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BIOACCUMULATION OF HEAVY METALS IN SEA TURTLES FROM THE WEST COAST OF INDIA

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ABSTRACT

The overall accumulation of Zinc in the olive ridleys ranged between 3.53 and 15.2 μ g/g (mean = 8.08+3.69 μ g/g) and from 1.53 to 40.4µg/g (mean=11.5±11.07µg/g) in the green turtles. The accumulation of Lead in the olive ridleys varied from BDL (Below Detectable Limit) to 0.226µ g/g (mean = 0.052+0.083µg/g) and from BDL to 1.39µg/g (mean=0.273+0.481µg/g) in the green turtles. Very low concentration of Lead was recorded in both the species and concentration was below the detectable limit in most of the tissues. The concentration of Cadmium in the olive ridleys ranged from 0.056 to $29.1\mu g/g$ (mean=6.97+8.79 μ g/g) and from 0.011 to $37.5\mu g/g$ (mean=5.54+10.56μg/g) in the green turtles. Higher concentration of Cadmium was in the olive ridley. In the olive ridleys the Copper ranged between 0.241 to 3.18μ g/g (mean= 0.81±0.776μg/g) and from 0.06 to 17μ g/g (mean 3.761+5.481 \mu g/g) in the green turtles. Mercury level ranged between BDL to 130 \mu g/g (mean=26.25+38.81 \mu g/g) in the olive ridley and in the green turtle it ranged from BDL to 50µg/g (mean=4.167+14.43µg/g) and the concentration of Mercury was higher in the tissues of olive ridleys than the green turtles. In view of the consumption of turtle meat and incidence of chelonotoxication, the present study is of relevance.

INTRODUCTION

Sea turtles are widely distributed in the tropical, sub-tropical and temperate waters. Recent reports have indicated that their populations are threatened by the problem of marine pollution (Stroelli et al., 1998). Heavy metals are one of the major pollutants and have the nature to concentrate in the tissues of animals and their concentrations vary according to biological process such as age, sex, reproduction and migration (Saeki et al., 1999). Reports are available on the detrimental effects of pollution on sea turtles particularly pollution by plastic debris, tar balls, persistent organochlorine compounds and heavy metals (Carr. 1987; Sakai et al., 2000) and these enter the turtle body mainly through food. According to Dara (1997), mercury, lead, copper, cadmium and zinc are generally known as "toxic heavy metals". Previous studies on the heavy metal analysis of higher vertebrates revealed that the distribution of these metals in the body of the animals varied depending on the type of species (Noda, 1995). In this context, the distribution of heavy metals in the body of sea turtles is expected to be more interesting, since they have unique tissues such as green fat and carapace, which can make the bioaccumulation to be more characteristic. Pollutants from coastal industries threaten the existence of sea turtles and many parts of Indian coasts are vulnerable to these problems. Hence, studies are necessary to establish the relationship between heavy metal accumulation and biological process among the sea turtles. Furthermore, investigations on heavy metal accumulation in green turtles are especially needed to elucidate the effects of the shift in feeding by younger juvenile turtles and adult ones. The former inhabit pelagic ocean and are considered to be carnivore, probably zooplankton eaters, while coastal adults are found to be herbivores (Carr, 1987 and Marguez, 1990).

MATERIALS AND METHODS

Samples of muscle, liver, heart and kidney of the green turtle (chelonia mydas) and the olive ridley (lepidochelys olivacea) turtles, which were caught incidentally and sold in the markets of Kattakada (Kerala) and Tuticorin (Tamil Nadu) were collected during December 1998, March 1999, July 1999 and December 2000. Carapace length range of the green turtle was from 103 cm to 120 cm and the total number of turtles sampled was 12. In the case of the olive ridley, the carapace length of the animals sampled ranged from 62 cm to 72 cm and totally 16 were sampled. The collected tissue samples were stored in polythene bags and kept at 4°C in the laboratory.

