

Hazardous Exposure of Ground-Living Small Mammals to Cadmium and Lead in Contaminated Terrestrial Ecosystems

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Abstract. The dietary exposure to cadmium and lead of two ground-living species of small mammals, *i.e.*, shrews *Sorex araneus* (Insectivora) and voles *Microtus agrestis* (Rodentia), was investigated and related to metal loads in target organs (kidneys and liver). The study was done in two natural areas polluted with cadmium and lead originating from urban and industrial metal sources. The average intake of cadmium by the herbivorous voles varied between 0.1 and 0.4 $\mu\text{g/g/day}$ and of lead between 2 and 10 $\mu\text{g/g/day}$. The carnivorous shrews showed a considerably higher metal intake rates, *i.e.*, cadmium 3 to 16 $\mu\text{g/g/day}$ and lead 19 to 53 $\mu\text{g/g/day}$, which was largely due to the consumption of contaminated earthworms (Oligochaeta). An average cadmium intake of 15 $\mu\text{g/g/day}$ or a lead intake of 20 $\mu\text{g/g/day}$ corresponded with critical renal metal loads of 120 $\mu\text{g/g}$ for cadmium and 25 $\mu\text{g/g}$ for lead, which are indicative of adverse health effects. The renal metal loads in shrews reached the critical level, but they remained far below this level in voles. The results indicate a greater risk of toxic exposure to cadmium and lead in soricine shrews than in microtine rodents.

Ground-living species of small mammals living in areas with soils contaminated with heavy metals generally show elevated levels of cadmium and lead in target organs (the kidneys and liver) (O'Neill *et al.* 1983; Andrews *et al.* 1984, 1989; Hunter *et al.* 1987; Ma 1989). The main route of exposure of wild mammals to heavy metals in a contaminated environment is through the consumption of food.

Studies on oral metal intake and organ metal loads in small mammals may provide useful information on the biological availability of heavy metals in contaminated terrestrial ecosystems. Hazard assessment of the effect of bioavailable metals in ecosystems can be derived from comparisons of metal intake rates and organ metal loads with critical levels associated with sublethal symptoms of metal intoxication. Critical renal levels of heavy metals in small mammals have recently been reported by Chmielnicka *et al.*

(1989) for cadmium and by Ma (1989) for lead, but critical levels of oral metal intake are unknown.

The above approach to hazard assessment was used in the present study done in two natural areas contaminated with cadmium and lead. Besides the measurement of organ metal loads in the animals, estimates were made of the average rate of daily intake of cadmium and lead in the two contaminated ecosystems. Two species of small mammals were investigated, *i.e.*, shrews *Sorex araneus* (Insectivora) and voles *Microtus agrestis* (Rodentia), which are carnivores and herbivores, respectively.

Materials and Methods

Sites

The study was carried out in two nature reserves, *i.e.*, one situated near the township of Budel and in another near the city of Arnhem, The Netherlands. These nature reserves were separated by a distance of about 100 km. Both had a podzolic type of sandy soil with a vegetation cover dominated by wild grasses (Faber and Ma 1986). The acidity of the soil (A_0 litter layer) corresponded with a pH- H_2O of 6.1 (pH-KCl 5.7) in the Budel area and a pH- H_2O of 3.5 (pH-KCl 3.0) at Arnhem. The organic matter content of the A_0 layer, as measured from loss-on-ignition, was 6.0 and 8.8 per cent, respectively.

The soils of the Budel and Arnhem sites were polluted by aerial deposition of cadmium and lead emitted by a metal smelter and urban-industrial sources, respectively (Ma *et al.* 1983). The A_0 layer of the soil in the Budel area had a mean \pm SD ($N = 11$) dry weight concentration of nitric-acid extractable cadmium of $5.5 \pm 0.8 \mu\text{g/g}$, with $2.9 \pm 0.6 \mu\text{g/g}$ present in the underlying 10 cm layer. The average concentration of lead in the two soil layers was 130 ± 30 and $90 \pm 21 \mu\text{g/g}$, respectively. The Arnhem area had a mean \pm SD ($N = 7$) soil content of cadmium of $1.2 \pm 0.2 \mu\text{g/g}$ in the A_0 layer and $0.3 \pm 0.1 \mu\text{g/g}$ in the underlying 10 cm layer, whereas the average soil concentration of lead was 177 ± 21 and $30 \pm 15 \mu\text{g/g}$, respectively.

