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Metals interaction tested in children's hair originating from industrial and rural areas

Interakcja metali badana we włosach dzieci zamieszkałych na terenach przemysłowym i rolniczym

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[(a)] *concept*
[(b)] *collection of material for research*
[(c)] *chemical analyses*
[(d)] *statistics*
[(e)] *working on text and references*
[(f)] *interpretation*

ABSTRACT

Introduction. Different biological samples (blood, gallstone, teeth, hair) serve as a biomarker of exposure to metals for many years. This method appeared to be useful not only in clinical medicine, but also in the studies on the environment. **Aim**. The study is to compare the amount of selected metals in children's hair residing in industrial and rural areas. **Material and methods**. Research of occurrence of 12 metals in children's hair at the age of 7, 10 and 14 living in an industrial (Nowy Bytom town) and a rural (Strumień town) areas has been presented. Determination of Pb, Cd, Ni, Co Na, K, Mg, Zn, Cu, Mn, Fe and Ca was carried out by atomic absorption spectrophotometry (AAS) using a spectrometer Perkin-Elmer 400. **Results**. In the case of seven-year old children, regardless of gender a common mechanism of co-occurrence was noticed for manganese and calcium, manganese and magnesium, calcium and magnesium, sodium and potassium. Apart from the correlation of metals for the seven-year-old-children mentioned, in case of ten-year old children, an additional correlation between calcium and zinc appears. **Conclusion**: The amount of some metals in the hair with the diversified possibility of interaction between the metals themselves and their relation to gender and age of children revealed different environmental exposure.

Keywords: hair, children, industrial area, agricultural area, interaction

STRESZCZENIE

Wstęp. Materiał biologiczny (krew, złogi pęcherzyka żółciowego, zęby, włosy) służy od lat jako biomarker ekspozycji na metale. Metoda ta okazała się być przydatna nie tylko w medycynie klinicznej ale również w badaniach nad środowiskiem. **Cel pracy.** Porównanie zawartości wybranych metali we włosach dzieci zamieszkałych na terenach przemysłowym i rolniczym. **Materiał i metody.** Zaprezentowano wyniki występowania 12 metali we włosach dzieci w wieku lat 7, 10 i 14, zamieszkujących tereny przemysłowy (Nowy Bytom) i rolniczy (Strumień). W celu dokonania oznaczeń Pb, Cd, Ni, Co, Na, K, Mg, Zn, Cu, Mn, Fe i Ca zastosowano absorpcyjną spektrometrię atomową. Wykorzystano spektrometr Perkin-Elmer 400. **Wyniki**. W przypadku dzieci 7- i 10-letnich dostrzeżono mechanizmy współwystępowania bez względu na płeć dla Mn i Ca, Mg i Mn, Ca i Mg, Na i K. Przy czym u dzieci 10 letnich dodatkowo wystąpiła współzależność pomiędzy Ca i Zn. **Wnioski**. Zawartość niektórych metali we włosach dzieci determinowana jest różną ekspozycją środowiskową, płcią i wiekiem badanych. Możliwa jest interakcja pomiędzy różnymi metalami.

Słowa kluczowe: włosy, dzieci, teren przemysłowy, teren rolniczy, interakcja

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INTRODUCTION AND AIMS

Content of various heavy metals is a frequent indicator of environmental exposure. Their concentrations have been identified in the foregoing literature primarily in occupational health protection.

Concentrations of lead were determined in the hair of 478 school children living in an industrial area and rural region of Spain. The average lead amount was higher in the industrial area (9.38 µg/g) [1].

Concentration of metals was also determined in the scalps of children living in an industrialized area and a rural region of Germany. The content of metals was as follows: Pb 2.7 µg/g, Cd 90.0 µg/g, Cu 10.6 µg/g and Zn 108.0 µg/g [2]. The lead content in hair of the gas station workers being 48.7±17.5 µg/g was significantly higher than that of the controls 17.2±8.1 µg/g [3].

In the hair of children living in the urban region of Mexico (close to the smelter plant complex) the range of cadmium in hair was 0.25–3.5 µg/g [4].

