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Toxicology

# Cadmium, lead and mercury concentrations and their influence on morphological parameters in blood donors from different age groups from southern Poland



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# ABSTRACT

Due to industrial development, environmental contamination with metals increases which leads to higher human exposure via air, water and food. In order to evaluate the level of the present exposition, the concentrations of metals can be measured in such biological materials as human blood. In this study, we assessed the concentrations of cadmium (Cd), mercury (Hg) and lead (Pb) in blood samples from male blood donors from southern Poland (Europe) born in 1994 (n = 30) and between 1947 and 1955 (n = 30). Higher levels of Pb were seen in the group of older men (4.48 vs 2.48 µg/L), whereas the Hg levels were lower (1.78 vs 4.28 µg/L). Cd concentrations did not differ between age groups (0.56 µg/L). The levels of Cd and Pb in older donors were significantly correlated (Spearman R 0.5135). We also observed a positive correlation between the number of red blood cells (RBC) and Hg concentrations in the older group (Spearman R 0.4271). Additionally, we noted numerous correlations among morphological parameters. Based on our results, we can state that metals influence the blood morphology and their concentrations in blood vary among age groups.

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# Introduction

Metals and their compounds found in all living organisms play many significant roles. Nevertheless, when their exposure is excessive they may cause toxic effects. Particularly toxic are cadmium, mercury and lead which are treated as xenobiotics [1]. Cadmium can enter the human body mostly by polluted air, cigarette smoking and plants which easily absorb this metal from the environment. The highest lead intake in adults is through lungs and gastrointestinal tract, almost 100% and 20%–70%, respectively [2]. In the case of mercury, the highest exposition occurs through vapors from amalgam dental fillings (an inorganic form) and diet, based on seafood and fish which deliver high amounts of MeHg [3,4]. The abovementioned metals may cause numerous negative human health effects. The main consequence of cadmium poisoning is kidney and

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skeletal damage whereas mercury and lead influence the central nervous system which is extremely dangerous in children. Järup indicated that they may evoke behavioral disturbances, learning and concentration difficulties and diminished intellectual capacity [5].

The concentration of metals can be measured in abiotic environment parts but recently biomonitoring is widely used. The evaluation of metal content in biological fluids and tissues (e.g. urine, blood and hair) can provide information about the level of intoxication and possible adverse health effects [6]. In contrast to urine, blood has more constant composition and is thought to be more useful for assessment of recent exposure in the environment [7]. Metal blood levels can be the indicators of environmental pollution and higher risk of xenobiotics exposure [8,9].

Having entered the bloodstream, most of metals bind to morphotic elements, especially erythrocytes (RBC). Alissa and Ferns claim that red blood cells bind 98–99% of lead and 95% of methylmercury [10]. Thus, metals can probably alter blood parameters. According to Vinodhini and Narayanan, the presence of these elements in blood can result in the decrease of hematocrit level in fish blood [11]. Moreover, higher blood levels of lead disturb hemoglobin synthesis, and therefore, decrease its concentration [9,11].

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#### Table 1

Limits of detection (LoD) and quantification (LoQ) established for blood samples analyses and recoveries for certified reference materials (CRM) with relative standard deviation (RSD) between duplicates of CRM analyses.

Metal	$\text{LoD}(\mu g/L)$	$\text{LoQ}(\mu g/L)$	CRM	Recovery [%]	RSD [%]
Cd	0.12	0.28	ERMCE195	91.2	4.9
Hg <sup>a</sup>	1.22	1.66	-	-	-
Pb	2.65	7.39	ERMCE195	94.3	7.7

<sup>a</sup> Appropriate mercury CRM was not found on the market.

Mean blood concentrations of cadmium and lead increase with age [12,13]. Sex differences are also noticeable [12,14]. It has been found out that children are more vulnerable to metal exposure [7]. However, little is known about the differences in blood heavy metal levels in adults of different ages. Additionally, the influence of metals on blood morphotic elements other than red blood cells is scarcely studied.

The main aim of our work was to investigate the concentrations of highly toxic metals in the blood of male donors from southern Poland. Apart from reporting the concentrations, we analyzed the possible influence of metal presence on blood morphology (number of blood cells and their characteristics), as well as, the relationships between metals and between blood parameters.

## Materials and method

Blood samples (n = 60) used in the study were taken from male blood donors in the blood donor center (Regionalne Centrum Krwiodawstwa i Krwiolecznictwa – RCKiK) in Krakow (southern Poland, Europe) in 2013. The blood was taken from donors with the main purpose of transfusion in RCKiK. Only a part of the blood which was not used in the transfusion was used in the research (the personal data of donors were not available in the study). We took samples from donors born in 1994 (the young men group; n = 30) and donors born between 1947 and 1955 (the older men group; n = 30). The blood was collected from the ulnar vein to a few vacuette tubes (with anticoagulant EDTA) for hematological and metal analyses.

Hematological parameters as hemoglobin concentration (Hb), hematocrit level (Hct), number of red blood cells (RBC), number of white blood cells (WBC), number of thrombocytes (PLT), mean corpuscular hemoglobin (MCH), mean corpuscular hemoglobin concentration (MCHC), mean corpuscular volume (MCV) were evaluated in all samples in RCKiK with ADVIA 70 analyzer.

Mercury concentrations were measured with cold vapor atomic absorption spectrometer (MA-2, Nippon Instruments Corporation) in 200  $\mu$ L of blood. Each sample was analyzed twice and the mean value was used in further data analysis. For cadmium and lead analyses 2 mL of each sample were mineralized in hot nitric acid (Ultranal 65%, POCh) in the open mineralization system (Velp Scientifica DK20). Concentrations were measured with electrothermal atomic absorption spectrometer (AAnalyst 800, PerkinElmer) in 10 mL solutions made from mineralized samples. The validity of the whole procedure was checked against the certified reference material (CRM; Table 1). Spikes and control samples were run every 10 analyses.

#### Statistical analysis

The data distribution for all variables was checked with the Shapiro Wilk test. The homogeneity of variance was evaluated with the Levene test. Since not all variables fulfilled the assumptions of parametric analyses, nonparametric tests were used. For the comparison of differences in all variables between age groups the Mann–Whitney *U* test was carried out. The relationships among



**Fig. 1.** Median concentrations of cadmium in blood of donors (n = 60) from two age groups. The differences were not statistically significant (Mann–Whitney *U* test p = 0.2804).

metals and between metals and hematological parameters were evaluated with Spearman R correlation factors. Significance level for difference test and correlation factor was established at 0.05. Excel 2010 PL (Microsoft) and Statistica 10 En (StatSoft) were used in all the analyses.

# Results

Median concentrations of Hg (p < 0.0001) and Pb (p = 0.0013) in blood varied statistically between age groups (Figs. 1–3). Among the three metals, the highest median concentration was noted for lead in the older men group ( $4.48 \mu g/L$ ), the lowest for cadmium in the young men group ( $0.48 \mu g/L$ ). The widest range occurred in the case of lead in the first age group ( $0.60-45.55 \mu g/L$ ). Higher levels of cadmium and lead were seen in the group of older men, whereas mercury level in this group was lower than in the young one. Statistically significant relationship occurred between Pb and Cd in the older men group. Other possible correlations were not statistically significant (Table 2).



**Fig. 2.** Median concentrations of mercury in blood of donors (n = 60) from two age groups. Differences were statistically significant (Mann–Whitney U test p < 0.0001).

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