Concentration of rare earths in dentine and enamel

A pilot study

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A neutron activation method for the determination of rare earths in hard tissues was developed and the concentrations of some of these elements in human dentine and enamel were determined. The results obtained for La, Sm, Tb and Yb are very low, indicating a total rare-earth content of the order of 0.1 ppm or less in the samples investigated. The observed La/Sm ratios seem to indicate a slight enrichment of the lighter rare earths relative to the heavier, as compared to average geological abundances.

Key-words: Activation analysis; dental enamel; dentin; lanthanum; samarium

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The existing knowledge of the concentration and distribution of the rare earths (lanthanum-lutetium) in the human organism is very limited. From the information available, however, it seems that they are present at a very low level. The rare earths are not known to be essential to living organisms (*Kyker*, 1962). The absorption from the gastrointestinal tract appears to be very low; values of less than 0.05 % are typical for mammals. The rare earths tend to accumulate in the skeleton, where they concentrate in the mineral fraction.

In a study of heavy elements in ocean sediments *Arrhenius, Bramlette & Picciotto* (1957) observed that fish skeleton debris contained several per cent of rare earths, in spite of the fact that the total concentration of the individual rare earths in ocean water is as low as 10^{-2} — $10^{-4}/\mu g/l$ (*Høgdahl, Melsom & Bowen*, 1968). It was shown that rare earths occurred in the apatite phase of the fishbone. This observation, together with the fact that some rare earths have been shown to be capable of decreasing the acid solubility of tooth enamel (*Manly & Bibby*, 1949; *Shresta, Mundorff & Bibby*, 1972), encouraged an investigation of the normal level of rare earths in dentine and enamel.

The only previous information available on the content of rare earths in human teeth seems to be from a semi-quantitative mass-spectrometric investigation, stating that the content of Y in enamel and dentine is in the range 1—10 ppm and that of La below 1 ppm (*Hardwick & Martin*, 1967). On the other hand, a more detailed investigation by *Lux* (1938) using

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X-ray fluorescence after chemical enrichment, indicated a content of 0.01 ppm or lower in animal bone for the elements Y, La, Ce, Nd, Gd and Dy.

The most sensitive analytical technique available at present for the determination of most rare earths appears to be neutron activation analysis. This technique has been used for the determination of a great number of trace elements in dental tissues (*Söremark & Samsahl*, 1962, 1964; *Lundberg et al.*, 1965; *Nixon*, 1969; *Retief et al.*, 1971), but so far no data on rare earths have appeared. In the present work a neutron activation method suitable for the determination of rare earths in mineralized tissues has been developed, and some results for samples of human enamel and dentine are presented and discussed.

MATERIALS AND METHODS

Collection and Preparation of Samples

Samples were obtained from 5 human wisdom teeth. Four of the teeth had erupted, while the fifth was impacted. From the latter only the dentine was examined. The teeth were devoid of fillings or substantial caries attacks. Two of the teeth were from male patients, and three of the teeth from female patients. The age of the patients varied from 21 to 72 years (Table I). Four samples of dentine and four samples of enamel were examined. After extraction ground sections were prepared with a diamond disc under constant water spray using a Gillings-Hamco Thin Sectioning Machine (*Gillings & Buonocore*, 1959). From the ground sections pieces of dentine and enamel were dissected out and subjected to analysis without grinding or ashing.

Neutron Activation Analysis

Attempts to determine some of the rare earths by non-destructive activation analysis using a technique similar to that described by *Retief et al.* (1971) failed because of interference from high activities of ^{32}P and ^{47}Ca – ^{47}Sc induced in the major elements. A radiochemical group separation of the rare earths from the irradiated sample followed by high-resolution γ-spectrometry would in most cases be sufficient to determine about ten of these elements, provided that they are present in sufficiently high concentrations. In the case of mineralized tissue, however, it is necessary to have a method giving very good separation from scandium in order to eliminate the large amounts of ^{47}Sc produced from calcium by the reaction ^{46}Ca(n, γ) ^{47}Ca$\xrightarrow{\beta^-}$ ^{47}Sc. As the procedures normally used for the rare-earth group separation do not discriminate efficiently enough against scandium, a method based on anion exchange separation in thiocyanate media (O. Johansen & E. Steinnes, unpublished work) was adapted to the present problem.