The samples were washed three times in metal free distilled water, dried for several days at 80° C in hot air oven until they reached constant weight. Then the samples were ground to powder using a glass pestle and stored in a desiccator at room temperature. During analysis, each tissue sample weighing 5 g was acid digested to a transparent solution with a mixture of nitric, perchloric and sulphuric acids in the proportion of 3:1 (Walting, 1981). The digested solution was filtered through millipore filter and again diluted with deionized water. The samples were made up to 25 ml with metal free distilled water. Digested and diluted samples were then subjected to metal quantification. Zinc, Cadmium and Copper and lead were directly determined by inductively coupled plasma spectrometer (model ICA -AES; Model Jobin Yvon-J7 24) after calibrating the instrument with suitable blank and a series of known standards for Zinc, Copper, Cadmium and Lead. The accuracy of the instrument (detection limits was 5 ng) was verified and equated by analyzing the reference standards. All metal concentrations are expressed in µg/g dry tissue weight, Mercury analyzer was used to determine mercury levels. Model ECIL MA 5800D1 was used for the analysis. Mercury levels were also expressed as µg/g dry tissue weight.

RESULTS

Heavy metal accumulation in the tissues of sea turtles

Zinc (Zn)

Analyses of various tissues of the turtles indicated that accumulation of Zn in the liver ranged between 3.76 and 8.34 μ g/g (mean = 5.42 \pm 2.53 μ g/g) in the olive ridley and between 4.92 and 18.2 μ g/g (mean = 9.48 \pm 7.54 μ g/g) in the green turtle. In the heart, the concentration ranged between 6.57 and 15.2 $\mu g/g$ (mean = 12.12+4.18 $\mu g/g$) in the olive ridley and from 1.53 to 16.1 $\mu g/g$ (mean = 9.06±7.29 $\mu g/g$) in the green turtle. In the muscle, the concentration ranged between 3.53 and 7.97 μ g/g (mean = 5.53 \pm 2.24 μ g/g) in the olive ridley and from 2.04 to 6.83 $\mu g/g$ (mean = 5.17±2.71 $\mu g/g$) in the green turtle. In the kidney, the concentration ranged between 6.26 and 11.1 μ g/g (mean = 9.04 \pm 2.5 μ g/g) in the olive ridley and from 5.34 and 40.4 μ g/g (mean = $22.24+17.56 \,\mu g/g$) in the kidney of the green turtle (Table 1).

Maximum accumulation of Zn was recorded in the kidney, liver and heart of the green turtles (40.4, 18.2 and 16.1 μ g/g, respectively) and in heart of the olive ridley (15.2 μ g/g). Accumulation of Zn varies considerably between the two species of sea turtles (Table 1).

Table 1. Concentration of zinc $\mu g/g$ (mean \pm standard deviation and ranges) in the tissues of the olive ridley Lepidochelys olivacea and the green turtle Chelonia mydas collected from Kattakada, Kerala and Tuticorin, Tamil Nadu

Tissues	Lepidochel	Chelonia mydas	
	Kattakada	Tuticorin	Tuticorin
Liver	5.42 ± 2.53 (3.76 - 8.34)	(5.42 - 5.42)	9.48 ± 7.54 (4.92 - 18.2)
Heart	12.12 ± 4.18 (6.57 - 15.2)	(12.3 - 12.3)	9.06 ± 7.29 (1.53 - 16.1)
Muscle	5.53 ± 2.24 $(3.53 - 7.97)$	(6.05 - 6.05)	5.17 ± 2.71 (2.04 - 6.83)
Kidney	9.04 ± 2.5 (6.26 - 11.1)	-	22.24 ± 17.56 (5.34 - 40.4)

Lead (Pb)

The accumulation of Pb in the liver ranged between below detectable level (BDL) and 0.14µ g/g $(\text{mean} = 0.047 \pm 0.082 \mu \text{ g/g})$ in the olive ridley and from BDL to $0.17 \mu \text{ g/g}$ (mean= 0.057 ± 0.987) in the green turtle. In the heart, the concentration of Pb in the olive ridley was below BDL in the green turtle and it ranged between BDL and $0.79\mu g/g$ (mean = $0.26\pm0.46 \mu g/g$). In the muscle, the Pb concentration ranged between BDL and 0.226 µg/g (mean = 0.075+0.13µg/g) in the olive ridley but in the green turtle, it was below BDL. In the kidney, the Pb concentration ranged from BDL to 0.21μ g/g (mean = $0.10\pm0.12\mu$ g/g) in the olive ridley and from BDL to 1.39μ g/g (mean = $0.77\pm0.70\mu$ g/g) in the green turtle (Table 2).

