

Recently it has been suggested that the activity of this enzyme has some function in phagocytosis and degradation of collagen²⁴. The same authors also speculated that alkaline phosphatase acts in conjunction with collagenase in collagen breakdown and is probably involved in the removal of some phosphate groups associated with the intact collagen before collagenase acts to disrupt a peptide bond, or, as alkaline phosphatase is associated with calcium transport, it is involved in the provision of calcium ions, necessary for collagenase activity²⁵. The demonstration of alkaline phosphatase activity in relation to collagen degra-

dation rather than to collagen synthesis in the connective tissue²⁴ does not seem to be compatible with the long-standing suggestion that alkaline phosphatase is related to collagen synthesis, unless of course it is involved in both synthesis and degradation. In any case, rapid synthesis of collagen and collagen degradation often go side by side. The gradual disappearance of alkaline phosphatase with the maturation of the collagen fibers in *Heteropneustes fossilis* may indicate that synthesis and degradation of collagen have ceased with the completion of the dermal repair.

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Retention of cadmium in a freshwater fish, *Channa punctatus*

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Summary. The study revealed a significant retention of cadmium in the liver and kidney of *Channa punctatus* exposed to various concentrations of cadmium. A linear relationship was observed between the period of exposure, the cadmium level in the medium and its retention in the target organs except for a slight decline in the quantity of cadmium in the liver when exposed to 0.01 ppm concentration.

Cadmium is known to enter the bodies of animals from the environment through the respiratory and gastrointestinal tracts². The major sites of retention seem to be the liver and kidney³. A good deal of information is available on the absorption, distribution and excretion of cadmium in mammals, especially rats; however, not much is known about the absorption of cadmium in aquatic animals, including edible fishes. A few reports can be cited, for example that of Smith et al.⁴ who studied acute effects of cadmium on the fish *Ictalurus punctatus*, and reported a higher accumulation of cadmium in the kidney than in the liver. Recently Banerjee et al.⁵ recorded a slightly higher quantity of cadmium in the liver than in the kidney of *Tilapia mossambica* and *Clarius batrachus* exposed to a very low dosage of cadmium chloride for 4 weeks.

Pandya⁶ carried out estimations of cadmium in the drinking water from 14 different centers at Ahmedabad and found cadmium concentrations ranging between 0.001 and 0.004 ppm with an average concentration of 0.002 ppm. Patel⁷ recorded cadmium concentrations between 0.001 and

0.007 ppm with an average value of 0.003 ppm in 30 samples collected from the river Sabarmati.

During the present investigation an air-breathing fish, *Channa punctatus*, was released in water containing different concentrations of cadmium to see whether this metal is retained in the target organs selected here viz., the liver and kidney and investigate the correlation between the concentration of cadmium in the medium, its retention in the body organs and the exposure period.

Materials and methods. Acquisition, care, maintenance and cadmium treatment of *Channa punctatus* have been described earlier⁸. The concentrations of cadmium and the time intervals at which the fishes were sacrificed were as follows:

Cadmium concentrations	Animals sacrificed at the end of
a) 0.01 ppm	1, 7, 14, 21, 28 and 35 days
b) 0.03 ppm	1, 7, 14, 21, 28 and 35 days
c) 0.05 ppm	1, 7, 14, 21, 28 and 35 days

6-8 specimens were sacrificed at specific intervals from each aquarium. Target organs (liver and kidney) were also studied from the control animals simultaneously. Pieces of liver weighing about 300 mg and pieces of kidney weighing about 150 mg were digested in separate Kjeldahl flasks with 5 ml of concentrated nitric acid, 5 ml of perchloric acid and 5 ml of hydrochloric acid. The flasks were then cooled and the contents diluted with 5 ml of double distilled water. This solution was transferred to a stoppered tube and stored in refrigerator. A standard solution with 0.5704 ppm of cadmium was prepared using pure cadmium metal. The tissue extracts (control as well as experimental samples) were analyzed for the determination of cadmium using an atomic absorption spectrophotometer (Perkin Elmer, 373) at the wave length of 228.7 nm. Cadmium was also analyzed in the aquarium water throughout the exposure period.

