

Assessing the Preservation of Biogenic Strontium in Fossil Bones and Tooth Enamel

K. A. HOPPE,^{a*} P. L. KOCH^b AND T. T. FURUTANI^c

^a Division of Ecosystem Sciences, University of California, Berkeley, California, USA

^b Department of Earth Sciences, University of California, Santa Cruz, California, USA

^c Department of Science and Mathematics, North Seattle Community College, Seattle, Washington, USA

ABSTRACT Analyses of the strontium isotope ratio ($^{87}\text{Sr}/^{86}\text{Sr}$) of vertebrate fossils can provide information about palaeobiological attributes such as habitat use and movement patterns. Diagenetic contaminants can alter the $^{87}\text{Sr}/^{86}\text{Sr}$ ratio of fossils, however, complicating palaeobiological interpretations. Several pretreatment protocols have been developed to separate diagenetic contaminants from biogenic Sr. While these methods can remove some diagenetic Sr, it has not been shown that any technique removes all contamination. The extent to which pretreatment removes diagenetic Sr can be quantified through analysis of the $^{87}\text{Sr}/^{86}\text{Sr}$ ratios of fossil marine mammal bones and teeth buried in sediments with non-marine diagenetic $^{87}\text{Sr}/^{86}\text{Sr}$ signatures. To do this, we examined Holocene seals recovered from archaeological sites in Greenland and California, as well as a Miocene whale from Maryland. Our results demonstrate that although pretreatment eliminated some contaminants from bone, a large percentage (up to 80%) of diagenetic Sr remained after treatment. In contrast, pretreatment does appear to remove nearly all ($\geq \sim 95\%$) diagenetic Sr from tooth enamel. Copyright © 2003 John Wiley & Sons, Ltd.

Key words: diagenesis; bone; enamel; strontium; Sr-87/Sr-86; fossils; pinnipeds

Introduction

Analysis of the strontium (Sr) component of vertebrate skeletal material has several applications in palaeoecological and palaeobiological research. The Sr isotope composition (i.e., $^{87}\text{Sr}/^{86}\text{Sr}$ ratio) of an animal's bones and teeth directly reflects the $^{87}\text{Sr}/^{86}\text{Sr}$ ratio of its environment (Lenihan *et al.*, 1967). Thus, an animal's $^{87}\text{Sr}/^{86}\text{Sr}$ ratio can be used to reconstruct patterns of habitat use (Elliott *et al.*, 1998) and migration (Sealy *et al.*, 1995; Hoppe *et al.*, 1999). Also, since Sr becomes depleted relative to calcium at higher levels in a foodweb, Sr/Ca ratios can be used to reconstruct

trophic levels and palaeodietary patterns (Sillen & Kavanaugh, 1982; Schoeninger, 1985).

While such applications make analyses of biogenic Sr potentially useful, interpreting the Sr signal recovered from fossil (or subfossil) material is often complicated by the fact that biological tissues undergo post-mortem alteration. During life, Sr substitutes for Ca in bones and teeth, both of which are composed of a carbonated hydroxyapatite ($\text{Ca}_9[(\text{PO}_4)_{4.5}(\text{CO}_3)_{1.5}](\text{OH})_{1.5}$) (Driessens & Verbeek, 1990). Bone is a relatively porous material composed of tiny hydroxyapatite crystals intermixed with $\sim 30\%$ (dry weight) organic matter. Tooth enamel is essentially non-porous and composed of relatively large crystals that include only minor amounts ($< 2\%$) of organic matter (Driessens & Verbeek, 1990). These structural differences result in differences in the

* Correspondence to: Division of Ecosystem Sciences, University of California, Berkeley, CA 94720-3110, USA.
e-mail: khoppe@nature.berkeley.edu

susceptibility of bone and enamel to post-mortem alteration (Sillen, 1986; Tuross *et al.*, 1989; Ezzo, 1992; Lee-Thorp & van der Merwe, 1991). Likewise, different chemical elements within each tissue alter at different rates (Price, 1989; Ezzo, 1992). Thus, the durability of each element must be assessed separately for each material analysed.

The durability of biogenic Sr in fossil and subfossil materials, especially bone, has been the subject of heated debate. Nelson *et al.* (1986) demonstrated that subfossil bone could be contaminated with diagenetic Sr. They analysed bones from Holocene marine mammals (which had a biogenic $^{87}\text{Sr}/^{86}\text{Sr}$ signal similar to that of the ocean) that had been preserved in terrestrial sediments (which had a non-marine diagenetic $^{87}\text{Sr}/^{86}\text{Sr}$ ratio) and concluded that biogenic Sr in bones was rapidly and completely replaced by diagenetic Sr. If, as suggested subsequently, diagenetic Sr is concentrated in secondary mineral phases that differ in solubility from biogenic hydroxyapatite (Sillen, 1986), it should be possible to isolate biogenic Sr by subjecting skeletal material to a series of sequential leaches in weak acid solutions (Sillen, 1986). Sillen (1986) demonstrated that the percent recovery of biogenic Sr improved when fossil bone was pretreated with a 0.1 N buffered acetic acid (pH = 4.5) solution for 25 sequential 1-min leaches. Additional work suggested that bones may retain biogenic Sr, at least on Holocene-Pleistocene time scales (Sealy *et al.*, 1991; Price *et al.*, 1994; Sillen & Sealy, 1995), although recovery of biogenic Sr in bones may not be possible on longer time scales (Koch *et al.*, 1992; Elliott *et al.*, 1998).