Further, in three Russian cities daily cadmium intake and health concerns of the working population and the children who resided close to the sources of contamination were assessed . In the surveyed workers of a storage battery factory the average cadmium content in hair was 99.3 µg/g; an average cadmium level in the hair of workers in a cadmium containing dyes plant was 25.1 µg/g [5].

In contrast to urine, blond hair serves as a record of exposure to heavy metals formed in a relatively short time and isolated from the general metabolic processes of the organism [6]. The co-occurrence of metals in the hair is a complex problem and depends on a number of factors. The presence of heavy metals in the hair may, to a certain extent, reflect environmental as well as biochemical characteristics of a given individual [7, 8].

Wright et al. [9] studied the potential associations between the hair metal levels and the neuropsychological function and behaviour of school-aged children. The mean hair metal levels were following: Mn: 471.5 µg/g, As: 17.8 µg/g, Cd: 57.70 µg/g. Children's general intelligence scores, particularly verbal IQ scores, were significantly related, inversely, to hair Mn and As levels, as were scores on tests of memory for stories and a word list. In some cases, a significant relation between Mn and As was found. More detailed analysis confirmed low scores for children for whom both Mn and As levels in the sample were above the median values. These results suggest further need for studies on neuropsychological consequences of developmental exposure to Mn and As particularly if they appear in the form of a mixture [9].

Khalique et al. (2005) investigated levels of 10 metals (Ca, Mg, Fe, Zn, Cu, Mn, Cd, Co, Cr and Ni) in the scalp hair of male and female donors. Calcium showed the highest concentration of 462 g/g in the hair of males and 870 µg/g in the hair of females followed by Zn. Content of Zn was almost equal 208 µg/g for boys and 251 µg/g for girls. For male donors Cd showed the lowest concentration (1.15 µg/g), for female donors from Chile country Co was at minimum level (0.92 µg/g). The order of decreasing metal concentration in the hair of male donors was: Ca > Zn > Mg > Fe > Cu > Mn > Ni > Cr > Co > Cd that for female donor sit was: Ca > Zn > Mg > Fe > Cu > Mn > Cr > Ni > Cd > Co. The female group exhibited in their hair enhanced levels of small quantity of selected metals except for Fe and Co as compared with the male counterparts. For the hair samples from males in Chile country a strong bivariate positive correlation was found between Fe and Zn (r = 0.841). As to Chilean females, strong positive relationship was observed for Ca–Mg (r = 0.617), Ca–Zn (r = 0.569), Ca–Mn (r = 0.565), Mg–Mn (r = 0.655), Cr-Cu (r = 0.655) and Cr-Ni (r = 0.685). The distribution of metals in the hair of donors with respect to different age groups was also investigated for both genders. The study showed that in the case of males, the concentration of all selected metals decreased with increasing age except for Cu, Co and Cr. However, for females the hair metal levels increased with age, except for Co for which the concentration decreased with age [10].

Knowledge on normal levels of concentration of trace elements (Cd, Cr, Cu, Hg, Pb, Se and Zn) in the population serve, among other things, in the designing of regulations of exposure limits and prevention of diseases caused by deficiency in essential trace elements. Values of trace elements concentrations in children's hair were: Cd 0.14 µg/g, Cr 0.22 µg/g, Cu 12 g/g, Hg 22 g/g, Pb 1.6 µg/g, Se 0.22 µg/g, and Zn 124 µg/g (in medians). Statistically significant differences correspond to the published values for non-exposed population [11].

Concentration of Cu, Zn, Cr and Br increase from birth to 8 years of age and then decrease. Fe, Mn and Sr strongly decrease up to 8 years of age and then remains almost constant. Concentration of elements depends on gender. In case of children the elements are: Fe, Zn and Br; in case of adolescents the elements are: Cu, Cr, and Br [12].

Available literature data concerning the co-occurrence is limited and do not comprise relevant information on the subject [13]. The scope of research programs usually includes determination of the content of several metals [14, 15, 16]. The problem, especially in the area of complex environmental exposure to selected heavy metals, may be tackled by means of bioindicating analysis in selected biological samples [17–21].

