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Analysis of human bone and teeth

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Abstract : The trace element analysis of four human bone samples of an individual of age 54 who died of an accident, were collected from different parts (tibia, parietal and occipital bone) of the human body by autopsy and analysed by particle induced X-ray emission technique Similarly, twenty-seven healthy human teeth samples from twelve individuals of the age group 35–57, were carried out by energy dispersive X-ray fluorescence spectroscopic technique at Institute of Physics, Bhubaneswar, India Elements like P, S, Cl, K, Ca, Mn, Fe, Cu, Zn, Sr and Pb were quantified in the human bone and teeth samples The role and uniformity of the trace elements, present in all the samples, in this study were presented and its effect on the statistical distribution of their concentrations is discussed

Keywords : Human bone, PIXE, EDXRF

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1. Introduction

Because most essential trace metals are present in biological specimens in very low concentrations, precise and accurate analysis is most essential if meaningful results are to be obtained. Mineral and trace elements are not distributed homogeneously in human bone and the distribution is not clearly solved. While in the substantia compacta (SC) and in the substantia spongiosa (SS) elements seem to be distributed nearly equally, concentrations of these elements in the transient zone between SC and SS are obviously altered [1]. Understanding the effects of trace metals on human health is as complex as it is fascinating. As mentioned earlier, the high concentrations may prove toxic, as also, depletion in the concentration of the essential trace elements may cause various metabolic instabilities due to enzyme dysfunction. In the era of rapid industrialization and technological advances, it is imperative to watch keenly for contamination of the environment and its vital composition from heavy metal wastes emanating out of industries. Many metabolic disorders in man are accompanied

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by alterations in the concentration of one or more trace elements in some body fluid, especially blood serum or plasma. It is thus important to update us with various techniques available for such determinations, their operational aspects, advantages/ disadvantages *etc.* [2].

The tooth is a bio-indicator of great interest because it contains information on exposure to elements that become deposited in the tooth material. The mineral tissue of the tooth consists of hydroxyapatite crystals with incorporated trace elements, which can provide information, concerning environmental influence and dietary habits. Trace elements may also pay a role in dental health [3].

2. Experimental techniques

2.1. Sample preparation :

The bone samples were collected from the City Hospital, Berhampur, India from an individual of age 54 who died of an accident. The samples were collected from different parts of the human body by autopsy. To keep contamination and loss of elements from the samples to a minimum, instruments were used for the collection of specimens and their handling prior to the analysis were acid cleaned and washed with Millipore water and then with alcohol. For Particle Induced X-Ray Emission (PIXE) analysis, after crushing the samples to fine powder, 150 mg of each of the specimens were added with high pure graphite (99.99%) powder in 1 : 1 ratio by mass and the mixture had to be ground thoroughly to make a homogeneous mixture. Pellets of size 13 mm diameter were obtained after putting the mixture in the KBr press [4]. Similarly for the Energy Dispersive X-Ray Fluorescence (EDXRF) analysis, 500 mg of the specimens were added with high pure cellulose powder in 1 : 1 ratio by mass and ground thoroughly to make a homogenous mixture and pressed in the KBr press to get pellets of size 25 mm diameter [5].

2.2. Data analysis :

For PIXE analysis, the bone samples were irradiated in vacuum inside the scattering chamber at the 3 MV tandem type horizontal pelletron accelerator with a proton beam of energy 3 MeV at Institute of Physics, Bhubaneswar, India. The data analysis was performed by using GUPIX-2000 software [5]. The EDXRF system at the Institute of Physics has been used for the analysis of human teeth samples. The spectra obtained for all the samples were evaluated using the AXIL computer program supplied by the International Atomic Energy Agency (IAEA). For both PIXE and EDXRF data acquisition, Si (Li) detector was used. A 50 μm aluminized mylar was used for PIXE data acquisition for low energy suppression. Target of a certified reference material was also prepared for analysis in the EDXRF system as standard [6].

3. Results and conclusion

In the human bone and teeth samples, Cl, Ca and P were quantified to be the major elements, whereas other elements like S, K, Mn, Fe, Cu, Zn, Sr and Pb were estimated in trace level in the present investigation. Because of their appreciable rate of bone renewal human bones can be expected to record lifelong intake of some trace elements The PIXE spectrum of human tibia bone is shown in Figure 1 and the EDXRF spectrum of a representative human teeth sample is shown in Figure 2

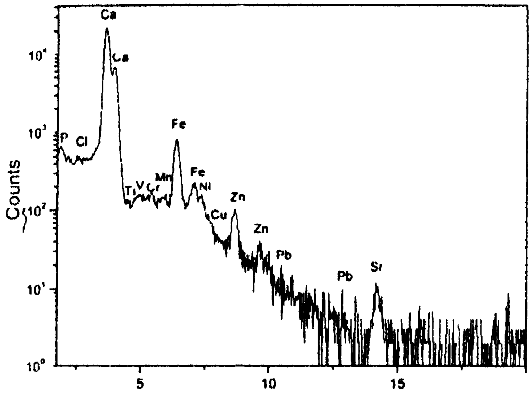

Figure 1. PIXE spectrum of tibia bone

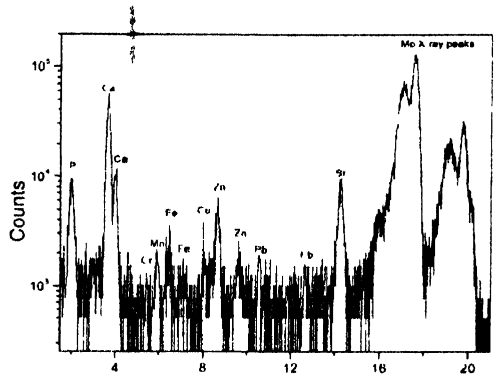

Figure 2. EDXRF spectrum of a teeth sample

Calcium and phosphorous .

