

# The concentrations of Pb, Cu, Co and Ni in extracted permanent teeth related to donors' age and elements in the soil

ROLF LAPPALAINEN & MATTI KNUUTTILA

Institute of Dentistry, University of Kuopio, Kuopio, Finland

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Extracted permanent molars, premolars or incisors were collected from 89 subjects aged 8 – 67 years living in a rural area of eastern Finland where large scale geochemical analyses of the soil have been carried out. A sample of 89 teeth was analyzed for Pb, Cu, Co and Ni by atomic absorption spectro-photometry. The concentration of Pb increased with age in the dentin but not in the enamel. The concentrations of Cu, Co and Ni remained unaltered with age. The mean concentrations ( $\mu\text{g/g}$ ) of Cu in the dentin were 9.1, 8.6 and 7.2 when the mean concentrations ( $\mu\text{g/g}$ ) in the soil were over 150, 50 – 150 and under 50, respectively. The concentration of Cu in the enamel was not dependent on the content in the soil. The mean concentrations ( $\mu\text{g/g}$ ) of Co and Ni were higher in the enamel (26.3 and 43.8) than in the dentin (13.8 and 31.4).

*Key-words:* Trace elements; enamel; dentin

Rolf Lappalainen, Institute of Dentistry, University of Kuopio, P.O.B. 138, SF-70101 Kuopio 10, Finland

The significance of deciduous teeth as an indicator of past and present exposures to Pb has been studied in connection with Pb poisoning (1), different environments of people (6, 10, 17, 19), as well as with domestic drinking water Pb concentrations (20). The concentration and accumulation of Pb in bones is complicated by the remodeling process (24). The concentration of Pb in the enamel of permanent teeth has not been found to increase with age and the Pb in the enamel has been found to be acquired prior to tooth eruption (3), possibly indicating early childhood exposure to Pb. Furthermore the age-related increase of Pb concentration in the dentin (5, 7), especially its

accumulation in the circumpulpal zone (5, 23), is well documented, but no detailed regression has been presented. The concentration of Pb in the entire dentin (root and crown) of permanent teeth may be suitable to reflect the exposure to Pb.

The concentrations of Cu, Co and Ni in deciduous or permanent teeth have been demonstrated in many previous studies (2, 4, 7, 11, 13, 14, 15, 18, 25, 26), but the relationship to age has been analyzed in young adults only by Derise & Ritchey (7). The possible correlation to the soil content has not been studied. The significance of Cu in bone metabolism and collagen crosslinking has been discussed by O'Dell (21). Co

has an important role in many enzyme reactions (28), and it interacts with Ni (9).

The aim of the present study was to determine the age-related Pb concentration in the enamel and the dentin of permanent teeth of a population in a rural area where the soil Pb content is known, and to compare it with the concentration of Pb in the teeth of an urban population. The relationship between the enamel and dentin concentrations of Cu, Co and Ni and those of the soil was also studied in the rural population, the inhabitants having spent most of their lives in the same area.

#### MATERIAL AND METHODS

The Geochemistry Department of the Geological Survey of Finland had carried out the geochemical mapping of the soil in the area from which the teeth were selected. The material of the soil samples was mostly glacial till, derived from the nearby bedrock. The total concentrations of Cu, Pb, Co and Ni of the samples were analyzed by direct reading optical emission spectrometer (ARL Model 31 000 Lausanne, Switzerland). The map (Fig. 1) based on 17 779 samples (6 samples per km<sup>2</sup>) was reconstructed from these data especially for this study. The distribution of Pb, Co and Ni varied so little that it was not possible to construe different soil concentrations.

The teeth from which all the analyses were made consisted of 89 permanent molars, premolars or incisors free of restorations and dental caries. The age of the donors varied from 8 to 67 years (Fig. 2 A). The teeth were collected from people who had lived most of their lives in the rural areas of eastern Finland.

The concentration of Pb was also determined from 50 dentin and enamel

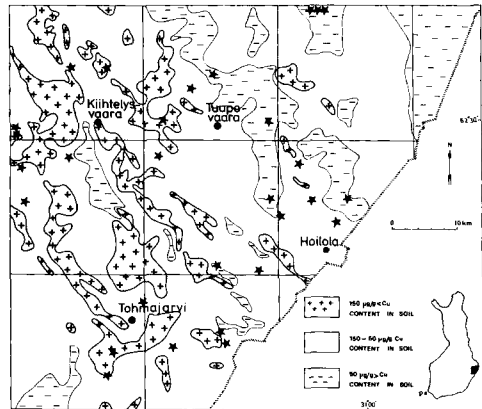


Fig. 1. Geochemical map of an eastern part of Finland presenting three different concentration levels of Cu in the soil. Location of villages from which the teeth were collected are indicated by stars (★) and four major villages by closed circle (●).