Present routine laboratory techniques concerning total metal content do not allow to distinguish which metals in hair samples are of endogenous and exogenous origin. Because of that, it is important to start research on the content of 12 metals in the hair of children who live in the area impacted by a steel plant (Nowy Bytom town) located in the center of the industrial region of South-West Poland (n = 83) and in the hair of children who live in Strumień town – 60 km away from the above industrial town (n = 70). Strumień town is situated near the Beskidy mountains and represents the rural region.

MATERIALS AND METHODS

Hair samples were cut from the back of the head according to the methodology published in the literature. 1 g of hair was taken for the analysis, then weighed with an accuracy of 0.0001 g and mineralized with spectrally pure concentrated nitric acid. After mineralization, the solution was introduced into a measuring flask (V = 25 cm^3) and filled with redistilled water to a level mark.

Measurements were carried out on 153 children by atomic absorption spectrophotometry (AAS) using a spectrometer Perkin-Elmer 400. The content of Na, K, Mg, Zn, Cu, Mn, Fe, Ca was measured by means of emission method and the content of Pb, Cd, Ni and Co by AAS (flame analysis). The accuracy of measurements was controlled by standard GBW 09101 Human Hair. The results differed among themselves maximum by 7% depending on an element.

The set of results obtained on the basis of a Student's test made it possible to define coefficients with a value higher than 0.45 and $p \leq 0.03$ as significant correlations.

RESULTS

Occurrence of metals in the hair of 7, 10 and 14 years old children constituted the subject of the research. Statistical analyses results of heavy metals content are shown in tables I–III. Moreover the co-occurrence of that relation between selected pairs of metals for children living in Nowy Bytom town (industrial area) was following: 7 years, 10 years and 14 years.

7 years old:

Mn–Ca {r = 0.57}, Mn–Mg {r = 0.60}, Ca–Mg {r = 0.84}, Na–K {r = 0.63}, Mn–Na {r = 0.48}, Mn–K {r = 0.54}, Cu–Ca {r = 0.48}, Cu–Fe {r = 0.66}, Ca–Zn {r = 0.46};

10 years old:

Mn–Ca {r = 0.62}, Mn–Mg {r = 0.53}, Ca–Mg {r = 0.63}, Ca–Zn {r = 0.54}; Na–K {r = 0.58}, Mn–Na {r = 0.51}, Mn–Pb {r = 0.53}, Ca–Pb {r = 0.63};

14 years old:

Fe–K {r = 0.62}, Ca–Mg {r = 0.77}, Na–K {r = 0.87}, Mn–Fe {r = 0.70}, Mn–K {r = 0.60}, F–Na {r = 0.57}, Cd–Co {r = 0.46}.

Table I. Characteristic of occurrence of selected metals in 7 year old children's hair (μg/g)
Tabela I. Charakterystyka występowania wybranych metali we włosach 7-letnich dzieci (μg/g)

Elements	NOWY BYTOM n = 30				STRUMIEŃ n = 25			
	Arithmetical mean ± standard deviation	Geometric mean	Range of changes	Coefficient variability [%]	Arithmetical mean ± standard deviation	Geometric mean	Range of changes	Coefficient variability [%]
Mn	3.53±0.38	2.94	0.90–11.30	11	1.5 1± 0.18	1.41	0.90–2.50	12
Cu	23.7±6.50	13.00	5.60–175.2	27	7.98±0.34	7.89.	5.20–9.10	4
Fe	44.20±10.50	33.80	14.4–403	24	47.8±2.30	47.30	38.3–58.0	5
Ca	836±121	612	147–2860	14	630±133	512	223–1605	21
Na	895±155	599	111–4855	17	784±128	673	247–128	16
Mg	70.10 v 9.20	53.40	13.60–241.5	13	41.4±12.2	31.0	14.40–154.0	29
Zn	137.3±8.10	128.40	39.0–272.5	6	409±200	223	96–2333	49
K	236±28	188	50–794	12	449±114	322	77.5–1102	25
Pb	28.6±2.60	22.90	0.25–80.10	9	2.74±1.98	0.76	0.25–22.2	69
Cd	1.31±0.32	0.31	0.05–6.50	24	0.61±0.23	0.21	0.05–2.00	36
Co	2.80±0.94	0.76	0.25–28.40	36	5.46±1.37	1.96	0.25–11.70	24
Ni	3.47±0.63	1.03	0.10–12.00	20	9.65±0.30	0.23	0.10–2.30	45

