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Size and Sex Related Study on Cadmium Accumulation in Different Organs of the Freshwater Field Crab, *Oziotelphusa senex senex* (Fabricius)

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Cadmium concentration, determined by AAS, increased significantly in haemolymph, hepatopancreas, gill and carapace of two size groups of crabs of both sexes after Days 1 and 2 of exposures to lethal and Days 1 and 15 of exposures to sublethal concentrations of cadmium. The increase was very high in the organs of crabs exposed to lethal concentrations, and was significantly more at Day 2 than at Day 1. Though there was an increase in accumulation from Day 1 to Day 15 in sublethal concentrations, it was significant only in hepatopancreas and carapace but not in haemolymph and gill. The level of accumulation of the metal was more in the organs of small crabs of both sexes, and was more in the organs of males than in those of females. In all these groups the degree of accumulation among the organs was in the order: gill > haemolymph > hepatopancreas > carapace in lethal and carapace > hepatopancreas > haemolymph > gill in sublethal concentrations. Thus, the level of accumulation of cadmium in the organs of carb was dependent upon the ambient concentration of cadmium, the period of exposure, and also the size and sex of the animal.

Key Words: Cadmium, Accumulation, *Oziotelphusa senex senex*, Lethal, Sublethal, Size, Sex

Introduction

Contamination of freshwater environment by cadmium escalated in recent years by the release of effluents into it from various sources like oil refineries, combustion of fuels, pesticides, degradation of tyres, phosphate fertilizers, mine leaching etc. (Engel & Fowler 1979, Shaikh & Smith 1984). A very important biological property of this metal is its tendency of bioaccumulation. Freshwater crustaceans, which play an important role in aquatic food webs, are proved to be very sensitive to cadmium, yielding to its detrimental effects (Marshall 1979). These effects are due to the accumulation of this metal in different body parts of the animal at various levels (Lake et al. 1979). The accumulated cadmium binds irreversibly to sulfhydryl groups of catalytic proteins and inhibit the normal biochemical activities (Cherian & Goyer 1978). However, the degree of inhibition depends on the amount of accumulation of metal in each organ of the animal. But, the information on accumulation of cadmium at organ level in the freshwater crustaceans exposed to lethal and sublethal concentrations of cadmium is scanty. Hence, it is felt worthwhile to undertake the present study in haemolymph, gill, hepatopancreas and carapace of the freshwater crab, *Oziotelphusa senex senex*. As the period of exposure, size and sex are considered as prominent factors influencing the rate of accumulation, the present study was carried out in the crabs of two size groups of both sexes at Days 1 and 2 in lethal and Days 1 and 15 in sublethal concentrations of cadmium.

Materials and Methods

Freshwater field crabs were collected from local fields and were acclimatized to laboratory conditi-

ons for a period of ten days. They were fed everyday with pieces of frog muscle and were divided into four experimental groups: small males and females (5 ± 1 g) and large males and females (20 ± 1 g). Batches of ten crabs from each group were exposed to lethal and sublethal concentrations of cadmium as determined by the methods of Finney (1971) and Carpenter (1975). Accordingly, 4.6, 6.5, 9.0 and 18.4 mg/l were the lethal concentrations and 0.92, 1.3, 1.8 and 3.68 mg/l were the sublethal concentrations to small male, small female, large male and large females respectively. Pure cadmium nitrate was used as the source of metal. Crabs in normal freshwater medium served as controls. The animals were maintained at room temperature of 29 ± 0.5°C. All the animals were starved for 24 hr prior to experimentation. Immediately after the stipulated period of exposure (1 & 2 days in lethal and 1 & 15 days in sublethal concentrations) the haemolymph was removed from the base of the chelate leg with the help of a hypodermal syringe, and the hepatopancreas, gill and carapace were carefully dissected out from each crab. The haemolymph was measured and the organs were weighed accurately and were kept in hot air oven at 85°C for 24 hr. Then they were digested with concentrated perchloric acid, nitric acid and sulphuric acid in 1:2:1 ratio (Humpries 1956). The digestates were neutralized with 20% sodium hydroxide and diluted with distilled water. Cadmium concentration in each sample was estimated by Atomic Absorption Spectrophotometer (Hitachi 170-30 model) at a wave length of 228.8 nm using cadmium cathode lamp, and was expressed as μg/100 ml in haemolymph and μg/g wet wt in other organs. The students 't' test was employed to determine the significance of the data obtained.