Since human bone and teeth contains hydroxyapatite $[Ca_{10}(PO_4)_6(OH)_2]$ as their main constituent, which contains higher amount of calcium and phosphorous in it, hence the higher amount of Ca and P is obvious in the samples. In the teeth samples, Ca and P amount were found to be much higher than the human bone samples. This may be due to the fact that the human tooth enamel contains higher concentration of hydroxyapatite. In the present study, Ca and P concentrations were estimated to be 28.1% and 13.6% respectively for tibia bone. In the parietal bone sample, concentrations of Ca and P were 21.9% and 7.2% respectively whereas for occipital bone sample, Ca and P concentration were estimated to be 24.9% and 9 2% respectively.

Moreover, the average concentrations of Ca and P for 27 human teeth samples were found to be 35.2% and 20.6% respectively. Comparing the concentration of Ca and P in the tibia bone to that of parietal and occipital bones, it was found that the Ca and P values are higher in tibia bone sample. This higher concentration may be due to presence of higher amount of hydroxyapatite in the tibia bone sample. Moreover, Ca/P ratio in hydroxyapatite should be 1.67. But, since the Ca/P ratio in the present study does not agree with that of the standard Ca/P ratio, it may be stated that the excess Ca concentration is due to some other complexes of Ca other than hydroxyapatite.

Calcium versus zinc, strontium and lead :

The strontium content of bones can also provide information about the diet of past populations [7]. Natural strontium levels in living bone are typically 150–200 ppm. But in the present study, the concentration of Sr in the samples is higher than the normal level (tibia bone : 258.7 ppm, occipital bone : 190.2 ppm, parietal bone : 218.6 ppm and average concentration of teeth : 285.6 ppm). An interesting observation has been found that the Ca/Sr ratio is directly proportional among all the human bone and teeth samples. However, the surprisingly positive Ca and Sr correlation must be of completely different origin because of the general fact that Ca is substituted by Sr and therefore a negative correlation between Ca and Sr is expected. The extent to which strontium is subject to diagenetic alteration is a matter of debate, though it is widely believed to be one of the least affected elements. Analysis of ancient diet using, for example, strontium-calcium ratios requires that elemental levels correspond to those present at the time of death *i.e.* that they reflect *in vivo* conditions. In a similar manner Ca/Zn concentration is also directly proportional among all the human bones and teeth samples. Zinc (tibia bone : 26.7 ppm, occipital bone : 1.3 ppm, parietal bone : 20.2 ppm and average concentration of teeth : 32.2 ppm) and strontium are more evenly distributed and are found to correlate quite well with the calcium concentration. Zinc plays an important role in the growth of new skeletal tissue and is localized in areas of bone formation. Zinc is also involved in the general metabolism of humans so that relations may exist between dietary intake, diseases, or occupational exposure and the bone mineral composition.

Since the present investigation is a preliminary study, our future work will concentrate on finding the role of every element in order to find conclusions regarding diet, physiology and contamination. Since Pb intake occurs *via* the walls of narrow bone channels inside the bone structure as is clearly visible in the PIXE spectra, so few ppm of Pb (tibia bone : 66.8 ppm, occipital bone: 36.3 ppm, parietal bone: 37.1 ppm and average concentration of teeth: 41.9 ppm) is present in the human bone and teeth samples. Ca/Pb concentrations are also directly proportional among each of the samples except the higher concentration of Pb in the tibia bone. Higher concentration of Pb in the tibia bone sample may be due to contamination with environmental lead containing materials or during sample preparation. Other elements like S, Cl, K, Mn, Fe and Cu were also estimated in the present study and are as shown in Figure 1 and Figure 2.

References

[1] P Wobrauschek, G Pepponi, C Streli, C Jokubonis, G Falkenberg and W Osterode, *Adv. X-Ray Anal.* **45** 478 (2002)

[2] *URL: http://www.ojhas.org/issue13/2005-1-1.htm*

[3] M L Carvalho, J P Marques, J Brito, C Casaca and A S Cunha *Nucl. Instrum. Meth.* **B196** 148 (2002)

[4] M Ashok, S Narayana Kalkura, V J Kennedy, A Markwitz, V Jayanthi, K G M Nair and V Vijayan *Int. J. PIXE* **12** 137 (2002)

[5] V Vijayan, T R Rautray, P K Nayak and D K Basa *X-Ray Spectrom.* **34** 128 (2005)

[6] V Vijayan, S N Behera, V Ṡ Ramamurthy, S Puri, J S Shahi, N Singh *X-Ray Spectrom.* **26** 65 (1997)

[7] R I Gilbert in *The Analysis of Prehistoric Diets* (eds.) R I Gilbert and J H Mielke (Orlando : Academic) p339 (1985)