At present, there is no quantitative evidence that pretreatment protocols completely isolate biogenic and diagenetic Sr. Furthermore, a uniform pretreatment protocol has not yet been established. Current protocols differ in details such as acid strength, sample reaction time, and even the component that is analysed (e.g., the Sr in leachates versus the Sr in powder residues). Finally, while mineralogical considerations suggest that tooth enamel should retain biogenic Sr better than bone (Lee-Thorp & van der Merwe, 1991; Koch *et al.*, 1997; Budd *et al.*, 2000), few workers have examined the effects of pretreatment on the $^{87}\text{Sr}/^{86}\text{Sr}$ ratios of enamel.

We quantified the amount of diagenetic Sr removed from bone and enamel by sequential leaches of weak acetic acid (0.1 N buffered and unbuffered solutions). We analysed subfossil material from Holocene marine mammals (pinnipeds) that had been preserved in terrestrial middens in West Greenland and California as well as a Miocene whale from Maryland. Initial biogenic $^{87}\text{Sr}/^{86}\text{Sr}$ ratios of skeletal materials were estimated from the ratios for contemporaneous seawater. Diagenetic $^{87}\text{Sr}/^{86}\text{Sr}$ ratios were estimated from measurements of the soluble Sr in the surrounding matrix (when available) or the $^{87}\text{Sr}/^{86}\text{Sr}$ ratios of local bedrocks. In addition, the mineralogy and crystallinity of skeletal materials were assessed using infrared spectrometry or X-ray diffraction.

Materials

Site number CA-MNT-234, California

We analysed subfossil material from fur seals (*Callorhinus ursinus*) recovered from a Holocene midden near Moss Landing in Monterey County, California (USA). Samples consisted of cortical bone taken from three limb bones, as well as enamel from three canine teeth (detailed sample description included in Hoppe, 1999). The surrounding matrix consisted of unconsolidated soil mixed with sand. Radiocarbon dates of associated shell material ranged between $1,630 \pm 70$ and $3,520 \pm 60$ radiocarbon years before present (C-14 yrs BP) (Breschini *et al.*, 1995). Samples are housed in the collections of the Moss Landing Marine Laboratory.

West Greenland

Samples were obtained from two separate archaeological excavations in west Greenland. One set of samples consists of two seal (*Phoca* sp.) bones excavated at a site on Godthåbs Fjord in southern west Greenland. These samples correspond to bones 743 and 747 analysed by Nelson *et al.* (1986), which have been dated to 920 ± 50 and 870 ± 40 C-14 yrs BP respectively. The other set of Greenland samples consists of seal (*Phoca*

sp.) teeth collected in 1982 from the Dorset Palaeoeskimo cultural levels (370 BC to 205 AD) at Qajâ, an Eskimo site in Jacobshavn Icefjord (Mohl, 1986). In order to obtain enough enamel to analyse, we combined material from several isolated teeth. Each sample consists of material from teeth that were recovered from a single level within the excavation and should have experienced the same diagenetic microenvironment. Bedrock at the site is Archaean gneiss from the Nagssugtoqidian orogenic belt (Kalsbeek & Taylor, 1999).

Pope's Creek, Maryland

Fragments of rib and one tooth were obtained from the skeleton of an early sperm whale (*Orycterocetus* sp.), specimen number 416221, recovered from a silty-sandstone unit of the Pope's Creek Sand Member of the Calvert Formation near Pope's Creek, Charles County, Maryland (USA). Biostratigraphic correlation suggests that this unit is late early Miocene in age (~16 to 18 million yrs BP) (Andrews, 1984). This specimen is housed in the National Museum of Natural History, Smithsonian Institution.

