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Stable Carbon and Oxygen Isotope Spacing Between Bone and Tooth Collagen and Hydroxyapatite in Human Archaeological Remains

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ABSTRACT Spacing between stable isotope values in bones and teeth is a valuable tool for examining dietary influences and diagenesis. This study examines carbon and oxygen isotope values from collagen and hydroxyapatite (structural carbonate and phosphate) in archaeological human bones and teeth to derive species-specific correlation equations and isotope spacing values. The $\delta^{13}C_{collagen}$ and $\delta^{13}C_{structural\ carbonate}$ in bone and dentin collagen show a strong correlation (R = 0.87, 0.90, respectively) with an average $\Delta^{13}C_{carb\text{-}coll}$ spacing of 5.4‰. The consistency of this isotope spacing with other large mammals and in humans with both low and high protein intake (as indicated by enriched $\delta^{15}N$ values) suggests a similar allocation of protein-derived carbon and whole diet-derived carbon to collagen and structural carbonates, respectively, as other terrestrial mammals regardless of absolute meat intake. The $\delta^{18}O_{structural\ carbonate}$ and $\delta^{18}O_{phosphate}$ show the strongest correlation in enamel (R = 0.65), weaker correlations in dentin (R = 0.59) and bone (R = 0.35), with an average $\Delta^{18}O_{carb\text{-}phos}$ of 7.8‰. This isotope spacing is slightly lower than previously reported for large mammals and limited available data for humans. The results potentially indicate species-specific fractionations and differing access to body water and blood-dissolved inorganic carbonates in the presence of collagen formation. The use of correlation between $\delta^{18}O_{structural\ carbonate}$ and $\delta^{18}O_{phosphate}$ to determine diagenetic state is not recommended. The strength of this correlation observed in bones and teeth is variable and alternate indicators of diagenetic state (i.e. C:N ratios of collagen) provide more robust and independent evidence of isotope preservation despite presence/absence of a strong isotope correlation. Published 2012. This article is a U.S. Government work and is in the public domain in the USA.

Key words: bone; carbon isotopes; collagen; hydroxyapatite; human; oxygen isotopes; tooth

Introduction

The use of stable isotopes to examine diet and provenance is common practice in archaeological and paleontological studies. Dietary and regional indications inherent in carbon, oxygen, and nitrogen isotope values of bones and teeth are applied to a myriad of terrestrial vertebrate remains (see reviews in Peterson & Fry, 1987; Ambrose, 1993; Koch *et al.*, 1994; Koch, 1998; MacFadden, 2000). Stable isotope studies of human remains are less common due to the paucity of robust sample sets and the desire to preserve rare or culturally sensitive specimens. Studies are often limited to a specific isotope analysis as opposed to a full suite of isotope indicators, with a few exceptions (Iacumin *et al.*, 1996a; Loftus & Sealy, 2012). Understanding of the spacing of isotope values between different components of bones and teeth in humans specifically is limited.

Inferences involving isotope spacing between different tissue and mineral fractions in humans often rely on data from large terrestrial non-human mammals that are more thoroughly studied and better understood. While these data are arguably comparable with humans to some extent, humans have a uniquely omnivorous diet that is often unaccounted for in non-human studies of diet-tissue isotope spacing with a few exceptions (Krueger & Sullivan, 1984; Lee-Thorp *et al.*, 1989; Hedges, 2003). This study focuses on an unprecedentedly large data set of 18th–19th century human remains to determine accurate human-specific stable isotope spacing values between bone collagen and hydroxyapatite in bones and teeth for individuals reliant on largely terrestrial diets. These data will allow more accurate correlations between isotope values of different tissues and mineral

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fractions within individuals consuming similar diets, potentially eliminating the need for a full suite of isotope analyses. This study also contributes to understanding pathways between elements in the diet and body tissues and has implications for archaeological diagenetic studies.

