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Lead in human teeth dentine as a bio-indicator of environmental exposure to lead

Ołów w zębinie ludzkich zębów jako bioindykator narażenia środowiska na ołów

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Abstract

Introduction. Health hazards caused by environmental exposure to lead is of concern in many places in the world. Lead is particularly harmful to young children who could be also exposed to lead *in-utero* through exposure of their mothers before pregnancy. There is no level of exposure to lead that is known to be without harmful effects to the organism. Identifying local sources of lead in the environment is a very important part of preventing exposure as there is no cure for lead poisoning. The usefulness of children's primary teeth and the permanent teeth of adults as an indicator of chronic environmental exposure to lead in polluted area have been studied.

Objective. The aim of the study was to assess the usefulness of teeth as an indicator of chronic environmental exposure to lead of children and adults in the Silesia Province (Poland), as a particularly polluted area.

Materials and method. A total of 71 primary teeth from children and 85 permanent teeth from adults were analyzed for lead content. The material was collected as a result of planned tooth extractions for medical reasons.

Results. The primary teeth were characterized by significantly higher concentrations of lead than the teeth collected from adults. The amount of lead in the teeth of adults varied depending on gender, but did not differ much from the place of residence. The lead content in the teeth was higher in the group of younger than older children, which may be an indication of prenatal exposure.

Conclusions. Children's primary teeth are more useful bio-indicator than adult teeth for determining environmental exposure to lead, and may also indicate the cumulative exposure to lead of their mothers.

Key words

lead exposure, environment, primary teeth, bio-indicator, permanent teeth

Streszczenie

Wprowadzenie. Zagrożenia zdrowia spowodowane środowiskowym narażeniem na ołów dotyczą wielu miejsc na świecie. Ołów jest szczególnie szkodliwy dla małych dzieci, które mogą być również narażone na kontakt z tym pierwiastkiem już w okresie prenatalnym w efekcie narażenia ich matek przed ciążą na ten metal. Nie istnieje taki poziom ekspozycji na ołów, o którym wiadomo, że nie ma szkodliwego wpływu na organizm. Identyfikacja lokalnych źródeł ołowiu w środowisku jest bardzo ważnym elementem zapobiegania narażeniu, ponieważ nie ma lekarstwa na zatrucie ołowiem.

Cel pracy. Celem pracy była ocena przydatności zębów jako wskaźnika przewlekłego środowiskowego narażenia na ołów dzieci i dorosłych w województwie śląskim, będącym obszarem szczególnie zanieczyszczonym.

Materiał i metody. W sumie przeanalizowano 71 zębów mlecznych od dzieci i 85 zębów stałych od dorosłych. Materiał został pobrany w wyniku planowanych ekstrakcji zębów ze względów medycznych.

Wyniki. Zęby mleczne charakteryzują się znacznie wyższą zawartością ołowiu niż zęby pobrane od osób dorosłych. Ilość ołowiu w zębach dorosłych jest różna w zależności od płci, ale nie różni się zbytnio od miejsca zamieszkania. Zawartość ołowiu w zębach jest wyższa w grupie dzieci młodszych niż starszych, co może świadczyć o ekspozycji prenatalnej.

Wnioski. Zęby mleczne dzieci są bardziej użytecznym wskaźnikiem biologicznym środowiskowego narażenia na ołów niż zęby osób dorosłych, mogą także wskazywać na skumulowaną ekspozycję matek na ołów.

Słowa kluczowe

zęby mleczne, środowisko, bioindykator, zęby stałe, narażenie na ołów

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INTRODUCTION

Environmental pollution with heavy metals is a major problem for many countries in the world. The most common heavy metal in the human environment is lead which can be found throughout the whole environment where people live [1]. This toxic heavy metal is found in the food people eat, the air people breathe, the water they drink and in the soil in which edible plants are cultivated on [2]. Additionally, children are exposed to lead when they walk and crawl on contaminated soil, and even when they play with some of their toys [3]. Lead enters the body through inhalation, ingestion and through dermal contact. In many cases, adults and especially children are exposed to lead from more than one source, making the risk assessment very difficult [4–7]. Once ingested, lead is absorbed through the digestive tract and distributed throughout the body via the bloodstream [8].