Analytical methods

Samples of cortical bone and tooth enamel were collected and powdered using a dental drill, or chipped off and ground in an agate mortar and pestle. The crystallinity and mineralogy of bone samples were examined using Fourier transform infrared spectroscopy (FTIR) or X-ray diffraction. Infrared spectroscopy was carried out using a Bruker IFS-66v FTIR spectrometer equipped with a liquid-He cooled Si bolometer, a globar source, and a KBr beam splitter (Department of Earth Sciences, University of California, Santa Cruz). All spectra were measured from 2000 to 400 cm^{-1} and were reported with 4 cm^{-1} resolution. Crystallinity was quantified using the infrared splitting factor, which is a measure of the extent of splitting between two phosphate-ion absorption peaks at 693 and 565 cm^{-1} (Weiner & Bar-Yosef, 1990). X-ray diffraction

patterns were measured on a Norelco wide-range vertical goniometer, which generated X-rays with a Spellman 60KV solid-state generator and a Cu-anode X-ray tube (Department of Earth Sciences, University of California, Santa Cruz). Each sample was scanned from 10° to 60° 2θ with a step size of 0.02 2θ and a counting time of 1 s.

Although different studies have used different strengths of acetic acid to pretreat fossils, evidence suggests that exposure to strong (≥ 1.0 N) acetic acid can cause bones to recrystallize (Sillen & Sealy, 1995; Neilsen-Marsh & Hedges, 1997; Hoppe, 1999). Therefore we only tested the effectiveness of the two protocols that use weak (≤ 1.0 N) acetic acid. Samples from Greenland and a sample split of the Maryland whale bone were processed using methods described by Sillen (1986) (referred to here as protocol one); samples were treated for 1 min with 0.1 N acetic acid buffered to a pH of 4.5, and this process was repeated 24 times. Samples lost ~20 to 30% of their original weight during this pretreatment. Bone and enamel samples from California and Maryland were pretreated using a different protocol (referred to as protocol two). These samples were reacted with 0.1 N acetic acid using a ratio of 1.0 ml of acid for every ~20 mg of powder. Reactions were allowed to continue for 20 min, then leachates were collected and the remaining powder was rinsed with deionized water. Bone samples were subjected to a series of 12 sequential leaches. At this point $\geq 70\%$ of the weight of the original bone had been dissolved and the residual powders for Holocene bones consisted primarily of organic matter, indicating that almost all of the original hydroxyapatite had been dissolved. Enamel samples were subjected to only eight sequential leaches, due to their smaller initial size. In addition to skeletal material, we also analysed soil and matrix samples, which were treated overnight in a solution of 1.0 N nitric acid. They were then centrifuged and the leachate was extracted for analysis.

After pretreatment, all samples were dissolved in 2.5 N HCl and Sr was extracted by standard ion exchange chromatography (Walker *et al.*, 1989). Samples were measured on a VG354-S thermal ionization mass spectrometer (Department of Earth Sciences, University of California, Santa Cruz). All measurements were referenced to a

value of $^{87}\text{Sr}/^{86}\text{Sr} = 0.71025$ for the NBS 987 Sr standard. Precision for each analysis was $\leq \pm 0.00002$ (2σ) unless otherwise noted.

Results

All samples of untreated Holocene bone consisted primarily of hydroxyapatite and still contained most ($\sim 30\%$ to $\sim 25\%$) of their original organic matter. Some samples, however, displayed a slight increase in crystallinity relative to modern bone. In contrast, untreated samples from the Miocene whale differed in both mineralogy and crystallinity from modern materials. Infrared analyses of Miocene bone revealed that this sample had increased crystallinity, as evidenced by an increased splitting of phosphate absorption peaks. In addition, the disappearance of the collagen peaks at 1655 cm^{-1} , 1540 cm^{-1} , and 1460 cm^{-1} indicated that organic matter had been lost (Weiner & Bar-Yosef, 1990; Weiner *et al.*, 1993; Stiner *et al.*, 1995). Increased carbonate peaks indicated that this sample contained secondary carbonate minerals or a secondary carbonated hydroxyapatite phase (LeGeros & Tung, 1983; LeGeros & LeGeros, 1984; Weiner *et al.*, 1993; Wright & Schwarcz, 1996), while the presence of fluorapatite was indicated by the fact that the 606 cm^{-1} phosphate peak is slightly stronger than the 567 cm^{-1} peak (Weiner *et al.*, 1993). X-ray diffraction confirmed that this sample consisted primarily of hydroxy- and fluorapatite. The enamel sample displayed similar evidence of a fluorapatite, but increases in crystallinity and carbonate content were less pronounced. Infrared analyses of the residual powder from Miocene

bone revealed a marked reduction in the strength of its carbonate peak, indicating that a carbonate phase had been selectively dissolved.

