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Differential diagenesis of strontium in archaeological human dental tissues

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Abstract

The investigation of prehistoric human migration from the measurement of Sr-isotope ratios within preserved tissue is critically dependent on the preservation of biogenic Sr. A number of recent studies have involved isotope ratio measurements on samples of archaeological tooth and bone, but doubt remains as to the extent of diagenesis in various skeletal tissues and the effectiveness of procedures designed to decontaminate them. We have compared Sr abundance and isotope ratios in enamel and dentine from archaeological teeth in order to assess the integrity of the biogenic Sr signal preserved within the respective tissues. We conclude that enamel appears, in most cases, to be a reliable reservoir of biogenic Sr, but that dentine, and by implication bone, is not. The diagenesis of dentine is highly variable even between burials within a single site. For the majority of teeth, dentine diagenesis was not simply by addition of soil-derived Sr, but involved substantial, sometimes complete, turnover of the original biogenic material. We suggest that, for most of the samples investigated, current decontamination techniques may not have been effective in isolating biogenic Sr from dentine. Similar considerations are likely to apply to archaeological and fossil bone, but the possibility arises to use dentine and enamel measurements to assess the effectiveness of decontamination procedures which may then be used for bone.

Keywords

Strontium isotopes, diagenesis, human teeth, archaeology, migration.

1. Introduction

Strontium isotope measurements of archaeological skeletal tissues have recently been used with the objective of reconstructing migration amongst ancient people (Sealy *et al.*, 1995; Ezzo *et al.*, 1997; Grupe *et al.*, 1997; Price *et al.*, 1998). The approach relies upon variations in the Sr isotope composition of rocks of different ages and lithologies. These ratios are reflected by associated soils. A proportion of soil Sr is available to plants and so enters the food chain (Sillen *et al.* 1998). As dietary Sr may substitute for Ca in the mineral phase of human skeletal tissues (Vukovic *et al.* 1998), their Sr isotope composition will reflect the geology of the place of residence during tissue formation or remodelling. Although Sr incorporation in human biological apatite *in vivo* is well accepted, its post mortem preservation in archaeological and fossil material is less certain.

Reconstructing migration requires biogenic Sr, derived from the diet *in vivo*, to be isolated from diagenetic Sr from the burial environment. Grupe *et al.* (1997) relied on the integrity of a biogenic Sr signal derived from compact femoral bone, but their approach has recently been criticised by Horn and Müller-Sohnius (1999) who believe that the data show increases in the Sr content of bone compared with enamel and shifts of Sr-isotope ratio which are characteristic of the diagenetic alteration of Sr. Grupe *et al.* (1999) have vigorously defended their methodology and the effectiveness of cleaning methods they, and others, have developed to remove diagenetic contamination prior to analysis. Here we compare the Sr content and isotopic composition of enamel and crown dentine from archaeological teeth in order to assess the integrity of these tissue types with respect to the preservation of biogenic Sr.

2. Sr incorporation in human skeletal tissue

The Sr isotope composition of human biological apatite measured after death reflects dietary signals from different periods of life depending on the tissue under consideration. The enamel of permanent teeth is formed over a short period, early in life, and incorporates trace elements from the childhood diet. It is not remodelled *in vivo* and is considered highly resistant to post-mortem diagenesis (Hillson 1996). Horn and Müller-Sohnius (1999) and Grupe *et al.* (1999) argue for some *in vivo* Sr uptake into enamel at its surface and at the enamel-dentine junction, but no such enrichment has been observed analytically (Brudevold and Söremark, 1967). We have found no evidence for Sr enrichment in enamel proximal to the tooth surface or the EDJ in high-sensitivity LA-ICP-MS studies of both ancient and modern teeth (Montgomery *et al.* In press). In any case, as Grupe *et al.* (1999) point out, only core enamel tissue is used for analysis.

