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Hair Lead and Body Lead Burden in Lead-Intoxicated Rats Fed Diets Enriched with Dietary Fibre

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Abstract

In the factorial 2^3 design male Wistar rats were fed ad libitum diets supplemented with dietary fibre preparations (fruit and maize preparations; 50 and 150 g/kg diet) and intoxicated with lead acetate per os (200 and 400 mgPb/kg diet) for 5 weeks. At the end of the study, rats were anaesthetised and hair, liver, kidney, spleen, and femoral bone were dissected for chemical analysis. It was found that: quantity of fibre in the diet significantly affected inner organ and tissue lead contents in the rats. Dietary lead levels influenced all analysed tissue and organ lead contents and the total body retention of lead in the rats. Positive correlations were found between hair lead and inner organs lead (r = 0.35) and bone lead (r = 0.35), and total body lead burden lead (r = 0.40).

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Keywords: lead, hair, tissues, correlation, rats

Introduction

The main source of adult human exposure to lead is food, which is believed to provide about 80-90% of daily dose. On the other hand, nutrition is an important factor modifying lead effects upon the organism and can be used in prevention of its toxicity. A number of studies carried out on animals have revealed that diets deficient in protein [1, 2], vitamin Bi [3, 4], vitamin C [5], vitamin E [6] and minerals such as calcium [7, 8], magnesium [9], iron [10], zinc [11], and copper [12, 13, 14] enhance toxicological effects caused by lead. Similarly, high-fat and high-protein diets are unfavourable for animals poisoned with lead [1]. On the other hand, dietary fibre features sorption capabilities that make it possible to deposit on its surface ions of various elements essential and nonessential for human organism. Therefore, taking into account physicians' and nutritionists' recommendations of increasing intake of dietary fibre in the diet and its possible protective effect against lead toxicity, we carried out investigations on laboratory rats fed diets supplemented with fibre intoxicated with lead. The results of this experiment were reported in our previous paper [15].

Recently, hair has been increasingly used as an index of exposure to heavy metals. Hair analysis has some advantages over blood tests since it is non-invasive, stressless and the concentration of elements in hair tissue is much higher than in body fluids, which simplifies analytical procedures and reduces the possibility of contamination. However, one must be aware of various endogenous and external factors influencing trace element deposition in hair which obscure interpretation of results.

In this study we investigated the relationship between lead levels in hair and inner organs, bone skeleton and total body lead content to evaluate the usefulness of hair mineral analysis in assessment of the total body lead burden.

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Materials and Methods

The study was carried out in 48 male Wistar rats (initial weight 80 g) in the factorial experiment (2^3). The experimental factors were:
A - source of dietary fibre (fruit, maize preparations), B - quantity of fiber supplementation (50 g/kg, 150 g/kg), C - dietary lead level (200, 400 mg/kg diet).

Rats, divided into 8 groups (6 rats in each) were housed individually into semi-metabolic cages and fed diets contained in 1 kg: 200 g casein, 80 g sunflower oil, 20 g lard, 500 or 400 g wheat starch, 100 g sucrose, 50 or 150 g fibre preparation (in the case of higher fibre supplementation, additional amount of fibre substituted for 100 g wheat starch), 40 g mineral mixture (NRC, 1976), and 10 g vitamin mixture (AOAC, 1975).

Lead as lead acetate trihydrate was introduced together with the mineral mixture. The source of fibre in diets were fruit (apple-raspberry pomace, 1:1, w/w) and maize preparations obtained according to the technology described by Aniola et al. [16].

At the end of the experiment, animals were weighed, anaesthetised (diethyl ether inhalation), and inner organs and femur bones were removed and hair samples were collected and stored at 253 K (- 20°C) until analysis. The content of lead in inner organs, femur and hair was determined using flame atomic absorption spectrometry method (Zeiss AAS-3 spectrometer, with deuterium BC) after wet digestion of samples with the mixture of spectra-pure concentrated acids (nitric, perchloric, 1:1, v/v, Merck). The approximate inner organs lead content was calculated as a sum of liver, kidneys and spleen lead contents. The approximate total bony skeleton lead content was estimated on the basis of the following assumptions: lead concentration in all sorts of bones is even and equals femur lead concentration, femur weight in an adult rat constitutes about 5% of the bony skeleton weight [11]. Approximate total body lead was calculated as a sum of inner organ lead and bony skeleton lead while hair lead was avoided due to difficulty with estimation of the total hair mass in the rat. All data were statistically analysed applying analysis of variance with the help of the Fisher test and computer program Statgraphics ver. 2.1.

Results

The influence of the source and quantity of dietary fibre on relative excretion of lead, and its accumulation in tissues, organs and total body lead in rats was discussed in our previous paper [15]. In this paper we analyzed the influence of dietary factors on the concentration of lead in tissues and organs and their correlation with hair lead content. The results are shown in Tables 1 and 2.

