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# Three case studies used to reassess the reliability of fossil bone and enamel isotope signals for paleodietary studies

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## Abstract

The emphasis on tooth enamel for extraction of stable light isotope signals from the mineral phase of archaeological and paleontological calcified tissues is based on the widespread understanding that enamel remains a relatively closed system, while bone does not. Twenty years ago, however, Sullivan and Krueger's groundbreaking study demonstrating the potential of stable carbon isotopes from the mineral phase relied entirely on bone apatite samples from archaeological sites. Further effort to test whether diagenetic effects in bone mineral may be circumvented remains important because bone apatite yields dietary information about adult life-stages beyond the discrete snapshots obtainable from enamel. In this paper we re-examine the grounds for exclusion of bone apatite as sample material, using case studies drawn from three sites which differ in age and depositional conditions. We use  $^{13}\text{C}/^{12}\text{C}$ ,  $^{18}\text{O}/^{16}\text{O}$ ,  $^{87}\text{Sr}/^{86}\text{Sr}$ , and Fourier transform infrared (FTIR) spectroscopy data from three sites (Reunion Rocks, Border Cave, and Makapansgat Limeworks) to show that, while enamel is not a closed system, it nevertheless retains biogenic isotopic signals. In addition, bone signals may be surprisingly well preserved where fossilisation pathways have induced 'enamel-like' crystallisation changes.

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**Keywords:** Makapansgat; Border Cave; Reunion Rocks; Bone chemistry; FTIR; Paleodiet; Carbon isotopes; Oxygen isotopes; Strontium isotopes

## Introduction

Twenty years ago Sullivan and Krueger published a seminal paper that pointed the way to routine use of the mineral phase of calcified tissues (Sullivan and Krueger, 1981) for dietary isotope studies. Using bone samples previously obtained from archaeological sites for radiocarbon dating, they showed that stable carbon isotope values extracted from a vigorously purified mineral phase were offset, but comparable in reliability, to values extracted from the purified organic phase (Fig. 1). All of their samples ranged in age from recent to just over 20,000 years.

Their findings were challenged on the grounds that mineral derived from other bones associated with fairly recent archaeological sites in Peru and Mexico did not yield similarly consistent results and seemed to be irreparably altered (Schoeninger and DeNiro, 1982). Although some questions arose about the comparability of the two sets of results, related to differing pre-treatment procedures and the effects of trophic level on the offset, the net effect was to cast carbon isotope values extracted from the mineral phase under suspicion. This general view was partially reversed when results of a study of consistent  $\text{C}_3$ -feeders showed that only small shifts occurred in their bone  $\delta^{13}\text{C}$  values over time (Lee-Thorp and van der Merwe, 1987). The results, however, seemed far more reliable for enamel, and it was this observation that was picked up and subsequently acted upon. A modelling study (Wang and Cerling, 1994) reinforced

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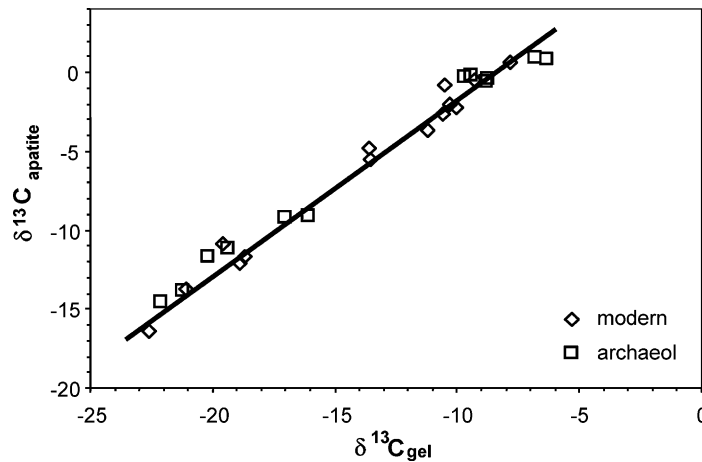


Fig. 1. Scattergram showing the linear relationship between  $\delta^{13}\text{C}_{\text{apatite}}$  and  $\delta^{13}\text{C}_{\text{collagen}}$ . Note that apatite is enriched in  $^{13}\text{C}$  by approximately 7–8‰ for both modern (diamonds) and archaeological (squares) bones. Redrawn from Sullivan and Krueger (1981).

the use of tooth enamel, emphasising the open, exchangeable properties of bone in contrast to the closed, sequestered nature of enamel. As a result, most isotopic studies, of older material in particular, shifted entirely to enamel, which henceforth became the sample material of choice. Results of phosphate-based oxygen isotope studies have tended to follow a similar route, and reinforced this general pattern (e.g., Ayliffe et al., 1994).