Human core enamel appears to be a reliable reservoir of childhood Sr, however, the reconstruction of human migration also requires the measurement of Sr ingested in later life. Rib (Sealy *et al.*, 1995), femur (Grupe *et al.*, 1997; Price *et al.*, 1998) and other bones (Latkoczy *et al.* 1998), which are continually remodelled *in vivo*, have been used in this way. These tissues, however, are known to be subject to diagenesis during burial and to require pre-treatment prior to analysis (Sealy *et al.*, 1991; Price *et al.*, 1992; Sillen and Sealy, 1995). Treatments involve washing or leaching in weak acids to remove diagenetic apatite which is considered more soluble than its physiologically constrained biogenic counterpart (Latkoczy *et al.* 1998). Procedures of this sort have been found to remove variable proportions of the Sr in fossil and archaeological bone samples prior to Sr-isotope analysis. Sealy *et al.* (1991) and Sillen and LeGeros (1991) found that their solubility profiling technique typically removed ~25-30% of the Sr from samples which were considered to preserve

biogenic material. Koch *et al.* (1992) used a variant of the same procedure to wash samples of fossil salmon bone and noted a decline in Sr/Ca ratios between early washes and final sample solutions of ~5-40%. Horn *et al.* (1994) used a similar pre-treatment to remove diagenetic Sr from a fossil stag's mandible. A weak acid leachate, which removed ~6% of the Sr from the sample, was of significantly different isotopic composition to the remaining material and considered diagenetic. Such procedures appear to remove diagenetic material, but doubt remains as to the integrity of the biogenic signal within the remaining (insoluble) proportion. The pre-treatment approach rests on an assumption that diagenetic Sr is largely additive and does not exchange to any significant extent with that ingested *in vivo* and preserved within the biogenic apatite.

The current study is primarily concerned with dental tissues. Both enamel and dentine have closely similar Sr levels *in vivo* (Underwood, 1977; Montgomery *et al.* In press). They also have common isotope ratios, regardless of changes in an individual's diet or place of residence, because both tissues form at essentially the same time (*in utero* or soon after birth for deciduous teeth and in early childhood for permanent teeth). Enamel is not remodelled after formation so that its Sr is retained throughout life. For dentine the situation is slightly more complex as the formation of secondary dentine *in vivo* provides a mechanism for some Sr incorporation into this tissue later in life. For the juveniles examined in this study, the accumulation of secondary dentine would have been negligible as the teeth analysed were formed only a short time before death. In adults the secondary dentine component can be minimised by careful sampling. In this study dentine samples were obtained from tooth crowns and tissues adjacent to the pulp cavity were carefully stripped and discarded to provide samples of predominantly primary dentine.

Here we present Sr abundance and isotope ratio data for enamel and dentine from a number of prehistoric, Romano-British and medieval individuals from the UK. These individuals were selected in the hope that at least some of them would prove to have enamel (childhood) Sr with an isotopic composition significantly different to that of their burial environment. From the considerations outlined above it is clear that such individuals would have had *in vivo* dentine Sr-isotope compositions closely similar to those of their enamel. We suggest that any differences which are now found to exist between the enamel and dentine samples obtained from individual archaeological teeth result from post-mortem diagenesis, primarily of the less stable dentine. Although dentine is not routinely measured in migration studies, it has a blood supply *in vivo* and a more porous structure than enamel making it a useful proxy for bone. The metabolism of Sr is known to be very similar for dentine and bone (Weiser *et al.* 1996).

3. Samples and methodology

Archaeological human tooth samples were obtained from four sites in the UK. Permanent teeth were obtained for one female (C) of Neolithic date and a Bronze Age male (E) buried in chalk soil at Monkton-up-Wimbourne, Dorset. Samples of both permanent and deciduous teeth were obtained from a further three juveniles (A, B & D) from the same burial group as the female. Four permanent teeth of Romano-British date were analysed. One (SK2A) derived from a female recovered from a soil-filled limestone sarcophagus at Mangotsfield near Bristol, three from two males (G318, G339) in chalk soil from a 4th century AD cemetery at Winchester. Finally, five permanent teeth from four medieval individuals buried in clay soil (B77, B89, B341 & B357) were analysed from the 12th-16th century AD Blackfriars cemetery in

Gloucester. Soil samples were taken from each of the sites. Aqueous (Millipore Alpha Q, <1ppb total heavy metal content) leaches of the soil samples were performed to extract soluble Sr representative of the burial environment. This is also considered representative of local food chain Sr in view of its bioavailability (Horn and Müller-Sohnius, 1999), although the relationship is not necessarily straightforward in regions of diverse geology (Sillen *et al.*, 1998).