Table II. Characteristic of occurrence of selected metals in 10 year old children's hair (μg/g)
Tabela II. Charakterystyka występowania wybranych metali we włosach 10-letnich dzieci (μg/g)

Elements	NOWY BYTOM n = 30				STRUMIEŃ n = 25			
	Arithmetical mean ± standard deviation	Geometric mean	Range of changes	Coefficient variability [%]	Arithmetical mean ± standard deviation	Geometric mean	Range of changes	Coefficient variability [%]
Mn	4.56±0.60	3.63	0.90–15.60	13	1.98 ±0.54	1.52	0.50–7.60	27
Cu	26.00±7.80	15.20	5.90–243	30	11.90 ±3.10	10.10	7.40–46.20	26
Fe	43.79 ±3.40	40.10	19.40–103	8	26.30 ±5.60	21.00	6.60–71.20	21
Ca	558 ±91	399	151–2287	16	685 ±150	555	188–2050	22
Na	698 ±115	479	63–3219	17	492 ±227	264	70–2919	46
Mg	57.9 ±12.0	38.30	11.40–394	21	50.50 ±15.40	37.00	13.60–208	31
Zn	162 ±13	152	86–522	8	216 ±32	196	119–459	15
K	217 ±31	160	40–741	14	209 ±123	95	27–1548	59
Pb	37.40v11.00	25.60	2.30–402	30	19.10 ±4.80	8.01	0.27–54.0	25
Cd	2.27 ±0.68	0.77	0.05–22.90	30	1.89 ±0.68	0.47	0.05–7.10	36
Co	1.17 ±0.23	0.66	0.25–4.70	20	10.20 ±2.10	4.78	0.25–20.50	20
Ni	6.07 ±0.92	2.87	0.10–19.30	15	3.35 ±1.32	0.59	0.10–12.10	39

Table III. Characteristic of occurrence of selected metals in 14 year old children's hair (μg/g)
Tabela III. Charakterystyka występowania wybranych metali we włosach 14-letnich dzieci (μg/g)

Elements	NOWY BYTOM n = 30				STRUMIEŃ n = 25			
	Arithmetical mean ± standard deviation	Geometric mean	Range of changes	Coefficient variability [%]	Arithmetical mean ± standard deviation	Geometric mean	Range of changes	Coefficient variability [%]
Mn	2.81 ±0.31	2.25	0.50-8.60	11	2.05 ±0.21	1.87	0.70-4.30	10
Cu	32.30 ±8.30	16.80	3.60-223.0	26	12.20 ±1.51	10.60	2.70-28.20	12
Fe	18.70 ±4.00	6.25	0.03-100.70	21	24.20 ±5.30	6.30	0.03-77.70	22
Ca	836 ±100	641	180-2382	12	940 ±113	828	310-2124	12
Na	487 ±62	363	38-1501	13	250 ±37	200	35-575	15
Mg	64.10 ±6.60	52.70	16.30-175.70	10	57.0 ±11.0	45.0	15.50-209.0	19
Zn	170 ±8	164	74-334	5	190 ±20	170	27-470	11
K	159 ±21	119	27.50-491.5	13	83.0 ±17.2	57.40	6.70-333	21
Pb	14.30 ±1.40	10.70	0.25-34.30	10	4.45 ±1.10	1.91	0.25-14.40	25
Cd	1.29 ±0.24	0.50	0.05-5.20	19	0.95 ±0.33	0.25	0.05-5.70	34
Co	1.02 ±0.31	0.47	0.25-9.80	30	1.46 ±0.42	0.71	0.25-6.80	29
Ni	1.90 ±0.60	0.37	0.10-15.40	30	1.34 ±0.31	0.60	0.10-4.20	24

For comparison – children living in the rural area:

7 years old

Mn–Ca {r = 0.88}, Mn–Mg {r = 0.73}, Ca–Mg {r = 0.93}, Na–K {r = 0.79}, Fe–Ni {r = 0.48}, Fe–Zn {r = 0.47}, Cu–Ca {r = 0.48}, Zn–Ni {r = 0.51}, Zn–K {r = 0.52}, K–Co {r = 0.68}, Pb–Ni {r = 0.56}, Na–Co {r = 0.47};

10 years old

Mn–Ca {r = 0.93}, Mn–Mg {r = 0.98}, Ca–Mg {r = 0.96}, Ca–Zn {r = 0.59}, Na–K {r = 0.98}, Mn–Zn {r = 0.66}, Fe–Na {r = 0.64}, Fe–Pb {r = – 0.49}, Fe–K {r = 0.57}, Fe–Co {r = 0.49}, Mg–Zn {r = 0.58}, Cd–Co {r = 0.49}, Cd–Ni {r = 0.58};

14 years old

Fe–K {r = 0.55}, Ca–Mg {r = 0.51}, Na–K {r = 0.71}, Zn–Cd {r = 0.56}, Mg–Pb {r = 0.69}, Fe–Pb {r = 0.47}, Cu–Zn {r = 0.46}, Mn–Ca {r = 0.78}, Pb–Ni {r = – 0.48}.

The tables I–III include common relations of occurrence of particular metals in both towns. Because the towns differ in the degree of air pollution one can conclude that the presented algebraic expressions concerning several elements describe the content of these metals as a result of characteristic metabolism of the metals in the examined children. It is possible to state that the mentioned relations do not include the effect of chemopressure. In the case of seven-year old children, regardless of gender, there is a common phenomenon of co-occurrence for: manganese and calcium; manganese and magnesium altogether. The content of calcium depends on the changing content of magnesium sodium and potassium. In the background of the above relation there are proportional changes between the mentioned elements. In the case of ten-year old children, apart from the correlations of metals presented for seven-year-old-children, an additional correlation between calcium and zinc appears. The comparison of expressions which describe the co-occurrence of these metals suggest that the content of these elements depends upon physiology and biochemical condition of calcium, magnesium, sodium, potassium and zinc.

DISCUSSION

In the case of older children (14 years old), relations between calcium and magnesium, sodium and potassium remain the same and additional correlation between iron and potassium appears. Characteristic feature for metal correlations is high correlation coefficient and in some cases these correlations are stronger for children living in the area not impacted by the steel plant (Strumień). It should be pointed out that 9 out of 12 correlations which occur in nature have much higher correlation coefficient to those appearing in the case of children living in the industrial town Nowy Bytom. Although metabolic activity of calcium, sodium, potassium, magnesium and zinc is similar in children's body additional interactions with other metals are still possible in the area of strong chemopressure. These metals may cause reduction of a physiological correlation coefficient of the above elements. High values of correlation coefficient justify the regression equations. Established regression equations confirm similar trends of common changes in the content: Na and K; Ca and Mg; Mn and Mg; Mn and Ca The significant coefficient correlation is within 0.63–0.87 in Nowy Bytom town and within 0.71–0.93 in Strumień town. For example, the tendency of changing the content of sodium in relation to potassium, in the children from Nowy Bytom town is described by the coefficient which increases 3.5 times for seven-year-old, 2.1 times for ten-year-old and 2.57 times for fourteen-year-old children. Children living outside the range of the steel plant's impact are characterized by the following coefficients: 0.98, 1.82, 1.51 respectively. Coefficients describing the change of calcium content in the function of the changes of magnesium content in hair, which amount to 4.8 in the case of ten-year-old children and 11.1 for seven-year-old children, may be another illustration of the problem. Relations of changes of manganese with calcium and manganese with magnesium are characterized by the following coefficient 10^{-2}–10^{-3}.

On the basis of previous research and interpretation of the absolute term – b in the expression $y = ax + b$, the value of the factor "b" may be interpreted as a physiological level of content for a given metal. For example, changes of calcium content in relation to magnesium in the case of fourteen year-old children from Strumien town is described by the expression $Ca = 5.32 \cdot Mg + 636.3$. Value 636.3 can be a statistical level of calcium content in the hair of population. The range of changes of calcium content among children from Strumien town is within 310–2124 µg Ca/g of hair, whereas the geometric mean of calcium content amounts to 828 µg Ca/g of hair, and arithmetic mean of calcium content is 940 ± 113 µg Ca/g of hair.