Sampling

The Common shrew *Sorex araneus* and the Field vole *Microtus agrestis* were snap-trapped in a trapping grid of about 1 ha. The animals were caught during three periods, *i.e.*, February-March

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Table 1. Body weights (g fresh weight) of *Sorex araneus* and *Microtus agrestis*^a

Site	Species	Parameter	Feb-Mar	May-Jun	Oct-Nov
Budel	<i>M. agrestis</i>	geom. mean	20.4 (32)	18.2 (32)	18.8 (39)
		95% conf. int.	18.7–22.4	15.8–21.0	17.1–20.7
		range	14.6–32.1	10.0–33.0	8.7–31.0
	<i>S. araneus</i>	geom. mean	6.0 (28)	8.4 (10)	7.4 (26)
		95% conf. int.	5.9–6.2	7.4–9.7	7.1–7.7
		range	5.5–6.9	6.6–11.0	6.1–8.5
Arnhem	<i>M. agrestis</i>	geom. mean	22.8 (23)	25.1 (27)	19.0 (37)
		95% conf. int.	20.8–25.0	21.4–29.4	18.3–19.8
		range	15.0–31.0	15.0–48.0	16.0–27.0
	<i>S. araneus</i>	geom. mean	6.4 (18)	7.6 (28)	6.9 (20)
		95% conf. int.	6.1–6.7	7.1–8.1	6.6–7.2
		range	5.5–7.7	6.0–11.0	5.8–7.8

^a Geometric means, 95% confidence intervals and ranges are shown, with the number of samples in parentheses

(prevernum), May-June (spring), and October-November (autumn). Average body weights of the animals during each sampling period are shown in Table 1. The voles were about three times heavier than the shrews. Consistent seasonal variations in mean body weight were not present.

Plants and invertebrates were sampled within a distance of 1.5 m from successful small mammal trappings. Plant samples consisted of green leaves and basal portions of the stems of species eaten as food by *Microtus*. Pitfalls were used to collect surface-active invertebrates eaten by *Sorex*, and hand-sorting of soil blocks (40 × 40 cm) excavated to a depth of 20 cm was applied to collect soil-inhabiting insect larvae and earthworms (*Lumbricus rubellus* and *Den-drodrilus rubidus*).

Analysis of Dietary Composition

The composition of the food ingested by the voles and shrews was determined by stomach dissection and microscopical examination of food particles. The results were expressed as a percentage dietary composition (PDC), which for *Microtus* consisted of the mean surface area of an identified food item relative the total surface area of all items. Similarly, the diet composition for *Sorex* was calculated from the occurrence of an identified item relative to the total score of all items (Pernetta 1976). The species of some food items, however, such as seeds, fungi, wood particles, and other plant and animal matter could not always be identified.

Metal Analysis

Samples of plants were dried at 70°C, homogenized, and digested in HNO₃ 65% p.a. Invertebrates with a hard exoskeleton were dried at 105°C and digested in a 1:1 mixture of HCl 30% suprapur and HNO₃ 50% suprapur. Soft-bodied invertebrates such as earthworms and spiders were dried at 105°C and digested in HNO₃ 65% suprapur. The same procedure as that used for soft-bodied invertebrates was used for the metal analysis of kidney and liver tissues of small mammals.

Total metal contents were determined by flame atomic absorption spectrophotometry (AAS), and graphite-furnace AAS was used to measure concentrations below the detection limit of flame AAS. Deuterium was used for background correction of Pb analysis, and the Smith-Hieftje method (IL Video Manual, Allied Analytical Systems, Andover, USA, 1983) was used for background correction in the analysis of Cd. All metal concentrations are given on a dry weight basis.

The methods used for metal analysis of plant and soil samples were checked by ring testing in the International Plant-analytical Exchange (IPE) and the International Soil-analytical Exchange (ISE), Agricultural University, Wageningen, The Netherlands, from which also reference standard materials were obtained. Certified soil reference material BCR nr.143 of the CEC Community Bureau of Reference was included as an additional soil reference standard. For the metal analysis of animal tissue, bovine liver nr.1577a of the U.S. National Bureau of Standards was used as a reference standard. Analyses of reference samples were run parallel to each series of samples under investigation. Analytical results were accepted if the measured reference was within the prescribed average value plus or minus two times its statistical standard deviation.

Metal Intake Rate

The average rate of metal intake was estimated for each sampling period as follows:

$$DMI_x = \sum_i \frac{AFR \cdot AMC_{xi} \cdot PDC_i}{ABW}$$

in which: DMI_x = average intake of a metal element X in µg per g fresh body weight per day;

AFR = average feeding rate in g dry weight per day;

AMC_{xi} = average concentration of metal element X in food item i in µg per g dry weight;

PDC_i = coefficient of dietary composition of food item i;

ABW = average body weight in g fresh weight.

