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**Characteristic Features of Toxic Metal Content in Hair Samples of Foreign Students at
RUDN University from different geographic regions**

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Abstract

1 *Background:* This study explores the regional variations in toxic metal accumulation among
2 RUDN University students from various global regions.
3

4 *Methods:* This comparative analysis examined hair samples from students hailing from different
5 regions, including Russia, Asia, the Middle East, Africa, and Latin America. The concentrations
6 of Aluminium (Al), Arsenic (As), Cadmium (Cd), Mercury (Hg), Lead (Pb), and Tin (Sn) were
7 measured in the hair samples. The data was then evaluated using regression models to assess the
8 link between the region of residence and toxic metal content in the hair.
9

10 *Results:* The analysis indicated significant regional variations in the levels of toxic metals in the
11 students' hair. The highest content of Al, Cd, and Pb was observed in students from Africa (13.542,
12 0.028, 0.794 $\mu\text{g/g}$) and Latin America (9.947, 0.025, 0.435 $\mu\text{g/g}$). Arsenic levels in students from
13 all regions exceeded that of Russian students by over two-fold. No substantial group differences
14 were found in the Sn content. The regression models suggested that residing in Asia, Africa, and
15 Latin America was a predictor of high Hg levels in hair (0.130, 0.096, 0.227 $\mu\text{g/g}$). Living in Africa
16 was significantly associated with higher Pb levels (0.794 $\mu\text{g/g}$), and living in Latin America was
17 close to significantly associated with the Cd level in the hair (0.025 $\mu\text{g/g}$).
18

19 *Conclusion:* This study confirmed an increased accumulation of toxic metals, especially Hg, Cd,
20 and Pb, in students primarily from Latin America and Africa. The findings highlighted the
21 importance of understanding the regional variations in toxic metal accumulation to address
22 associated health risks and the potential impact on students' well-being and academic performance.
23 These insights may guide the development of targeted interventions to reduce exposure to toxic
24 metals in students from various regions around the world.
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1. Introduction

The intensive development of mining and heavy industry, road and air transport, the burning of coal and other materials over the past century has inevitably led to environmental pollution with toxic metals [1-7]. Toxic metals, including mercury (Hg), lead (Pb), cadmium (Cd), aluminum (Al), arsenic (As), and tin (Sn), have been widely recognized as harmful substances that can accumulate in the human body and cause a variety of health issues, such as neurotoxicity, nephrotoxicity, and cardiovascular diseases [8-19]. In many instances, exposure to toxic metals results in severe behavioral disorders, decreased intelligence, and learning ability [20-22]. The accumulation of these toxic elements in humans varies significantly across different regions of the world, which is primarily due to the differences in environmental pollution levels and exposure pathways in these areas [23, 24].

To assess the levels of toxic elements in the human body, various analytical methods involving urine, blood, and hair samples have been employed [25, 26]. Among these methods, hair analysis offers several advantages. Hair samples are non-invasive, easier to collect, and require minimal processing and storage conditions compared to blood and urine samples [27]. Moreover, while blood analysis provides information on the current state of toxic element exposure, hair analysis reveals long-term accumulation patterns [28-34].

Understanding the long-term accumulation of toxic metals in the human body is essential for several reasons. Firstly, it allows for a more comprehensive evaluation of chronic health effects that might arise from prolonged exposure to these elements [35, 36]. Secondly, this information can inform the development of targeted interventions aimed at reducing exposure to toxic metals and mitigating their associated health risks [37, 38]. Lastly, identifying regional variations in toxic metal accumulation can help address the potential impact of these elements on the health and academic performance of foreign students, who might be more vulnerable to the adverse effects

of toxic metals due to their unique stressors and adaptation challenges [39, 40]. To the best of our knowledge, there are no data on the study of the content of toxic metals: Aluminum (Al), Arsenic (As), Cadmium (Cd), Mercury (Hg), Lead (Pb), and Tin (Sn) in the hair of students from different climatic and geographical regions of the world (Asia, Africa, the Near and Middle East, Latin America) who arrived at the same time in the new identical environmental conditions of the Moscow metropolis. There are only fragmentary studies on the content of individual elements in the literature.