The level of Pb was very less and it was below detectable limit in majority of the tissues of both the species. Very meager accumulation of Pb was detected in the liver of the olive ridley (0.14 $\mu g/g$) and in the green turtle (0.17 $\mu g/g$). Similarly, very low value of Pb was recorded in the heart of the green turtle $(0.79\mu g/g)$ and in the muscle of the olive ridley $(0.226\mu g/g)$ (Table 2).

Cadmium (Cd)

The accumulation of Cd in the liver of the olive ridley ranged from 1.38 to 15.1 μ g/g (mean = $6.46 \pm 7.51 \mu g/g$) and from 0.504 to $7.740 \mu g/g$ (mean = $3.511 \pm 3.76 \mu g/g$) in the green turtle. In the heart, the Cd level ranged between 0.273 to $7.62\mu g/g$ (mean = $2.79\pm4.17\mu g/g$) in the olive ridley and in the green turtles it was between 0.019 to $37.5\mu g/g$ (mean = $12.55\pm21.6\mu g/g$). In the muscle of the olive ridley, the Cd level ranged from 0.097 to $7.13\mu g/g$ (mean = $2.45\pm4.94\mu g/g$). The level of Cd in the muscle of green turtles ranged from 0.011 to $7.77\mu g/g$ (mean $2.75\pm4.35\mu g/g$). In the kidney of the olive ridley, the Cd ranged from 6.94 to $29.10\mu g/g$ (mean = $18.74\pm11.15\mu g/g$) and in the green turtle it was 0.064 to $7.730\mu g/g$ (mean = $3.36\pm3.94\mu g/g$) (Table 3).

Table 2. Concentration of lead μg/g (mean ± standard deviation and ranges) in the tissues of the olive ridley Lepidochelys olivacea and the green turtle Chelonia mydas collected from Kattakada, Kerala and Tuticorin, Tamil Nadu

	Species			
Tissues	Lepidochelys olivacea		Chelonia mydas	
	Kattakada	Tuticorin	Tuticorin	
Liver	0.04 ± 0.082 (BDL-0.14)	(0.144)	0.057 ± 0.987 (BDL - 0.17)	
Heart	BDL	BDL	0.26 ± 0.46 (BDL-0.79)	
Muscle	0.075± 0.13 (BDL - 0.226)	BDL	BDL	
Kidney	0.10 ± 0.12 (BDL - 0.21)	BDL	0.77 ± 0.70 (BDL-1.39)	

Maximum accumulation of the Cd $(37.5\mu g/g)$ was found in the heart of the green turtle, followed by the kidney of the olive ridley $(29.10\mu g/g)$. A moderate concentration of 15.1 and 7.740 $\mu g/g$ were recorded in the liver of the olive ridley and the green turtle respectively. Among the two species of sea turtles, the green turtles recorded more accumulation of Cd than the olive ridley (Table 3).

Copper (Cu)

The level of Cu in the liver ranged between 0.668 and $3.18\mu g/g$ (mean = $1.829\pm1.26\mu g/g$) in the olive ridley turtles and between 0.26 and 17μ g/g (mean = $9.06\pm8.39\mu g/g$) in the green turtle. In the heart, the Cu level ranged from 0.62 to $0.83\mu g/g$ (mean = $0.708\pm0.11\mu g/g$) in the olive ridley and from 0.019 to $37.5\mu g/g$ (mean = $12.55\pm21.60\mu g/g$) in the green turtle. In the muscle, the Cu accumulation ranged between 0.241 and $0.59\mu g/g$ (mean = $0.381\pm0.184\mu g/g$) in the olive ridley and from 0.96 to $2.6\mu g/g$ (mean = $1.02\pm1.36\mu g/g$) in the green turtle. The level of Cu in the kidney ranged between 0.27 and $0.71\mu g/g$ (mean = $0.44\pm0.23\mu g/g$) in the olive ridley and between 0.766 and $9.92\mu g/g$ (mean = $3.99\pm5.13\mu g/g$) in the green turtles (Table 4).