samples collected from people living in the town of Kuopio. The teeth were washed with distilled water and the enamel was separated from the dentin as described by Derise et al. (7). Enamel and dentin samples (500 mg) were dried at 105°C to constant weight, powdered and ashed with a solution of HCL and HNO<sub>3</sub> (2 : 1), and the volume was adjusted to 10 ml as described by Lappalainen & Knuuttila (15). The concentrations of Pb, Cu, Co and Ni were measured with a Perkin-Elmer Atomic absorption spectrophotometer, Model 372 (Perkin-Elmer Corporation, Norwalk, Connecticut, USA) according to the manufacturer's directions. The absorbances were registered by a recorder (REC 1, Pharmacia Fine Chemicals, Switzerland). Mean percent recoveries in repeated assays of the dentin samples were: Pb 96.0; Cu 95.2; Co 104.2; Ni 106.0.

The significances were tested with Student's t-test. The relationship between age and Pb concentration was evaluated using unweighted linear regression analysis. The regression coefficient for Pb was determined with 95 per cent confidence limits.

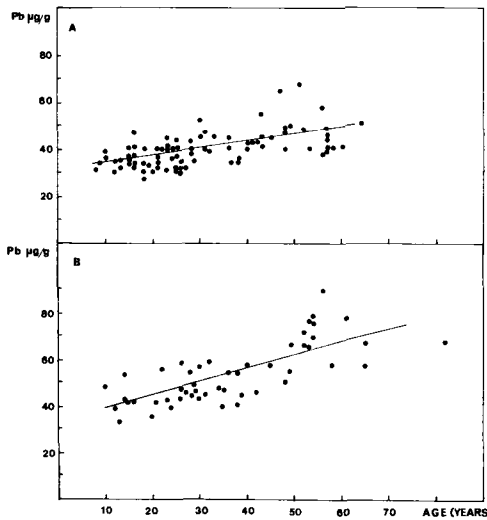


Fig. 2. Scattergram of concentration of Pb ( $\mu\text{g/g}$ ) in the dentin of permanent teeth versus age. The rural samples (A) came from the area presented in Fig. 1. The regression equation was  $Y = 30.8 + 0.30x$  ( $r = 0.58$ ). The urban samples (2 B) came from the town of Kuopio. The regression equation was  $y = 32.7 + 0.58x$  ( $r = 0.74$ ).

## RESULTS

### Lead

The concentration of Pb in the dentin of people living in the non-industrialized rural area, where the mean content of Pb in the soil was  $26 \mu\text{g/g}$  increased  $3.1 \mu\text{g/g}$  per 10 years (Fig. 2 A). The regression coefficients with 95 per cent confidence limits were 0.42 and 0.20. A slightly greater accumulation ( $5.8 \mu\text{g/g}$  per 10 years of age) was observed in the industrialized urban area (town of Kuopio) about 200 km from the rural area studied (Fig. 2 B). There was, however, no statistically significant difference between the rates of accumulations of the two populations. The mean concentration ( $\mu\text{g/g} \pm \text{S.D.}$ ) of Pb in the enamel was  $64.3 \pm 16.1$  in the rural sample and  $56.6 \pm 11.0$  in the urban sample. No age-related changes were found in the enamel. There were no statistically significant differences between males and females with regard

to the concentrations of Pb in dentin or enamel.

### Copper

The concentration of Cu in the dentin and in the enamel, when its concentration ( $\mu\text{g/g}$ ) in the soil was less than 50, 50 – 150 or over 150, is shown in Table 1. The concentration was significantly higher ( $p < 0.001$ ) in the enamel than in the dentin in all the three areas. The teeth from the area containing more than  $150 \mu\text{g/g}$  had a significantly higher ( $p < 0.001$ ) concentration of Cu in the dentin than those from the area containing less than  $50 \mu\text{g/g}$  soil-Cu. No such difference was found in the enamel. The concentrations of Cu were similar in the teeth of males and females and there were no age-related changes in either the enamel or the dentin.

### Nickel and cobalt

The concentrations of Ni and Co in the dentin and the enamel are shown in Table 1. There were significantly higher concentrations ( $p < 0.001$ ) of Ni and Co in the enamel than in the dentin. The mean concentrations of Ni and Co in the soil are also presented in Table 1. No significant correlation between the concentrations of Ni and Co and the age of donors could be observed.