In this study we examined the levels of toxic metals in hair samples of first-year students of RUDN University from various regions, such as Russia, Asia, the Middle East, Africa, and Latin America [41, 42]. The study underscores the importance of comprehending regional differences in toxic metal accumulation and implementing appropriate measures to safeguard the well-being and academic success of foreign students [26, 43].

2. Material and methods

This study was carried out in accordance with the Declaration of Helsinki of the World Medical Association (1964) and its subsequent amendments. All subjects took part in the study voluntarily on the principle of informed consent. The study involved 310 students (40% men, 60% women; mean age 22.7 ± 7.1 years). All subjects were first-year students of the RUDN University. The research involved students from Russia (Moscow and the Moscow region), Southeast Asia (Bangladesh, Vietnam, China, Cambodia, Nepal, Laos, Malaysia, Myanmar, Thailand), Near and Middle East (Iraq, Iran, Yemen, Lebanon, Syria, Palestine, Jordan, Turkey), Africa (Guinea-Bissau, Guinea, Equatorial Guinea, Congo, Ghana, Angola, Zambia, Zimbabwe, Cameroon, Kenya, Ivory Coast, Tanzania, Chad, Benin, Burkina Faso, Rwanda, Sierra Leone) and Latin America (Bolivia, Brazil, Venezuela, Colombia, Mexico, Panama, Peru, Ecuador). The composition of the student cohort by region when entering at RUDN university was as follows: Russia (n = 60), Asia (n = 63), Near and Middle East (n = 81), Africa (n = 55) and Latin America (n = 51). The exclusion criteria were the presence of somatic diseases, as well as specific diets

1 (vegetarianism, raw food diet) and smoking. Immediately after arrival, the subjects were taken hair
2 samples, which is an informative bioindicator substrate in assessing the long-term exposure to
3 toxic metals.
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6 Hair samples were taken from the occipital region using stainless steel scissors pre-treated
7 with ethanol (RUDN University, Moscow, Russia). Only the proximal section of the hair 1–2 cm
8 long was used for analysis (this part of the hair corresponds to the recent time period of the
9 students' life in their region of origin). Subsequently, the proximal parts of the hair strands were
10 washed with acetone, followed by washing three times with distilled deionized water and drying
11 in air. Next, the samples were ground in the presence of 62% nitric acid and placed in a Berghof
12 SpeedWave-4 DAP-40 system (Berghof Products + Instruments GmbH, Germany) for microwave
13 decomposition. Determination of the content of aluminum (Al), arsenic (As), cadmium (Cd),
14 mercury (Hg), lead (Pb) and tin (Sn) in the hair of first-year students was carried out by mass
15 spectrometry with inductively coupled argon (Ar) plasma on a NexION 300D spectrometer
16 (PerkinElmer Inc., USA). The system was calibrated using Universal Data Acquisition Standards
17 Kit (PerkinElmer Inc., USA). Yttrium and rhodium solutions (10 ppb) were used for internal online
18 standardization (Pure Single-Element Standard, PerkinElmer Inc. USA). GBW09101 hair
19 (Shanghai Institute of Nuclear Research, Shanghai, China) was used as a certified reference
20 material of human hair.
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43 Data processing was performed using the Statistica 10.0 software package (Statsoft, OK,
44 USA). Due to the absence of a Gaussian distribution of data on the content of metals in hair, the
45 median and the corresponding boundaries of the 25–75 centile interval were used as descriptive
46 statistics. The significance of group differences at $p < 0.05$ was assessed using the nonparametric
47 Mann–Whitney U test. For a comparative analysis of the influence of regions (independent
48 predictors) on the level of toxic metals in the hair (dependent variable), we performed a multiple
49 regression analysis adjusted for gender and age of the participants.
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61 **3. Results and discussion**

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3.1. Analytical figure of merits

Certified reference material of human hair GBW09101 was used for laboratory quality control (**Table 1**). The recovery rates for all studied toxic chemical elements were within the range of 90–110%.