The Sullivan and Krueger (1981) study and subsequent work (Krueger and Sullivan, 1984) also set in motion another important line of investigation concerning the nature of the isotopic relationships between diet and bone collagen or bone mineral—whether these relationships were dependent on dietary quality. A detailed discussion of these investigations is beyond the scope of this paper. Suffice it to point out here, that as currently understood, dietary information derived from isotopic composition of the organic and mineral phases is not equivalent, because collagen preferentially reflects isotopic composition of protein in the diet, while the mineral reflects that of the entire, integrated diet (Ambrose and Norr, 1993; Tieszen and Fagre, 1993). This means that although both phases reliably reflect diet, they provide different perspectives. Hence, some questions are best addressed using analysis of both phases, for instance where there are questions about differences among sections of a population in access to high-quality foods (e.g., animal foods).

A number of studies concentrating on either the isotope values, crystallographic indicators, or both (e.g., Nelson et al., 1986; Person et al., 1995; Wright and Schwarcz, 1996) have underscored the difficulties frequently encountered in obtaining reliable isotope determinations from both the mineral and the organic phases of even recent archaeological bone. Many of

these studies have been done on rather poorly preserved material from tropical or low to mid-latitude sites, where the combination of heat, moisture, and high-frequency bacterial interference promote rapid alteration and degradation of calcified tissues. Even in cooler areas, combinations of wetting cycles and bacterial action soon destroy most bones. Calcified tissues subject to such conditions are best thought of as on the pathway to destruction.

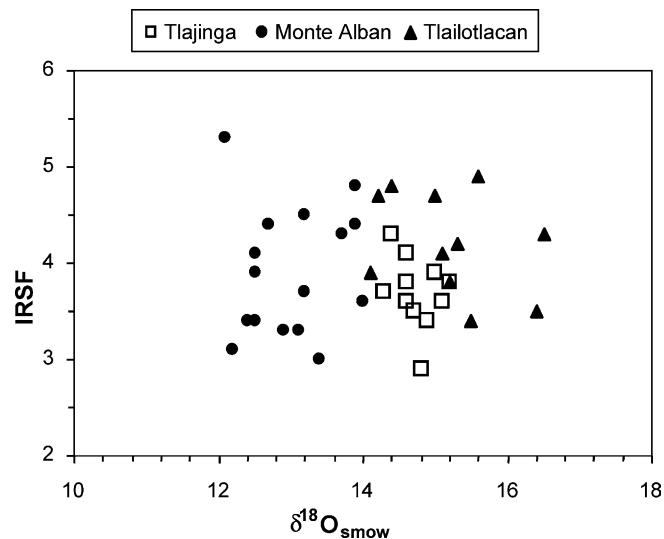
In spite of the susceptibility of bone to destruction, there are however, some good reasons to question the near exclusive reliance on enamel and abandonment of bone apatite in stable light isotope studies. This point was made by Harold Krueger several years ago (pers. comm.). Trace element studies of well-preserved fossils have continued to rely on bone, with some of the more successful, if not yet widely practised, adopting an approach based on differing solubilities of ‘compartments’ in fossilised bone (Sillen, 1989). Testing these procedures satisfactorily has remained problematic, but the demonstration of predictable differences in strontium isotopes between riverine- and veld-dwelling fossil animals at Swartkrans (ca. 1.7 my) provided a successful independent test showing that detectable biogenic strontium remained in situ in bone (Sillen et al., 1998). A closer re-examination of the bone apatite  $\delta^{13}\text{C}$  values from the original fossil  $\text{C}_3$  animal data set presented by Lee-Thorp and van der Merwe (1987) suggested that many were more reliable than recognised earlier (i.e., they conformed to predictable isotopic values based on their diets, where known). The higher “success” rate seemed to extend back at least to specimens of Late Pleistocene age (Lee-Thorp, 2000). Some of the observations used in that reassessment include material from the important South African Middle Stone Age site of Border Cave, to

which we return below. The apparent contradictions between these various observations, and those made on the often poorly preserved material from more recent archaeological sites, prompted the suggestion that bones and teeth that self-evidently survived long periods had followed a different pathway—a pathway to preservation, rather than to destruction.

A further important consideration resides in the methods that have been used to detect and characterise chemical or crystallographic alteration of enamel and bone. One regularly employed approach is the determination of a ‘crystallinity index,’ which denotes the degree of crystal imperfection and strain as well as crystal size. This index is derived from either the infrared splitting factor (IRSF), or from X-ray diffractometry. The infrared splitting factor is calculated from the degree of separation, or splitting, between the two equivalent phosphate ( $\text{PO}_4$ ) peaks at  $605$  and  $560\text{ cm}^{-1}$ , and therefore it is most directly related to symmetry in the phosphate domain. Almost all studies show a rapid postmortem increase in bone crystallinity (Person et al., 1995; Tuross et al., 1989; Wright and Schwarcz, 1996), which is often correlated with other indices such as loss of the organic phase (e.g., Person et al., 1995; Sillen and Parkington, 1996). While this approach is certainly useful for many purposes, including delimiting spatial zones of irregular preservation across archaeological sites (Weiner and Bar-Yosef, 1990), and detecting intrusive material (Sillen and Morris, 1996), it is not clear that higher crystallinity necessarily implies significant alteration of isotope values for either bone or enamel.

For instance, White et al. (1998) showed that predictable, differentiated oxygen isotope values were observed in bone phosphate from the Valley of Mexico and the Valley of Oaxaca in spite of significantly increased IRSF (Fig. 2). Variable IRSF did not appear to affect the isotope results enough to obscure the goal of the study.

