±1.83	66.16±1.84	61.14±2.14	51.54±2.62	64.28±1.93	59.32±1.82	49.17±2.08	63.47±2.08	57.06±1.44
<i>P. cylindrica</i>		45.03±1.79	54.46±1.75	51.37±1.83	52.73±1.27	65.95±1.67	62.38±1.78	50.30±1.79	64.33±2.16	60.84±2.19	48.32±1.66	63.08±1.63	58.84±1.74
<i>L. corrianus</i>	DNA	1.05±0.078	1.47±0.043	1.24±0.042	1.30±0.054	1.84±0.038	1.65±0.052	1.38±0.042	2.09±0.048	1.72±0.041	1.52±0.070	2.34±0.073	2.09±0.060
<i>L. marginalis</i>		1.03±0.070	1.45±0.052	1.21±0.041	1.29±0.049	1.89±0.052	1.70±0.041	1.37±0.040	2.01±0.048	1.74±0.041	1.50±0.056	2.30±0.048	2.12±0.072
<i>P. cylindrica</i>		1.05±0.047	1.49±0.041	1.22±0.049	1.26±0.041	1.83±0.050	1.71±0.051	1.31±0.064	2.05±0.049	1.73±0.038	1.49±0.047	2.33±0.063	2.13±0.048
<i>L. corrianus</i>	RNA	3.28±0.12	4.51±0.33	3.54±0.15	5.41±0.23	6.26±0.24	6.07±0.29	6.69±0.15	8.75±0.26	8.16±0.35	4.97±0.28	6.88±0.37	6.04±0.13
<i>L. marginalis</i>		3.18±0.11	4.41±0.22	3.52±0.18	5.38±0.25	6.26±0.26	5.98±0.22	6.69±0.20	8.72±0.27	8.05±0.30	4.85±0.25	6.81±0.16	6.13±0.31
<i>P. cylindrica</i>		3.08±0.17	4.42±0.26	3.53±0.18	5.31±0.18	6.37±0.33	5.88±0.29	6.72±0.32	8.70±0.37	8.06±0.27	4.89±0.19	7.02±0.41	6.09±0.41

± indicate Standard Deviation

digestive glands and whole soft body tissues were removed and dried at 70⁰ to 80⁰C in the oven till the constant weight of dry tissues were obtained. From each powder total protein, DNA and RNA content were estimated. The total protein content of the tissues was estimated by method of (Lowry *et al.*, 1951). DNA content of the tissue was estimated by using Diphenylamine method of Burton (1956). RNA content of the tissue was estimated by following Orcinol method of Volkin and Cohn (1954). The results are presented in the Table 3.

RESULTS AND DISCUSSION

The mean values of Zn concentrations were 0.1008, 0.0573 and 0.0812 (mg/l), Cu concentrations were 0.0193, 0.0128 and 0.0145 (mg/l), Pb concentrations were 0.0250, 0.0207 and 0.0228 (mg/l), Cd concentrations were 0.0070, 0.0054, and 0.0068 (mg/l) respectively in summer, monsoon and winter seasons in surface waters of Gangapur reservoir. The mean values of Zn concentrations in whole soft body tissues of *Lamellidens corrianus* were 353.34, 213.24 and 248.61 (µg/g), Cu concentrations were 91.19, 64.56

and 78.71 ($\mu\text{g/g}$), Pb concentrations were 97.67, 64.56 and 82.35 ($\mu\text{g/g}$), and Cd concentrations were 11.78, 5.92 and 7.88 ($\mu\text{g/g}$) respectively. The mean values of Zn concentrations in whole soft body tissues of *Lamellidens marginalis* were 359.15, 225.09 and 254.70 ($\mu\text{g/g}$), Cu concentrations were 98.26, 74.42 and 85.11 ($\mu\text{g/g}$), Pb concentrations were 95.37, 62.53 and 79.23 ($\mu\text{g/g}$) and Cd concentrations were 12.51, 6.37 and 8.48 ($\mu\text{g/g}$), while in *Parreysia cylindrica* the mean values of Zn concentrations were 367.81, 230.91 and 255.70 ($\mu\text{g/g}$), Cu concentrations were 91.63, 69.82 and 82.35 ($\mu\text{g/g}$), Pb concentrations were 93.62, 61.27 and 77.47 ($\mu\text{g/g}$) and Cd concentrations were 13.67, 6.45 and 9.03 ($\mu\text{g/g}$) during summer, monsoon and winter seasons respectively. The results indicate that the Zn, Cu, Pb and Cd concentration were highest in surface water in summer season and lowest in monsoon season at gangapur reservoir. It was also observed that the mean concentrations of Zn, Cu, Pb and Cd were highest in summer season and lowest in monsoon season in whole body tissue of three bivalve species, *Lamellidens corrianus*, *Lamellidens marginalis* and *Parreysia cylindrica* sampled from the gangapur reservoirs. The mean concentrations of Pb was highest in *Lamellidens corrianus*, Cu was highest in *Lamellidens marginalis*, while Zn and Cd were highest in *Parreysia cylindrica* at studied reservoir in three seasons. In the present seasonal study the lowest total protein, DNA and RNA contents were observed in different soft body tissues like mantle, gills, digestive glands and whole soft body tissue of three species of bivalves sampled during summer season, might be due to bivalves were exposed to higher level of pollutant in summer than winter and monsoon seasons. The observed low level of protein contents in different tissues indicate that, environmental stress reduces the rate of protein synthesis or increase the proteolysis to cope with the high energy demands under toxicants stress (Vincent *et al.*, 1995; Waykar and Lomte, 2001).

DNA content is significantly influenced due to the stress condition caused by exposure of pollutants. The metals generate reactive oxygen species (ROS) and other reactive intermediates or react directly with DNA (O'Brien *et al.*, 2003) and cause damage to DNA through inhibition of DNA repair enzymes (Hartwig *et al.*, 2002), or by binding to histone protein (Bal *et al.*, 2000). The low level of RNA in soft body tissues of bivalves might be due to damage in DNA, poor rate of synthesis of enzymes necessary for transcription or increased catabolism of RNA due to their abnormalities on binding to abnormal. The cellular degradation, rapid histolysis and decreased rate of protein synthesis are the possible reasons (Andhale and Zambare 2011). Shaikh (2011) reported the seasonal changes in protein content in different tissues of *Lamellidens marginalis* from Pravara river. Seasonal changes in DNA and RNA content in different tissues of *Lamellidens marginalis* from Nathsagar dam at Paithan was reported by Pardeshi (2011). This work provides information regarding the heavy metal level in surface water in three bivalve species and biomarker responses in relation to heavy metal accumulation at gangapur reservoir of Nasik.

Conclusion

The study clearly demonstrate that the mean values of Pb and Cd in surface water of gangapur reservoir were higher than WHO recommended limits for drinking water standard; whereas those of zinc and copper were within the limits. Therefore, this study indicates that the surface water of

gangapur reservoir were polluted by heavy metals Pb and Cd. Therefore, it is suggested that there is need of treating water by central water treatment plant and removing heavy metals before utilizing this water for crop and drinking purpose. Heavy consumption of other aquatic animals from the reservoir by humans, as in the current situation, is therefore, at a risk of health implications. Hence, it is recommended that regular monitoring are needed to maintain water quality.

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