AFR values of 2.5 g/day for shrews and 12 g/day for voles (Hunter *et al.* 1987) were used, although lower rates of 6 to 7 g/day have been reported for voles (Hansson 1971; Ferns 1976; Phillipson *et al.* 1983). For unidentifiable food items, AMC values were replaced by the average value of all identified items. AMC values for food items of *Sorex* were not available in the cold season when soil fauna could not be obtained in sufficient amounts for chemical analysis. ABW values were used as shown for each period in Table 1.

Results

Dietary Composition and Metal Contamination

Microscopic analysis of stomach contents showed that the *Microtus* voles fed mainly on grasses, although some herbs were taken as well during the growing season (Table 2).

Table 2. Percentage dietary composition (PDC) of bulk food items for *Microtus agrestis* and the concentration ($\mu\text{g/g}$) of cadmium and lead

Site	Food item	Feb-Mar			May-Jun			Oct-Nov		
		PDC	Cd	Pb	PDC	Cd	Pb	PDC	Cd	Pb
Budel	<i>Holcus lanatus</i>	55	0.5	14	39	0.2	2.2	38	0.2	1.6
	<i>Agrostis capillaris</i>	40	1.1	22	9	0.3	1.9	11	0.8	5.6
	<i>Poa spp.</i>	0	—	—	6	0.3	3.8	16	0.8	4.0
	<i>Elymus repens</i>	0	—	—	2	0.4	4.3	17	0.1	2.5
	<i>Cytisus scoparius</i>	0	0.4	3.8	7	0.2	3.2	0	—	—
	<i>Lychnis flos-cuculi</i>	0	—	—	5	0.6	8.0	0	—	—
	Undeterm. material	5	—	—	32	—	—	18	—	—
Arnhem	<i>Deschampsia flexuosa</i>	100	0.4	14	74	0.2	4.7	74	0.4	4.2
	<i>Vaccinium myrtillus</i>	0	—	—	7	0.2	2.1	6	0.5	5.3
	<i>Hypnum cupressiforme</i>	0	—	—	9	1.0	75	3	0.7	43
	Undeterm. material	0	—	—	10	—	—	17	—	—

Table 3. Percentage dietary composition (PDC) of bulk food items for *Sorex araneus* and the concentration ($\mu\text{g/g}$) of cadmium and lead

Site	Food item	Feb-Mar			May-Jun			Oct-Nov		
		PDC	Cd	Pb	PDC	Cd	Pb	PDC	Cd	Pb
Budel	Lumbricidae	28	—	— ^a	38	62	160	36	109	108
	Coleoptera (adults)	17	—	—	8	7.7	19	15	7.7	12
	Coleoptera (larvae)	24	—	—	6	7.9	25	15	9.5	20
	Diptera (larvae)	10	—	—	0	—	—	6	3.0	7.5
	Lepidoptera (larvae)	5	—	—	18	3.2	8.0	4	4.5	11
	Arachnida	14	—	—	9	74	31	8	16	12
	Opilionida	1	—	—	1	16	18	7	40	18
	Undeterm. material	1	—	—	21	—	—	9	—	—
Arnhem	Lumbricidae	42	—	—	45	13	295	52	38	144
	Coleoptera (adults)	12	—	—	1	2.7	14	11	1.1	13
	Coleoptera (larvae)	17	—	—	8	4.5	65	1	3.0	37
	Diptera (larvae)	7	—	—	0	—	—	0	—	—
	Lepidoptera (larvae)	4	—	—	7	0.6	3.2	11	0.2	4.9
	Arachnida	17	—	—	9	17	9.0	7	8.8	10
	Opilionida	1	—	—	1	6.8	33	9	6.1	19
	Undeterm. material	0	—	—	31	—	—	10	—	—

^a Soil fauna could not be collected in sufficient amounts for chemical analysis

Some amounts of mosses (*Hypnum cupressiforme*) were consumed by voles living at the Arnhem site, but generally the contribution of dicotyledons and bryophytes to the total diet remained relatively small. The main grasses eaten by voles consisted of species of *Holcus*, *Agrostis*, and *Deschampsia*.

The average concentrations of cadmium and lead in the food items of *Microtus* are shown in Table 2 as well. The concentration of lead in the grasses showed a seasonal variation with highest levels occurring in the period just before the growing season. A similar pattern has been described in other reports on studies of metals in grasses (Badsha and Badsha 1988; Hunter *et al.* 1987). *H. cupressiforme* contained markedly elevated levels of lead compared with the average concentration present in mono- or dicotyledons, which reflected the specific preference of mosses for accumulation of aerial metal emissions.