Samples of 10—40 mg and standards of La, Sm, Tb and Yb were irradiated for one week in the JEEP II reactor (Kjeller, Norway) at a thermal neutron flux of about $2 \cdot 10^{13}$ n cm^{-2} s^{-1}. After five days' delay the samples were fused with 2 g sodium hydroxide pellets in a nickel crucible in the presence of La (10 mg) and Sm (1 mg) carriers. The fusion cake was released from the crucible with water and centrifuged, and the supernatant solution was discarded. After two washings with water the precipitate was dissolved in 10 ml of 6 M HCl. The rare-earth fluorides were then precipitated with concentrated hydrofluoric acid, the precipitate was centrifuged and washed twice with an aqueous solution of Ca^{2+} (1 mg/ml). The fluoride precipitate was dissolved in 8 ml 2 M HCl + 10 ml boric acid solution (30 mg/ml H_3BO_3), 2 ml 5 M NH_4SCN were added, and the solution was applied to an anion exchange column pre-equilibrated with 1 M HCl (Dowex 1—X8, 100—200 mesh, 1.0 cm diameter, 10 cm height). The column was washed with two portions of 5 ml 1 M HCl, and the combined eluate was adjusted to pH 5 with 5 M ammonium acetate after addition of 10 mg Sc »hold-back» carrier. The rare earths were precipitated with a fluoride solution of pH 5, filtered, washed, and prepared for γ-spectrometry measurements with a system consisting of a 20 cm^3 Ge(Li) detector connected to a 2000 channel analyzer. The radioisotopes and γ-energies used are listed in Table I.

After the γ-activity measurements the chemical yield of the added carriers was determined by re-irradiation, after dissolving the precipitate in a 6 M nitric acid solution containing 30 mg/ml H_3BO_3 and diluting to an appropriate concentration. The yields were of the order of 40—60 %, and were very similar for La and Sm within each sample, indicating that no appreciable fractionation of the rare earths takes place during the separation.

The nuclides ^{140}La and ^{153}Sm are among the species formed by thermal neutron fission of ^{235}U, and uranium may therefore interfere with the determination of La and Sm if present in appreciable amounts. By the use of a neutron activation method based on alkaline decomposition of the samples and extraction of ^{239}U ($t_{1/2}$ = 23.5 min) into tributyl phosphate from 5 M HNO_3, it was found that the present samples contained less than 0.01 ppm U, which means that there is no significant interference. This is in accordance with the investigation of *Hamilton* (1971), who found a mean value of 0.007 ppm U in teeth.

RESULTS AND DISCUSSION

Results of the analyses are given in Table I. In three cases the enamel and dentine were taken from the same teeth. It was possible to obtain positive results only for La and Sm. For the other two rare earths investigated only upper limits could be given. Some samples were analysed in duplicate, and the agreement seems satisfactory considering the low concentrations present.

The observations made in this study indicate that the »normal» rare-earth content of enamel and dentine are very low (Table I). From the existing knowledge of the systematics of natural rare-earth distributions and the observed La/Sm ratios it can be inferred that the total rare-earth content is probably of the order of 0.1 ppm or less in the samples investigated.

The incorporation of rare earths in the teeth could be assumed to take place in two ways: (1), through gastrointestinal absorption and transport through the circulatory system; (2), direct adsorption on the tooth surface from the oral cavity. The present results seem to support the first alternative, since the concentration of the elements determined is generally somewhat higher in the dentine than in the enamel, in spite of the fact that the percentage of inorganic matter in the dentine is lower, and it is usually not exposed to the oral environment.

In nature the rare earths act as a coherent group chemically, but their distribution pattern may be modified by extensive geological fractionation pro-

Table I. *Concentration of some rare earth elements (PPM) in dentine and enamel determined by neutron activation analysis*

Sample	Age of patient	La	Sm	Tb	Yb	La/Sm
1 Dentine	22	0.0153	0.00103	< 0.005	< 0.01	14.8
1 Enamel		0.0041	0.00029	< 0.005	< 0.01	14.1
2 Dentine	21	0.0137 0.0136	0.00168 0.00174	< 0.005	< 0.01	8.0
2 Enamel		0.0055 0.0059	0.00075 0.00075	< 0.005	< 0.01	7.6
3 Dentine	25	0.057* 0.016	0.00190 0.00179	< 0.005	< 0.01	8.7
4 Enamel	72	0.0052 0.0030	0.00438* 0.00045	< 0.005	< 0.01	9.1
5 Dentine	21	0.0072	0.00070	< 0.005	< 0.01	10.3
5 Enamel		0.0077	0.00107	< 0.005	< 0.01	7.2
Radioisotope measured		^{140}La	^{153}Sm	^{160}Tb	^{175}Yb	
γ-energy used (keV)		1596	103	298	396	