Given these contradictions, it may be suggested that the indicators for diagenesis currently in use, while demonstrating diagenesis in certain respects, do not adequately demonstrate *isotopic* alteration. It may turn out that each case, or site, will have to be tested on an individual basis. Nevertheless, we believe that it is worth further attempts to develop tests for screening fossil bone apatite for isotopic analyses. Although it might be argued that enamel represents a far easier, and more reliable ‘target’ sample material, there are many instances where bone is more appropriate or where only bone is available for analysis. Perhaps the most compelling reason for pursuing the issue of bone reliability is related to the period of life history represented by bone. Dietary and behavioral information obtained from enamel isotope signals is necessarily restricted to the earlier stages of an individual’s life, whereas that obtained from bone represents an average signal, mostly from adulthood (although this will vary between and within elements). This distinction presents obvious opportunities for investigation of dietary, and sometimes environmental, patterns during different stages of an individual’s life (Cox and Sealy, 1997; Sealy et al., 1995). Analysis of both bone and enamel mineral can provide



broader behavioral information than is available from enamel alone.

### This study

In the belief that examination of older, fossilised bone that has *demonstrably* followed the pathway to preservation may offer new clues towards screening tests, we examine isotope and structural data from bone and enamel at three sites. They are the Reunion Rocks elephant site, the Border Cave Middle Stone Age site, and Makapansgat Limeworks Member 3. They represent, in a simplified sense, a very wet, a very dry, and a highly calcareous, alkaline site, respectively. Because the data have been drawn partly from studies directed towards other purposes, we do not have exactly comparable information from each of these sites. Most notably, only one individual was found at the Reunion Rocks site in contrast to the other two sites.

### Reunion Rocks site

A large proportion of the skeleton of a single elephant, submerged in seawater at low tide and encased in hardened coastal aeolianite ('beachrock'), was discovered at Reunion Rocks on the east coast of South Africa. A single age determination based on Th/U concentration gave a tentative age of ca. 130,000 years—i.e., the elephant died at about the time of the onset of the last Interglacial period or perhaps earlier (Ramsay et al., 1993). Whatever the exact age, it is certain that it was encased in the aeolianite or submerged under seawater

for a good part of the last 100,000 years. Hence the skeleton has been consistently exposed to seawater, or deposits of marine origin, for most of its history. Since several skeletal elements were discovered, it provided a good opportunity to compare preservation of various tissues (enamel, dentine, tusk, and bone) and to compare different methods for extracting purified, and hopefully biogenic, components. Furthermore, burial of a terrestrial animal in marine sediments provided the opportunity to test whether the trace element strontium was indeed biogenic, or not, using the well-known strontium isotope signal for seawater.

Both the standard isotopic purification procedure (elimination of organics using sodium hypochlorite solution and removal of secondary carbonates and soluble apatites using weak acetic acid) (Koch et al., 1997; Lee-Thorp and van der Merwe, 1987), and the 'solubility profile' (Sillen, 1989) procedures were applied. Strontium isotope data were obtained following standard procedures for measurement of strontium isotopes in calcified tissues (Sealy et al., 1995). The tissues were examined by standard infrared spectroscopy before and after the procedures. In addition, FTIR spectra were obtained for untreated bone and enamel samples.

The results for carbon and strontium isotopes show that in all cases (whatever the procedure used), only enamel reflected anything resembling the original signals for  $\delta^{13}\text{C}$ ,  $\delta^{18}\text{O}$ , and  $^{87}\text{Sr}/^{86}\text{Sr}$  (Fig. 3). Hoppe (2000) also reported that while most of the original strontium (Sr) is preserved in enamel, biogenic Sr was unrecoverable from bone, even using the solubility profiling method. Although Sr/Ca in bone seems to reach biologically reasonable levels during the solubility profile procedure,  $^{87}\text{Sr}$  values of 0.7092 are essentially the same as seawater

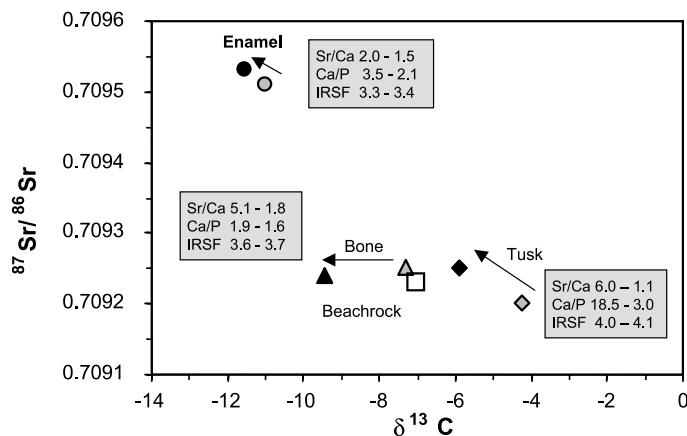


Fig. 3. Plot of  $^{87}\text{Sr}/^{86}\text{Sr}$  versus  $\delta^{13}\text{C}$  for enamel (circles), bone (triangles), and tusk (diamonds) of the Reunion Rocks Pleistocene elephant, showing the shifts before (shaded) and after (dark) standard acetic acid application. The range of values for Sr/Ca, IRSF, and Ca/P is given in associated text blocks before and after acid wash. Results from the solubility profile technique, pooled for  $^{87}\text{Sr}$  determinations, are not shown but essentially indicate the same pattern in more detail.