Table 3. Concentration of cadmium $\mu g/g$ (mean \pm standard deviation and ranges) in the tissues of the olive ridley Lepidochelys olivacea and the green turtle Chelonia mydas collected from the markets of Kattakada, Kerala and Tuticorin, Tamil Nadu

Tissues	Species			
	Lepidochelys olivacea		Chelonia mydas	
	Kattakada	Tuticorin	Tuticorin	
Liver	6.46 ± 7.51 (1.38 - 15.1)	n) yiga in kalan Lipti Linun linun L	3.511 ± 3.76 0.504-7.740	
Heart	2.79 ± 4.17 (0.273 - 7.62)	emplical har	12.55 ± 21.6 $(0.019 - 37.5)$	
Muscle	2.45 ± 4.94 (0 097 - 7 13)	no e Januares	2.75 ± 4.35 (0.011 - 7.77)	
Kidney	18.74 ± 11.15 $(6.94 - 29.10)$		3.36 ± 3.94 (0.064 - 7.730)	

Among the two species, the tissues of green turtles concentrated more amount of Cu than that of the olive ridleys (Table 4).

Table 4. Concentration of copper $\mu g/g$ (mean \pm standard deviation and ranges) in the tissues of the olive ridley Lepidochelys olivacea and green turtle Chelonia mydas collected from the markets of Kattakada, Kerala and Tuticorin, Tamil Nadu

Tissues	Species			
	Lepidochelys olivacea		Chelonia mydas	
	Kattakada	Tuticorin	Tuticorin	
Liver	1.829 ± 1.26 $(0.668 - 3.18)$	BDL - 1.77	9.06 ± 8.39 (0.26-17)	
Heart	0.708 ± 0.11 $(0.62 - 0.83)$	BDL - 5360	12.55 ± 21.60 $(0.019 - 37.5)$	
Muscle	$0.381 \pm 0.184 \\ (0.241 - 0.59)$	BDL - 0.2430	1.02 ± 1.36 $(0.96 - 2.6)$	
Kidney	$0.44 \pm 0.23 \\ (0.27 - 0.71)$	BDL - 0.3310	3.99 ± 5.13 (0.766 - 9.92)	

Mercury (Hg)

The level of Hg in the liver of the olive ridley ranged between BDL and $105\mu g/g$ (mean = 39.66 $\pm 57.01 \mu g/g$) and from BDL to $50 \mu g/g$ (mean = $16.66 \pm 28.86 \mu g/g$) in the green turtle. In the heart, the Hg level ranged between BDL and $20\mu g/g$ (mean = $6.66\pm11.54\mu g/g$) in the olive ridley and in the green turtle it was BDL. In the muscle of the olive ridley, Hg level ranged from BDL to 20µg/g $(\text{mean} = 6.66 + 11.5 \,\mu\text{g/g})$ and in green turtle it was BDL. In the kidney of olive ridley, Hg level ranged from BDL to $45\mu g/g$ (mean = $23\pm22.51\mu g/g$) and in the green turtle it was BDL (Table 5).

Table 5. Concentration of mercury $\mu g/g$ (mean \pm standard deviation and ranges) in the tissues of the olive ridley Lepidochelys olivacea and the green turtle Chelonia mydas collected from the markets of Kattakada, Kerala and Tuticorin, Tamil Nadu

Tissues	Species			
	Lepidochelys olivacea		Chelonia myda:	
	Kattakada	Tuticorin	Tuticorin	
Liver	39.66 ± 57.01 (BDL - 105)	BDL - 130	16.66 ± 28.86 (BDL – 50)	
Heart	6.66 ± 11.54 (BDL - 20)	BDL - 20	BDL To accuse and	
Muscle	6.66 ± 11.5 (BDL - 20)	BDL	BDI	
Kidney	23 ± 22.51 (BDL - 45)	BDL-40	BDI	

Heavy metal accumulation among the species

Combined values of heavy metal accumulation in all the tissues of the olive ridley and the green turtle are shown in (Table 6). The accumulation of Zn in the olive ridley ranged between 3.53 and $15.2 \mu g/g$ (mean = $8.08 + 3.69 \mu g/g$) and from 1.53 to $40.4 \mu g/g$ (mean = $11.5 \pm 11.07 \mu g/g$) in the green turtle. The concentration of Zn showed higher levels in the green turtle when compared with that of the olive ridley.

