

Effects of Moulting, Age, and Sex on the Accumulation of Heavy Metals in the Otter (*Lutra lutra*) in Finland

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Populations of the European otter (*Lutra lutra*) have declined in western and middle Europe (Foster-Turley et al. 1990). In Finland there have not been any strong decreases in otter populations, although during the 1970s - 1980s the population densities declined. Adverse effects of contaminants in the aquatic environment may be associated with these population declines (Erlinge 1972, Borg 1975, Mason 1989). Mercury contamination has been suggested to be a cause for the decline of otter populations in Sweden (Erlinge 1972, Borg 1975), and mercury poisoning has been reported also in the American river otter (*Lutra canadensis*) (Wren 1985).

Although mercury is considered to be very hazardous to the otter, very little is known about the effects of age and sex on the accumulation of mercury in the tissues of the otter. Mercury concentrations are high in the hair of otters exposed to mercury pollution (Halbrook et al. 1994). This suggests that the elimination of mercury via hair during the moulting can be important for mammals like the otter. No data on the efficiency of this elimination route have been reported in otters. In birds, however, the excretion of mercury into feathers is an effective protective mechanism against the continued accumulation of mercury (Burger et al. 1994).

The aims of the present study are (1) To determine the relationships of age and sex on the accumulation of mercury, cadmium, lead, zinc and copper in the muscle, kidney, liver and fur of the European otter and (2) To estimate the role of moulting in the elimination of mercury and other heavy metals.

MATERIALS AND METHODS

The otters (n = 39, 17 juveniles, 11 adult females and 11 adult males) of this study were collected in 1986-2000. The otters were either roadkills, found dead in fishing nets, muskrat or beaver traps, or killed with permission at fish ponds. The carcasses were stored at -20°C.

The animals were weighed and their length was measured from the rostrum to the anus. The otters were skinned and the moulting stage was determined. Muscle,

kidney, liver and hair samples were collected, their fresh weight was measured and the samples were dried overnight at 105 °C. The dried samples were weighed and digested in a microwave digestion unit (8 ml HNO₃, 2 ml H₂O₂). Mercury concentrations were measured by a gold-film mercury analyser (Jerome Inst. Corp. Model 511, Jerome, Arizona, USA) and the cadmium, copper, zinc and lead concentrations in an atomic absorption spectrophotometer with graphite atomizer (Hitachi Z-9000, Hitachi Ltd., Tokyo, Japan).

The age of the animals was determined from the root of the lower canine tooth. The root was fixed in 4 % neutral formalin for 48 h and decalcified for one week in 6 % HNO₃, dehydrated and embedded in paraffin. Then the root was cut into 10 µm thick sections with a microtome, stained in Mayers hematoxylin and eosin, and the age was determined from the number of the cementum rings. Age was estimated also for juvenile otters (less than one year of age). In Central Europe the otter generally gives birth throughout the year (Jenkins & Harper 1982). The material of this study (thickness of the dentine, the size of the animals) indicated, however, that in Eastern Finland the juveniles are generally born in the spring or in the early summer. Thus the time interval in months from May until the death of the animal was used as an age estimate for the juveniles.

The weights of dried pelts of 10 otters of different body weight categories were measured. A pelt sample of 10-15 cm² was taken from the ventral skin, weighed and the hair cut off as close to the root as possible. The proportion of the weight of the hair to the total weight of the pelt sample was calculated and from this proportion the total hair weight of the pelt and the proportion of the hair weight to the total body weight were determined. The time interval from the last moult was calculated in months. April-June was used to represent the time of the spring moult and Sep 1st - Oct 15th the autumn moult. Thus the animals that had died in July were given the time interval value 0. The subsequent months were given the values as follows: Aug to Oct 1, Nov 2, Dec 3, Jan 4, Feb 5, Mar 6 and Apr 7. These values were used to calculate the interval from the last moult.

The multiple comparisons were performed with the one-way analysis of variance (ANOVA) followed by the *post hoc* Student-Newman-Keuls test. Paired comparisons were performed with the Student's t-test and the correlations calculated with the Spearman's correlation coefficient (r_s). The p value less than 0.05 was considered to be statistically significant.

RESULTS AND DISCUSSION

Mercury concentrations in the tissues of adult otters were higher than in the young otters (ANOVA, $p < 0.05$, Table 1). Mercury levels in the livers of young animals increased during the first year of their life (Fig. 1a). Mercury concentrations in the hair of otters were at the adult level after the first spring moult (Fig. 2a). In adult

Table 1. Heavy metal concentrations (mean \pm SE) in different age groups of the otters (hair (H) $\mu\text{g/g}$ dry weight; liver (L), kidney (K) and muscle (M) $\mu\text{g/g}$ wet weight).