To assess the degree of diagenetic alteration in each sample, we estimated original, biogenic $^{87}\text{Sr}/^{86}\text{Sr}$ ratios as well as the ratio of diagenetic Sr at each site. Since all of the skeletal materials we analysed come from marine mammals, the biogenic $^{87}\text{Sr}/^{86}\text{Sr}$ ratio of each sample should reflect the ratios of contemporaneous seawater. Modern pinnipeds that feed in offshore environments, such as the fur seals analysed from California (Burton & Koch, 1999), should have $^{87}\text{Sr}/^{86}\text{Sr}$ ratios equal to the average ratio of modern seawater (0.70920) (Table 1). Pinniped species that feed near shore in Greenland, such as harbour seals (*Phoca vitulina*), likewise display values similar to modern seawater, although some individuals have slightly elevated $^{87}\text{Sr}/^{86}\text{Sr}$ ratios (0.70927), perhaps due to use of brackish water habitats (Nelson *et al.*, 1986). Since average seawater $^{87}\text{Sr}/^{86}\text{Sr}$ ratios have not changed significantly in the last several thousand years (Hodell *et al.*, 1991), we would expect Holocene pinnipeds to display biogenic $^{87}\text{Sr}/^{86}\text{Sr}$ ratios similar to their modern counterparts. Uncertainties in the biostratigraphic assignment of the Miocene whale result in uncertainties on estimates of its initial biogenic $^{87}\text{Sr}/^{86}\text{Sr}$ ratio, though it most likely ranged between 0.70861 and 0.70871 (Hodell *et al.*, 1991).

The $^{87}\text{Sr}/^{86}\text{Sr}$ ratios of diagenetic Sr at the California and Maryland sites were estimated by analysis of the matrix from each site (Table 1). Since Greenland matrix samples were not available, we estimated diagenetic $^{87}\text{Sr}/^{86}\text{Sr}$ ratios at these sites using published $^{87}\text{Sr}/^{86}\text{Sr}$ ratios of local

Table 1. Measured $^{87}\text{Sr}/^{86}\text{Sr}$ ratios of matrix samples and estimated $^{87}\text{Sr}/^{86}\text{Sr}$ ratios of biogenic and diagenetic Sr at each site

Site	Matrix	Biogenic Sr	Diagenetic Sr
Moss Landing (CA)	0.70884	0.70920	0.70884
Godthåbs Fjord (Greenland)		0.70926 ¹	0.75365–0.75376 ³
Qajâ (Greenland)		0.70926 ¹	$\geq 0.72476^4$
Pope's Creek (MD)	0.70911	0.70861–0.70871 ²	0.70911

¹ Value for modern harbour seals (*Phoca vitulina*) from southwest Greenland (Nelson *et al.*, 1986).

² The $^{87}\text{Sr}/^{86}\text{Sr}$ ratio of seawater during the late early Miocene (Hodell *et al.*, 1991).

³ Estimated based on $^{87}\text{Sr}/^{86}\text{Sr}$ ratio of first acid leach from sample 747 (this study) and bones from co-occurring terrestrial animals measured by Nelson *et al.* (1986).

⁴ Minimum $^{87}\text{Sr}/^{86}\text{Sr}$ ratio for Archean bedrock in the Disko Budt region (Kalsbeek & Taylor, 1999).

bedrocks. Assuming that the first acid leach from samples contains Sr from highly soluble diagenetic minerals (Sillen, 1986), the $^{87}\text{Sr}/^{86}\text{Sr}$ ratio of this leachate can provide a minimum estimate of the diagenetic $^{87}\text{Sr}/^{86}\text{Sr}$ ratio. In addition, terrestrial animals consume terrestrial plants, which in turn uptake Sr that is soluble in soil solutions, and thus provide an independent estimate of the isotopic composition of terrestrial Sr. The first leachates from the Godthåbs Fjord bone 747 had a value of 0.75365, which is similar to the highest $^{87}\text{Sr}/^{86}\text{Sr}$ ratio measured for co-occurring terrestrial animals (0.75376) by Nelson *et al.* (1986). This suggests that the first leachate consists primarily of diagenetic Sr. In contrast, the value for the first leachate from Qajâ enamel sample 1119 was similar to the corresponding value for untreated enamel (Figure 2) and differed from values measured for regional bedrock (Kalsbeek & Taylor, 1999). This suggests that the first acid leachate from the Qajâ enamel samples did not contain significant diagenetic Sr. We used the lowest values for regional bedrock as a minimum estimate for the diagenetic $^{87}\text{Sr}/^{86}\text{Sr}$ ratio at Qajâ.

All untreated samples had $^{87}\text{Sr}/^{86}\text{Sr}$ ratios that differed from expected biogenic ratios and the amount of diagenetic Sr in all samples decreased during pretreatment (Figures 1 and 2, Table 2).

