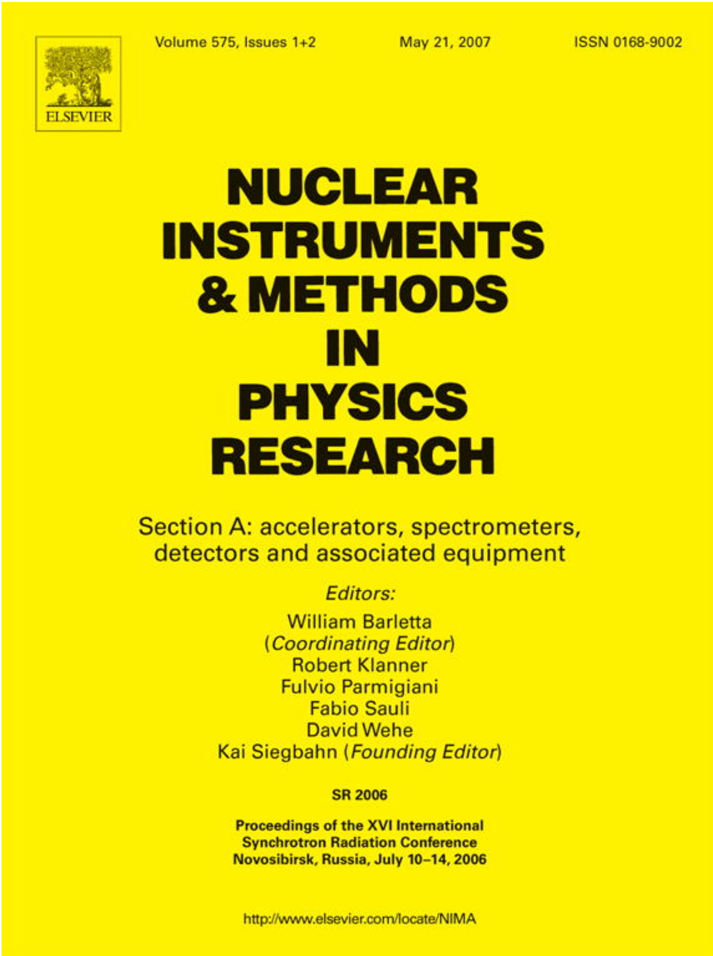
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Using of Synchrotron radiation for study of multielement composition of the small mammals diet and tissues

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Abstract

The Synchrotron radiation X-ray Fluorescence analysis (SRXRF) was used for estimation of "geochemical selection" of elements by small mammals, which belong to different trophic groups and inhabit polluted and background areas (the Middle Ural). The concentrations of K, Ca, Ti, V, Cr, Mn, Fe, Co, Ni, Cu, Zn, As, Br, Rb, Sr, Y, Cd, Pb in the diet and into hepar of a herbivorous (*bank vole*) and carnivorous (*Laxmann's shrew*) small mammals were compared. Herbivores play a particular role in chemical elements translocation between trophic levels, limiting element transition to consumers of the consequent levels. Whereas, insectivores concentrate most elements in their tissues under the same conditions.

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Keywords: SRXFR; AAS; Multielemental composition; Small mammals; Environmental pollution

1. Introduction

Numerous data on the content of different chemical elements in separate components of natural ecosystems have been gathered to the present time. However, information about migration of elements through food chains is still insufficient. Most frequently it concerns a small set of elements, as a rule, ecotoxicants. The knowledge of microelemental contents of populations and animal associations is essential, since it helps to reveal the mechanisms of forming biogenous cycles. Finally, the regulation of influence of industrial manufactures on environment is based on the latter. The aims of the present study were to estimate "geochemical selection" of elements by small mammals belonging to different trophic groups, and inhabiting both polluted and background areas.

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2. Materials and methods

Small mammals were trapped with snap-traps at two sites in the Middle Ural in July 2004: in polluted (1–2 km from cooper-smelting plant, impact zone) and background areas (30 km, the regional level). Captured animals were identified to species and sex. Only mature individuals were included into the analysis.