Whole teeth were mechanically pre-cleaned and washed in water (Millipore Alpha Q). The surface enamel was mechanically removed to a depth exceeding 100µm using a tungsten carbide burr (DFS, Riedenberg, Germany) and discarded. A layer of dentine was similarly removed from the pulp cavity to ensure that secondary dentine, if present, was minimised. Core enamel and primary dentine samples were then separated using an acid-leached stainless steel dental saw and tungsten carbide burrs (DFS, Riedenberg, Germany). Samples were sealed in acid-leached teflon (Savillex, Minnetonka, MN) containers and transferred to clean (class 100, laminar flow) laboratories at the NERC Isotope Geosciences Laboratory (NIGL).

Samples were then washed in water (Millipore Alpha Q), followed by a 5 minute wash in acetone (teflon distilled) and final water rinse. Samples were weighed into pre-cleaned teflon beakers and washed for 7 minutes in ~1.5M Teflon-distilled HCl to remove surface any contamination acquired in handling. Tissue fragments were then dissolved in 16M Teflon-distilled HNO₃. Samples were spiked with ⁸⁴Sr and Sr isolated by anion exchange chromatography. Sr isotope ratios were measured on a MAT262 Thermal Ionisation Mass Spectrometer (Finnigan Corp., San Jose, CA) at NIGL. Samples were analysed using Ta filaments with a silica gel-phosphoric acid emitter. Results were corrected for fractionation monitored by repeat analysis of the NBS 987 standard which gave a mean ⁸⁷Sr/⁸⁶Sr value of 0.710200±32 (n=10, 2σ). Sr concentrations were determined using the isotope dilution method. The mean Sr blank was 300pg (range 33-400pg).

4. Results

⁸⁷Sr/⁸⁶Sr isotope ratio measurements of aqueous leachates of soils from the archaeological sites are presented in Table 1 and results for the enamel and dentine of each tooth are given in Table 2 together with Sr concentrations determined by the isotope dilution method. The Sr isotope compositions of all of the enamel samples measured are very significantly different to those of their burial environments. Dentine Sr isotope compositions are closer to those of the soils, but still significantly different from them.

A standard measure of the difference between the Sr isotope ratios of the tissues of interest and the soil of the burial environment is useful in order to compare teeth from different sites. Ezzo *et al.* (1997), following DePaolo and Wasserburg (1977), calculated the parameter ε⁸⁷Sr to express the difference between the ⁸⁷Sr/⁸⁶Sr ratios of their samples and those of the 'bulk earth'. In this case we are interested in a standard measure of the difference in ⁸⁷Sr/⁸⁶Sr, and therefore ε⁸⁷Sr, between dental tissue samples and the specific soils in which each was buried. We have therefore calculated the parameter Δε⁸⁷Sr where:-

$$\Delta\epsilon^{87}\text{Sr} = \left[\frac{R_t - R_s}{R_s} \right] 10^4 \quad (1)$$

where R_t is the measured $^{87}\text{Sr}/^{86}\text{Sr}$ ratio of the tissue of interest and R_s is the measured $^{87}\text{Sr}/^{86}\text{Sr}$ ratio of the burial soil. Calculated values of $\Delta\epsilon^{87}\text{Sr}$ for each tissue sample are given in Table 2 and the relationship between $\Delta\epsilon^{87}\text{Sr}$ and Sr concentration is illustrated in Figure 1.