## DISCUSSION

The age-related accumulation of Pb in the dentin, but not in the enamel, and the considerable variation of its concentration in the enamel justify the use of dentin-Pb in assessing long-term Pb exposure. Earlier studies (3, 5) have shown high concentrations of Pb in the surface of enamel and in circumpulpal

dentin. A possible increase of Pb concentration in the ameloblasts and the odontoblasts may be one reason for this accumulation. Besides the effect of Pb on the formation of organic matrix, it can also cause defects in lattices. Le-Geros & Quirolgico (16) have demonstrated an expansion of a-axis of apatite by Pb, possibly causing an increased solubility of apatite crystals. This is also supported by Brudevold et al. (3) who found that there was an association between high enamel Pb and high prevalence of caries.

Earlier studies (1, 6, 10, 17, 19, 20, 22) on deciduous teeth have shown some differences in Pb concentration due to environmental factors. The regression coefficients obtained in this study also indicate differences between rural and urban populations. A comparison of these results with earlier findings concerning whole teeth (15, 24, 29) is not relevant because of the effect of enamel Pb. The discrepancy between the results of different workers dealing with studies of Pb concentrations in whole permanent teeth (2, 15, 24, 29) may be explained by methodological differences. The concentrations presented in this study agree with those of Derise & Ritchey (8), although it was not possible to compare regression coefficients.

The methodological differences, e.g. the use of bromoform separation of enamel and dentin (25, 26), may also give lower concentrations of Cu (13, 14) compared to the concentrations found earlier (8, 11, 15) and those presented in this study. Fosse & Justesen (11) have shown geographical differences in Cu concentrations of deciduous teeth in Norway. These results indicate that the differences of Cu content in the soil are detectable in the dentin of permanent teeth. This may give some evidence that tooth-Cu reflects amount of absorption of this ion even if this has been doubted (11). Cu has been found to have an effect on crystallization of apatite (16) and collagen synthesis (12).

The concentration of Co was significantly higher in the enamel than in the dentin. This could be related to its role in the mineral phase, although by reacting with thiol groups of aminoacids and proteins, and by having an effect on erythropoiesis, lipid metabolism and vitamin B<sub>12</sub> (27) Co might primarily affect the organic matrix in the mineralization. The concentration of Ni was also higher in the enamel than in the dentin, indicating, most likely, the diadocia of Ni and Co, which is well known in mineralogy.

Table 1. The concentrations ( $\mu\text{g/g}$ ) of Cu, Ni and Co in permanent teeth of rural population related to the concentration in the soil

| Trace element | Content in the soil $\mu\text{g/g}$ | Dentin         |                  |             | Enamel         |                 |              |
|---------------|-------------------------------------|----------------|------------------|-------------|----------------|-----------------|--------------|
|               |                                     | No. of samples | Mean $\pm$ S. D. | Range       | No. of samples | Mean $\pm$ S.D. | Range        |
| Cu            | 20 - 50                             | 20             | 7.2 $\pm$ 1.4    | 9.9 - 4.8   | 88             | 12.3 $\pm$ 3.8  | 28.5 - 5.7   |
|               | 50 - 150                            | 26             | 8.6 $\pm$ 2.1    | 14.2 - 5.0  |                |                 |              |
|               | 150 $\rightarrow$                   | 19             | 9.1 $\pm$ 2.2    | 16.9 - 6.6  |                |                 |              |
| Ni            | mean 62.6                           | 89             | 31.4. $\pm$ 4.1. | 44.1 - 23.0 | 88             | 43.8 $\pm$ 13.4 | 74.4. - 20.8 |
| Co            | mean 18.5.                          | 89             | 13.8 $\pm$ 2.2.  | 22.0 - 10.0 | 88             | 26.3 $\pm$ 9.1  | 42.4 - 14.4  |

