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Full Length Research Paper

Trace elements in Egyptian teeth

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Teeth are reported to be suitable indicators of heavy metals exposure from environment and nutritional status. Inductively coupled plasma mass spectrometry (ICP-MS) is used to compare the trace element content of children's primary teeth and permanent adult teeth. Primary teeth were collected from 64 children and permanent teeth collected from 112 adult with age range of 40-60 years. The data were assessed statistically using t-tests. The permanent teeth contained significantly greater concentrations of Na, Mg, Al, Fe, Ni, Cu, Sr, Cd, Ba, Pb and U, and significantly less Mn, Co, As, Se, Mo and Bi than the children teeth. Moreover, comparison between heavy element concentrations in healthy and caries teeth pulps show that the mean concentrations of Na, Al, K, Cr, Mn, Co, Cu, Zn, Mo, Ag, Bi and U are lower in caries than healthy teeth pulps. On the other hand, the mean concentrations of Mg, Cd and Pb are higher in caries samples than healthy teeth pulps.

Key words: ICP-MS, teeth, heavy metals.

INTRODUCTION

Biomonitoring of trace elements in human teeth has become an important tool for nutritional and environmental status (Brown et al., 2004; Haavikko et al., 1985; Guillard et al., 1992; Alexander et al., 1993; Al-Mahroos and Al-Saleh, 1997). Primary teeth in particular, are easily obtained since they naturally exfoliate as the permanent teeth erupt. Variation in the trace element content of teeth has been demonstrated (Brown et al., 2004). Modes of human trace element intake may be diverse ranging from the normal ingestion of food and water, to the deliberate consumption of soils and dermal absorption. The presence and/or absence of trace elements in the environment will influence the availability of such elements to man (Brown et al., 2004). For example, in the rural areas of Finland, the concentrations of zinc and magnesium in dentine have been related to that in soil (Lappalainen et al., 1981), while dental fluorosis has been linked to fluoride and lead concentrations in drinking water (Brown et al., 2004; Alexander et al., 1993).

It has been reported that the concentration of lead in teeth can be used as an index of environmental pollution (Gulson, 1994). Lead is preferentially incorporated and stored in calcified tissue such as teeth (Begerow et al., 1994) and a tooth lead concentration above 4 mg/kg has been suggested as indicating a toxic body load (Al-Mahroos and Al-Saleh, 1997; Carvalho et al., 2001). The importance of the location of trace elements in teeth, particularly with respect to lead, has been investigated. These studies have used various techniques including microprobes and have highlighted the systematic but inhomogeneous distribution of trace elements within enamel and dentine (Budd et al., 2000).

Several other studies demonstrate that it is possible to detect nutritional deficiencies through the chemical composition of teeth and that such deficiencies can have an important effect on the critical growth period of the dentition (Lappalainen et al., 1981; Gulson, 1994; Begerow et al., 1994; Budd et al., 2000; Frank et al., 1990; Bercovitz and Laufer, 1992; Budd et al., 1998; Montgomery et al., 1999; Alvarezet al., 1990). Differences in the availability of foods and food choice, often based on social and cultural practices, may be important factors in determining trace element intake in man and complicate the modeling of intake.

The pulp occupies the center of each tooth and consists of soft connective tissue (Figure 1). Every person normally has a total of 52 pulp organs (Murray, 1990); 32 in the permanent and 20 in the primary teeth. They have a number of morphologic characteristic that are similar. Each pulp organ resides in a pulp chamber surrounded by dentin containing the peripheral extensions

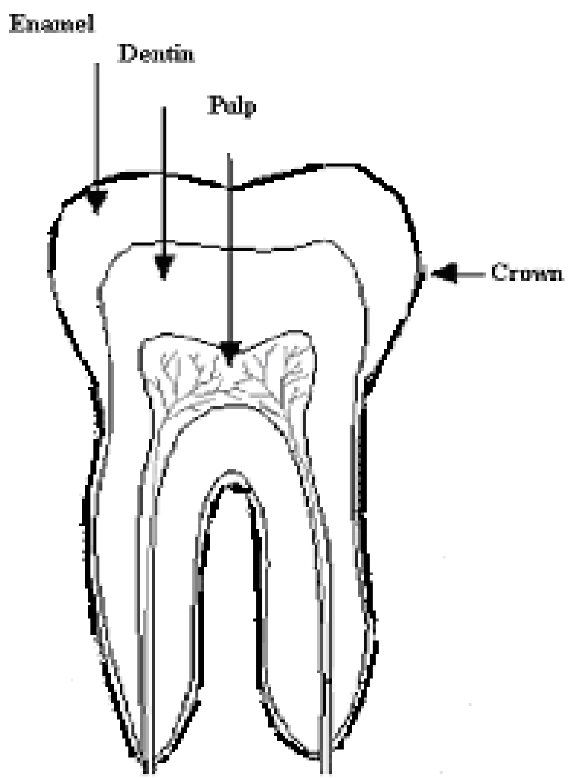

Figure 1. Schematic diagram of human molar.