Lead is a cumulative toxic substance that primarily affects the liver and kidneys. Once in the body, lead also affects multiple body systems, such as the central nervous system and the endocrine and reproductive systems. There is no level of exposure to lead that is known to be without harmful effects to the organism. Lead is particularly harmful to young children [7, 9] who are also exposed to lead *in utero* through exposure of their mothers, with adverse impacts on neurobehavioral development. Exposure to lead both before and during pregnancy can also be extremely harmful. Mother's exposure to lead present in the environment before pregnancy could result in the later release of the lead stored in her bones during pregnancy. It increases blood lead levels and poses risks to both the mother and unborn child. It has been shown to cause bleeding, miscarriage, premature birth and low birth weight, as well as malformations. Usually, the amount of lead in a mother's blood is similar to the levels found in the foetus. Lead exposure during pregnancy could affect the baby's growth, as well as their future ability to see, hear and learn [10].

There is no cure for lead poisoning. Once lead has been in the body for a prolonged period of time, it is very difficult to remove as it is deposited in the bones, prior to which it causes a great deal of neurodevelopmental damage [11]. In 2019, the Institute of Health Metrics and Evaluation (IHME) estimated that lead exposure accounted for over 902,000 deaths and 21.7 million years of healthy life lost (measured in Disability Adjusted Life Years – DALYs) worldwide due to the long-term effects of lead exposure on health [12].

Identifying local sources of lead in the environment is a very important part of preventing exposure because of the lack of a cure for lead poisoning. Based on numerous studies, it can be concluded that recent exposure to lead can be measured in blood samples, while cumulative exposure can be measured in teeth or bones [13–15]. Health hazards caused by environmental exposure to lead concern many places in the world, including Poland, where the highest concentrations of heavy metals are found in the southern part of the country. This applies in particular to the Silesia Province, due to many years of heavy industry activity in this area, related to the extraction and processing of non-ferrous metal ores [16].

Children are a group particularly vulnerable to lead as they are more exposed to pollutants in their environment than adult. This is connected with the fact that they do not have fully developed respiratory and immune systems. Moreover, they spend relatively more time outdoors than adults, and are characterized – especially young children – by specific behaviours that increase the risk of exposure to lead, such as putting dirty hands or objects in the mouth [6, 17].

Due to the toxicity of lead and its ability to accumulate in organisms, it is necessary to assess the environmental exposure of people living in contaminated areas to this metal. The methods used to assess the risk include heavy metals accumulation monitoring in the organisms of inhabitants of areas contaminated with these elements (based on the analysis of their content in hair, nails, blood, urine, bones and teeth) [18–21]. However, not all methods are appropriate for the assessment of chronic human environmental exposure to heavy metals. Although blood and urine are often used as an indicator of human exposure to lead, the half-life of lead in blood or urine is relatively short (approx. 28–30 days), and therefore they are not reliable indicators of long-term exposure [22]. In the case of hair and nails, the half-life of lead is longer; however, due to contaminants entering the hair and nails during cosmetic procedures (hair dyeing, hair and nails varnishing, masks applying, etc.) they are not good biological indicators [23, 24]. The best bio-indicators of environmental exposure to heavy metals, including lead, especially in the case of chronic exposure starting from early development, are bones and the teeth, both of which have a high affinity for lead. However, human bone is a material that is difficult to access, and the possible sampling for research is a very invasive procedure. Teeth, however, are a material very similar to bone tissue and are considered part of the human skeleton, and are easily accessible material in a minimally invasive manner [25–27]. The accumulation of heavy metals in the teeth begins already in the period of tooth bud formation. Studies have shown that the content of lead in the teeth does not change for a long time, and the measurements of its concentration in the teeth of the inhabitants of polluted areas and the population not exposed to toxic environmental factors confirmed that teeth may be an appropriate research material to determine the level of environmental exposure to lead [18, 28]. Studies of human teeth of different races, genders and ages have shown that human dentin can provide chronological information on exposure, and are a reliable bio-indicator of environmental pollution [26].