Concentrations of 18 chemical elements (K, Ca, Ti, V, Cr, Mn, Fe, Co, Ni, Cu, Zn, As, Br, Rb, Sr, Y, Cd, Pb) in diet ($n = 20$) and in hepar ($n = 20$) of a herbivorous (bank vole, *Clethrionomys glareolus,* Shreber, 1780) and a insectivorous (Laxmann's shrew, *Sorex caecutiens,* Laxmann, 1788) small mammals were compared. Samples of hepar and stomach content of animals were dried at 70 °C to constant mass. The processes of preparation of samples for chemical measures were described in detail earlier [1]. Samples were analyzed as tablets of 10 mm in diameter and of 30 mg in weight each. The elemental compositions of these samples (excluding Cd) were measured by the Synchrotron radiation X-ray Fluorescence analysis (SRXRF) method at the station of elemental analysis of the Budker Institute of Nuclear Physics, Siberian Branch of the RAS (VEPP-3 storage ring) [2]. The excitation energy was 21 keV. Processing of X-ray emission spectra was carried out with the help of AXIL program. The quantitative determination of elements was accomplished on the base of the external-standard methods (Russian Standard grass-mix SBMT-02). Cd was determined by means of atomic absorption spectrometer (ASS-6, Carl Zeiss) in the laboratory of population ecotoxicology of the Institute of Plant and Animal Ecology RAS. Chemical analysis was performed after digesting of 0.1 g of each sample in 4:1 HNO_3: H_2O mixture by microwave decomposition.

Data were analyzed by means of Microsoft Excel applied programs. The data were $\log_{10}$ transformed to meet the underlying assumption of homogeneity of variance and normality of distribution. Statistical analysis was performed with Student's test of significance ($p<0.05$) to evaluate observed differences in elemental concentrations (geometric means).

3. Results

3.1. Composition of small mammals' rations of two trophic groups

Concentrations of most part of elements in animals rations of both groups were between 0.1 and 1000 ppm, the ranges of concentrations were about the same (Table 1). At the background site, food of shrews contained more Co, Fe and Zn as compared to rations of the voles. Under pollution regime, levels of Ca, Fe, Co, Cu, Zn, As, Pb in food of bank vole significantly increased (up to eight times). Elemental composition of shrew's diet also changed: levels of Pb and Cu increased, whereas Rb and Y decreased significantly. This might be accounted for by changes in shrew's diet composition in polluted areas. To be precise, some groups of invertebrate animals disappeared (*Lumbricidae, Enchytraeidae, Diplopoda, Mollusca*) or sharply diminished in their numbers (*Carabidae, Staphylinidae, Arachnidae, Diptera*), whereas abundance of the others (*Elateridae*) sharply enhanced. The representatives of the order *Coleoptera* (*Elateridae, Staphylinidae and Carabidae*) played a great role in the diet of animals from the polluted areas. These insects are characterized by low bioaccumulation of elements.

3.2. Composition of small mammal's tissues of two trophic groups

We consider that hepar to be an organ which reflects the accumulation of chemical elements in the organism. Enhanced levels of Co, As and Pb in vole's diet lead to increase of their content in hepar. Meanwhile an increase in concentrations of essential Fe, Cu, Zn is not accompanied by significant increase of these elements in hepar, this is caused by homeostatic barrier. In the hepar of shrews from polluted sites, only Cd, Pb and Ti were highly concentrated.

Obtained data allow us to evaluate the role of ecological factors in the formation of biogenous cycles, which include small mammals of different trophic levels. In the case, increase of some element concentration in the ration (at polluted sites) led to its proportional increase in an accumulating medium (hepar), then a direct (without limitation) translocation to the upper trophic level would occur. Contrarily, if the elemental content in an accumulating organ decreased under the same conditions, its flow to the upper level would be limited by gastrointestinal tract level barrier.

Most elements in the bank vole, including toxic ones (V, Cr, Mn, Ni and Pb), and a number of physiologically essential were discriminated by its barrier. Thus transition from the producers level (vegetable diet) to the primary consumers level (carnivorous diet) is limited. We found that in the food of bank vole the typical toxicants (As and Cd) only were concentrated. The reverse effect was observed in insectivorous that inhabit polluted areas: concentrations of most part of elements in shrew's hepar increased as compared with their diet. The exceptions were only Cu, Zn, As and Y.

Ecotoxicological effect of environmental pollution is most probably defined not by concentrations of individual pollutants, but by total exceeding of toxicant concentrations in food and tissues above background levels. The most frequently as a basis for evaluation of toxic load is taken summarized stands out above background concentrations of elements and it is calculated as

$$S_n = (1/n)\Sigma(C_i/C_f),$$

where C_i and C_f and the concentrations of elements that are measured at polluted and background areas, respectively, n the number of calculated elements. The load will be calculated for different causes (supplies toxicants with diet, accumulation elements in tissues) [3]. Exceeding of joint toxically influence took place in the all variants in polluted areas. The maximum load is formed by broad spectrum of elements in rations of phytofags (Ti, V, Cr, Co, Cu, Zn, As, Cd, Pb). "Food load" for shrews is two times lower and is formed by Cu, Cd, and Pb. Despite of marked differences, hepar of both species in polluted environments was exposed to about the same toxic influence.