of the cells that formed it. Ide-Ektessabi et al. (2004) investigated trace element distribution in tooth layer and pulp in two teeth by X-ray fluorescence using synchrotron radiation microbeams. They (Ide-Ektessabi et al., 2004) found high concentrations of calcium, lead, and zinc on the border of dentine and pulp. Pinheiro et al. (1999) studied elemental concentrations in the different tooth regions (enamel, dentine crown, dentine root and pulp) in nine teeth by the synchrotron microprobe to determine environmental influence and dietary habits. Carvalho et al. (2002) analyzed forty molar teeth restored with dental amalgam by a synchrotron microprobe to evaluate the diffusion of its major constituents, Cu, Zn and Hg, throughout the tooth structures. Curson and Crocker (1978) determined Zn and Pb in teeth pulp.

The aim of this study is to compare the toxic and essential elements concentrations in primary and permanent teeth collected from Al-Kanayat city, Egypt.

MATERIALS AND METHODS

Samples and sample preparation

Primary teeth were collected from children living in the El-Kanayat City, Egypt. Teeth collected from children of age ranges from 5 -12 years with either gender and from teeth of all types (incisors, canines and molars). 112 permanent teeth were collected from adults with age range from 40 to 60 years. Where possible, the teeth used for comparison are caries free. The teeth were soaked in hydrogen peroxide to remove connective tissue and then washed with deionized water and dried overnight. Each tooth was grinded and weighed. Teeth were digested in microwave digestion system (milestone 1600) using 10 ml nitric acid.

Teeth crowns were cut and their pulps collected directly after their take off from the adults. Teeth pulps were washed by 1% ultra pure nitric acid and dried in 60°C. Samples were digested in microwave digestion system using mixture of 250 µl of HNO_3 and 100 µl of H_2O_2. After digestion, samples were cooled in water bath and diluted to 1 ml using deionized water (18.2 MΩ).

Instrumentation

An inductively coupled plasma mass spectrometer (JMS-PLASMAX2) was used for trace elements measurements. Optimization of the ICP-MS was performed using ^{89}Y standard solution of 10 ppb in 2% HNO_3. An identical 2% HNO_3 blank solution was used as background sample as well as to clean the tubes between the measurements. A micro concentric nebulizer with a desolvation introduction system (ARDIUS, CETAC, USA) was used in the measurements. Table 1 (Brown et al., 2004) shows the operating parameters of ICP-MS which was used in the measurements.

RESULTS AND DISCUSSION

The results of the analysis of primary and permanent teeth are summarized in Table 2. The table contains essential elements (such as Cu, Mg, and Zn), possibly essential (Sr) and those that does not have known biological role and/or are potentially toxic (Al, Cd, Ba, Pb, U). The data were assessed by use of a one-way analysis of variance (ANOVA) using Tukey's test at the 95% confidence level. A P value of less than 0.05 was considered statistically significant. Outlying values have been omitted from the analysis in order to make general conclusions. The outlier data has been rejected using D-value [$D= (X_n - \bar{X})/S$; X_n is a value, $\bar{X}$ is the mean, and S is the standard deviation]. The data showed that the mean concentrations (ppm) of Na, Mg, Al, Fe, Ni, Cu, Sr, Cd, Ba, Pb and U were greater in the permanent teeth compared with the primary teeth. The reverse was observed for Mn, Co, As, Se, Mo and Bi.

The pulp contains the tooth's nerve and blood vessels. The nerve endings inside the pulp send signals to the brain about what is going on the tooth. The blood vessels feed the tooth and keep it alive and healthy. The actual concentrations often provide information on deficiency or disease states, or whether poisoning or contamination has occurred. The most widely encountered trace elements include Al, Cd, Ba, K, Li, Mg, Mn, Na, Pb and Sr. Some of these, for example, Cd and Pb, are perceived as potentially toxic elements. Table 3 shows heavy elements concentrations (ppm) in teeth pulps. A standard reference material, namely, bovine blood, level 2 (NIST966), is used for quality assurance.