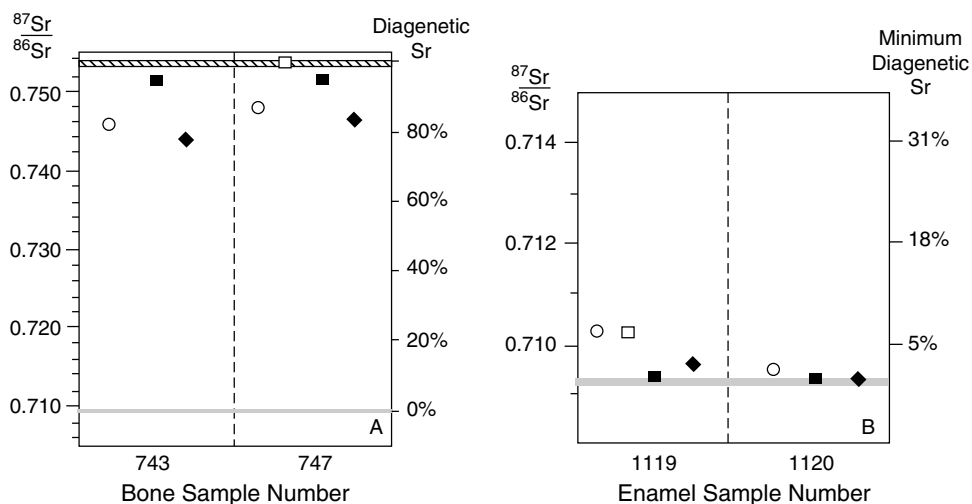


Figure 2. The $^{87}\text{Sr}/^{86}\text{Sr}$ ratio of Holocene bone and enamel samples from west Greenland harbour seals (*Phoca sp.*) treated with 25 leaches of 0.1 N buffered acetic acid (pH = 4.5). A) Bone samples from Godthåbs Fjord. B) Enamel samples from Qajâ. ○ untreated powders (note: values for untreated bone from Nelson *et al.*, 1986); □ first leachate; ■ 25th leachate; ◆ residual powder remaining after treatment. The grey bar represents initial biogenic $^{87}\text{Sr}/^{86}\text{Sr}$ ratios. The striped bar represents the $^{87}\text{Sr}/^{86}\text{Sr}$ ratio of surrounding matrix.

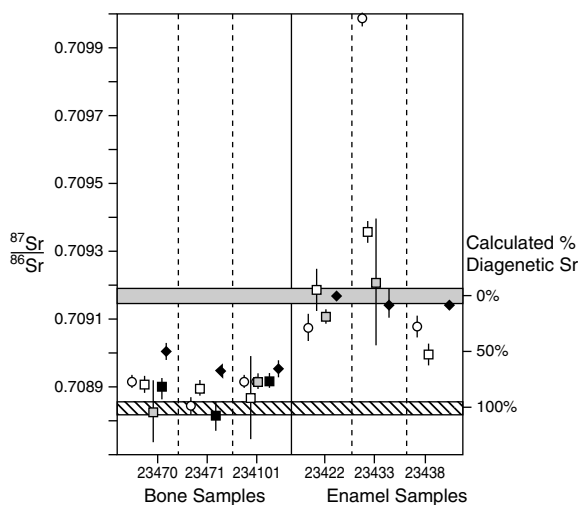


Figure 1. The $^{87}\text{Sr}/^{86}\text{Sr}$ ratio of Holocene bone and enamel samples from northern fur seal in California. All samples treated with sequential leaches of 0.1 N acetic acid (unbuffered). ○ untreated powders; □ fourth leachate; ■ eighth leachate (last enamel leachate); ◆ residual powder remaining after treatment. The grey bar represents original biogenic $^{87}\text{Sr}/^{86}\text{Sr}$ ratios. The striped bar represents the $^{87}\text{Sr}/^{86}\text{Sr}$ ratio of surrounding matrix, the presumed diagenetic endmember. Note: the $^{87}\text{Sr}/^{86}\text{Sr}$ ratios of diagenetic Sr for enamel sample 23433 is unknown.

Each sample, however, displayed different initial degrees of diagenetic contamination and

Table 2. $^{87}\text{Sr}/^{86}\text{Sr}$ ratios of bone and tooth enamel samples from a late early Miocene sperm whale before and after pretreatment

Treatment stage	Bone	Estimated % diagenetic Sr ¹	Enamel	Estimated % diagenetic Sr ¹
Untreated	0.70881	33%	0.70884 ± 0.0001	40%
Treated with 0.1 N acetic acid (unbuffered)				
4th acid leach	0.70881	33%	0.70876 ± 0.00005	22%
8th acid leach	0.70873	16%	0.70868	4%
Residue (8 leaches)	0.70876 ± 0.00004	22%	0.70875	20%
Residue (12 leaches)	0.70876	22%		
Treated with 0.1 N buffered acetic acid (pH = 4.5)				
25th acid leach	0.70890	53%		
Residue	0.70874	18%		

¹ Errors at least ±5%, due to uncertainties in the $^{87}\text{Sr}/^{86}\text{Sr}$ ratios of contemporaneous ocean water.

responded differently to pretreatment. For example, untreated enamel samples contained an estimated 1% to 40% diagenetic Sr. Corresponding bone samples usually contained higher amounts of diagenetic Sr (~35% to ~95%). The one exception to this pattern was enamel sample 23433 from Moss Landing, CA, which had ratios much higher than those expected for either biogenic Sr or diagenetic Sr derived from local soil (Figure 1). However, sample 23433 was associated with a rare lithic artifact that differed in composition from local rocks and likely altered the $^{87}\text{Sr}/^{86}\text{Sr}$ ratio of diagenetic fluids in the near-by area (Breschini *et al.*, 1995).