Table 1

Changes in four FTIR-derived indicators—the infrared splitting factor (IRSF), proportion of “B” to “A” site carbonate (BAI), estimation of carbonate content (CO<sub>3</sub>%) modified after LeGeros (1991), and the phosphate (605 cm<sup>-1</sup>)–phosphate (560 cm<sup>-1</sup>) index (PPI)

	Modern		Makapan M3		Border Cave		Reunion Rocks	
	NT	AW	NT	AW	NT	AW	NT	AW
BONE, <i>n</i> =	4	4	3	5	2	3	1	1
IRSF	2.95	3.11	3.57	3.65	3.27	3.30	3.62	3.72
BAI	1.23	nm	nm	3.93	2.32	2.34	2.98	nm
CO <sub>3</sub> %	6.91	6.01	7.64	4.79	8.14	7.90	7.03	5.64
PPI	0.85	0.86	0.95	0.91	0.87	0.85	1.04	0.99
ENAMEL, <i>n</i> =	7	3	3	nm	2	2	1	1
IRSF	3.58	4.00	3.63	nm	3.90	3.60	3.40	3.40
BAI	3.33	3.40	3.83	nm	5.20	2.26	2.25	nm
CO <sub>3</sub> %	3.23	3.30	3.17	nm	3.00	3.94	5.66	3.96
PPI	0.86	0.83	0.86	nm	0.86	0.85	0.89	0.88

Means are given for modern, Makapansgat, and Border Cave samples. NT denotes no treatment, AW the sodium hypochlorite/acetic acid wash outlined in the text, and ‘nm’ indicates no measurement. Indices for modern, Makapansgat, and Border Cave samples are averaged; *n* is given above each column.

and show that the strontium content has thoroughly exchanged with seawater. Enamel <sup>87</sup>Sr/<sup>86</sup>Sr is significantly different to all other tissues, including dentine of the same tooth. Lower than expected Ca/P is observed for bone, suggesting loss of calcium or addition of phosphate, while Ca/P for tusk is highly variable suggesting multiple Ca-containing phases. Ca/P for enamel is initially high but returns to biogenic levels after the acetic acid wash, suggesting elimination of a calcium carbonate phase. The IRSF for all tissues except enamel were higher than modern values for that tissue (Fig. 3 and Table 1) and application of the acetic acid wash induced slightly higher IRSF, as observed for the modern samples in Table 1, and elsewhere (e.g., Koch et al., 1997; Sponheimer and Lee-Thorp, 1999a). The acid wash did not appear to shift IRSF in fossil enamel. CO<sub>3</sub>% in untreated bone is similar to modern levels. CO<sub>3</sub>% is higher than expected for enamel and remained moderately high even after acid wash. Compared to the modern tissues, B-carbonate/A-carbonate index (BAI) is higher for untreated bone but lower than expected for enamel, possibly indicating some rearrangement of carbonate ions for enamel (Table 1). We report a new index (PPI), the ratio of the phosphate peaks at 605 and 560 cm<sup>-1</sup>, which we believe is sensitive to changes or realignment of phosphate ions in calcified tissues. Although changes are subtle, PPI shows a larger increase above typical modern levels for bone (+0.09) than for enamel (+0.03) (Table 1).

Hence, almost all structural, and all isotopic, indicators show that bone apatite is highly altered. The original strontium content must have been replaced as evidenced by the <sup>87</sup>Sr values. Enamel, however, maintains near-biogenic isotopic values in spite of altered BAI and slightly higher carbonate content. These

observations conform to suggestions arising from experimental and modelling studies about the importance of hydrology and porosity for preservation (Hedges and Millard, 1995; Wang and Cerling, 1994).

### Border Cave

Border Cave is in the Lebombo Mountains on the border between South Africa and the mountain kingdom of Swaziland. It contains a long sequence of material, most of which can be assigned to the African Middle Stone Age (MSA) (Beaumont et al., 1978). The sequence yielded a number of hominid specimens that have been claimed to represent early examples of anatomically modern humans (Beaumont et al., 1978), but their importance has been weakened by controversy over their proveniences (Sillen and Morris, 1996). The site is extremely well-protected and very dry. Temperatures inside the cave fluctuate only slightly on diurnal or seasonal timescales. Uranium series determinations show very little movement of uranium (Grün et al., 1990), suggesting the pervasiveness of aridity through time. A peculiar feature within the deposits is the occurrence of spatially extensive, thick layers of ash (stratigraphically denoted as 1WA, 2WA, etc.).