The accumulation of Pb in the olive ridley turtle varied from BDL to 0.226 g/g (mean = 0.052 $+0.083\mu g/g$) and from BDL to $1.39\mu g/g$ (mean = $0.273\pm0.481\mu g/g$) in the green turtle. Very low concentration of Pb was recorded in both species. Concentration was not detectable in most of the tissues of both the species. The concentration of Cd in the olive ridley ranged from 0.056 to 29.1 $\mu g/g$ (mean = 6.97±8.79 $\mu g/g$) and from 0.011 to 37.5 $\mu g/g$ (mean = 5.54±10.56 $\mu g/g$) in the green turtle. Comparatively higher concentration of Cd was recorded in the olive ridley than in the green turtle. In the olive ridley turtle, concentration of the Cu ranged between 0.241 to 3.18µg/g (mean = $0.81\pm0.776\mu g/g$) and from 0.06 to $17\mu g/g$ (mean = $3.761\pm5.481\mu g/g$) in the green turtle. Remarkable difference in the accumulation of Cu was noted between the two species of sea turtles. Hg level ranged between BDL to $130\mu g/g$ (mean = $26.25\pm38.81\mu g/g$) in the olive ridley and from BDL to $50\mu g/g$ (mean = $4.167\pm14.43\mu g/g$) in the green turtle. The olive ridley turtle exhibited higher level of Hg concentration than the green turtle.

Table 6. Combined value of heavy metal concentration $\mu g/g$ (mean \pm standard deviation and ranges) in the olive ridley and green turtles collected from the markets of Kattakada, Kerala and Tuticorin, Tamil Nadu

Species	Zinc	Lead	Cadmium	Copper	Mercury
Olive ridley (n=16)	8.08 ± 3.69	0.052 ± 0.083	6.97 ± 8.79	0.81 ± 0.776	26.25 ± 38.81
	(3.53–15.2)	(BDL 0.226)	(0.056-29.1)	(0.241-3.18)	(BDL -130)
Green turtle (n=12)	11.5 ± 11.07	0.273 ± 0.481	5.54 ± 10.56	3.761 ± 5.481	4.167 ± 14.43
	(1.53-40.4)	(BDL-1.39)	(0.011-37.5)	(0.06-17)	(BDL-50)

BDL = Below Detectable Limit

DISCUSSION

Davenport and Wrench (1990) while reporting the metal level in the leatherback turtle, states 'for most heavy metals (Hg, Cd, Pb, Zn, Ni), the liver appears to contain the highest levels' and 'this observations is in line with the trend in other marine vertebrates'. Sakai et al. (1996) also agreed with the above opinion in that 'heavy metal concentrations and burdens were high in liver, kidney, pancreas and hard tissues, while low in brain and fat tissue. Higher concentration of Zn was noticed in the tissues of muscle, heart, liver and kidney in the present study (mean 5.42 to 12.12 µg/g) in the olive ridley. With regard to the green turtle, the mean values ranged from 5.17 to 22.24µg/g in the above four tissues. In both the species, kidney exhibited higher level of accumulation of zinc. Sakai et al. (2000) found Zn accumulation to be always higher in fat tissue than in any other tissues. They attribute this to the pigmented proteins, which seem to play in Zn storage in the body. The present study showed higher level of Pb accumulation in the kidney and low level in the heart, liver and muscle of the olive ridley and in the green turtle the mean lead level ranged from BDL to 0.77µg/g in different tissues. These values are comparable to those of Sakai et al. (2000). The lead is a non-beneficial element but toxic to humans. Even at very low level of concentration of lead in human beings, it can cause encephalopathy, anaemia and renal problems (FAO/WHO, 1972). In the present observations, higher Cd concentration was observed in the kidney, liver and heart of the olive ridley as well as in the green turtle. The mean concentration in the olive ridley ranged from 2.79 to 18.74µg/g in the heart and kidney. In the green turtle, higher level of 12.5µg/g was noticed in the heart and 3.51 g/g observed in liver. In the studies carried out by Aguirre et al. (1994) and Sakai et al. (2000), very high levels of 9.30µg/g