The information on the concentration of trace elements

Table 1. Experimental conditions used in the measurements.

Forward RF power	950 W
Reflected RF power	< 2 W
Coolant gas flow rate	14 L/min
Auxiliary gas flow rate	0.3 L/min
Nebulizer gas flow rate	0.95 L/min.
Sample uptake rate	0.6 L/min.
Accelerating voltage	6 kV applied on the sampler and skimmer
Mass resolution (m/Δm)	300
Sampling depth	5 mm
Introduction system	Desolvation introduction system (ARDIUS)
Optimization	Maximum ion intensity of 10 ppt of ^{89}Y

Table 2. Heavy metals concentrations (ppm) in primary and permanent teeth samples.

Element	Primary teeth		Permanent teeth	
	$\bar{X}$ ± SD	Range	$\bar{X}$ ± SD	Range
Na	5454±950	(4211-7280)	9000±120	(7054-10030)
Mg	1755 ± 340	(1043-2750)	2800 ± 478	(1500 - 3700)
Al	17.9±12.3	(3-69.5)	51.4±18.2	(27.5 - 84)
Cr	0.04± 0.01	, (0.005 - 2)	0.05±0.03	(0.03-0.11)
Mn	5.5±2	(2.5 - 8.5)	0.27±0.11	(0.09-1.2)
Co	0.54± 0.12	(0.006 - 1.11)	0.003±0.002	(0.0009-0.005)
Fe	80.1± 16.5	(28.9 - 135)	94±23	(65.3-122)
Ni	1.66±0.46	(1.16-2.16)	5.54±1.05	(3.78-7.04)
Cu	6.4 ± 4.8	(2 -11.3)	9.2 ±11.4	(1.4 - 26.1)
Zn	133 ± 30	(85 - 166)	178±44.6	(124.6-235.7)
As	0.82±0.07	(0.023 - 0.14)	0.02±0.007	(0.014-0.027)
Se	10.5±1.57	(7.23-13.56)	0.34±0.09	(0.28-0.44)
Sr	87±11.3	(60.9-114)	101.2±24.3	(70.2-130)
Mo	1.8±0.29	(1.02- 2.33)	0.044±0.012	(0.031-0.067)
Ag	0.08± 0.03	(0.004 – 1.9)	0.009±0.002	(0.005-0.012)
Cd	0.00011±0.00001	(0.00007-0.00014)	0.012±0.004	(0.0084-0.016)
Ba	7.8±3.2	(2.06 -13.8)	9.5±5.4	, (5.11- 17.97)
Pb	1.2±0.89	(0.34-4.01)	6.26±1.24	(0.6-9.23)
Bi	23±2.34	(16.3-28.5)	0.22±0.09	(1.10-3.01)
U	0.005±0.002	(0.001- 0.052)	0.011±0.005	(0.0012-0.041)

in teeth pulps is scarce and contradictory (Pinheiro et al., 1999; Carvalho et al., 2002; Curson and Crocker, 1978). Carvalho et al. (2002) measured the concentration of Mn, Fe, Cu Zn, and Pb in restored and healthy teeth pulp samples by synchrotron microprobe. The concentration of Mn, Fe, Cu Zn, and Pb in restored pulp samples are 5±3, 16±6, 43±20, 570±120, and 43±25, respectively. The concentrations of Mn, Fe, Cu Zn, and Pb in healthy samples are 1.7±1.2, 21±11, 2.3±1.4, 286±50, and 46±15, respectively.

From Table 3, it was observed that magnesium is an abundant element in caries samples. Magnesium (Mg) mean concentration in healthy and caries sample is 2733±589 and 6285±1981 ppm, respectively. Sodium (Na) is important for nerve and muscle functions, blood pressure control (Underwood, 1971). Potassium is the main base ion of the fluid in the body's cells. Along with sodium, it is important to the electrical potential of the nervous system and, therefore, for the efficient functioning of nerve and muscle. Sodium (Na) mean

Table 3. Concentration of heavy metals (ppm) in permanent healthy and caries teeth pulps.