Isotope spacing in archaeology

Archaeological bones and teeth contain several components that record isotope information and represent a lifetime average of isotopic input. Collagen is found in both well-preserved bones and tooth dentin. This durable protein contains carbon and nitrogen incorporated directly from the diet (Hedges, 2003). Hydroxyapatite, the inorganic mineral in bones and teeth, contains both phosphate and structural carbonate (-CO_3) substituted in the -PO_4 and -OH locations in this mineral. Both phosphates and structural carbonates preserve oxygen isotopes incorporated through drinking water (Luz & Kolodny, 1985; Bryant & Froelich, 1995; Kohn, 1996). Structural carbonates also preserve carbon isotopes which, similar to collagen, are incorporated from the diet (Hedges, 2003). All isotope values are measured and reported in standard delta notation:

$$\delta X = \left[\left(R_{sample} - R_{standard}\right)/\left(R_{standard}\right)\right] * 1000$$

where X represents the system of interest (i.e. ^{13}C, ^{15}N, ^{18}O) and R represents the ratio of interest (i.e. $^{13}C/^{12}C$, $^{15}N/^{14}N$, $^{18}O/^{16}O$). The carbon standard is Vienna Pee-Dee Belemnite (i.e. V-PDB). The nitrogen standard is atmospheric air. There are two internationally accepted standards for oxygen; this manuscript references all oxygen values to Vienna Standard Mean Ocean Water (i.e. V-SMOW). This study examines the carbon and nitrogen isotopes of bone and tooth collagen ($\delta^{13}C_{collagen}$, $\delta^{15}N_{collagen}$), carbon isotopes of structural carbonate ($\delta^{13}C_{structural\ carbonate}$), and oxygen isotopes in structural carbonates and phosphates ($\delta^{18}O_{structural\ carbonate}$, $\delta^{18}O_{phosphate}$). The different carbon and oxygen components are compared for correlations and spacing ($\Delta^{13}C_{carb\text{-}coll}$, $\Delta^{18}O_{carb\text{-}phos}$).

Carbon isotopes are typically applied to archaeological data sets to determine dietary input due to distinct $\delta^{13}C$ values inherent in plants using the C3 versus C4 photosynthetic pathways (Smith & Epstein, 1971; O'Leary, 1988; Heaton, 1999). Once digested, carbon pathways to the body differ according to the ultimate tissue or mineral into which carbon is incorporated. Previous studies suggest the source of carbon in collagen is mostly dietary protein with lesser contributions from dietary carbohydrates or lipids (Krueger & Sullivan, 1984; Lee-Thorp *et al.*, 1989; Ambrose & Norr, 1993; Tieszen & Fagre, 1993). More recent research suggests a complex system where consumer collagen carbon isotopes are largely influenced by direct routing of particular amino acids from prey collagen with limited secondary synthesis of others from precursor amino acids or additional dietary components (Howland *et al.*, 2003; Jim *et al.*, 2004; Froehle *et al.*, 2010). This results in an overall diet-collagen fractionation of ~2–5‰ in herbivorous mammals (van der Merwe, 1982; Balasse *et al.*, 1999; Roth & Hobson, 2000; Hedges, 2003; Jim *et al.*, 2004; Warinner & Tuross, 2010; review by Koch, 1998) and ~1–2‰ in carnivorous and omnivorous mammals (Bocherens *et al.*, 1991; Hilderbrand *et al.*, 1996; Bocherens & Drucker, 2003; Coltrain *et al.*, 2004; Fox-Dobbs *et al.*, 2007; Coltrain, 2009). The source of carbon in structural carbonates is blood-dissolved inorganic carbon (DIC) derived primarily from carbohydrates and lipids with a smaller contribution from dietary protein resulting in a $\delta^{13}C$ representative of the whole diet (Tieszen & Fagre, 1993; Hedges, 2003; Zazzo *et al.*, 2010). The diet-mineral fractionation for structural carbonates is ~12–15‰ in large herbivorous mammals (Passey *et al.*, 2005; Zazzo *et al.*, 2010; reviews by Koch, 1998; Kohn & Cerling, 2002; Hedges, 2003).

Given these fractionations, the $\Delta^{13}C_{carb\text{-}coll}$ observed in mammals is ~5–7‰ (Krueger & Sullivan, 1984; Lee-Thorp *et al.*, 1989; Ambrose & Norr, 1993; Hedges, 2003; Jim *et al.*, 2004; Warinner & Tuross, 2010), with limited data for humans with a mostly terrestrial diet (Iacumin *et al.*, 1996a; Loftus & Sealy, 2012). It should be noted that this average $\Delta^{13}C_{carb\text{-}coll}$ value of ~5–7‰ is often derived from rodents and swine which arguably have a different metabolism, physiology, and diet than humans. Considering that blood- DIC contains contributions from protein, carbohydrates, and lipids, the varying ratio of these contributions in different diets and different metabolisms may produce a species-specific and comparatively unique $\Delta^{13}C_{carb\text{-}coll}$ value in humans.