	n	Juveniles	n	Adult females	n	Adult males
H Hg	17	5.9 \pm 1.17 ^a	11	20.0 \pm 2.45 ^b	11	29.5 \pm 3.53 ^b
L Hg	12	1.2 \pm 0.28 ^a	10	3.8 \pm 0.49 ^b	9	5.5 \pm 1.13 ^b
K Hg	12	0.4 \pm 0.10 ^a	10	1.3 \pm 0.21 ^b	9	1.4 \pm 0.24 ^b
M Hg	12	0.4 \pm 0.09 ^a	10	0.8 \pm 0.12 ^b	9	1.3 \pm 0.25 ^b
H Cd	17	0.005 \pm 0.004	11	0.038 \pm 0.036	11	0.036 \pm 0.025
L Cd	12	0.10 \pm 0.029 ^a	10	0.58 \pm 0.118 ^b	9	0.60 \pm 0.117 ^b
K Cd	12	0.39 \pm 0.112 ^a	10	1.77 \pm 0.265 ^b	9	1.84 \pm 0.261 ^b
H Cu	17	8.4 \pm 0.44	11	7.7 \pm 0.75	11	7.0 \pm 0.88
L Cu	12	51.3 \pm 0.60	10	33.0 \pm 7.25	9	35.8 \pm 7.76
H Zn	17	85.1 \pm 18.95	11	95.4 \pm 56.90	11	76.4 \pm 45.70
L Zn	12	132.3 \pm 19.50	10	101.0 \pm 10.94	9	135.9 \pm 19.91

Values with dissimilar superscripts differ at $p < 0.05$ (Student-Newman-Keuls test).

males the hair mercury concentration was higher than in the adult females (t-test, $p < 0.05$). In other tissues the differences between the sexes were not significant, although the concentrations tended to be higher in the males than in the females. The hair mercury concentrations correlated with liver ($r_s = 0.77$, $p < 0.01$), kidney ($r_s = 0.75$, $p < 0.01$) and muscle ($r_s = 0.74$, $p < 0.01$) mercury concentrations. The correlation was the most significant between muscle and liver ($r_s = 0.89$, $p < 0.001$). In adult animals increasing age did not have any further effect on the tissue mercury concentrations (Fig. 1-2a). A positive correlation was found, however, between the fur age (time in months from the last moult) and the liver mercury concentration in adult otters ($r_s = 0.57$, $p < 0.05$, Fig. 2b).

The liver and kidney cadmium concentrations were higher in adult otters than in young otters (ANOVA, $p < 0.05$, Table 1) but in the adults further ageing did not have any significant effect on cadmium concentrations. The muscle cadmium concentrations were below the detection limit (0.001 $\mu\text{g/g}$). There were no differences in cadmium concentrations between adult females and males. Nor were there any age- or sex-related differences in the copper and zinc concentrations. The lead concentrations were nearly always below the detection limit ($< 0.002 \mu\text{g/g}$) in all tissues.

In adult males (body weight about 8 kg) the relative hair weight of the total body weight was about 3.2 % and in the adult females (body weight about 5.5 kg) 3.7 %. The theoretical mean elimination rate of mercury via hair in a complete moult

calculated from these data was about 7.3 mg in adult males and about 4.2 mg in adult females (Table 2).

Table 2. Estimates of the total weight of hair and of the amount of mercury eliminated in a complete moult of adult male and female otters.

	Males	Females
Body weight (kg)	8.0	5.5
Hair weight / body weight (%)	3.2	3.7
Weight of hair lost in a complete moult (g)	256	204
Mean mercury concentration of hair ($\mu\text{g/g}$)	29.5	20.0
Elimination of Hg in a complete moult (mg)	7.3	4.1

The maximum liver, kidney and muscle mercury concentrations of this study (11.9, 2.7 and 3.1 $\mu\text{g/g}$) were below the levels considered to be toxic for the otter. Severe intoxication or death occur if the liver mercury concentration exceeds 30 $\mu\text{g/g}$ (O'Connor & Nielsen 1981, Wren 1985, Kruuk & Conroy 1991). Previously liver mercury concentrations of otters in Finland have been 0.05-31 $\mu\text{g/g}$ (Skaren 1992), in Spain 3.9-17.5 $\mu\text{g/g}$ (Hernandez et al. 1985), and in the Czech Republic in an individual otter as high as 55.6 $\mu\text{g/g}$ (Gutleb et al. 1998). On the other hand, the hair mercury concentrations in this study (0.7- 61.3 $\mu\text{g/g}$) were quite high. According to Halbrook et al. (1994) in American river otters in Georgia the hair mercury concentration has been 0.5-54.6 $\mu\text{g/kg}$. The muscle mercury concentrations in Georgia were as high as 0.7-10.3 $\mu\text{g/g}$ (in this study 0.01-3.1 g/g).