Another illustration characterizing the population of children from Strumien town is data concerning sodium and potassium. Value for Na 124.8 g /g of hair is assumed as a physiological level of sodium. The range of changes of sodium is within 35–575 µg Na/g of hair and arithmetic means of the content are 200 and 250 µg Na/g of hair respectively.

By analyzing the values of the absolute terms = [b] and their statistics (tab. I–III), it is possible to assume that the physiological level of this group of metals is as follows: manganese in interaction with calcium shows the value 2.0 in the case of seven-year-old and 2.6 in ten-year-old children living close to the steel plant, whereas in Strumień town, the content of manganese in relation to calcium is 0.34 and 0.77 for ten- and seven-year-old children respectively. In the case of interaction with magnesium, a physiological level of manganese for ten-year-old children amounts to 0.22 µg Mn/g of hair and for Mn 1.07 µg /g of hair for seven-year-old children. The next group consists of elements whose interaction is of a more complex nature. It is assumed that it results from the changes of metal content in the air due to long-range emission. The significant relationships, for various age groups of children from Nowy Bytom town are as follows:

7 years old – manganese and sodium, manganese and potassium, magnesium and zinc, copper and iron, calcium;

10 years old – manganese and magnesium, manganese and lead, calcium and lead;

14 years old – manganese and iron, manganese and potassium manganese and potassium, iron and sodium, cadmium and cobalt;

In the case of children living 60 km from the steel plant Strumien town, the correlation is as follows:

7 old year – lead and nickel, potassium and cobalt, zinc and potassium, zinc and nickel, copper and calcium, iron and zinc, manganese and zinc;

10 old year – cadmium and nickel, cadmium and cobalt, magnesium and zinc, iron and cobalt, iron and potassium, iron and lead, iron and sodium, iron and nickel;

14 old year – lead and nickel, manganese and calcium, copper and zinc, iron and lead, magnesium and lead, zinc and cadmium.

The co-occurrence of these metals is characterized by high correlation coefficients. Therefore the interpretation of the absolute term may be similar i.e. absolute numbers present a statistical level of content of a given metal above which an orderly interaction between selected metals take place. Obtained results and their comparison, taking into consideration the age and the place of living, indicate that in the area of high chemopressure a shorter list of characteristic effects may be expected. In the case of the rural area which is 60 km away from the plant, hair can be used as a bioindicator (biomarker) of long-range emission effects of particular metals. Lower correlation between the content of one metal parallel to the changes of another metal in the hair of children living close to the plant is a result of much bigger range of changes of these metals in hair and their lower coefficient of variation. Lower variation coefficients of occurrence of 12 metals in the hair of children from Nowy Bytom town point to a high permanent environmental hazard. With the observed levels of metal content in the air and consequently in hair, a specific type of interaction between given metals is not sufficiently explained, both in their antagonistic and synergetic interaction.

Therefore, the statistical analysis and dependencies noted for children at different ages living in the industrial areas defined corresponding average environmental exposure. The above mentioned conclusions are of regional importance. Characteristic way of changes which is a result of interaction and chemopressure of particular metals is limited to the areas with similar environmental hazard. This specific way of metal co-occurrence in hair is very often observed in the case of other elements in a number of ecosystems. In the light of the above presentation it is reasonable to claim that the use of hair as a biomarker of contamination of natural environment with metals requires a pilot study for a given element. Other research projects take into the consideration the age and content of bioavailable metals in the air [22, 23, 24].

CONCLUSION

1. Hair is recommendable to use as a biomarker for a given metal. The range of application relates from the possibility of various interactions between elements and discrimination processes.
2. Only the case of 7 year old children was exceptional because the content of many biologically important metals was compatible, for example, (Mn–Ca, Mg, Na, K; Cu–Ca, Fe).
3. The region with varied chemopressure compounds of metals shows characteristic levels of content in the hair.

Diversified possibility of interaction between the metals and relation to the gender and age of children can decrease or increase metals content.

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