Table 1. Laboratory quality control with certified reference material of human hair GBW09101.

| Region | Certified Value, $\mu\text{g/g}$ | Obtained Value, $\mu\text{g/g}$ | Recovery Rate, % | LoD, ppb | BEC, ppb |
|--------|----------------------------------|---------------------------------|------------------|----------|----------|
| Al | 23.2 | 23.3 ± 2.48 | 101 | 0.13 | 0.26 |
| As | 0.198 | 0.194 ± 0.014 | 97 | 0.001 | 0.001 |
| Cd | 0.072 | 0.066 ± 0.006 | 92 | 0.0016 | 0.0006 |
| Hg | 1.06 | 1.15 ± 0.29 | 108 | 0.012 | 0.034 |
| Pb | 3.83 | 4.13 ± 0.28 | 108 | 0.0006 | 0.0007 |
| Sn | n.s. | 0.32 ± 0.02 | - | n.s. | n.s. |

LoD – limit of detection; BEC – background equivalent concentration; ppb – part per billion; n.s. – not specified.

3.2. Hair analysis for toxic metals

The results of the study indicate significant differences in the content of toxic metals in hair samples of first-year students from different regions. For example, the aluminum content (**Table 2**) in hair of African students is 2.4 times higher than that of Russian students, 2.9 times higher than that of Asian students, and 2.5 times higher than that of students from countries of the Nearest and Middle East (NME). Hair samples from students of Latin America also showed higher levels of aluminum compared to students from other countries. Hair samples from Latin American students contain 78% more aluminum than Russian students, 114% more than Asian students, and 88% more than students from the NME region. At the same time, the content of aluminum in the hair of African and Latin American students does not differ significantly.

The content of other elements (arsenic, cadmium, mercury, lead, and tin) in hair samples of first-year students is also characterized by pronounced regional differences (**Tables 2**). For example, the lowest levels of arsenic are observed among Russian students. At the same time, the lowest content of cadmium is typical for representatives of Asian and NME countries, being 2 times lower than the control values. The highest levels of cadmium are found in hair samples from students from Africa and Latin America. The content of cadmium in African students is 4.6 times higher than in Asian students and 5.6 times higher than in NME students. A similar situation is observed in the case of Latin American students (cadmium levels are 4.1 times higher than those for Asian students, and 5 times higher than for students from NME countries). Despite two-fold differences, the level of cadmium in the hair of Russian students is quite high and does not significantly differ from that of African and Latin American students. The content of mercury in hair of students from Latin America exceeds the level of mercury of Russian, Asian and NME students by 118, 75 and 136%, respectively (**Table 2**). The maximum lead content was registered in Africans: their level is higher in comparison with Russian (by 2.7 times), Asian (by 4.5 times) and NME (by 3.8 times) first-year students. Unlike other metals, the content of tin in students' hair is not characterized by significant differences. However, it should be noted that there is a slight trend towards an increase in the level of tin in hair of Latin American and African students.

Table 2. Levels of aluminum, arsenic, cadmium, mercury, lead, and tin in hair ($\mu\text{g/g}$) of foreign students and students from Russia.

| Region | Al | As | Cd | Hg | Pb | Sn |
|--------|---------------------------|----------------------------|----------------------------|----------------------------|----------------------------|---------------------------|
| Russia | 5.59 (3.614÷ 8.443) | 0.015 (0.009÷ 0.020) | 0.014 (0.006÷ 0.022) | 0.104 (0.064÷ 0.164) | 0.290 (0.128÷ 0.532) | 0.177 (0.06÷ 0.407) |
| Asia | 4.632 (3.544÷ | 0.037 (0.023÷ | 0.006 (0.004÷ | 0.130 (0.064÷ | 0.173 (0.122÷ | 0.069 (0.049÷0.2) |