Pretreatment eliminated most contaminating Sr ($\geq 95\%$) from enamel samples. After pretreatment, the $^{87}\text{Sr}/^{86}\text{Sr}$ values for all enamel samples, including sample 23433, converged on ratios expected for biogenic Sr. Differences were observed in the behaviour of samples during treatment, however. The residual powder remaining after treatment contained the most biogenic Sr for Holocene samples from California and for Greenland sample 1120 (Figures 1 and 2), whereas the last leachate contained the most biogenic Sr for Greenland sample 1119 and Miocene whale enamel (Table 2). No systematic variations were observed between the results obtained for Holocene enamel samples using different pretreatment protocols.

Although the $^{87}\text{Sr}/^{86}\text{Sr}$ ratios of bone samples likewise became less diagenetic with pretreatment, each sample responded differently to treatment and all sample splits after pretreatment

contained significant diagenetic Sr. For Holocene bones, the most biogenic Sr was found in the residual powders left after treatment. For the Miocene whale bone, however, the most biogenic sample split varied depending on the pretreatment protocol; the residual powder contained the highest percentage of biogenic Sr for protocol one, whereas the eighth leachate yielded the most biogenic Sr for protocol two. However, all sample splits collected for protocol two, except for the fourth leachate, yielded $^{87}\text{Sr}/^{86}\text{Sr}$ ratios that were similar (within error) to each other and to the ratio of the residue collected for protocol one (Table 2). Most importantly, all bone samples retained significant biogenic Sr after pretreatment; the most biogenic splits from bone contained still ~15% to 80% diagenetic Sr (average = 60%).

Discussion

Our results confirm that pretreatment of bones and teeth with sequential rinses in weak acetic acid selectively removes some diagenetic Sr. However, several systematic differences were found in the response to pretreatment. First, pretreatment was less effective at removing diagenetic Sr from bone than from enamel. Second, the residual powder contained the most biogenic Sr for some samples, whereas the last leachate had the most biogenic Sr for others. Third, although both protocols that we tested were equally effective at removing diagenetic Sr, they sometimes differed in which splits yielded the most biogenic Sr.

These differences appear to result from variations in the type and degree of diagenetic alteration in each sample. Diagenetic Sr can be incorporated in fossils in several ways, including: 1) pore-filling by secondary minerals; 2) recrystallization or remineralization of hydroxyapatite; 3) direct exchange with Sr or Ca in the original hydroxyapatite crystals; and 4) absorption in microcracks or onto the surfaces of original hydroxyapatite crystals (Nelson *et al.*, 1986). Treatment with sequential 0.1 N acetic acid should remove Sr in secondary minerals and/or dissolve material absorbed onto surfaces, but it will not isolate a purely biogenic component if Sr has pervasively been incorporated into hydroxyapatite by recrystallization or exchange (Nelson *et al.*, 1986; Sillen, 1986; Tuross *et al.*, 1989). Among the samples we studied, bone and enamel displayed very different degrees of diagenetic contamination and different responses to pretreatment, so we discuss potential diagenetic mechanisms in each material separately.

Pretreatment appears to isolate biogenic Sr in enamel samples under controlled conditions when the type of diagenetic alteration is identified prior to protocol pretreatment choice. Sillen (1986) suggested that diagenetic Sr in fossils can be present largely in secondary carbonates and apatitic mineral phases and that sequential leaching should separate diagenetic Sr in a predictable fashion. Initial leachates would contain Sr from soluble diagenetic minerals (carbonate and highly-carbonated hydroxyapatite), late stage leachates would contain Sr from biogenic hydroxyapatite, and residual powder would contain Sr from more crystalline diagenetic apatites (fluorapatite and chlorapatite). The results for enamel from the Miocene whale fit this model; pretreatment selectively removed a highly carbonated apatite phase, leaving behind a residue consisting of hydroxy- and fluorapatite. However, while the model suggested by Sillen (1986) seems to apply for this enamel sample, no other enamel samples produced results consistent with Sillen's (1986) model. All but one of the Holocene enamel samples displayed the greatest percentage of biogenic Sr in residual powders, rather than in late stage leachates. This suggests that these samples did not contain significant amounts of diagenetic

fluorapatite, and that the most insoluble mineral present was biogenic hydroxyapatite. We note that Holocene tooth 1119 did yield residual powder with a small amount of diagenetic Sr (~2%). As this tooth contained no fluorapatite, it is likely this low-level contamination was from an insoluble mineral phase in the surrounding matrix.