OBJECTIVE

The aim of the study was to assess the usefulness of teeth as an indicator of chronic environmental exposure to lead of children and adults in the Silesia Province (Poland), as a particularly polluted area. When assessing the usefulness of teeth as an exposure indicator, it is necessary to examine the amount of lead accumulation in the teeth of children and adults, depending on age, gender and place of residence.

MATERIALS AND METHOD

The research material consisted of children's primary teeth and permanent teeth obtained from adults. Primary teeth were removed by root resorption with further tooth exfoliation. Permanent teeth were removed for prophylactic, orthodontic or surgical indications. The material was collected at the Academic Centre of Dentistry and Specialized Medicine in Bytom, Upper Silesia, Poland, after obtaining consent from the parents/legal guardians of the children, and directly from the adult patients. This study protocol was approved by the Ethical Review Board of Medical University of Silesia in Katowice (Approval No. KNW/0022/KB/84/1/2015).

The lead content in the teeth was analyzed in the Analytical Laboratory at the Department of Environmental Health, Faculty of Health Sciences in Bytom, Medical University of Silesia in Katowice (Poland). All teeth were thoroughly cleaned and washed with redistilled water, then dried and ground in a vibratory grinder. The mass obtained from the entire tooth sample was weighed, placed into a Teflon vessel, and 10 ml of spectrally pure concentrated nitric acid V (Merck, Germany) added, followed by pressure digestion in a Magnum II microwave mineralizer (Ertec, Poland). The mineralization process took 15 minutes, and the parameters were as follows: 100% power, pressure 45–42 bar and temperature 295–300°C. After completion of the mineralization process, the test material was transferred to a disposable 50 ml volumetric flask, and the missing amount was supplemented with ultrapure water. The lead (Pb) content in the samples was determined by atomic absorption spectrophotometer (AAS) Savant AA Sigma (GBC, Australia).

Statistical analysis. The data were statistically analyzed using Statistica 13 software (StatSoft, Poland). The collected data were evaluated for normal distribution by the Shapiro-Wilk test with a confidence level of 95%. The linear regression model, using the non-parametric independence R-Spearman test, was used to investigate the relationship between age, place of residence and the concentration of lead in the teeth. All analysis were carried out using a threshold of statistical significance set at a value of $p \leq .05$.

RESULTS

All samples of primary teeth analyzed in the study came from children aged 2–14 years who lived in Bytom (52 samples: 33 from girls and 19 from boys); the samples of permanent teeth were from adults aged 18–40 years (85 samples: 45 from women and 40 from men), residents of two neighboring cities: Bytom (43 samples) and Zabrze (42 samples). The results of the chemical analysis of the tested tooth samples are presented in Tables 1–4, together with descriptive statistics (minimum, maximum, mean, median, standard deviation).

The determined concentrations of lead in the permanent teeth ranged from below or equal to the limit of quantification (≤ LOQ) to 3.64 μg/g. The average lead concentration obtained for permanent teeth (from the two cities) was 0.49 μg/g. The average concentration of Pb in the teeth of women and men from Bytom was at the same level (0.49 μg/g); the highest concentration of Pb in the female population was recorded in the tooth of a 40-year-old resident of Bytom (1.41 μg/g), and in the case of men it was a tooth taken from a 24-year-old resident of that city (2.37 μg/g). In the case of Zabrze residents, the highest concentration of Pb among women was found in the tooth of an 18-year-old (1.60 μg/g), and among men – in a 51-year-old resident (3.64 μg/g); the mean concentration for women was 0.41 μg/g and for men 0.63 μg/g. In the case of Bytom, 37% of the samples exceeded the average lead content obtained for this city, and 12% in the case of samples from Zabrze. The dispersion of the concentration values was similar in all cases, and the median ranged from 0.33–0.37 μg/g (Tab. 1).