Of importance for this study is the presence of a large faunal sequence of visually well-preserved bones and teeth. In all bone specimens tested no measurable collagen is preserved. Some specimens including teeth are dark and brittle and appear burnt. Carbon isotope values for both browsers and grazers obtained earlier suggested that preservation of stable carbon isotopes in the enamel mineral was good (Lee-Thorp, 2000). In a study aimed at establishing the contextual relationships of the

hominid samples using nitrogen content and IRSF, Silen and Morris (1996) showed that, apart from an upper, recent layer, all the bones in their study had relatively high IRSF, which was observed to increase irregularly with age. Another study employing FTIR to determine the amounts of  $\text{CO}_3$  remaining in various structural positions within the mineral structure (based on the ratio of carbonate in the “B” (phosphate) to “A” (hydroxyl) positions (BAI), as well as IRSF) concluded that 2 Border Cave enamel samples seemed highly altered (Sponheimer and Lee-Thorp, 1999a).

Bearing in mind that rigorous acetic acid procedures used in the earlier isotopic study of the Border Cave material (Lee-Thorp, 2000) likely produced oxygen isotope artifacts or offsets, we repeated carbon and oxygen isotope analyses of the available faunal samples using gentler methods. The new samples include bone samples, three of which are paired with enamel from the same animal. The results corroborate the earlier conclusion of excellent preservation of expected  $\text{C}_3$  and  $\text{C}_4$  carbon isotope signatures in enamel and the bone samples, too, fall within the expected ranges (Fig. 4). The oxygen isotope results are noisy and harder to interpret, perhaps not unexpectedly since the climatic ranges covered in the age span are great, covering as they do large parts of an interglacial–glacial cycle. The results for all the bushpigs (*Potamochoerus*), however, stand out as being systematically and significantly depleted compared to the other fauna (ANOVA;  $p < 0.01$ ). Although we are not aware of published  $\delta^{18}\text{O}$  values for this particular suid, several studies have shown consistently depleted values for another suid, the warthog (*Phacochoerus*) (Bocherens et al., 1996; Sponheimer and Lee-Thorp, 2001). The suggestion is that the depleted  $\delta^{18}\text{O}$  values are related to a reliance on underground rootstocks where plant-water

values are low (Sternberg, 1989). This explanation would also hold for *Potamochoerus*, although underground rootstocks are likely to be only one of a number of contributing factors. Hence the inference is that enamel  $\delta^{18}\text{O}$  values, at least, preserve biogenic differences.

FTIR parameters for bone confirm the crystallographic shifts noted earlier but show that variability is high and therefore, on average, some of the shifts are lower than previously observed (Table 1). Both bone and enamel have slightly higher IRSF prior to acid washing, but enamel IRSF is reduced after an acid wash. Differences are observed in the proportions of carbonate occupying the “B” (phosphate) site within the crystal structure, compared to carbonate occupying the “A” (hydroxyl) site. The untreated Border Cave enamel samples measured earlier ( $n = 2$ , Sponheimer and Lee-Thorp, 1999a) showed a markedly higher BAI compared to modern samples or even other older fossils we have examined (including far older Makapansgat). This may imply preferential removal of “A” site carbonate, possibly via replacement with fluorine ions (Sponheimer and Lee-Thorp, 1999a). The acid-washed enamel specimens measured here ( $n = 3$ ) show a modest decrease in BAI. Unfortunately, the untreated and acid-washed sample sets are not the same specimens and cannot be used to discern shifts caused by the acid wash. The variability observed in BAI, however, does point to high variability in this index. BAI in both untreated and acid-washed bone samples is a little higher than that observed for modern samples. The amount of carbonate in the bones is higher than for modern samples, but only slightly higher for enamel. Altered BAI suggests some alterations in the carbonate domain, which are dissimilar in bone and enamel, but unaltered PPI suggests few changes in the phosphate domain.

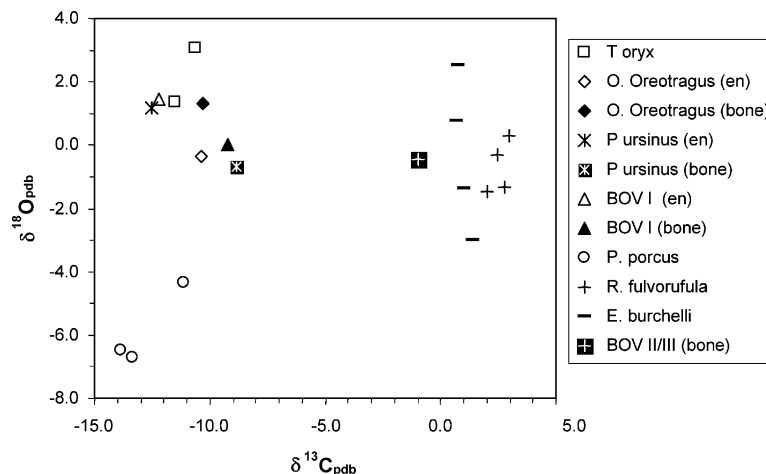


Fig. 4.  $\delta^{13}\text{C}$  values for enamel (shaded) and bone (dark) apatite of Border Cave fauna plotted against  $\delta^{18}\text{O}$ , showing consistent separation between  $\text{C}_3$ - and  $\text{C}_4$ -feeders.