Table 3 shows the dietary composition of the *Sorex* shrews. A large proportion of the food consisted of soft-bodied prey, including Oligochaeta and the larvae of Coleoptera, Diptera, and Lepidoptera. Lumbricid earthworms predominated among the soil-dwelling invertebrates,

Table 4. Estimated average daily metal intake (DMI) of cadmium and lead in *Sorex araneus* and *Microtus agrestis*

Site	Species	Period	DMI ($\mu\text{g/day}$)	
			Cadmium	Lead
Budel	<i>Microtus agrestis</i>	Feb-Mar	0.4	10.1
		May-Jun	0.2	2.1
		Oct-Nov	0.3	1.9
	<i>Sorex araneus</i>	Feb-Mar	—	—
		May-Jun	11.3	23.0
		Oct-Nov	15.9	18.6
Arnhem	<i>Microtus agrestis</i>	Feb-Mar	0.2	7.4
		May-Jun	0.1	6.3
		Oct-Nov	0.3	4.9
	<i>Sorex araneus</i>	Feb-Mar	—	—
		May-Jun	3.3	53.0
		Oct-Nov	8.0	30.2

whereas spiders and beetles were the most important surface-active invertebrates eaten. Harvestmen were taken in substantial numbers, mainly in the autumn season.

Table 3 shows the concentration of cadmium and lead

Table 5. Mean concentration ($\mu\text{g/g}$ dry weight) of cadmium in target organs of *Sorex araneus* and *Microtus agrestis*^a

Site	Species	Organ	Parameter	Feb-Mar	May-Jun	Oct-Nov
Budel	<i>M. agrestis</i>	kidney	geom. mean	2.2 (16)	1.0 (16)	2.7 (19)
			95% conf. int.	1.5-3.2	0.65-1.6	1.8-4.2
			range	0.70-6.8	0.48-5.4	0.91-16
		liver	geom. mean	0.33 (16)	0.19 (16)	0.57 (20)
			95% conf. int.	0.24-0.44	0.14-0.25	0.32-1.0
			range	0.12-0.85	0.07-0.48	0.07-9.8
	<i>S. araneus</i>	kidney	geom. mean	200 (14)	81 (5)	126 (13)
			95% conf. int.	148-270	11-588	91-173
			range	46-392	15-406	59-284
		liver	geom. mean	268 (13)	77 (5)	180 (13)
			95% conf. int.	219-328	8.5-701	140-231
			range	140-510	16-541	92-329
Arnhem	<i>M. agrestis</i>	kidney	geom. mean	0.28 (11)	0.09 (14)	0.22 (18)
			95% conf. int.	0.25-0.33	0.04-0.23	0.11-0.42
			range	0.01-0.80	0.01-1.3	0.22-0.38
		liver	geom. mean	0.10 (12)	0.12 (13)	0.13 (19)
			95% conf. int.	0.08-0.11	0.09-0.14	0.11-0.15
			range	0.06-0.14	0.06-0.22	0.07-0.23
	<i>S. araneus</i>	kidney	geom. mean	51 (9)	14 (14)	31 (10)
			95% conf. int.	41-62	7.5-25	26-38
			range	35-75	3.0-56	19-41
		liver	geom. mean	45 (9)	10 (14)	30 (10)
			95% conf. int.	33-61	5.7-18	23-39
			range	19-66	2.1-49	18-54

^a Geometric means, 95% confidence intervals and ranges are shown, with the number of samples in parentheses

present in the various food items of *Sorex*. Earthworms accumulated cadmium and lead and contributed these metals to a greater extent than any other group of invertebrates.

Metal Intake Rate

The average rates of intake of cadmium and lead in *Sorex* and *Microtus* are given in Table 4. The daily metal intake showed strong interspecific variation, with the levels many times higher in shrews than in voles. Values for shrews in February-March could not be calculated because of the absence of active soil fauna during the cold season.

The average daily intake of cadmium by *Sorex* was greater at the Budel than at the Arnhem site. The shrews showed a higher daily intake of lead at the Arnhem site than at the Budel site. *Microtus*, however, showed relatively little difference in the average intake of cadmium and lead between sites. The elevated lead intake in voles during February-March was due predominantly to the increased concentration of lead in grasses during that period.

Metal Load of Target Organs

Average metal concentrations present in the kidneys and liver of *Microtus* and *Sorex* are shown in Tables 5 and 6 for cadmium and lead, respectively. The two species differed markedly in the tissue accumulation of cadmium and lead, with *Sorex* showing metal levels in the kidneys and liver many times higher than those in *Microtus*. Mean concentrations of tissue cadmium generally tended to be lowest in the May-June period. This was also found with respect to the tissue level of lead, although only in animals of the Budel

site. Voles showed the highest level of tissue lead in the February-March period.