In contrast, the diagenetic Sr that remained in all bone samples after treatment was too abundant to represent contamination of the samples with traces of matrix Sr. Some of the diagenetic Sr in bones that is removed by pretreatment is concentrated in secondary minerals or absorbed onto crystal surfaces. This interpretation is supported by the fact that the Miocene whale bone contained secondary minerals, some of which were selectively removed by pretreatment. The fact that pretreatment did not remove all diagenetic Sr from bone samples demonstrates that the contaminant was incorporated in essentially all the hydroxyapatite phases present in these fossils. Some untreated bone samples were more crystalline than modern bone, confirming that they had recrystallized post-mortem. This process would certainly introduce diagenetic Sr to the hydroxyapatite structure. However, not all bone samples showed large increases in crystallinity. For example, X-ray diffraction analyses of Holocene-age Greenland bone samples revealed little or no change in crystallinity and no evidence of secondary minerals. Yet ~80% of the diagenetic Sr in these samples remained after pretreatment. It thus appears that diagenetic Sr can be incorporated into bone through direct exchange with the Sr in ground waters, as suggested by Nelson *et al.* (1986). This process contaminates all hydroxyapatite pools in bones, thus making it impossible to isolate a biogenic pool by selective leaching.

In summary, our results demonstrate that pretreatment can successfully eliminate most ($\geq 95\%$) diagenetic contamination from Holocene and Miocene enamel, but that corresponding bones retained a significant amount (15 to 80%) of diagenetic Sr after pretreatment. This suggests that biogenic $^{87}\text{Sr}/^{86}\text{Sr}$ ratios of bones may not be recoverable, even on a Holocene time scale. Given the greater durability of enamel samples and their better response to pretreatment methods, we recommend that studies of biogenic Sr use enamel

and avoid bone. In addition, sample response to acid treatment varied depending on mineralogy. Thus, the mineralogy and crystallinity of all samples should be determined before pretreatment, and protocols should be modified accordingly (e.g., residual powders be discarded when fluorapatite is present).

Acknowledgements

This research was supported by grants from the National Science Foundation (EAR-9316371 and EAR-9725854), the Leakey Foundation, and the Geological Society of America. Technical assistance was provided by D. Bohaska, R. Burton, P. Holden, C. Janousek, H. Scott, and Q. Williams. We would especially like to thank M. DeNiro for access to Greenland specimens and valuable comments on the manuscript.

References

- Andrews GW. 1984. Miocene diatomaceous beds of the Calvert Formation at Popes Creek, Charles Country, Maryland. In *Cretaceous and Tertiary Stratigraphy, Paleontology, and Structure, Southwestern Maryland and Northeastern Virginia*, Prederiksen NO, Krafft K. (eds). American Association of Stratigraphic Palynologists Field Trip Volume and Guidebook 1984: 175–180. AASP Foundation, College Station, TX, USA.
- Breschini GS, Haversat T, Davis MK, Gibson RO, Huddleston RW, Jackson TL, King JH, Langenwaller PE, Miksicek CH, Rondeau MF, Rondeau VL, Runnings AL. 1995. *Archaeological Evaluation of CAMNT-234, at the site of the Proposed Mass Landing Marine Laboratory, Moss Landing, Monterey County, California*. Society of Professional Archaeologists.
- Budd P, Montgomery J, Barreiro B, Thomas RG. 2000. Differential diagenesis of strontium in archaeological human dental tissue. *Applied Geochemistry* **15**: 687–694.
- Burton RK, Koch PL. 1999. Isotope tracking of foraging and long distance migration in northeast Pacific pinnipeds. *Oecologia* **119**: 578–585.
- Driessens FCM, Verbeeck RMH. 1990. *Biomaterials*. CRC Press: Boca Raton, FL.
- Elliott TA, Forey PL, Williams CT, Werdelin L. 1998. Application of the solubility profiling technique to recent and fossil fish teeth. *Bulletin Société géologique de France* **169**: 443–451.
- Ezzo JA. 1992. A test of diet versus diagenesis at Ventana Cave, Arizona. *Journal of Archaeological Science* **19**: 23–37.
- Hodell DA, Mueller PA, Garrido JR. 1991. Variations in the strontium isotopic composition of seawater during the Neogene. *Geology* **19**: 24–27.
- Hoppe KA. 1999. *Biogeochemistry and Paleoecology of Late Pleistocene Proboscideans from the Southern United States*. Ph.D. dissertation. Princeton University, Princeton.
- Hoppe KA, Koch PL, Carlson RW, Webb SD. 1999. Tracking mammoths and mastodons: reconstruction of migratory behavior using strontium isotope ratios. *Geology* **27**: 439–442.
- Kalsbeek F, Taylor PN. 1999. Review of isotope data for Precambrian rocks from the Disko Bugt region, West Greenland. *Geology of Greenland Survey Bulletin* **181**: 41–47.
- Koch PL, Halliday AN, Walter LM, Stearley RF, Huston TJ, Smith GR. 1992. Sr isotopic composition of hydroxyapatite from recent and fossil salmon: the record of lifetime migration and diagenesis. *Earth and Planetary Science Letters* **108**: 227–287.
- Koch PL, Tuross N, Fogel ML. 1997. The effects of sample treatment and diagenesis on the isotopic integrity of carbonate in biogenic hydroxylapatite. *Journal of Archaeological Science* **24**: 417–429.
- Lee-Thorp JA, van der Merwe NJ. 1991. Aspects of the chemistry of modern and fossil biological apatites. *Journal of Archaeological Science* **18**: 343–354.
- LeGeros RZ, LeGeros JP. 1984. Phosphate minerals in human tissues. In *Phosphate Minerals* Nriago JO, Moore PD (eds). New York: Springer-Verlag.
- LeGeros RZ, Tung MS. 1983. Chemical stability of carbonate- and fluoride-containing apatites. *Caries Research* **17**: 419–429.
- Lenihan JMA, Loutit JF, Martin JH (eds). 1967. *Strontium Metabolism*. Academic Press: London, 345.
- Mohl J. 1986. Dog remains from a Paleoeskimo settlement in West Greenland. *Arctic Anthropology* **23**: 81–89.
- Neilsen-Marsh CM, Hedges REM. 1997. Dissolution experiments on modern and diagenetically altered bone and the effect on the infrared splitting factor. *Bulletin Société géologique de France* **168**: 485–490.
- Nelson BK, DeNiro MJ, Schoeninger MJ, De Paolo DJ. 1986. Effects of diagenesis on strontium, carbon, nitrogen and oxygen concentration and isotopic composition of bone. *Geochimica et Cosmochimica Acta* **50**: 1941–1949.
- Price TD. 1989. Multi-element studies of diagenesis in prehistoric bone. In: *The Chemistry of Prehistoric Human Bone*, Price, TD (ed.). Cambridge, Cambridge University Press.