Results and discussion. Retention of cadmium was studied in the liver and kidney of normal fish *Channa punctatus* as well as those exposed to 0.01, 0.03 and 0.05 ppm of cadmium. As seen from the accompanying tabulated statement the cadmium residues in the liver and kidney of the normal fish were found to be 0.22 µg/gm (ppm) and 0.31 µg/gm (ppm) respectively. The high level of cadmium found in the normal fish could be due to the presence of cadmium in the water in which the fish had lived (Pandya⁶, Patel⁷).

The quantity of cadmium increased gradually up to 21 days in the livers of the fish exposed to 0.01 ppm of cadmium. Thereafter a slight decline was observed in the residual quantity, continuing until 35 days. Earlier studies have shown a temporary regeneration of the liver cells during

3-4 weeks of exposure of the fish to 0.01 and 0.03 ppm of cadmium^{8,9}. It may be interesting to note that this period (decline of cadmium content in the liver) coincides with the temporary regeneration of the hepatic cells reported earlier in the fish *Channa punctatus* exposed to 0.01 ppm of cadmium. There was, however, no decline in the quantity of cadmium at any stage when the fishes were exposed to 0.03 ppm concentration. Both with 0.03 and 0.05 ppm dosages a gradual increase in cadmium accumulation was recorded in the target organs throughout the period of exposure.

In the kidney the quantity of cadmium was observed to increase gradually throughout the period of experimentation and in proportion to the dosage of cadmium in the medium. Working on the absorption of cadmium in fishes, Smith et al.⁴, Banerjee et al.⁵, and Coombs¹⁰ have also recorded a significant increase in the amount of cadmium both in the liver and kidney.

Cadmium content was also analyzed in the aquarium water throughout the exposure period and the results were shown in table 2. The concentration of cadmium in the water of all 3 aquaria showed a gradual decrease. At the end of 35 days when the experiment was discontinued, the concentrations of cadmium found in the water were 0.007, 0.021, 0.037 ppm in the aquaria containing nominal concentrations of 0.01, 0.03 and 0.05 ppm.

Compared with the kidney, liver accumulated a higher amount of cadmium during the 1st week of exposure. Thereafter it was the kidney which started accumulating a larger quantity of this metal. At the end of 35 days the increases in cadmium concentration were respectively 122%, 168% and 186% of the normal value in the liver with 0.01, 0.03 and 0.05 ppm cadmium in the medium. The corresponding figures for the retention of cadmium in the kidney were 161%, 196% and 212% respectively of the normal value. It is suggested that a higher retention of cadmium in the liver than the kidney during the early period is due to the involvement of the liver in the detoxification process. On the other hand, a higher retention of this metal in the kidney can be explained as being due to the latter serving as a terminal point before the final discharge of foreign substances from the body.

Table 1. Retention of cadmium (ppm) in the target organs of *Channa punctatus* exposed to cadmium

Exposure period in days	Organs	Concentrations of cadmium in ppm		
		0.01	0.03	0.05
1	L	0.24 ± 0.03	0.25 ± 0.03	0.28 ± 0.03
	K	0.32 ± 0.03	0.33 ± 0.03	0.35 ± 0.04
7	L	0.27 ± 0.04	0.28 ± 0.03	0.30 ± 0.03
	K	0.33 ± 0.03	0.37 ± 0.03	0.41 ± 0.04
14	L	0.29 ± 0.03	0.29 ± 0.02	0.31 ± 0.03
	K	0.42 ± 0.05	0.44 ± 0.04	0.46 ± 0.04
21	L	0.31 ± 0.03	0.32 ± 0.03	0.34 ± 0.03
	K	0.40 ± 0.05	0.49 ± 0.04	0.51 ± 0.05
28	L	0.28 ± 0.03	0.36 ± 0.03	0.38 ± 0.04
	K	0.47 ± 0.04	0.53 ± 0.05	0.59 ± 0.06
35	L	0.27 ± 0.03	0.37 ± 0.03	0.41 ± 0.04
	K	0.50 ± 0.04	0.61 ± 0.06	0.66 ± 0.06
Normal fish			Liver (L)	0.22 ± 0.02
			Kidney (K)	0.31 ± 0.04

Values represent the mean ± SE for a minimum of 6 animals.

Table 2. Cadmium content (ppm) in the aquarium water

Exposure period in days	Nominal concentrations of cadmium (ppm)		
	0.01	0.03	0.05
1	0.013	0.031	0.052
7	0.011	0.028	0.049
14	0.009	0.025	0.044
21	0.008	0.026	0.040
28	0.008	0.023	0.038
35	0.007	0.021	0.037

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