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Toxic and essential elements in children's blood (<6 years) from Kinshasa, DRC (the Democratic Republic of Congo)

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ABSTRACT

In this study we determined the concentration of 9 trace elements (As, Cd, Cu, Hg, Mn, Mo, Pb, Se and Zn) in whole blood of children ($n = 100$, 64 girls, 36 boys and median age: 36 months) using inductively coupled plasma mass spectrometry (ICP-MS). The proportion of children potentially deficient in essential elements or poisoned by toxic elements was evaluated. The aging effects on the concentration of these elements were also investigated. The median values were 3.17 μg/L (As), 0.15 μg/L (Cd), 1.1 mg/L (Cu), 2.1 μg/L (Hg), 10.4 μg/L (Mn), 17.7 μg/L (Mo), 8.7 μg/dL (Pb), 10.7 μg/L (Se) and 5.0 mg/L (Zn). The concentration of many elements (As, Cd, Hg, Mn, Pb and Zn) showed significant age variations but not sex influence. Regarding levels of the essential elements (Cu, Mn, Mo, Se and Zn), B-Cu, B-Mn, B-Se and B-Zn were in the normal range, whereas exceeded levels were observed for B-Mo. None of these children was deficient in essential elements. Except B-Cd, all toxic elements showed exceeded blood levels. The proportion of children potentially poisoned by toxic elements varies from 10% ($n = 10$) to 95% ($n = 95$) and depends on toxic element: 95% for As, 10% for Hg and 35% for Pb. The main health concerns emerging from this study are the high As, Hg and Pb exposures of the Kinshasan children requiring further documentation, corrective actions and the implementation of appropriate regulations.

Introduction

There is need for knowledge of trace element levels in different population groups. One application is as reference values that can be used for detection of changed environmental exposure situations. Moreover, the levels of toxic elements may be close to concentrations where adverse effects can occur. The levels of essential elements may be used to discover deficiency, and in some cases toxicity [1]. It is of importance to investigate various categories within the population and to identify specific risk groups.

Children are more likely to be exposed to environmental sources because of their parental exposures, hand-to-mouth behavior and physiology. Because of this special susceptibility, children are more vulnerable to the effects of trace elements than adults [2].

Information on the levels of many trace elements in biological tissues in children is scarce and for many non-essential elements, baseline in the general population of Kinshasa [the capital of Democratic Republic of Congo (DRC)], and especially in children, are lacking. The present study originated from that observation and its main objective was to evaluate the blood levels of nine trace elements from ≤6 years old children in the city of Kinshasa [i.e., arsenic (As), cadmium (Cd), copper (Cu), manganese (Mn), mercury (Hg), molybdenum (Mo), lead (Pb), selenium (Se) and zinc (Zn)].

Materials and methods

Study design

This survey was organized by the laboratory of environmental chemistry at the University of Kinshasa. Because Kinshasa does not have a register of population, we applied a systematic sampling approach [3] in order to obtain a representative sample of the Kinshasa population. The methods used for sampling of study population are described in detail by Tuakuila et al. [4,5]. The survey selected 100 children aged 1–5 years (91% of the target number was reached).

This study was approved by the Congolese Committee of Medical Ethics and the study results will be informed back to individuals sample donors with proper explanations.

Analysis of blood metals

Blood specimens were collected in metal-free tubes (EDTA anticoagulated) in the local health centers after careful cleaning of the

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skin at the venipuncture site. After sampling, the tubes were frozen and transported in a cool box to the Louvain centre for Toxicology and Applied Pharmacology (Brussels, Belgium), for analysis. In all blood samples, nine trace elements: total arsenic (B-As), cadmium (B-Cd), copper (B-Cu), manganese (B-Mn), mercury (B-Hg), molybdenum (B-Mo), lead (B-Pb), selenium (B-Se) and zinc (B-Zn)) were quantified by means of inductively coupled argon plasma mass spectrometry (ICP-MS) with an Agilent 7500cx ICP-MS using a Babington nebulizer, following 1:10 dilution in a basic diluent: 1-butanol (2%, w/v), EDTA (0.05%, w/v), Triton X-100 (0.05%, w/v), NH_4OH (1%, w/v), internal standards (Sc, Ge, Rh and Ir) and MilliQ water (ISO 15189 accredited method). Table 2 reports the limits of detection (LOD) for all trace elements. For quality control purposes, internal controls and reference materials were run together with the samples on a daily basis.

Statistical analysis

Data analyses were conducted with the SAS Software package version 9.2 (SAS Institute Inc., Cary, NC). For all trace elements, Geometric means (GM), arithmetic means (AM), percentiles (P) and range were calculated. The limit of detection (LOD) divided by 2 was used for imputation of values lower than the LOD. The Kolmogorov–Smirnov test was used to verify the normality of each distribution and parametric tests were used for the analysis of normally distributed variables. The Pearson correlation test was used to examine the effects of aging on trace element levels. Stepwise multiple linear regression analyses of log-transformed data were used to estimate the influence of independent variables (age and sex) on the trace element levels (Stepwise procedure, criteria: F probability to enter ≤0.05 and F probability to remove ≥0.10). A p-value lower than 0.05 was considered as statistically significant for all tests.