Table 1. Characteristics of lead content [μg/g] in the teeth of the inhabitants of Bytom and Zabrze, Silesian Province, Poland

	Gender	No. of samples	Min.	Max.	Median	Mean ± SD
Adults (residents of Bytom)	F	20	≤ LOQ	1.41	0.37	0.49 ± 0.37
	M	23	≤ LOQ	2.37	0.33	0.49 ± 0.48
Adults (residents of Zabrze)	F	25	≤ LOQ	1.60	0.34	0.41 ± 0.35
	M	17	≤ LOQ	3.64	0.34	0.63 ± 0.84

*SD – standard deviation; **F – female; M – male; ***LOQ – limit of quantification (0.05 μg/g)

The mean concentration of lead in the primary teeth was 1.24 μg/g; 1.35 in the case of girls, 1.05 in the case of boys. Standard deviations 1.51 and 1.31, respectively, and the median was 0.71 (girls) and 0.68 (boys). The range of Pb concentrations in deciduous teeth was ≤ LOQ – 6.34 μg/g. The average concentration of lead in girls' teeth was therefore about 23% higher than in boys' teeth. Regardless of gender, the range of lead concentrations in primary teeth was very large and more than 1/3 of the samples had a concentration exceeding the average Pb content (>1.24 μg/g – 37% of samples). The most contaminated sample, in which Pb was determined at the level of 6.34 μg/g, exceeded the average content of this element in the analyzed samples up to five times. Such a high concentration of Pb was determined in the tooth of a 6-year-old girl. An equally high concentration of this element was found in the primary tooth of a 7-year-old girl (5.49 μg/g) and boy (5.34 μg/g) (Tab. 2, 3).

By comparing the mean concentrations of lead determined in permanent and deciduous teeth, higher values were

Table 2. Pb content in teeth of female children and adults living in Bytom, Silesian Province, Poland

No.	Group	Age [years]	No. of samples (n)	Concentration range [μg/g]	Mean ± SD [μg/g]	Median [μg/g]
1	children	2	1	1.28	-	-
2		3	2	1.24 - 1.31	1.28 ± 0.04	1.28
3		4	2	0.51 - 0.63	0.57 ± 0.08	0.57
4		5	4	0.46 - 1.61	1.04 ± 0.56	1.04
5		6	4	≤ LOQ - 6.34	1.88 ± 2.98	0.57
6		7	4	≤ LOQ - 5.49	2.48 ± 2.65	1.76
7		8	3	1.43 - 3.57	2.25 ± 1.15	1.76
8		9	5	≤ LOQ - 2.46	1.02 ± 0.91	0.71
9		10	4	0.28 - 2.64	1.09 ± 1.09	0.72
10		11	1	0.55	-	-
11		12	2	0.31 - 0.87	0.59 ± 0.39	0.59
12		14	1	0.09	-	-
13	adults	18	1	0.80	-	-
14		19	2	0.36 - 1.17	0.77 ± 0.57	0.77
15		20	3	≤ LOQ - 0.21	0.10 ± 0.09	0.05
16		22	4	0.24 - 0.79	0.41 ± 0.25	0.31
17		23	1	0.77	-	-
18		24	3	0.37 - 0.56	0.45 ± 0.09	0.43
19		25	4	0.14 - 0.86	0.36 ± 0.33	0.22
20		28	1	0.48	-	-
21		40	1	1.41	-	-

*SD – standard deviation; **LOQ – limit of quantification (0.05 μg/g)

Table 3. Pb content in teeth of male children and adults living in Bytom, Silesian Province, Poland