Humans in this study are examined for a correlation between $\delta^{13}C_{collagen}$ and $\delta^{13}C_{structural\ carbonate}$ (i.e. variability of $\Delta^{13}C_{carb\text{-}coll}$) and potential differences explainable by dietary input. The $\delta^{15}N_{collagen}$ is used as a qualitative indicator of protein consumption. The $\delta^{15}N_{collagen}$ values become more enriched with increased consumption of animal proteins due to the ~3–4‰ trophic shift inherent in terrestrial food chains (DeNiro & Epstein, 1981; Minagawa & Wada, 1984; Schoeninger & DeNiro, 1984; Sutoh *et al.*, 1987; Post, 2002; Bocherens & Drucker, 2003). A relatively enriched $\delta^{15}N_{collagen}$ value should indicate higher protein input from animal flesh (and therefore higher carbon input from animal flesh) as opposed to dietary input from plants and carbohydrates.

Oxygen isotopes are typically used as proxies for meteoric drinking water in mammals due to the direct correlation between $\delta^{18}O$ of local meteoric drinking water, body water, and $\delta^{18}O_{phosphate}$ or $\delta^{18}O_{structural\ carbonate}$ with subtle variations in fractionation according to species and climate variables (Longinelli, 1984; Luz *et al.*, 1984; Luz & Kolodny, 1985; Levinson *et al.*, 1987; D'Angela & Longinelli, 1990; Bryant & Froelich, 1995; Kohn, 1996; Daux *et al.*, 2008). The relationships between $\delta^{18}O_{phosphate}$, body water, and drinking water in mammals are fairly well understood with several published correlation equations (Longinelli, 1984; Luz *et al.*, 1984; Levinson *et al.*, 1987; Daux *et al.*, 2008). Less well understood is the relationship between $\delta^{18}O_{structural\ carbonate}$ and $\delta^{18}O_{phosphate}$, and relationships between $\delta^{18}O_{structural\ carbonate}$ and body/drinking water. Previously reported fractionation between $\delta^{18}O_{structural\ carbonate}$ and $\delta^{18}O_{phosphate}$ (i.e. $\Delta^{18}O_{carb\text{-}phos}$) is ~9–10‰, and fractionation between $\delta^{18}O_{structural\ carbonate}$ and body water is ~26.3–27.0‰ in mammals (Bryant *et al.*, 1996; Iacumin *et al.*, 1996b; Martin *et al.*, 2008; Pellegrini *et al.*, 2011), with limited data for humans specifically (Iacumin *et al.*, 1996a). This study examines the $\delta^{18}O$ correlation between phosphate and structural carbonate in an effort to accurately predict $\delta^{18}O_{phosphate}$ values from $\delta^{18}O_{structural\ carbonate}$. The former require more expensive offline chemical purifications, and the most expedient mass spectrometry methods to analyze $\delta^{18}O_{phosphate}$ typically show poorer reproducibility (see Methods). It is tempting to streamline the chemical and mass spectrometry procedures and use the better understood phosphate equations to obtain $\delta^{18}O_{phosphate}$ from $\delta^{18}O_{structural\ carbonate}$. However, this calculation cannot be performed without observed consistency in $\Delta^{18}O_{carb\text{-}phos}$ and a value specific for humans.

This study analyzes a set of 18th–19th century North American archaeological remains. Individuals from this time period had a more localized diet than modern humans thus eliminating variability due to global dietary influences inherent in most modern diets. All individuals resided primarily in the eastern United States which shows a limited range in the baseline $\delta^{15}N$ values of vegetation of approximately −4.0 to 0.0‰ with a few isolated exceptions (Handley *et al.*, 1999; Nadelhoffer *et al.*, 2004; Billings & Richter, 2006; Pardo *et al.*, 2007; Templer *et al.*, 2007). These similar and localized diets allow the use of $\delta^{15}N$ values as independent measures of protein input as opposed to an indication of provenance. This sample set is also relatively recent which greatly reduces the likelihood of post-mortem diagenetic alteration, although all samples are individually examined for diagenetic alteration in the course of study.