4. Conclusion

Ability of use of the SRXFR method for detection of multielemental composition of biological objects in the small volumes was shown in our study. It was concluded that phytofags play a particular role in limiting of element transition to consuments of consequent levels, whereas insectivorous extend as concentrators of the most part of studied elements.

Table 1
Elemental composition[a] of the diet and hepar of individuals of the bank vole and Laxmann's shrew, inhibiting background (A) and polluted (B) areas

Element	Concentration of element (ppm)								P
	Bank vole				Laxmann's shrew				
	Stomach content		Hepar		Stomach content		Hepar		
	A (1)[b]	B (2)	A(3)	B (4)	A (5)	B (6)	A (7)	B (8)	
K	26837	17585	12268	9993	14301	7753	10254	11266	
	12740–31200	10413–25427	9920–15670	8565–10844	8075–24786	5347–12499	9269–12362	10105–1222	
Ca	5945	2286	703	233	7316	2906	232	269	a
	4090–7749	1057–3658	205–711	168–41	2215–20436	1254–4796	179–279	218–329	
Ti	29.8	104	4.6	5.6	375	48	5	9.9	d
	21–39	18–250	2.7–11.7	3.8–8.2	40–576	20–80	3.6–9.0	7.8–12.9	
V	0.4	0.8	0.02	ns	2.2	0.3	0.001	0.02	
	0.27–0.43	0.05–1.83	0.01–0.05		0.43–6.17	0.23–0.39	0.0–0.002	0.01–0.02	
Cr	175	1110	7	10.8	383	5061	11	28	
	72–393	36–3061	5.5–9.9	4.3–17.2	27–939	73–1137	7.2–16.0	10.0–54.4	
Mn	263	334	16	15	312	131	32.6	31	g, h
	198–359	99–615	13–20	9–24	101–592	110–160	28–38	20–37	
Fe	474	2171	497	1296	2933	1233	1053	1379	a, b, e
	329–644	785–3947	234–742	707–2041	666–6720	883–1641	564–2099	796–1918	
Co	0.2	0.7	0.2	0.4	0.7	0.4	0.36	0.4	a, b, e
	0.14–0.21	0.27–1.24	0.11–0.22	0.29–0.65	0.22–1.42	0.32–0.47	0.23–0.63	0.30–0.53	
Ni	21	26	0.6	0.6	11.8	6	0.4	0.5	f
	17.6–29.0	7.6–39.3	0.48–0.91	0.44–0.82	4.1–19.0	4.8–8.1	0.02–0.58	0.25–0.71	
Cu	18	118	12.7	13.5	29	84	14.7	18	a, c
	17–21	63–214	11–16	11–17	17–58	65–98	14–16	15–21	
Zn	111	316	103	99.8	268	348	82	85	a, b, f, g
	100–132	132–424	96–110	85–117	177–426	287–464	74–88	82–89	
As	0.4	2.8	0.2	2.3	1.2	0.8	2.0	0.8	b, h
	0.02–1.22	2.71–3.11	0.17–0.29	0.81–5.02	0.3–35	0.26–1.25	0.9–3.2	0.28–1.2	
Br	10.5	85	8.8	39	17	15.5	7.6	10.4	b, h
	8–13	19–307	6–13	10–63	12–23	11–21	6–9	9–12	
Rb	37	15	30	14.7	16	4.9	14.9	5.8	b, c, d, f, g, h
	15–52	9–22	21–37	10–20	8–29	3–6	12–19	4–8	
Sr	27	13.6	0.4	0.3	24	33	0.3	2.4	
	20.2–45.3	1.36–23.3	0.19–0.71	0.22–0.52	7.4–63	7.0–82.7	0.11–0.52	0.06–9.4	
Y	2.0	2.2	1.7	0.9	1.9	1.3	0.7	0.4	b, c, d, f, g, h
	1.0–2.8	1.35–3.6	1.1–2.1	0.58–1.2	0.85–3.8	0.58–2.52	0.63–0.88	0.33–0.53	
Pb	23.7	306	0.9	3.6	100	169	0.8	13	a, b, c, d, f
	14–38	155–481	0.38–1.17	0.88–5.67	23–192	116–217	0.24–1.39	3.45–25.4	
Cd	1.7	6.3	1.4	11.2	7.5	7.3	17.8	54	a, e
	0–14.4	0.59–16.8	0–8.9	0–16	1.54–19.8	0.89–16.1			

Different letters denote significant differences within groups ($p<0.05$): a—1–2; b—3–4; c—5–6; d—7–8; e—1–5; f—2–6; g—3–7; h—4–8; ns—non-significant.

[a]Average means concentration of element and minimum–maximum values.

[b]Different numbers denote number of group for comparison with Student's test.

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