Rats fed high-fibre diet vs. low-fibre diet had lower liver lead concentration (by 35%) whereas high dietary lead increased the liver lead content (by 28%). The kidney lead content increased (by 23%) with a dietary lead level of 400 mg/kg. Spleen lead content was influenced both by source of fibre and lead level in the diet. A significant decrease in spleen lead in the rats was associated with diet containing maize preparation while increasing lead content in this organ followed high dietary lead. The hair lead concentration decreased (by 40%) in the rats fed diet supplemented with maize fibre as well as high-fibre diets (by 50%). The inner organs lead content, femur lead, the total bony skeleton lead, and total body retention of lead were influenced independently by quantity of fibre in the diet and dietary lead level. Rats fed high-fibre diets resulted in lower femur lead and bony skeleton lead (by 30%), the inner organs lead content (by 25%) as well as total body retention of lead (by 30%), whereas high dietary lead level produced a significant inverse effect on these parameters.

For evaluation of relationships between hair lead levels and lead body pools linear regression analysis was performed. As shown in Table 2, there were significant

Table 1. Influence of experimental factors on tissue lead content of rats parameters and its statistical evaluation (mean values for 6 rats) [after 15].

Lead content	Experimental factor		
	A source of fibre (dietary fibre preparation) fruit maize	B quantity of fibre (% of diet mass) 5% 15%	C dietary Pb (mg Pb/kg diet) 200 400
Liver (µg/g WM)	2.32 2.34	2.82** 1.84	2.05** 2.61
Kidney (µg/g WM)	11.6 11.0	12.3 10.4	10.1* 12.4
Spleen (µg/g WM)	2.19** 1.60	2.00 1.79	1.62** 2.12
Hair (µg/g DM)	19.8* 12.1	20.9** 10.6	16.3 15.5
Femur (µg/g DM)	119 109	135** 94	100** 129
Inner organs Pb content (µg/rat)	56.4 56.7	64.4** 48.7	48.0** 65.0
Bony skeleton (mg/rat)	1.86 1.65	2.05** 1.46	1.51** 2.01
Total body (mg/rat)	1.93 1.71	2.12** 1.51	1.56** 2.07

Interactions:
Liver Pb: AB**, BC', ABC**; Kidney Pb: AB*; Hair Pb: AB**; Femur Pb: AB*
Inner organs Pb: AB**, BC*; Bony skeleton Pb: AB**, Total body Pb: AB*
LEGEND:
DM - dry mass, WM - wet mass
*,** - statistically significant at $p < 0.05$, $p < 0.01$

Table 2. Relationship between hair lead and inner organs, bone and the total body lead in rats (regression coefficient, p).

Parameter	Inner organs lead content (μg)	Bony skeleton lead content (mg)	Total body lead content (mg)
Hair lead (μg/g)	r = 0.35 p < 0.01	r = 0.35 p < 0.05	r = 0.40 p < 0.01
Regression equation	Y = 0.23x + 2.38	Y = 0.0067x + 3.52	Y = 0.0074x + 1.47

positive correlations between hair lead content and its concentration in the inner organs ($r = 0.35$, $p < 0.01$), bones ($r = 0.35$, $p < 0.05$) and the whole organism ($r = 0.40$, $p < 0.01$).

Discussion

In literature information on the relationship between quantity and fibre composition in the diet and tissue lead distribution in animals intoxicated with lead is scant. We found that high-fibre diet intake was associated with lower lead content in the liver, kidneys, inner organs, hair, femur, (bone), and the total body retention of lead in the rats. On the other hand, high dietary lead level (400 mg/kg) increased lead content in all analysed tissues and organs [15]. The kidney lead content was four-fold that of the liver, which shows increasing accumulation of this metal in renal cortex that was also observed by Flora [14]. The highest lead concentration was found in bone tissue (femur), where the majority of lead is stored in the organism.

In order to evaluate the usefulness of hair lead analysis in assessment of lead exposure some authors calculated the relationship between hair lead and other body lead pools in animals and humans. Zachwieja et al. [17], Foo et al. [18], Olejnik et al. [19] reported a significant positive correlation between lead levels in hair and blood in children and adults. Hac and Krechniak [20] studied the relationship between lead levels in bone and hair of rats treated orally with lead acetate in drinking water for 12-14 weeks. The authors found a high positive correlation between the concentration of lead in desiccated bone and hair of the rats ($r = 0.876$). In our study the coefficient r is also positive but of lower value ($r = 0.35$) which may be caused by the differences in doses of lead ingested, duration of the experiment as well as the protective role of dietary fibre introduced to the animal's diets. The results obtained in this experiment indicate that in the case of continuous exposure to lead via alimentary tract, the content of this element in hair seems to reflect its deposition in the inner organs and bone, thus hair may serve as a biopsy material for evaluation of the total lead body accumulation over several weeks or months.

However, one must be cautious with the assessment of the dose ingested since various dietary, non-dietary and external factors affect the content of lead in hair tissue.

Conclusions

The hair lead concentration correlates with the inner organs lead, bone lead and the total body lead burden in rats fed diets enriched with dietary fibre and chronically exposed to oral lead, thus hair lead may reflect total body lead accumulated over the time of hair growth.

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