According to the results of this study, the excretion of mercury via hair at moulting could be the most important route for the elimination of the highly toxic methylmercury in otters. The concentrations of mercury in otter tissues increase continuously after the moult (Fig. 1-2a). An adult male otter can eliminate in one complete moult about 7.3 mg of mercury (Table 2). The otter, however, has two annual moults. The spring moult is a complete moult lasting about three months, but the autumn moult is an incomplete moult (Harper & Jenkins 1982). In these moults the male otter could eliminate 7.5-10 mg of mercury. This is more than the theoretical total amount of mercury in the body (about 7 mg) calculated from the mean values of this work. The liver and muscle mercury concentrations of the juveniles increase to the adult level before the first spring moult. The hair mercury concentrations of juveniles, however, increase to the adult level only after the first moult. There was no further accumulation of mercury after one year of age. The elimination rate via hair thus seems to be efficient enough in eastern Finland to keep the tissue mercury levels at a fairly low level independent of the age of adult animals. The lower mercury concentrations in the fur of adult females is probably caused by the passage of mercury across the placenta during pregnancy and into milk during the suckling period.

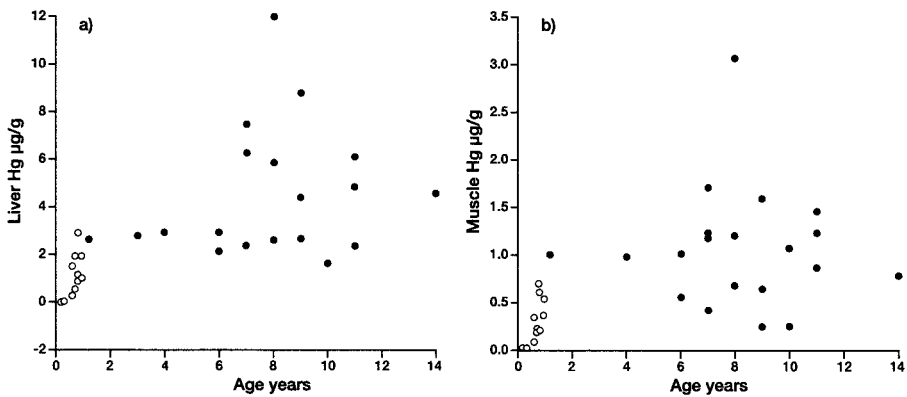


Figure 1 a-b. Liver (a) and muscle (b) mercury concentrations of the juvenile (○) and adult (●) otters.

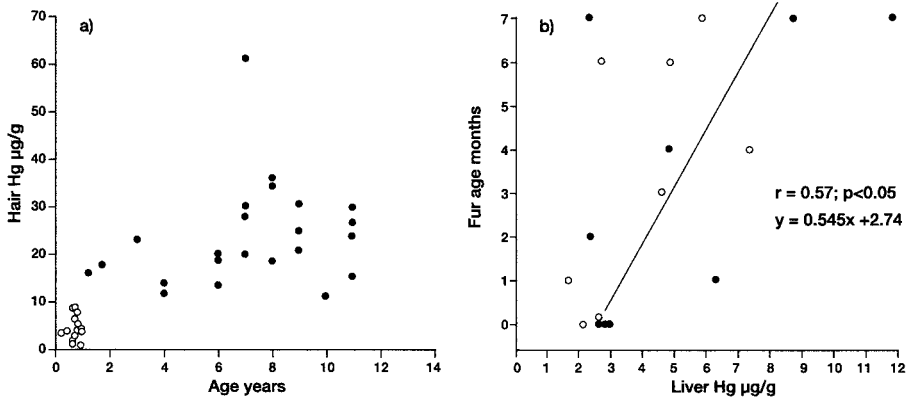


Figure 2 a-b. Hair mercury concentrations (a) of the juvenile (○) and adult (●) otters and the correlation between the fur age and the liver mercury concentrations (b) of the adult female (○) and male (●) otters.

The liver and kidney cadmium levels also showed the expected increase from juveniles to adults. Cadmium concentrations were the highest in the kidney, where cadmium accumulates in humans, too. Cadmium concentrations in hair were quite low, but some cadmium could be eliminated via fur. The muscle cadmium concentrations were below the detection limit. The significance of fur as an elimination route for copper and zinc seems to be small. Copper and zinc are both essential for organisms and their availability is quite high.

Mercury concentrations in the fur of otters were about five times higher than in the liver of otters. This observation is different from data gathered in marine mammals. In adult Saimaa ringed seals (*Phoca hispida saimensis*) – another Finnish aquatic

predator – the liver mercury concentration is about 50 µg/g and the hair mercury concentration much lower, about 15 µg/g (Hyvärinen et al. 1998). Marine mammals counteract mercury toxicity by the storage and immobilization of mercury in the liver by selenium (Koeman et al. 1973). The strategy of the otters, however, is the elimination of mercury via the fur. As a semiaquatic mammal the environmental exposure of the otter to mercury could have been high even before industrialization. Thus the capacity for the elimination of mercury via hair could have developed during evolution. A similar adaptation to high mercury levels has been observed in birds. In great skuas (*Catharacta skua*) the mercury concentrations in growing feathers correlates significantly with the concentrations in blood (Bearhop et al. 2000). In conclusion: the European otter is able to excrete mercury via hair in significant amounts. In the adult otter this prevents the accumulation of mercury at concentrations encountered in Finland with no increase in the tissue mercury levels after one year of age.

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