EFFECT OF CADMIUM AND MERCURY ON LYSOSOMAL ACID PHOSPHATASE ACTIVITY IN THE KIDNEY OF MYXINE GLUTINOSA.

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After exposure to heavy metals highest concentrations of these environmental pollutants are found in the liver and kidney in fish as well as in mammals (Hilmy et al., Comp. Biochem.Physiol. 81C:139-143,1985). At the cellular level, it has been demonstrated by Madsen (Kidney Int. 18:445-453,1980) that heavy metals such as mercury accumulate predominantly in the lysosomes. Therefore, the aim of the present study was to investigate the effect of cadmium and mercury on a particularly active intralysosomal enzyme, the acid phosphatase (AcPase).

Hagfish were caught at the coast of Maine and maintained as previously described (Fels et al., Bull. MDIBL 25:18-19,1985). The animals ranged in length from 29 to 41 cm (body weight 31-71 g). Homogenates of 40-60 glomeruli and 4 cm long pieces of the archinephric duct (AND) were prepared with Ringer solution (Riegel, J. exp. Biol. 73:261-277, 1978) after pressure controlled perfusion of the hagfish to remove the blood from the kidneys. AcPase activity was measured by a highly sensitive ultramicroassay utilizing 4-methylumbelliferyl phosphate as a stubstrate (Olbricht et al., Am. J. Physiol. 247:F252-F259,1984). $CdCl_2$ or $HgCl_2$ were added directly into the reaction mixture. The measurement of protein was carried out according to the method of Bradford (Anal. Biochem. 72:248-254,-1976).

Linear kinetics regarding time were obtained. AcPase activity was lower in the glomeruli than in the AND (Table 1). Samples of the AND that had been taken from the posterior region near the cloaca revealed higher activity by the factor 2 than samples that had been taken 1-4 cm distal from the heart. In order to investigate the effect of heavy metals on AcPase activity it was necessary to test the enzyme reaction without EDTA in the assay. Table 1 shows that the AcPase activity is not altered in the absence of EDTA.

Table 1: Acid phosphatase activity (µmol/30 min/mg prot.) in glomeruli and archinephric duct (AND) of the Atlantic hagfish, Myxine glutinosa.

	Glomeruli	Proximal AND	Distal AND
With EDTA	2.17 ± 1.25 (7)	3.08 ± 0.99 (4)	6.34 ± 4.02 (4)
Without EDTA	-	2.93 ± 1.40 (5)	6.22 ± 4.75 (6)

Means ± S.D. (numbers of animals in parentheses).

Cadmium has no significant effect on AcPase activity in the AND because the mean values presented in Table 2 are mostly close to the controls (100%). Mercury inhibits renal AcPase by more than 60% at concentrations above 10^{-6} M.

Table 2: Effect of cadmium and mercury on renal AcPase *in vitro* (activity in % of control, $\bar{x} \pm$ S.D., numbers of animals in parentheses).

Cd^{2+}	Proximal AND	Distal AND
10^{-4} M	110 ± 61.8 (6)	101 ± 34.8 (7)
$5x10^{-5}$ M	95 ± 32.8 (4)	104 ± 26.8 (5)
10^{-5} M	97 ± 31.7 (6)	105 ± 29.5 (7)
10^{-6} M	98 ± 30.2 (6)	122 ± 43.3 (7)
10^{-7} M	106 ± 43.3 (6)	128 ± 58.7 (7)
Hg^{2+}		
10^{-3} M	28 ± 8.6 (5)	34 ± 7.3 (5)
10^{-4} M	34 ± 10.1 (5)	41 ± 6.3 (5)
10^{-5} M	38 ± 6.6 (5)	39 ± 18.8 (5)
10^{-6} M	87 ± 17.0 (5)	101 ± 2.8 (5)
10^{-7} M	98 ± 5.6 (5)	102 ± 4.5 (5)

In agreement with our mean values, AcPase activity was not significantly changed *in vitro* by cadmium concentrations up to 10^{-3} M in the teleost fish *Mugil cephalus* (Hilmy et al., Comp. Biochem. Physiol. 81C:145-153,1985). Kidney and liver of cadmium-injected mice also showed no significant decrease in AcPase activity (Mego et al., Biochem. Pharmacol. 24:12271232,1975). However, AcPase activity decreased in all organs that have been investigated in cadmium-exposed molluscs (Evtushenko et al., Comp. Biochem. Physiol. 83C:371-376,1986).

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