Figure 1 shows that the dentine Sr concentration is consistently elevated with respect to the enamel in the archaeological teeth in contrast to modern samples. The highly significantly different Sr isotope ratios between the two tissues for most of the teeth is also apparent. Dentine Sr-isotope ratios are closer to the respective soil values and therefore have lower values of $\Delta\epsilon^{87}\text{Sr}$ in almost all cases. One exception, G318, has closely similar enamel and dentine strontium concentrations and isotope ratios suggesting that the tooth is essentially unaffected by diagenesis of either tissue. SK2A also has similar enamel and dentine Sr isotope ratios, but has a dentine Sr concentration more than three times that of the enamel. This tooth was in a very much poorer state of preservation than the others with substantial demineralisation of both tissues. We suggest diagenetic alteration of both enamel and dentine in this case. The great majority of samples have enamel Sr concentrations similar to those reported for modern samples not subject to burial post-mortem (Underwood 1977). This suggests that their enamel is a relatively reliable and stable reservoir of biogenic strontium with which dentine can be compared.

Examination of the $\Delta\epsilon^{87}\text{Sr}$ data (Table 2) indicates that one sample (357) has dentine which appears to be isotopically equilibrated with the soil ($\Delta\epsilon^{87}\text{Sr} = 0$). This suggests complete exchange of the biogenic Sr with soil-derived Sr as a result of diagenesis. Even in this tooth however, the enamel remains very significantly different from the soil and dentine ($\Delta\epsilon^{87}\text{Sr} = 21$), indicative of good preservation of biogenic strontium in this, more resistant, tissue. For most of the teeth, dentine Sr isotope ratios are systematically shifted towards soil values by diagenesis giving lower values of $\Delta\epsilon^{87}\text{Sr}$ in dentine than enamel. However, the great majority of the teeth also have significantly higher Sr concentrations in dentine than enamel so that much of the diagenetic strontium would appear to be additive. The extent to which diagenetic strontium in the dentine is additive, as opposed to exchanged with the original biogenic material, can be estimated by calculating the change from the original (enamel) ratio which would occur as a result of the addition of soil-derived Sr to achieve the measured dentine concentration.

$$R_{dp} = \frac{C_e R_e + (C_d - C_e) R_s}{C_d} \quad (2)$$

where R_{dp} is the predicted $^{87}\text{Sr}/^{86}\text{Sr}$ ratio of the dentine, C_e is the measured Sr concentration of the enamel, C_d is the measured Sr concentration of the dentine, R_e is the measured $^{87}\text{Sr}/^{86}\text{Sr}$ ratio of the enamel, and R_s is the measured $^{87}\text{Sr}/^{86}\text{Sr}$ ratio of the soil.

Predicted Sr isotope ratios and values of $\Delta\epsilon^{87}\text{Sr}$ are given in Table 3 and plotted in Figure 2. Some teeth have measured differences between enamel and dentine $\Delta\epsilon^{87}\text{Sr}$ which are slightly less than those predicted, probably because the original (biogenic) dentine Sr concentrations were a little higher than the estimate based on the enamel composition. In these cases, diagenetic Sr appears to have been essentially additive with little or no turnover of the biogenic Sr within the dentine. For most samples however, the measured differences between the enamel and dentine are

greater than those which would have occurred as a result of adding diagenetic (soil-derived) Sr. For these samples diagenesis of the dentine appears to have involved partial replacement of the biogenic Sr with soil-derived material. The data may be used to estimate the magnitude of turnover (T) of the biogenic Sr with diagenetic material in the dentine:-

$$T = \left[\frac{R_d - R_{dp}}{R_s - R_{dp}} \right] 10^2 \quad (3)$$

where T is the proportion (%) of the original (biogenic) Sr in the dentine replaced by diagenetic Sr and R_d is the measured $^{87}\text{Sr}/^{86}\text{Sr}$ ratio of the dentine.