No.	Group	Age [years]	No. of samples (n)	Concentration range	Mean ± SD	Median
				[µg/g]		
1		3	2	≤ LOQ - 2.19	1.12 ± 1.51	1.12
2		5	1	≤ LOQ		
3		6	5	≤ LOQ - 1.06	0.32 ± 0.44	0.05
4	Children	7	1	5.34	-	-
5		8	1	2.55	-	-
6		9	4	0.24 - 1.92	1.15 ± 0.78	1.22
7		10	3	0.47 - 0.81	0.65 ± 0.17	0.68
8		12	2	0.15 -1.41	0.78 ± 0.89	0.78
9		18	2	0.20 - 0.78	0.49 ± 0.41	0.49
10		20	5	0.16 - 0.38	0.28 ± 0.07	0.29
11		21	1	≤ LOQ		
12		22	1	0.24	-	-
13		24	3	0.21 - 2.37	1.05 ± 1.15	0.57
14	Adults	25	3	0.19 - 0.76	0.43 ± 0.29	0.33
15		26	2	0.27 - 0.85	0.56 ± 0.41	0.56
16		27	2	0.55 - 0.87	0.71 ± 0.22	0.71
17		28	2	0.70 - 0.35	0.53 ± 0.24	0.53
18		30	1	≤ LOQ		
19		33	1	0.58	-	-

*SD – standard deviation; **LOQ – limit of quantification (0,05 µg/g)

obtained in the teeth of children than in the teeth of adult residents of Bytom. The concentration of 0.49 µg/g, which was the mean value for the adult population, was exceeded in as many as 63% of the children's tooth samples (Tab. 4).

Table 4. Characteristics of the lead content [µg/g] in the teeth of the inhabitants of Bytom (Silesian Province, Poland)

	Gender	No. of samples	Min.	Max.	Median	Mean ± SD
Children (< 18 years old)	F	33	≤ LOQ	6.34	0.71	1.35 ± 1.51
	M	19	≤ LOQ	5.34	0.68	1.05 ± 1.31
Adults (≥ 18 - 40 years old)	F	20	≤ LOQ	1.41	0.37	0.49 ± 0.37
	M	23	≤ LOQ	2.37	0.33	0.49 ± 0.48

*SD – standard deviation; **F – female, M – male; ***LOQ – limit of quantification (0.05 µg/g)

Taking into account the age of the inhabitants and Pb concentration in their teeth, a statistically significant relationship ($p<.05$) was obtained for the male population from Zabrze (Fig. 1). In other cases, no statistical relationship was found ($p >.05$).

DISCUSSION

Environmental contamination by lead has been a matter of great concern for several decades in many countries, and is still a current threat in many regions. One such region where human exposure to lead may cause adverse health effects and young children are particularly at risk, is the Silesian Province in southern Poland [29, 30].

A study by Ben Said et al. [31] confirmed that human teeth provide good chronological information on long-term

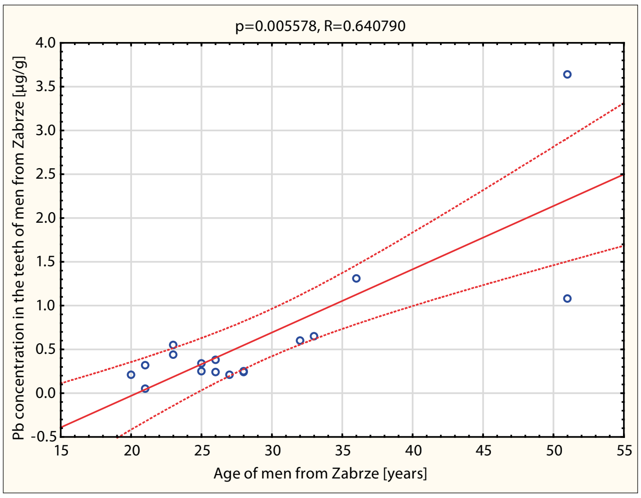

Figure 1. Relationship between the age of men from Zabrze and lead content in their teeth