* The high value is probably due to contamination during the preirradiation handling procedure

cesses. A point of interest might be whether the ratios of individual rare earths supplied through the food are maintained through the various processes in the human body where these elements are involved. Some information on this point may be gained by considering La/Sm ratios. The average La/Sm ratio of the human diet is not known, but as a first approximation one might consider it to be similar to that of sedimentary rocks or natural waters. Recent analyses of sediments have yielded La/Sm ratios of the order of 5—6 (*Haskin et al.* 1966). In water samples from eleven Norwegian rivers the average La/Sm ratio was found to be 4.7 (B. Salbu & E. Steinnes, unpublished work). If a figure of about 5 is representative for the La/Sm ratio in an average diet, the present results indicate a slight enrichment of the light rare earths relative to the heavier members of the group during the processes leading to incorporation of the rare earths in dental tissues (Table I). On the other hand, *Wester* (1964), in a study of human heart tissue, reported La/Sm values in the range 0.1—1.0 in eight different cases, which would presumably indicate a considerable enrichment of the heavy rare earths. Evidently more information has to be obtained before the behaviour and possible fractionation of the rare earths in the human body can be satisfactorily understood.

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REFERENCES

Arrhenius, G., Bramlette, M. N. & Picciotto, E. 1957. Localization of radioactive and stable heavy nuclides in ocean sediments. Nature (Lond.) 180, 85—86

Gillings, B. & Buonocore, M. 1959. The preparation of thin serial sections of undecalcified tissues. J. Dent. Res. 38, 1156—1165

Hamilton, E. I. 1971. The concentration and distribution of uranium in human skeletal tissues. Calc. Tiss. Res. 7, 150—162

Hardwick, J. L. & *Martin, C. J.* 1967. A pilot study using mass spectrometry for the estimation of the trace element content of dental tissues. Helv. Odont. Acta 11, 62—70

Haskin, L. A., *Frey, F. A.*, *Schmitt, R. A.* & *Smith, R. H.* 1966. Meteoritic, solar and terrestial rare-earths distribution. In: *Physics and Chemistry of the Earth*, Vol. 7. Pergamon Press, Oxford, New York, pp 167—321

Høgdahl, O. T., *Melsom, S.* & *Bowen, V. T.* 1968. Neutron activation analysis of lanthanide elements in sea water. In: *Trace Inorganics in Water* (D. W. Hood, ed.). Advances in Chemistry Series No. 73, American Chemical Society, Washington, D.C., pp 308—325

Kyker, G. C. 1962. Rare earths. In: *Mineral Metabolism* (C. L. Comar, F. Bronner, eds.), Vol. II, Part B. Academic Press, New York and London, pp 499—541

Lundberg, M., *Söremark, R.* & *Thilander, H.* 1965. Gamma-ray spectrometric analysis of some elements in coronal dentine of unerupted (impacted) human teeth. Odontologisk Revy 16, 97—100

Lux, H. 1938. Die Häufigkeit der seltenen Erden in Tierknochen. Z. anorg. allg. Chem. 240, 21—30

Manly, R. S. & *Bibby, B. G.* 1949. Substances capable of decreasing the acid solubility of tooth enamel. J. Dent. Res. 28, 160—171

Nixon, G. S. 1969. Trace element content of the hard dental tissues and dental plaque. Caries Res. 3, 60—74

Retief, D. H., *Cleaton-Jones, P. E.*, *Turkstra, J.* & *De Wet, W. J.* 1971. The quantitative analysis of sixteen elements in normal human enamel and dentine by neutron activation analysis and high-resolution gamma spectrometry. Arch. Oral Biol. 16, 1257—1267

Shresta, B. M., *Mundorff, S. A.* & *Bibby, B. G.* 1972. Enamel dissolution: I. Effects of various agents and titanium tetrafluoride. J. Dent. Res. 51, 1561—1566

Søremark, R. & *Samsahl, K.* 1962. Analysis of inorganic constituents in dental calculus by means of neutron activation and gamma-ray spectrometry. J. Dent. Res. 41, 596—602

Søremark, R. & *Samsahl*, K. 1964. Gamma-ray spectrometric analysis of the concentrations of Cr, Ag, Fe, Co, Rb, and Pt in normal human enamel. Acta Odont. Scand. 22, 255—259

Wester, P. O. 1964. *Concentration of 24 trace elements in human heart tissue determined by neutron activation analysis.* Report AE-146. AB Atomenergi, Stockholm