Results and Discussion

The data presented in tables 1 and 2 revealed a significant increase in cadmium concentration in haemolymph, hepatopancreas, gill and carapace of the crabs of both sexes and sizes at the two exposure-periods in lethal and sublethal concentrations of cadmium. However, this increase was predominantly higher in the organs of crabs exposed to lethal concentrations compared to sublethal ones. It could be due to the speedy diffusion of the metal into the soft parts of animal from the highly concentrated external medium. The less amount of metal in the organs of crabs exposed to sublethal concentrations indicates that the accumulation of cadmium in these is concentration-dependent, as reported in the mollusc, *Trichomya hirsuta* (Klumpp & Burdon-Jones 1982).

The amount of cadmium accumulated in the organs of crabs increased very rapidly over time of exposure to lethal concentrations but, in sublethal concentrations the increase from Day 1 to Day 15 was significant only in hepatopancreas and carapace. The significant increase in accumulation of the metal from Day 1 to Day 2 in lethal concentrations could reveal the dysfunctioning of detoxification mechanisms and metal elimination processes involved with in these animals. Whereas, the less increase in sublethal concentrations from Day 1 to Day 15, particularly in haemolymph and gill, could be due to the synthesis of metal binding proteins or metallothioneins to sequester the metal and reduce its toxicity (Cherian & Goyer 1978) and existence of some regulatory mechanisms for elimination and accumulation (Katz 1979).

In both the size and sex groups of crabs, the degree in the rate of accumulation of cadmium within the four organs studied was concentration dependent. Thus, the % increase of cadmium accumulation was in the order: gill > haemolymph > hepatopancreas > carapace in lethal concentrations and carapace > hepatopancreas > gill > haemolymph in sublethal concentrations.

Gills being in direct contact with the surrounding water, more accumulation of metal in it is possible in the crabs exposed to toxicologically effective concentrations (Hutchinson et al. 1975). Similar high concentrations were also reported in this organ of aquatic forms exposed to metals and correlated to its structural deformities and impaired physiological functions (Stagg & Shuttleworth 1982, Venkataramana & Radhakrishnaiah 1987). Contrary to lethal concentrations, the very less accumulation of metal in gills of crabs exposed to sublethal concentrations could be due to active

Table 1 *Cadmium concentration in haemolymph* (μg/100 *ml*), *hepatopancreas, gill and carapace* (μg/g *wet wt*) *of* ***O. senex senex*** *at* 1 *and* 2 *days of exposures to lethal concentrations of cadmium* (*each value is a mean of* 10 *estimations*)

Organ	Small males			Small females			Large males			Large females		
	C	1	2	C	1	2	C	1	2	C	1	2
Haemolymph	1.20	10.86	13.02	1.16	8.41	10.73	1.00	6.33	8.29	1.20	5.58	6.47
SD (±)	0.10	1.06	1.22	0.08	0.94	1.02	0.12	0.80	0.96	0.10	0.62	0.60
%		(805)	(985)		(625)	(825)		(533)	(729)		(365)	(439)
Hepatopancreas	1.08	4.51	5.34	1.20	3.26	4.29	1.20	3.03	3.86	1.30	2.85	3.48
SD (±)	0.15	0.60	0.73	0.31	0.84	0.63	0.20	0.42	0.33	0.08	0.36	0.42
%		(317)	(394)		(172)	(258)		(153)	(222)		(119)	(168)
Gill	0.81	7.58	9.13	0.80	6.28	7.84	0.90	6.02	8.41	1.00	5.64	6.73
SD (±)	0.06	1.01	1.20	0.06	0.52	0.58	0.06	0.50	0.80	0.02	0.62	0.56
%		(836)	(1027)		(685)	(880)		(569)	(834)		(464)	(573)
Carapace	1.10	2.39	3.20	1.00	3.01	3.46	1.16	3.66	4.02	0.98	3.98	4.46
SD (±)	0.14	0.32	0.20	0.06	0.24	0.40	0.02	0.37	0.52	0.07	0.21	0.41
%		(117)	(191)		(201)	(246)		(216)	(247)		(306)	(355)

The differences between C (Control) and 1 or 2 days ($P < 0.01$) and between 1 and 2 days ($P < 0.05$) are statistically significant
The data in parentheses show % increase over control *C*, Control; *P*, Level of significant, *SD*, Standard deviation

Table 2 *Cadmium concentration in haemolymph* (μg/100 *ml*), *hepatopancreas, gill and carapace* (μg/g *wet wt*) *of* ***O. senex senex*** *at* 1 *and* 15 *days of exposures to sublethal concentrations of cadmium* (*each value is a mean of* 10 *estimations*)