| | | | | | | |
|---------|--------------------------|---------------------|-----------------------|-------------------------|-------------------------|---------|
| | 6.663) | 0.052) ¹ | 0.012) ¹ | 0.172) | 0.465) | |
| NME | 5.297 | 0.030 | 0.005 | 0.078 | 0.207 | 0.046 |
| | (3.733÷ | (0.019÷ | (0.003÷ | (0.043÷ | (0.122÷ | (0.025÷ |
| | 7.265) | 0.049) ¹ | 0.015) ¹ | 0.158) | 0.38) | 0.093) |
| Africa | 13.542 | 0.039 | 0.028 | 0.096 | 0.794 | 0.433 |
| | (8.145÷ | (0.033÷ | (0.006÷ | (0.068÷ | (0.28÷ | (0.16÷ |
| | 18.215) ^{1,2,3} | 0.057) ¹ | 0.055) ^{2,3} | 0.143) ³ | 1.826) ^{1,2,3} | 0.704) |
| Latin | 9.947 | 0.038 | 0.025 | 0.227 | 0.435 | 0.443 |
| America | (5.755÷ | (0.026÷ | (0.004÷ | (0.135÷ | (0.113÷ | (0.088÷ |
| | 12.059) ^{1,2,3} | 0.055) ¹ | 0.097) ^{2,3} | 0.456) ^{1,2,3} | 1.582) | 0.898) |

The data are presented as median and 25–75 centile interval; ^{1,2,3} – reliability of differences at $p < 0.05$ relative to the regions: 1 – Russia, 2 – Asia, 3 – NME countries.

We performed multiple linear regression analysis to further analyze the relationship between the region of origin of first-year students and the level of metals in their hair (**Table 3**). We found that geographic differences have the strongest effect on mercury and lead levels in hair, and this is broadly consistent with group comparison results. In particular, living in Asia, Africa, and Latin America is a predictor of high mercury levels in hair. Living in Africa is also significantly associated with higher levels of lead. At the same time, male sex is a negative predictor of the level of metal content in the hair. In the case of cadmium, the direct relationship between residence in Latin America and the level of the metal in hair is close to significant. At the same time, age has a significant effect on the content of cadmium in hair. Between living in different regions and the content of aluminum, arsenic, and tin in hair, we did not reveal any significant relationships using the regression models.

Table 3. Regression analysis of the relationship between the level of metals in hair samples of first-year students and the region of their residence.

| Predictor | Al | As | Cd | Hg | Pb | Sn |
|---------------|--------|-------|-----------------------|------------------------|------------------------|--------|
| Male sex | -0.072 | 0.118 | -0.056 | -0.224 ($p = 0.001$) | -0.138 ($p = 0.042$) | -0.039 |
| Age, years | -0.001 | 0.038 | 0.322 ($p < 0.001$) | -0.030 | -0.027 | -0.006 |
| Russia | 0.005 | 0.097 | 0.078 | 0.153 | 0.023 | 0.190 |
| Asia | 0.016 | 0.220 | 0.074 | 0.258 ($p = 0.043$) | 0.085 | 0.053 |
| NME | 0.118 | 0.207 | -0.007 | 0.234 | 0.103 | 0.153 |
| Africa | 0.151 | 0.182 | 0.099 | 0.324 ($p = 0.004$) | 0.295 ($p = 0.011$) | 0.024 |
| Latin America | 0.078 | 0.106 | 0.190 ($p = 0.058$) | 0.373 ($p < 0.001$) | 0.112 | 0.054 |

The data are presented as regression coefficients (β), for significant relationships, the corresponding p values are indicated.