This can be simply rearranged to give the proportion of the total dentine Sr which is diagenetic (D) and which would have to be removed by sample pre-treatment prior to analysis (Table 3). Turnover of the biogenic Sr is highly variable. Only six samples, all from chalk burials, were essentially unaffected ($T \leq 4$), but only one of these (G318) appeared not to have been subject to substantial accumulation of diagenetic Sr. Diagenetic Sr accounted for ~34-79% of total dentine Sr for the other five. These estimates may be compared with the ~25-30% reduction in Sr typically reported for acid sample pre-treatments (Sealy *et al.* 1991). A further nine samples, from both chalk and clay soils, appear to have exchanged ~15-76% of their biogenic Sr with that derived from the soil. This implies proportions of diagenetic Sr in the dentine of up to ~95%. Two samples (357 and SK2A) would appear to have been subject to complete exchange with diagenetic Sr making recovery of any biogenic material impossible.

5. Conclusions

We conclude that diagenetic alteration of Sr in the dentine of the archaeological teeth examined was common but highly variable, bearing no clear relationship with tooth type or burial conditions. Only one tooth appeared to be essentially unaffected by diagenesis. A further five appear to have preserved their original, biogenic, Sr but to have accumulated 34-79% of their current dentine Sr from the burial environment post mortem. It is likely that the remaining samples exchanged varying proportions (15-100%) of their biogenic dentine Sr with soil derived material. Unlike enamel, dentine would appear to be unreliable as a reservoir of biogenic Sr. We suggest that acid pre-treatments of the sort discussed by Sealy *et al.* (1991) and Grupe *et al.* (1999) would not have been successful in removing diagenetic material for the majority of the dentine samples we have investigated. We believe the same considerations are likely to apply to archaeological and fossil bone.

On a positive note, it seems likely that the measurement of dentine Sr abundance and isotopic composition could be used to monitor the effectiveness of sample pre-treatment procedures providing there has been only addition and not significant diagenetic replacement of the biogenic Sr. We would expect that a sample pre-treatment regime which successfully retrieved dentine ratios matching those of enamel could then be used on cortical bone from the same individual to isolate biogenic Sr ingested in later life.

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Figure 1.

Sr concentrations plotted against $\Delta\epsilon^{87}\text{Sr}$ for enamel (open symbols) and dentine (filled symbols) samples. Diamond - Mangotsfield; Triangles - Blackfriars; Squares - Monkton-up-Wimbourne; Circles - Winchester. Aqueous soil leaches from the four sites are represented by the dashed line at $\Delta\epsilon^{87}\text{Sr} = 0$. Measurement errors, $0.45 \epsilon^{87}\text{Sr}$ units (2σ), are smaller than symbols.

Figure 2.

Measured $\Delta\epsilon^{87}\text{Sr}$ values for enamel and measured and predicted values for the dentine. Predicted values of dentine $\Delta\epsilon^{87}\text{Sr}$ relate to the change expected from the diagenetic addition of soil-derived Sr. Aqueous soil leaches from the four sites are represented by the dashed line at $\Delta\epsilon^{87}\text{Sr} = 0$. Measurement errors, $0.45 \epsilon^{87}\text{Sr}$ units (2σ), are smaller than symbols.

Table 1

TIMS Sr isotope ratio measurements of aqueous leachates of soils from the archaeological sites. External reproducibility of $^{87}\text{Sr}/^{86}\text{Sr}$ from repeat analysis of the NBS 987 standard was ± 0.000032 (2σ).

Site	$R_s; ^{87}\text{Sr}/^{86}\text{Sr}$
Blackfriars	0.70998
Mangotsfield	0.71045
Monkton	0.70765
Winchester	0.70756

Table 2

Sr isotope ratio measurements by TIMS and Sr concentration data. External reproducibility of $^{87}\text{Sr}/^{86}\text{Sr}$ from repeat analysis of the NBS 987 standard was ± 0.000032 (2σ), which gives an error of ± 0.45 (2σ) in $\epsilon^{87}\text{Sr}$ units. Sr concentrations were determined using the isotope dilution method. The mean Sr blank was 300pg (range 33-400pg).