Organ	Small males			Small females			Large males			Large females		
	C	1	15	C	1	15	C	1	15	C	1	15
Haemolymph	1.21	2.42	2.58	0.96	1.68	1.72†	1.00	1.46	1.49†	1.20	1.26*	1.30†
SD (±)	0.09	0.21	0.13	0.08	0.10	0.27	0.12	0.16	0.18	0.11	0.13	0.10
%		(100)	(113)		(75)	(79)		(46)	(49)		(0.5)	(8.3)
Hepatopancreas	0.93	2.49	3.24	0.86	2.20	2.47	0.80	1.87	2.18	0.83	1.63	1.94
SD (±)	0.09	0.12	0.09	0.06	0.14	0.13	0.06	0.11	0.15	0.03	0.08	0.12
%		(168)	(204)		(156)	(187)		(134)	(173)		(96)	(134)
Gill	1.00	1.62	1.68†	1.10	1.47	1.60†	1.06	1.26	1.32†	0.98	1.18	1.23†
SD	0.08	0.12	0.09	0.05	0.11	0.08	0.13	0.07	0.12	0.06	0.10	0.14
%		(62)	(68)		(34)	(46)		(19)	(24)		(20)	(26)
Carapace	0.88	1.26	3.21	0.66	1.47	2.96	0.74	1.72	3.48	0.86	1.86	4.33
SD (±)	0.07	0.08	0.22	0.04	0.16	0.18	0.09	0.08	0.23	0.06	0.10	0.21
%		(43)	(265)		(123)	(348)		(132)	(370)		(116)	(405)

The differences between C (Control) and 1 or 15 days, as also between 1 and 15 days, are statistically significant ($P < 0.05$)
*Not significant from control data; †Not significant from Day 1 data ($P > 0.05$)
The data in parentheses show % increase over control *C*, Control; *P*, Level of significance; *SD*, Standard deviation

transport of metal to detoxifying and/or elimination sites. The gills also can eliminate the metal and actively participate in detoxification (Noel-Lambot et al. 1978). High accumulation of metal in haemolymph, the circulating fluid, of the crabs exposed to lethal concentrations could be due to the metabolic impairment of detoxification and metal elimination centres. The less accumulation in sublethal concentrations could indicate the regular transfer of the metal to other parts of the body for further course of action.

Relatively less accumulation of cadmium in hepatopancreas, the primary site for detoxification (Hutterer et al. 1969), is an evidence for ill functioning of this organ of crabs under lethal stress. In sublethal concentrations, high cadmium accumulation could be correlated to its well augmented detoxification mechanism. The deposition of a significant portion of cadmium from haemolymph onto hepatopancreas for detoxification was also reported in *Carcinus maenas* (Wright & Brewer 1979). Probably, hepatopancreas of *O. senex senex* possess some cadmium binding proteins having the characteristics of metallothionein to detoxify the accumulated cadmium as in the case of *Carcinus maenas* (Jennings et al. 1979). Carapace was reported as the site for deposition of cadmium on continuous exposure of the crab to metal medium (Wright 1977). The less accumulation in it in *O. senex senex* exposed to lethal could be due to the functional failure of the organs like hepatopancreas which play a role on the deposition of cadmium onto carapace. High amounts of cadmium in it at 15 days of exposure to sublethal concentrations clearly document the active role of hepatopancreas. The deposition could also take place by active adsorption as suggested by Jennings & Rainbow (1979).

Irrespective of sex, cadmium accumulation was significantly more in the organs of small crabs than of large ones in both lethal and sublethal concentrations. Further, irrespective of size, the organs of male crabs accumulated more cadmium than those of females. More accumulation of metal in the organs of small and male crabs, though the external medium concentration was significantly less, is a reflection of their high metabolic activity (Ambore & Venkatachari 1978) as reported in green mussel, *Mytilus viridis* (Hutagalung 1989). The rapid turnover of cellular material and the increase in body weight would dilute the metal concentration in the old and female specimens. Further, the lesser cell surface area in large ones and the presence of more less active tissues in females could prevent greater diffusion of the metal thereby they could tolerate fairly high concentrations of cadmium than the small and male animals. The same level of tolerance was also reported in *O. senex senex* exposed to zinc (Victoriamma & Radhakrishnaiah 1986). The present results reveal that the accumulation of cadmium in the organs of crab is not only dependent on metal concentration but also on its size and sex and the length of exposure period.

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