Thus, the survey of first-year students at RUDN University shows that foreign students are characterized by higher rates of accumulation of toxic metals in their hair. In particular, the maximum levels of mercury content are observed in students who arrived from Latin America, and the maximum level of lead is typical for African students. Differences in the content of arsenic and cadmium in hair samples of students from different regions are less pronounced. The revealed differences in the patterns of metal accumulation in the body of subjects from different regions most likely reflect characteristic features of environmental pollution by metals in the mentioned countries [1]. High levels of lead in African students are consistent with the ubiquity of lead sources in Africa, while mercury and arsenic emissions are more localized [44]. Hrubá et al. have recently shown that Moroccan blood lead levels are twice as high as those in European countries, China, and Ecuador [38]. This is also consistent with the data obtained in the Ngueta group that children living near the Sahara are characterized by a significant excess of blood lead levels compared to the United States [45]. Lead sources in Africa are primarily gold [46] and lead-zinc [44] mines. The direct routes of lead entry into the human body can be associated with water, soil, grain, and dust pollution [47]. Environmental pollution with cadmium is also an important problem in a number of African countries, including Nigeria [48]. As for arsenic, emissions of this toxicant are not a widespread problem in Africa, and only in some cases, gold mining operations are

1 associated with arsenic emissions into the environment and its subsequent negative impact on
2 human health [49].
3

4 Despite the leading role of Asian countries (primarily China) in the release of mercury into
5 the atmosphere, the maximum content of mercury in hair is characteristic of students from Latin
6 America. This observation is consistent with previously found by Hrubá et al. high concentrations
7 of mercury and lead in Ecuador [38]. The leading role of mercury as an inorganic pollutant in the
8 Latin American region may be due to the prevalence of artisanal gold mining [50]. For example,
9 Streets et al. state that the direct contribution of Latin America to the total release of mercury into
10 the atmosphere is 10% [51]. Along with mercury, environmental pollution with arsenic is also an
11 urgent medical and social problem in the Latin American region, and it is associated with high
12 incidence of pathology caused by arsenic exposure [52].
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27 Living in Asia is also a significant predictor of metal accumulation in the body, associated
28 with increased concentrations of mercury and cadmium. This observation is consistent with the
29 recent work of Dix-Cooper and Kosatsky, who reported higher concentrations of mercury, lead
30 and cadmium in whole blood in women that arrived from Asia in Canada compared with the
31 Canadian population [26]. Some of us also recently conducted a survey of residents of the Republic
32 of Bangladesh and found higher levels of mercury and arsenic in hair compared to Russian ones
33 [53]. Moreover, the reported concentrations of toxic metals in Asian first-year students are in close
34 agreement with those of Guangzhou college students published by Zhang et al. [54]. Despite
35 significant preventive activities, metal pollution remains a significant problem in Central and
36 Southeast Asia [55]. The risk of excessive exposure to mercury also remains for residents of the
37 Near and Middle East, and this is consistent with the observed annual increase in mercury
38 emissions in the region during the period from 2000 to 2015 [51]. The development of the
39 metalworking industry in the Middle East also makes a significant contribution to environmental
40 pollution with cadmium and lead [56]. Considering the identified stress reactions in first-year
41 foreign students of RUDN University [57], excessive accumulation of metals due to environmental
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1 pollution of the regions from which these students came might significantly reduce the adaptive
2 reactions of the students through the implementation of a wide range of toxicity mechanisms [58,
3 59]. This negative effect, in turn, might contribute to an increase in illness and a decrease in the
4 effectiveness of teaching foreign students and should be the subject of future studies.
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9 Our findings revealed increased accumulation of different toxic metals in students hailing
10 from different geographic regions. It seems natural that the detected increased accumulation
11 mirrors the specific features of environmental pollution with metals in these countries, which can
12 result from factors such as industrial activities, mining, and inadequate waste management
13 practices [60, 61].
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21 **4. Conclusion**

22 The results of this work can be considered from the following few perspectives.
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28 Firstly, we found an increased accumulation of toxic metals, especially mercury, lead,
29 cadmium, in the body of first-year students, who arrived primarily from Africa and Latin America.
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34 Secondly, we confirmed that the differences in the patterns of metal accumulation in the
35 students from different regions most likely reflect the specific features of environmental pollution
36 with metals in these countries. Probably, this increased accumulation of toxic metals might have a
37 negative impact on the health status and effectiveness of teaching foreign students. Therefore,
38 further research is needed, focused on assessing the direct contribution of excess toxic metals to
39 the health indicators of RUDN University students, and this project is underway in our group.
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55 conditions”).
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