heavy metal exposure, and also demonstrated that the place of residence is the most important factor influencing metals concentrations. The studies by Malara et al. [32] confirmed this dependence on the example of two cities of the Silesian Province. The lead concentrations in the impacted third molars and the bones surrounding the teeth were significantly higher for people living in the relatively polluted Ruda Śląska region than for people living in Bielsko-Biała region with a relatively clean environment. Mean concentration of lead in impacted wisdom teeth and in the bone surrounding the impacted teeth was, respectively: samples from people living in Bielsko-Biała (n = 33) – 10.29 µg/g (range: 6.24–14.26 µg/g), samples from people living in Ruda Śląska (n = 34) – 13.23 µg/g (range: 6.26–18.10 µg/g) [32]. In own study, the average concentration of lead in the teeth of adult residents of Bytom (n = 43) and Zabrze (n = 42) was lower than in the studies conducted by Malara et al. [32]; 0.49 µg/g and 0.50 µg/g, respectively. The obtained concentrations did not differ significantly between the analyzed cities. Bytom and Zabrze are neighbouring cities, unlike the distant Ruda Śląska and Bielsko-Biała. The level of environmental pollution in Bytom and Zabrze is on a comparable level.

Outside the territory of Poland, Kamberi et al. [33] determined the concentration of Pb in 86 human permanent teeth extracted from residents of three different geographical regions. The study included 31 permanent teeth from residents of Mitrovica (Kosovo), 32 from Klina (Kosovo) and 23 from Graz (Austria). The highest lead level was found in teeth extracted from Mitrovica residents (22.3 µg/g), followed by Klina (3.2 µg/g) and Graz (1.7 µg/g). Lead levels in teeth from Mitrovica residents were significantly higher ($p <.0001$) than in the other two groups. The researchers emphasized that this may be due to differences in lead pollution in the analyzed regions [33]. In the study by Al-Qattan & Elfawal [34], the concentration of lead was determined in 398 permanent teeth of inhabitants of Kuwait, aged 11–74 years. A significant correlation was found between dentin lead levels and gender. The mean dentin lead concentration was significantly higher in males than in females (6.8 ± 4.7 and 5.6 ± 4.6, respectively) [34]. The concentration of Pb was also significantly higher ($p = .000$) in men (8.45 ± 0.98 µg/g) than in women (3.48 ± 0.83 µg/g) in studies by Fernández-Escudero et al. [35].

In own study, the average concentration of Pb in the population of women and men from Bytom was the same (0.49 µg/g). In the case of Zabrze residents, higher concentrations of lead were recorded in the teeth of men (0.63 µg/g) than women (0.41 µg/g). The example of Zabrze is in line with the results obtained in the Al-Qattan & Elfawal [34] and Fernández-Escudero et al. [35]. In the Amr study [36], 112 permanent teeth collected from adults (aged 40–60 years) and primary teeth collected from 64 children (aged 5–12 years) living in Egypt were tested for lead content. In permanent teeth, the average Pb content was 6.26 ± 1.24 ppm (range: 0.6–9.23) and in deciduous teeth was 1.2 ± 0.89 ppm (range: 0.34–4.01). Thus, in the cited studies, permanent teeth were characterized by a higher concentration of Pb than primary teeth. Inverse results were obtained in own study – the lead content in primary teeth of children from Bytom was 2.5 times higher than in the teeth of adult residents.