The average concentration of tissue cadmium was higher in animals in the Budel area than in those living in the Arnhem area. Differences between sites with respect to the tissue level of lead were apparent only in *Sorex* during the late season.

Discussion

The concentration level of cadmium in target organs was two orders and those of lead one order of magnitude higher in *Sorex* than in *Microtus*. The strong interspecific difference in organ metal load reflected the average daily intake of cadmium and lead which was higher for shrews than for voles. The major source for the intake of metals by shrews consisted of lumbricid earthworms, which have also been implicated in the metal exposure of other insectivores such as the mole *Talpa europea* (Ma 1987). Earthworms, however, have not been mentioned in the diet of shrews by Hunter *et al.* (1987), who based their conclusion on fecal analysis. However, the chance of underrating soft-bodied earthworms is greater in fecal analysis than in stomach analysis as used in our study (Phillipson *et al.* 1983).

Cadmium intoxication leading to subclinical symptoms have been described in small mammals at a critical renal concentration of 30 $\mu\text{g/g}$ fresh weight (Chmielnicka *et al.* 1989), which corresponds with a dry weight concentration of 119 $\mu\text{g/g}$ assuming a kidney dry matter content of 25.3% (Ma 1987). *Sorex* living at the contaminated Budel site showed a renal cadmium load above the critical level, especially during the late season. The risk of cadmium intoxication for the herbivorous *Microtus*, however, seemed negligible in view of the low cadmium level in the target organs.

Table 6. Mean concentration ($\mu\text{g/g}$ dry weight) of lead in target organs of *Sorex araneus* and *Microtus agrestis*^a

Site	Species	Organ	Parameter	Feb–Mar	May–Jun	Oct–Nov
Budel	<i>M. agrestis</i>	kidney	geom. mean	5.9 (16)	2.6 (16)	4.7 (19)
			95% conf. int.	5.0–6.9	2.4–2.9	3.8–5.7
			range	3.2–8.1	1.9–3.8	3.0–14
		liver	geom. mean	1.7 (18)	0.76 (16)	1.2 (20)
			95% conf. int.	1.5–1.9	0.66–0.87	0.92–1.5
			range	0.90–1.5	0.50–1.2	0.50–4.1
	<i>S. araneus</i>	kidney	geom. mean	35 (14)	58 (5)	23 (13)
			95% conf. int.	20–59	27–124	13–41
			range	12–178	39–143	7–156
		liver	geom. mean	2.8 (13)	5.4 (5)	2.0 (13)
			95% conf. int.	2.1–3.8	3.4–8.6	1.4–2.9
			range	1.3–6.6	3.5–8.7	1.0–8.7
Arnhem	<i>M. agrestis</i>	kidney	geom. mean	5.2 (11)	2.9 (14)	2.4 (18)
			95% conf. int.	4.4–6.2	2.3–3.8	2.0–2.8
			range	3.4–7.6	1.2–4.8	1.5–4.7
		liver	geom. mean	1.5 (12)	0.86 (13)	1.0 (19)
			95% conf. int.	1.2–2.0	0.62–1.2	0.81–1.2
			range	0.60–3.1	0.30–1.9	0.40–2.3
	<i>S. araneus</i>	kidney	geom. mean	60 (9)	47 (14)	39 (10)
			95% conf. int.	31–116	33–66	31–47
			range	15–323	21–269	25–54
		liver	geom. mean	4.7 (9)	4.5 (14)	4.2 (10)
			95% conf. int.	3.1–7.3	3.4–6.0	2.9–6.2
			range	1.4–7.8	1.1–7.6	2.1–9.2

^a Geometric means, 95% confidence intervals and ranges are shown, with the number of samples in parentheses

For the exposure of small mammals to lead a critical level of 25 $\mu\text{g/g}$ (Ma 1989) in target organs (kidneys) has been reported. This level was exceeded by *Sorex* at both the sites. Lead levels in target organs of *Sorex* were somewhat higher at Arnhem than at Budel, reflecting the highest average daily lead intake by shrews at the Arnhem site. Concentration of lead in *Microtus* remained below the critical level, which is consistent with the lower daily intake of lead in this species.

According to the hazard assessment approach used in this study, the Budel area was polluted with cadmium and lead and the Arnhem area with lead, to an extent such as to be hazardous for insectivores. The latter group of small mammals appears to be more sensitive indicators of the toxicity of bioavailable cadmium and lead in terrestrial ecosystems than herbivores.

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