### Makapansgat Limeworks Member 3

The Limeworks Member 3 fauna is believed to be about 3 million years old based on biostratigraphy (e.g., Delson, 1984; Reed, 1996; Vrba, 1985), or a little older, based on geomagnetic reversals (Partridge, pers. comm.). Member 3 (the ‘grey breccia’) is highly fossiliferous, and is believed to have become consolidated from hyena accumulations subsequently exposed to solutions rich in carbonate within sections of an extensive karstic formation. Hence, during earlier stages of formation Member 3 was not as dry and protected as it now appears. The faunal assemblage includes a high proportion of extinct species, including at least one hominid species, *Australopithecus africanus*.

We have reported the results of enamel-based isotopic and FTIR studies of the faunal assemblage and hominids elsewhere (Sponheimer and Lee-Thorp, 1999a,b). The results indicated that surprisingly little alteration in carbon or oxygen isotopes had occurred in enamel carbonate, since predictable patterns of browsing and grazing were observed. The FTIR parameters showed little evidence of increased crystallinity (IRSF) but did show increases in BAI. These changes suggest that some rearrangement has occurred in the carbonate domains. Determination of the same data for paired bones reveals that half of the bones (total  $n = 6$ ) conform to expected carbon isotope patterns, while the other half do not (Fig. 5). Apart from more frequent occurrence of calcite peaks (which are easily removed after acid washing), FTIR spectra parameters for Limeworks fossil bones resemble those of fossil enamel, although the carbonate content and PPI are higher (Table 1). Acid-washed fossil bone has lower  $\text{CO}_3\%$  than untreated or acid-washed modern bone, and therefore, there is no evidence of wholesale incorporation of exogenous  $\text{CO}_3$  ions into bone from the Makapansgat environment. This is not to say that isotopic and ionic exchange have not occurred, however, as the original

isotopic signals are observed in only 50% of the bones analysed.

### Discussion

What inferences can we draw from these three examples of long-term ‘preservation?’ We admit immediately to being hampered by the fact that our datasets are not thoroughly comparable across the three sites. Nevertheless, some features stand out. One is that higher IRSF, or increased crystallinity, is not a good predictor of carbonate isotopic alteration in either bone or enamel in these sites. Rather, modestly raised IRSF may be a good predictor or possibly even a prerequisite, for isotopic preservation in bone apatites. We note that reasonable carbon isotope values, in particular, occur in those instances where bone most “resembles” enamel in crystallographic features. Shifts in PPI appear intriguing. Higher bone PPI in the Reunion Rocks elephant and to a lesser extent in Makapansgat bones seem to correspond with poor or poorer, respectively, isotopic preservation. No fossil enamel samples reached the high PPI seen in some of the bones. Since changes in this index reflect shifts in the local environment of phosphate ions, they may provide a sensitive indication of large-scale recrystallisation. This possibility merits investigation with larger numbers of samples of different ages and taphonomic histories.

A major difference between the site showing the most reasonable results for carbon isotopes in bone (Border Cave) and the site showing poor results (Reunion Rocks elephant) lies in the hydrology. Makapansgat occupies an intermediate position. As suggested by White and Hannus (1981), the presence of calcium in alkaline soils buffers buried bone mineral from rapid degradation and dissolution. The mechanism of calcium buffering was removed in the case of the Pleistocene elephant, but not in Border Cave and Makapansgat. Another feature of

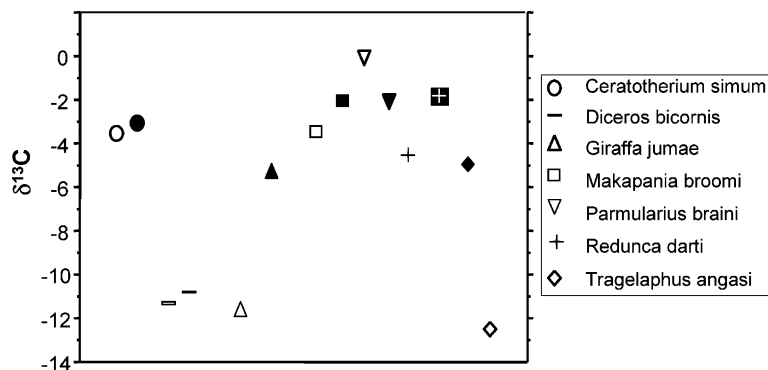


Fig. 5. A comparison of  $\delta^{13}\text{C}$  bone (dark) and enamel (unshaded) apatite values for seven paired tooth and bone specimens from Makapansgat Limeworks Member 3. All enamel  $\delta^{13}\text{C}$  values accurately reflect faunal diets—browsers are relatively depleted in  $^{13}\text{C}$  and grazers are near 0‰, whereas half of the bone analyses conform to this pattern.

the burial environment, in combination with buffering, may be the speed of removal of the organic phase. A reasonable prediction for a “preservation” pathway would be relatively slow removal of collagen in concert with a relatively fast increase in crystallinity. This is the trajectory suggested for bones from the archaeological site of Elands Bay Cave on the Western Cape coast of South Africa, as shown in a curve of % nitrogen (representing the organic component) versus IRSF (Sillen and Parkington, 1996).