Results

Populations investigated

The 100 children included (64 girls, 36 boys) in this study were between 1 and 60 months of age, with a median of 36 months. The median age by sex was 36 months and 37 months for girls and boys, respectively (Table 1).

Trace element levels

The AM and GM levels of trace elements among 100 children as a whole are summarized in Table 2 together with percentiles (P) and range. The median blood (P50) values of trace elements were 3.17 μg/L (As), 0.15 μg/L (Cd), 1.1 mg/L (Cu), 2.1 μg/L (Hg), 10.4 μg/L (Mn), 17.7 μg/L (Mo), 8.7 μg/dL (Pb), 10.7 μg/dL (Se) and 5.0 mg/L (Zn). Six (6%) Cd levels were less than their LOD.

In multivariable analyses, age (continuous log-variable) was the parameter significantly associated with blood concentrations (after logarithmic conversion) of trace elements: As, Cd, and Pb ($p \leq 0.01$) and also for Hg, Mn and Zn ($p \leq 0.05$) with a limited range for R^2 (0.042–0.192) (Table 3). The analysis showed that the sex was not an influential variable for all elements.

Table 1
Demographic characteristics of the participants.

Number of subjects	100
Age, months[a]	36 [1–60]
Sex	
Boys, *n* (%)	64 (64%)
Girls, *n* (%)	36 (36%)

[a] Median age [Range].

Influence of aging on trace element levels in blood

A positive correlation was observed between age and log As ($r = 0.37$, $p \leq 0.01$), age and log Cd ($r = 0.43$, $p \leq 0.01$), age and log Hg ($r = 0.24$, $p = 0.01$), age and log Pb ($r = 0.33$, $p \leq 0.01$) and age and log Zn ($r = 0.20$, $p = 0.04$) (Table 4). A negative correlation was also observed between age and log Mn ($r = -0.23$, $p = 0.02$).

Discussion

The evaluation of essential and toxic elements in human fluids is accepted as a useful tool in both scientific research and the diagnoses of disease [6].

During the data sampling, great care was taken to select a representative sample of the Kinshasan children but the exact representativeness of our sample was not checked because Kinshasa does not have a reliable register of population. There is, however, no reason to suspect a selection bias.

In studies of trace elements, numerous errors may be introduced during the procedures from sample collection to the ultime detection of the analyte. The evaluation of data is dependent on the normal range of biological variability and on the end use of the results. According to results of the present work, the level ranges are not wide, and the levels in line of previous studies, or higher, or lower, showing that contamination was not a general problem.

Children are more susceptible to toxic exposure than adults because they have proportionally more intake of food

Table 2
Blood concentrations of selected trace elements in children from Kinshasa, DRC ($n = 100$, <6 years old).

Parameter	*N*	LOD	*N* < LOD	Range	Selected percentiles			AM (95% CI)	GM (95% CI)
					P5	P50	P95		
As (μg/L)	100	0.23	0	1.10–14.68	1.48	3.17	8.82	4.08 (3.59; 4.58)	3.52 (3.17; 3.92)
Cd (μg/L)	100	0.04	6	<LOD–1.24	<LOD	0.15	0.30	0.16 (0.13; 0.19)	0.13(0.11; 0.15)
Cu (mg/L)	100	0.6	0	0.8–2.2	0.9	1.1	1.7	1.2 (1.14; 1.23)	1.2 (1.1; 1.2)
Hg (μg/L)	100	0.1	0	0.4–10.4	0.8	2.1	6.0	2.5 (2.2; 2.9)	2.1 (1.9; 2.4)
Mn (μg/L)	100	0.5	0	4.3–64.0	5.7	10.4	22.2	12.4 (10.7; 14.1)	10.9 (10.0; 12.0)
Mo (μg/L)	100	0.1	0	1.4–36.3	3.6	17.7	30.6	16.1 (14.2; 18.0)	12.7 (10.9; 14.8)
Pb (μg/dL)	100	0.1	0	1.5–22.0	3.6	8.7	15.9	9.5 (8.8; 10.3)	8.7 (8.0; 9.5)
Se (μg/dL)	100	0.8	0	7.4–14.4	8.5	10.7	13.6	10.9 (10.6; 11.2)	10.8 (10.5; 11.1)
Zn (mg/L)	100	0.7	0	2.9–8.7	3.4	5.0	6.9	5.0 (4.8; 5.2)	4.9 (4.7; 5.1)

N = Sample size.
LOD = limit of detection.
N < LOD: number of sample below the LOD.
P5 – 5th percentile, P50 – 50th percentile = median, P95 – 95th percentile.
AM = arithmetic mean (CI = 95% confidence interval).
GM = geometric mean (CI = 95% confidence interval).