- Price TD, Johnson CM, Ezzo JA, Ericson J, Burton JH. 1994. Residential mobility in the prehistoric southwestern United States: a preliminary study using strontium isotope analysis. *Journal of Archaeological Science* **21**: 315–330.
- Schoeninger MJ. 1985. Trophic level effects on $^{15}\text{N}/^{14}\text{N}$ and $^{13}\text{C}/^{12}\text{C}$ ratios in bone collagen and strontium levels in bone mineral. *Journal of Human Evolution* **14**: 515–525.
- Sealy J, Armstrong R, Schrire C. 1995. Beyond lifetime averages: tracing life histories through isotopic analysis of different calcified tissues from archaeological human skeletons. *Antiquity* **69**: 290–300.
- Sealy JC, van der Merwe NJ, Sillen A, Krueger FJ, Chargar HW. 1991. $^{87}\text{Sr}/^{86}\text{Sr}$ as a dietary indicator in modern and archaeological bone. *Journal of Archaeological Science* **18**: 399–416.
- Sillen A. 1986. Biogenic and diagenic Sr/Ca in Plio-Pleistocene fossils of the Omo Shungura Formation. *Paleobiology* **12**: 311–323.
- Sillen A, Kavanaugh M. 1982. Strontium and paleodietary research: a review. *Yearbook of Physical Anthropology* **25**: 67–90.
- Sillen A, Sealy JC. 1995. Diagenesis of strontium in fossil bone: a reconsideration of Nelson *et al.* (1986). *Journal of Archaeological Science* **22**: 131–320.
- Stiner MC, Kuhn SL, Weiner S, Bar-Yosef O. 1995. Differential burning, recrystallization, and fragmentation of archaeological bone. *Journal of Archaeological Science* **22**: 223–237.
- Tuross N, Behrensmeier AK, Eames ED. 1989. Sr increases and crystallinity changes in taphonomic and archaeological bone. *Journal of Archaeology* **16**: 661–672.
- Walker RJ, Carlson RW, Shirey SB, Boyd FR. 1989. Os, Sr, Nd, and Pb isotope systematics of southern African peridotite xenoliths: implications for the chemical evolution of subcontinental mantle. *Geochimica et Cosmochimica Acta* **53**: 1583–1595.
- Weiner S, Bar-Yosef O. 1990. States of preservation of bones from prehistoric sites in the Near East: a survey. *Journal of Archaeological Science* **17**: 187–196.
- Weiner S, Goldberg P, Bar-Yosef O. 1993. Bone preservation in Kebara Cave, Israel using on-site Fourier transform infrared spectrometry. *Journal of Archaeological Science* **20**: 613–627.
- Wright LE, Schwarcz HP. 1996. Infrared and isotopic evidence for diagenesis of bone apatite at Dos Pilas, Guatemala: palaeodietary implications. *Journal of Archaeological Science* **23**: 933–944.