The subtle changes observed in proportions of carbonate and phosphate occupying different sites seem to offer tantalising clues to fossilisation pathways, but their precise interpretation is still uncertain. One way forward might be the execution of careful fossilisation simulations, designed, not just to emulate difficult burial conditions, but in particular those conditions that we now know are most likely to result in isotopically well-preserved fossils.

Wright and Schwarcz (1996) demonstrated that we cannot assume that even recent archaeological bone mineral retains a biogenic signal. This study suggests that the converse is also true, that we cannot assume that ancient bone mineral does not preserve its original isotopic signature. Clearly some bone mineral, even when its organic fraction has been completely supplanted, does preserve accurate paleodietary and paleoenvironmental information. Indeed, while it has traditionally been assumed that structural preservation (both organic and crystallographic) is the sine qua non of isotopic preservation, it may be that just the opposite is true: that ancient bone mineral can only retain its isotopic signal if it is “locked in” by dissolution/reprecipitation or recrystallisation phenomena under appropriate conditions. Admittedly, we are still a long way from being able to regularly use the stable isotopes in bone mineral as a window into the past. Nevertheless, bone mineral might allow us to study paleodiets when collagen is not preserved. Moreover, when collagen is preserved, using bone mineral and collagen in tandem should provide a more complete picture of ancient diets. Thus, further research into the isotopic integrity of ancient bone seems warranted.

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### References cited

- Ambrose, S.H., Norr, L., 1993. Experimental evidence for the relationship of the carbon isotope ratios of whole diet and dietary protein to those of bone collagen and carbonate. In: Lambert, J.B., Grupe, G. (Eds.), *Prehistoric Human Bone: Archaeology at the Molecular Level*. Springer-Verlag, Berlin, pp. 1–37.
- Ayliffe, L.K., Chivas, A.R., Leakey, M.G., 1994. The retention of primary oxygen isotope compositions of fossil elephant skeletal phosphate. *Geochimica et Cosmochimica Acta* 58, 5291–5298.
- Beaumont, B., de Villiers, H., Vogel, J., 1978. Modern man in sub-Saharan Africa prior to 49000 year b.p.: a review and evaluation with particular reference to Border Cave. *South African Journal of Science* 74, 409–419.
- Bocherens, H., Koch, P.L., Mariotti, A., Geraads, D., Jaeger, J.-J., 1996. Isotopic biogeochemistry (<sup>13</sup>C, <sup>18</sup>O) of mammalian enamel from African Pleistocene hominid sites. *Palaeos* 11, 306–318.
- Cox, G., Sealy, J.C., 1997. Investigating identity and life histories: isotopic analysis and historical documentation of slave skeletons found on the Cape Town Foreshore, South Africa. *International Journal of Historical Archaeology* 1, 207–224.
- Delson, E., 1984. Cercopithecoid biochronology of the African Plio-Pleistocene: correlation among eastern and southern hominid-bearing localities. *Courier Forschungsinstitut Senckenberg* 69, 199–218.
- Grün, R., Beaumont, P.B., Stringer, C., 1990. ESR dating evidence for early modern humans at Border Cave in South Africa. *Nature* 344, 537–539.
- Hedges, R.E.M., Millard, A.R., 1995. Measurements and relationships of diagenetic alteration of bone from three archaeological sites. *Journal of Archaeological Science* 22, 201–209.
- Hoppe, K.A., 2000. The biogeochemistry and paleoecology of late pleistocene proboscideans from the southern United States. PhD Dissertation, Princeton University, Princeton, NJ.
- Koch, P.L., Tuross, N., Fogel, M.L., 1997. The effects of sample treatment and diagenesis on the isotopic integrity of carbonate in biogenic hydroxylapatite. *Journal of Archaeological Science* 24, 417–429.
- Krueger, H.W., Sullivan, C.H., 1984. Models for carbon isotope fractionation between diet and bone. In: Turnlund,