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## REFERENCES

- Altshuller, L.F., Halak, D. B., Landing, B. H. & Kehoe, R.A. Deciduous teeth as an index of body burden of lead. *J. Pediat.* 1962, 60, 224 – 229
- Attramadal, A. & Jonsen, J. The content of lead, cadmium, zinc and copper in deciduous and permanent teeth. *Acta Odontol. Scand.* 1976, 34, 127 – 131
- Brudevold, F., Aasenden, R., Srinivasian, B. N. & Bakhos, Y. Lead in enamel and saliva, dental caries and the use of enamel lead biopsies for measuring past exposure to lead. *J. Dent. Res.* 1977, 56, 1165 – 1171
- Brudevold, F. & Steadman, T. A study of copper in human enamel. *J. Dent. Res.* 1955, 34, 209 – 216
- Brudevold, F., Steadman, T. & Smith, F.A. Inorganic and organic components of tooth structure. *N.Y. Acad. Sci.* 1960, 85, 125 – 217
- Burde, B. & Shapiro, I.M. Dental lead, blood lead and pica in urban children. *Arch. Environ. Health* 1975, 30, 281 – 284
- Derise, N.L. & Ritchey, S.J. Mineral composition of normal human enamel and dentin and the relation of composition to dental caries. II Microminerals. *J. Dent. Res.* 1974, 53, 853 – 858
- Derise, N.L., Ritchey, S. J. & Furr, A.K. Mineral composition of normal human enamel and dentin and the relation of composition to dental caries: I Macrominerals and comparison of methods of analyses. *J. Dent. Res.* 1974, 53, 847 – 852
- Fiedler, H., Hoffman, H.D. Über die Wirkung von Nicklel (II) – L-glutamat und versiedenen Kobaltkomplexen auf das Verhalten einigen Lipidkomponenten bei Kaninchen. *Acta Biol. Med. Ger.* 1970, 25, 389 – 398
- Fosse, G. & Justesen, N.-P.B. Lead in deciduous teeth of Norwegian children. *Arch. Environ. Health* 1978, 33, 166 – 175
- Fosse, G. & Justesen, N.-P. B. Zinc and copper in deciduous teeth of Norwegian children. *Intern. J. Environ. Studies* 1978, 13, 19 – 34
- Harris, E. D., Gonnerman, W.A., Savage, J. E. Connective tissue amine oxidase II Purification and partial characterisation of lysyl oxidase from chic aorta. *Biochim. Biophys. Acta* 1974, 332 – 344
- Helle, A. & Haavikko, K. Macro- and micro-mineral levels in deciduous teeth from different geographical areas corrected with caries prevalence. *Proc. Finn. Dent. Soc.* 1977, 73, 87 – 98
- Lakomaa, E. & Rytömaa, I. Mineral composition of enamel and dentin of primary and permanent teeth in Finland. *Scand. J. Dent. Res.* 1977, 85, 89 – 95
- Lappalainen, R. & Knuutila, M. The distribution and accumulation of Cd, Zn, Pb, Cu, Co, Ni, Mn and K in human teeth from five different geological areas of Finland. *Arch. Oral Biol.* 1979, 24, 363 – 368
- LeGeros, R.Z. & Quirolgico, G. Trace elements: Their effects on the crystal growth of apatites. *J. Dent. Res.* 1977, 56, (Special issue A), A52
- Lockeretz, W. Lead content of deciduous teeth of children in different environments. *Arch. Environ Health* 1975, 30, 583 – 587
- Losee, F.L., Curzon, M.E.J. & Little, M.F. Trace element concentrations in human enamel. *Arch. Oral Biol.* 1974, 19, 467 – 470
- Mackie, A.C., Stephens, R. & Townshend, A. Tooth lead levels in Birmingham children. *Arch. Environ. Health* 1977, 32, 178 – 185
- Moore, M.R., Campbell, B.C., Meredith, P.A. Beattie, A.D., Goldberg, A. & Campbell, D. The association between lead concentrations in teeth and domestic water lead concentrations. *Clin. Chem. Acta* 1978, 87, 77 – 83
- O'Dell, B.L. Biochemistry of copper. In Burch & Sullivan (Eds) Symposium on trace elements. *Med. Clin. North Am.* 1976, 60, 687 – 703
- Rytömaa, I. & Tuompo, H. Lead levels in deciduous teeth. *Naturwiss.* 1974, 61, 363
- Shapiro, I.M., Dobkin, B., Tuncay, O.C. & Needleman, H.L. Lead levels in dentine, and circum-pulpal dentine of deciduous teeth of normal and lead poisoned children. *Clin. Chim. Acta* 1973, 46, 119 – 123
- Strehlow, C.D. & Kneip, T.J. The distribution of lead and zinc in the human skeleton. *Am. Ind. Hyg. Assoc. J.* 1969, 30, 372 – 378
- Söremark, R. & Samshal, K. Gamma-ray spectrometric analysis of elements in normal human enamel. *Arch. Oral Biol.* 1961, 6, 275 – 283
- Söremark, R. & Samshal, K. Gamma-ray spectrometric analysis of elements in normal human dentine. *J. Dent. Res.* 1962, 41, 603 – 606
- Taylor, A. & Marks, V. Cobalt. A review. *J. Human Nutr.* 1978, 32, 165 – 177
- Underwood, E.J. Cobalt. *Nutrit. Rev.* 1975, 33, 65 – 69
- Wilkinson, D.R. & Palmer, W. Lead in teeth as a function of age. *Int. Lab.* 1975, 41 – 46