Many studies have shown that children's teeth can be a good indicator of long-term exposure to lead, and some researchers have shown that primary teeth are a better indicator of environmental exposure to lead than blood, urine, hair or nails [25, 37]. According to the analysis of data obtained in the study, the determined lead concentrations in primary teeth varied, but the average values are comparable to the values obtained by other researchers. In own study, the mean Pb concentration was 1.24 µg/g, which is consistent with the results of studies conducted by Arruda-Neto et al. [18] in contaminated areas in Brazil where the mean concentration of lead in children's deciduous teeth was 1.28 µg/g. A slightly higher mean value of Pb (1.37 µg/g) was obtained by Tvinnereim et al. [38], who studied the concentration of Pb in the teeth of children living in Norway. A study evaluating the content of Pb in the teeth of 6-year-old children from Kraków (Poland), conducted by Barton [25], showed that the average lead content was 1.60 µg/g. A higher concentration than that obtained in own study may be correlated with the place of residence. Kraków is one of the most polluted cities in Poland and Europe in terms of $PM_{2.5}$ concentration [39]. Karahalil et al. [40] collected 297 shed deciduous teeth from 263 children (4–15 years) to determine the level of Pb exposure in Ankara and Balikesi, Turkey. The concentration range was 1.30 µg/g (Ankara-Centre) to 1.77 µg/g (Balikesir-Centre). The authors of the study did not observe a statistically significant difference between urban and suburban regions [40]. A very high concentration of lead was obtained in deciduous teeth of children in Jordan with the mean concentration of Pb obtained for 320 primary teeth (170 from boys and 150 from girls) – 30.26 µg/g [41].

The lead content in children's primary teeth in study groups from different places often varied by gender. Analysis of the teeth of children living in Norway [18] showed that the concentration of lead in the teeth of boys was about 20% higher than that in the teeth of girls. Own research showed the opposite result, higher concentrations of lead were determined in girls' teeth by about 23%, compared to the content of this element in boys' teeth. No difference was observed between boys and girls and the accumulation of lead in teeth in the studies conducted by Rahman & Yousuf [28], Karahalil et al. [40] and Alomary et al. [41].

The latest UNICEF and Pure Earth report [10] emphasizes that children are also exposed to lead *in-utero* through exposure of their mothers, with adverse impacts on neurobehavioural development that are comparable to those from childhood lead exposures. Karahalil et al. [40] proved in their research that the Pb content in shed primary teeth is a reliable measure of cumulative lead exposure in children as it is an indicator of the degree of exposure over several years, from *in-utero* life to loss of the tooth. A study undertaken in Karachi, Pakistan, to evaluate chronic lead exposure in children by measuring lead levels in shed primary teeth (309 teeth in total), showed that the accumulation of lead in teeth was not correlated with chronological age [28]. Own study also did not show any statistical significance ($p > .05$). Fernández-Escudero et al. [35] showed in their research that the Pb concentration in 150 permanent teeth from patients of Spanish nationality was significantly related with age ($p < .05$). In the authors' own research, a statistically significant correlation between age and the concentration of lead in permanent teeth was shown only for the population of men living in the city of Zabrze ($p = .005$; $r = .6$). In other cases, no statistical relationship was found.

The rate of human exposure on xenobiotics present in the environment is usually difficult to assess. According to Kamberi et al. [19] the content of trace elements in human teeth is a more suitable indicator to demonstrate environmental pollution rather than fitological and zoological sample analysis in ecological studies.

CONCLUSIONS

1. In the light of the conducted research, it can be concluded that human teeth dentine may be a bio-indicator of environmental exposure to lead.
2. It should be noted that children's primary teeth are more useful bio-indicators than adult teeth for determining environmental exposure to lead, because they may also indicate cumulative exposure to lead *in-utero*.
3. The relatively easy collection of samples, their storage and determination in terms of the content of trace elements (including lead), supports the importance of teeth in screening tests aimed at determining individual or population exposure to heavy metals.
4. Constant bio-monitoring of people exposed to lead, especially children, living in polluted areas, with the use of non-invasive methods of sampling for research is necessary for the proper targeting of activities in the field of primary prevention.

Conflicts of interest

The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

Declaration of originality and Ethics approval

This study has not been published elsewhere, nor has it been simultaneously submitted elsewhere for publication All Tables and Figures are the original work of the authors and no permissions were required. The first Author, as the dentist responsible for obtaining the tooth samples, received permission from the Bioethical Commission to conduct research within the scope described in the presented work.

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