Element	Healthy teeth pulps			Caries teeth pulps		
	X ± SD	Range	Element to calcium ratios (×10^{-4})	X ± SD	Range	Element to calcium ratios (×10^{-4})
Na	1793±342,	(1206-3071)	217	1385±705,	(2114-4753)	114
Ca	82336±8980	(53961-164840)	1	121814±1718,	(89365-157238)	1
Mg	2733±589	(1869-4948)	332	6285±1981,	(4728-10125)	516
Al	1029±172	(599-1630)	125	1413±422,	(1153-2501)	116
K	1309±152	(814-2899)	159	1088±387,	(1008-2453)	89
V	2.31±0.12	(1.22-2.95)	0.28	2.92±0.78,	(2.08-5.14)	0.24
Cr	42.40±6.29	(26.7-63)	5.15	33.99±9.14,	(26.08-56)	2.79
Mn	24.04±4.23	(13.68-39.4)	2.92	29.84±9.81,	(22.32-53)	2.45
Fe	5.85±1.12	(4.62-9.8)	0.71	1.22±0.36,	(0.97-2.16)	0.1
Cu	270±44	(185-435)	32.84	234±59	, (172-396)	19.22
Zn	1383±254	(946-2781)	168	1303±318,	(987-2309)	107
Rb	1.15±0.12	(0.89-1.9)	0.14	2.56±0.68,	(1.87-4.02)	0.21
Sr	82.09±17.05	(68.3-131)	9.97	106±28	(87-188)	8.75
Mo	2.31±0.19	(1.3-3.25)	0.28	3.29±0.99,	(2.07-5.28)	0.27
Ag	55.91±10.41	(32.1-90)	6.79	26.07±6.84	(21-39)	2.14
Cd	1.98±0.27	(1.63-3.2)	0.24	6.70±1.18	(4.53-10.9)	0.55
Ba	18.94±2.05	(15.2-33)	2.3	34.60±8.43	(26.5-63)	2.84
Pb	29.23±4.13	(17.5-46.9)	3.55	77±21.2	(56-135)	6.4
Bi	35.98±3.47,	(20.9-62.6)	4.37	12.55±3.67	(9.13-19.9)	1.03
U	0.91±0.38	(0.66-1.6)	0.11	0.97±0.28,	(0.72-1.06)	0.08

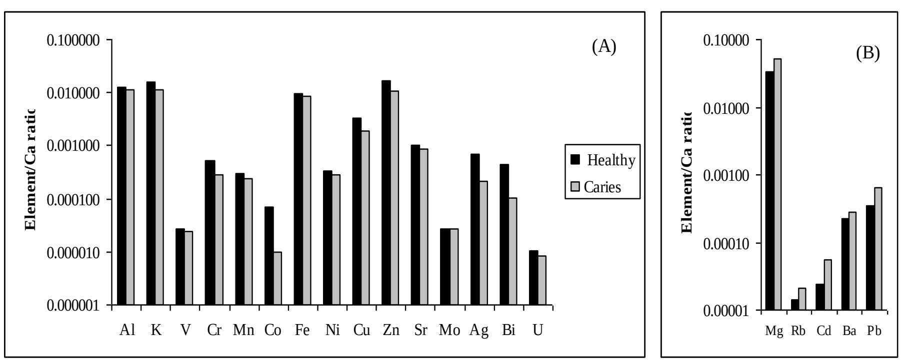

Figure 2. Comparison between elements/calcium concentration ratios in teeth pulps.

concentrations are 1793 ± 342 and 1385 ± 705 ppm in healthy and caries samples, respectively. Potassium (K) mean concentrations are 1309 ± 152 and 1088 ± 387 ppm in healthy and caries, respectively. It is observed that Na and K concentrations in the caries samples are lower than healthy samples.

Zinc is an essential element to humans (Stürup, 2000). It is required for the synthesis of protein and collagen, which is great for wound healing and a healthy skin, and also necessary for a healthy immune system (Sandström, 1996). Published work shows that Zn concentration in teeth pulp are 541 ppm in miner and 583 ppm in fisherman (Pinheiro et al., 1999), 286 in healthy teeth and 570 ppm in restored teeth (Carvalho et al., 2002), and are in the range of 218-520 ppm in healthy teeth pulps (Curson and Crocker, 1978). Measured Zn concentrations in the present teeth pulp samples are 1383±254 ppm in healthy samples and 1303±318 ppm in caries samples. Normalizing the mean concentrations of elements to calcium concentration shows that Al, K, V, Cr, Mn, Co, Fe, Ni, Cu, Zn, Sr, Mo, Ag, Bi and U are lower in caries samples than healthy samples (Figure 2).

On the other hand, the mean concentrations of Mg, Rb, Cd, Ba and Pb are higher in caries samples than healthy samples.

Conclusion

Toxic elements (Cd, Pb, and Mg) are higher in permanent teeth compared with primary teeth. Moreover, toxic elements are higher in caries teeth pulps compared with healthy teeth pulps.

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