and 5.58µg/g of cadmium respectively, were noticed. In the case of the leatherback and loggerhead turtles also, higher Cd levels was recorded by Caurant et al. (1994). In the present study the mean level of Cu in the olive ridley in different tissues, such as muscle, heart, liver and kidney varied between 0.381 and 1.829 \mu g/g. Higher concentration of 1.829 and 0.708 \mu g/g was noticed in the liver and heart, respectively. In the green turtle, mean Cu level varied from 1.02 to 12.55μg/g and higher concentration was noticed in the heart (12.55μg/g) and liver (9.06μ g/g). Accumulation of copper in green turtle recorded in the present study could be compared with the earlier works i.e., 50.2µg/g (Sakai et al., 2000) and 87.6µg/g (Aguirre et al., 1994). Similar values were noticed in the loggerhead by Caurant et al. (1994). Higher Hg concentration (39.66μg/g) was noticed in the liver of the olive ridley in the present study, when compared to 0.01 to 0.28µg/g in the tissues of the green turtle observed by Sakai et al. (2000). Mercury is known to be accumulated in the higher trophic level of organisms in the food chain (Honda et al. 1987). Since main food items are lower trophic organisms such as the benthic organisms (conchs, clams, crabs, shrimps etc.) for the loggerhead turtle, sea grasses (weeds) for the green turtle, and jellyfish for the leatherback turtle (Marquez, 1990), accumulation of Hg is less in these turtles. The higher level of accumulation of mercury in the olive ridley may be due to its feeding on a variety of fishes. The above observation proves the existence of species-specific distribution of heavy metals among the turtles depending on their feeding habits. Cd accumulation in marine mammals and seabirds has been considered to be related to their feeding habits (Noda et al., 1995; Kim et al., 1996). The unique trend of Cd accumulation with growth of the green turtles might be associated with the shift in the feeding habits in their life stage. During the pelagic stage, the juvenile green turtles are likely to feed on zooplankton, while during the coastal stage, green turtles mainly feed on sea grasses and seaweeds. This shift in feeding may explain Cd accumulation in the green turtle. Cd concentrations in sea plants ranged from 0.099 to 1.14µ g/g dry weight with a mean of 0.581µg/g dry weight (Fujise, 1987). Cd concentrations in zooplankton were generally higher than those of sea plants (Fujise, 1987). In the amphipods the mean Cd levels were comparable to the concentration found in the liver of green turtles (Sakai et al., 2000). The apparent difference in the levels of Cd in zooplankton and sea plants is an explanation for the specific Cd accumulation found in green turtles during different stages of growth. Rapid Cd accumulation found in pelagic juvenile stage is also suggested because Cd concentrations in eggs were quite low (Sakai et al., 2000). After returning to coastal areas, the green turtles start to feed on sea grass and seaweeds, which contain comparatively low concentrations of Cd. Thus, Cd concentrations in green turtles decrease because the amount of Cd intake is smaller than the amount of excretion. A wide range of Cd concentration in the liver suggests variable rates of shift in feeding habits by individual green turtles, also due to shorter residence time of Cd in the liver than that in the kidney (Schuhammer, 1987). The decrease in Zn concentrations correlated with Cd levels in the kidney is deemed to arise from the decline of metallothionein concentrations because of less Cd intake with a growth. Cu concentrations in the kidney also decreased with growth similar to Zn. As stated above, Cu concentrations in the liver of green turtle were much higher than those in the kidney. On the contrary, Zn levels were almost similar in the liver and kidney. Metallothionein is able to find Zn, Cu and Cd while its affinity is higher for Cu than that for Zn or Cd. In view of consumption of turtle meat regularly in some areas, the present study on the estimation of heavy metals in the tissues of the olive ridley and the green turtle is of relevance.

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