Site	Sample	Type	Age at death	Enamel			Dentine		
				C_e ; Sr (ppm)	R_e ; $^{87}\text{Sr}/^{86}\text{Sr}$	$\Delta\epsilon^{87}\text{Sr}$	C_d ; Sr (ppm)	R_d ; $^{87}\text{Sr}/^{86}\text{Sr}$	$\Delta\epsilon^{87}\text{Sr}$
Blackfriars	77	P ₁	17-25	65.89	0.71197	28.1	184.69	0.71023	3.6
Blackfriars	89(1)	P ₁	13-14	70.05	0.70888	-15.5	243.98	0.70971	-3.8
Blackfriars	341R	P ₁	15-20	58.54	0.71423	60.0	242.88	0.71026	4.0
Blackfriars	341L	P ₁	15-20	56.11	0.71429	60.7	273.03	0.71019	3.0
Blackfriars	357	P ₂	17-25	56.37	0.71146	21.0	386.56	0.70997	0.0
Mangotsfield	SK2A	P ₂	Adult	36.76	0.70995	-7.0	117.77	0.71008	-5.2
Monkton	A	M ¹	~5	71.44	0.70878	16.0	207.34	0.70782	2.5
Monkton	A	c ¹	~5	74.68	0.70955	27.0	230.24	0.70788	3.3
Monkton	B	P ¹	8-9	56.80	0.70928	23.1	184.28	0.70789	3.5
Monkton	B	c ¹	8-9	103.25	0.70844	11.3	247.63	0.70790	3.7
Monkton	C	P ²	30-35	54.90	0.71007	34.3	161.98	0.70866	14.3
Monkton	D	C ¹	9-10	54.50	0.70897	18.7	249.59	0.70792	3.9
Monkton	D	c ¹	9-10	72.19	0.70849	11.9	281.99	0.70809	6.3
Monkton	E	P ₁	Adult	84.88	0.70804	5.5	198.97	0.70781	2.3
Winchester	G318	P ¹	Adult	56.35	0.70828	10.1	58.97	0.70834	10.9
Winchester	G339A	M ³	Adult	93.44	0.70915	22.4	110.91	0.70862	14.9
Winchester	G339B	P ₂	Adult	79.18	0.70931	24.7	127.30	0.70863	15.1

Table 3

The predicted Sr-isotope ratios and $\Delta\epsilon^{87}\text{Sr}$ values of dentine from the addition of soil derived strontium assuming that enamel and dentine Sr concentrations were equal *in vivo*. Measured values of $\Delta\epsilon^{87}\text{Sr}$ for dentine are generally smaller than the values predicted. The data can therefore be used to estimate the proportion (T) of the original (biogenic) Sr replaced by diagenetic Sr in the dentine and the proportion (D) of the total dentine Sr which is diagenetic and which would need to be removed prior to analysis.

Sample	Predicted dentine $R_{dp}; ^{87}\text{Sr}/^{86}\text{Sr}$	Predicted dentine $\Delta\epsilon^{87}\text{Sr}$	Turnover T (%)	Diagenetic Sr D (%)
77	0.71069	10.0	64	87
89(1)	0.70966	-4.4	15	76
341R	0.71100	14.5	72	93
341L	0.71086	12.5	76	95
357	0.71019	3.1	100 ^a	100
SK2A	0.71029	-2.2	100 ^b	100
A (perm)	0.70803	5.5	55	84
A (decid)	0.70826	8.7	62	88
B (perm)	0.70815	7.1	50	85
B (decid)	0.70798	4.7	22	67
C (perm)	0.70847	11.6	0 ^c	66
D (perm)	0.70793	4.1	4	79
D (decid)	0.70786	3.1	0 ^c	74
E (perm)	0.70781	2.4	2	58
G318	0.70825	9.7	0 ^c	4
G339A	0.70890	18.9	21	34
G339B	0.70865	15.4	2	39

^a calculated value of T = 101%.

^b sample thought to be subject to enamel diagenesis and T estimated at 100%.

^c smaller than predicted changes in dentine Sr isotope ratios leading to negative calculated values of T.