- J.F., Johnson, P.E. (Eds.), *Stable Isotopes in Nutrition*. ACS Symposium Series 258, American Chemical Society, Washington, DC, pp. 205–222.
- Lee-Thorp, J.A., van der Merwe, N.J., 1987. Carbon isotope analysis of fossil bone apatite. *South African Journal of Science* 83, 71–74.
- Lee-Thorp, J.A., 2000. Preservation of biogenic carbon isotope signals in Plio-Pleistocene bone and tooth mineral. In: Ambrose, S.H., Katzenberg, M.A. (Eds.), *Biogeochemical Approaches to Paleodietary Analysis*. Kluwer Academic/Plenum Press, New York, pp. 89–115.
- LeGeros, R.Z., 1991. *Calcium Phosphates in Oral Biology and Medicine*. Karger Press, Basel.
- Nelson, B.K., DeNiro, M.J., Schoeninger, M.J., DePaolo, D.J., Hare, P.E., 1986. Effects of diagenesis on strontium, carbon, nitrogen and oxygen concentration and isotopic composition of bone. *Geochimica et Cosmochimica Acta* 50, 1941–1949.
- Person, A., Bocherens, H., Saliège, J.-F., Paris, F., Zeitoun, V., Gérard, M., 1995. Early diagenetic evolution of bone phosphate: an X-ray diffractometry analysis. *Journal of Archaeological Science* 22, 211–221.
- Ramsay, P.J., Smith, A.M., Lee-Thorp, J.A., Vogel, J.C., Tyldsley, M., Kidwell, W., 1993. 130 000-year-old fossil elephant found near Durban, South Africa: preliminary report. *South African Journal of Science* 89, 165–166.
- Reed, K., 1996. The paleoecology of Makapansgat and other African Plio-Pleistocene hominid localities. PhD Dissertation, State University of New York at Stony Brook, Stony Brook, NY.
- Schoeninger, M.J., DeNiro, M.J., 1982. Carbon isotope ratios of apatite from bone cannot be used to reconstruct diets of animals. *Nature* 297, 577–578.
- Sealy, J.C., Armstrong, R., Schrire, C., 1995. Beyond lifetime averages: tracing life histories through isotopic analysis of different calcified tissues. *Antiquity* 69, 290–300.
- Sillen, A., 1989. Diagenesis of the inorganic phase of cortical bone. In: Price, T.D. (Ed.), *The Chemistry of Prehistoric Human Bone*. Cambridge University Press, Cambridge, pp. 211–229.
- Sillen, A., Morris, A., 1996. Diagenesis of bone from Border Cave: implications for the age of the Border Cave hominids. *Journal of Human Evolution* 31, 499–506.
- Sillen, A., Parkington, J.E., 1996. Diagenesis of bones from Elands Bay Cave. *Journal of Archaeological Science* 23, 535–542.
- Sillen, A., Hall, G., Richardson, S., Armstrong, R., 1998.  $^{87}\text{Sr}/^{86}\text{Sr}$  ratios in modern and fossil food-webs of the Sterkfontein Valley: implications for early hominid habitat preference. *Geochimica et Cosmochimica Acta* 62, 2463–2473.
- Sponheimer, M., Lee-Thorp, J.A., 1999a. The alteration of enamel carbonate environments during fossilisation. *Journal of Archaeological Science* 26, 143–150.
- Sponheimer, M., Lee-Thorp, J.A., 1999b. Isotopic evidence for the diet of an early hominid, *Australopithecus africanus*. *Science* 283, 368–370.
- Sponheimer, M., Lee-Thorp, J.A., 2001. The oxygen isotope composition of mammalian enamel carbonate: a case study from Morea Estate, Mpumalanga Province, South Africa. *Oecologia* 126, 153–157.
- Sullivan, C.H., Krueger, H.W., 1981. Carbon isotope analysis of separate chemical phases in modern and fossil bone. *Nature* 292, 333–335.
- Sternberg, L.S.L., 1989. Oxygen and hydrogen isotope ratios in plant cellulose: mechanisms and applications. In: Rundel, P.W., Ehleringer, J.R., Nagy, K.A. (Eds.), *Stable Isotopes in Ecological Research*. Springer, New York, pp. 124–141.
- Tieszen, L.L., Fagre, T., 1993. Effect of diet quality on the isotopic composition of respiratory  $\text{CO}_2$ , bone collagen, bioapatite and soft tissues. In: Lambert, J.B., Grupe, G. (Eds.), *Prehistoric Human Bone: Archaeology at the Molecular Level*. Springer-Verlag, Berlin, pp. 121–155.
- Tuross, N., Behrensmeier, A.K., Eanes, E.D., Fisher, L.W., Hare, P.E., 1989. Molecular preservation and crystallographic alterations in a weathering sequence of wildebeest bones. *Applied Geochemistry* 4, 261–270.
- Vrba, E.S., 1985. Ecological and adaptive changes associated with early hominid evolution. In: Delson, E. (Ed.), *Ancestors: the Hard Evidence*. Alan R. Liss, New York, pp. 63–71.
- Wang, Y., Cerling, T.E., 1994. A model for fossil tooth and bone diagenesis: implications for paleodiet reconstruction from stable isotopes. *Palaeogeography, Palaeoclimatology, Palaeoecology* 107, 596–606.
- Weiner, S., Bar-Yosef, O., 1990. State of preservation of bones from prehistoric sites in the Near East: a survey. *Journal of Archaeological Science* 17, 187–196.
- White, E.M., Hannus, L.A., 1981. Chemical weathering of bone in archaeological soils. *American Antiquity* 48, 316–322.
- White, C.D., Spence, M.W., Stuart-Williams, H., Le, Q., Schwarcz, H.P., 1998. Oxygen isotopes and the identification of geographical origins: the Valley of Oaxaca versus the Valley of Mexico. *Journal of Archaeological Science* 25, 643–655.
- Wright, L., Schwarcz, H.P., 1996. Infrared evidence for diagenesis of bone apatite at Dos Pilas: paleodietary implications. *Journal of Archaeological Science* 23, 933–944.