Methods and materials

Samples

Samples consist of 18th–19th century North American human remains. Archaeological sites are located primarily in the mid- and north-eastern United States with two exceptions in New Mexico (Table 1). All individuals resided in the eastern United States although several are buried in the west due to death during military service.

Chemical extraction

Specimens were exhumed, mechanically cleaned, documented, and catalogued. All specimens were stored without chemical treatments or bone consolidants. Samples were prepared mechanically using a common rotary tool. Whole bone plugs or tooth roots were separated for collagen analyses. Bone, dentin, and enamel were crushed to a coarse powder for phosphate and structural carbonate analyses.

Bone and dentin collagen was extracted according to modified methods of Longin (1971), DeNiro & Epstein (1978), and Bocherens *et al.* (1991). Whole bone or tooth roots (~200 mg) were sonicated in ultra-pure water and

Table 1. Sample sites, locality, and time period

Site	Site Location	Time period	n
Congressional Cemetery	District of Columbia	~1850–1900	34
Trinity Catholic Church	District of Columbia	~1800–1850	23
Woodville Cemetery	Delaware	1790–1850	10
Walton Family Cemetery	Connecticut	~1750–1830	20
Glorieta Pass	New Mexico	1862	38
First African Baptist Church	Pennsylvania	1824–1842	9
Parkway Gravel	Delaware	1800–1900	5
Fort Craig	New Mexico	1854–1877	58

rinsed to remove extraneous dirt and labile salts. Samples were decalcified in 0.6 M hydrochloric acid at 4^0C for several days, with fresh acid added daily, until the reaction completed; average reaction time was 3–5 days. After rinsing in ultra-pure water, the remaining crude protein was soaked in 0.125 M sodium hydroxide for 18 h to remove humic and fulvic acids. Samples were rinsed and reacted in 0.03 M hydrochloric acid at 95^0C for 18 h to separate hot water soluble and insoluble phases. The resulting supernatant was lyophilized to extract purified collagen.

Phosphate in the hydroxyapatite was extracted according to the method of Dettman *et al.* (2001). Bone and tooth powders (~20 mg) were soaked in 2 M hydrofluoric acid solution overnight to liberate phosphate ions. The solution was diluted and buffered with 20% ammonium hydroxide before adding 2 M silver nitrate. The resulting silver phosphate precipitate was rinsed with ultra-pure water and dried (60^0C).

Structural carbonate in the hydroxyapatite was extracted according to modified methods of Bryant *et al.* (1996). Bone and tooth powders (~20 mg) were soaked in 2–3% sodium hypochlorite overnight to remove organic substances. After rinsing in ultra-pure water, samples were soaked in 1 M acetic acid solution buffered with 1 M calcium acetate (pH ~ 4.5) for 4 h to remove secondary carbonate phases. The remaining material was rinsed in ultra-pure water and dried (60^0C).

Mass spectrometry methods

All analyses were conducted on Thermo Delta V mass spectrometers at the Smithsonian OUSS/MCI Stable Isotope Mass Spectrometry Laboratory. Collagen was weighed into tin capsules (~0.5 mg) and combusted in a Costech 4010 elemental analyzer (EA) producing CO_2 and N_2 gases which were introduced to the mass spectrometer via a Conflo IV interface and measured for $\delta^{13}C_{collagen}$ and $\delta^{15}N_{collagen}$. Silver phosphates were weighed into silver capsules (~0.5 mg) and thermally decomposed in a Thermo temperature conversion EA to CO gas which was introduced to the mass spectrometer via a Conflo IV interface and measured for $\delta^{18}O_{phosphate}$. Structural carbonate samples were acidified in 102% phosphoric acid (density $\geq$1.92) at 25^0C for 18 h in a pure helium environment producing CO_2 gas which was introduced to the mass spectrometer via a Gas Bench II system and measured for $\delta^{13}C_{structural\ carbonate}$ and $\delta^{18}O_{structural\ carbonate}$.

Collagen samples were linearly corrected to a house acetanilide standard and the urea UIN-3 standard (Schimmelmann *et al.*, 2009), both of which are calibrated to international standards USGS-40 and USGS-41. All $\delta^{13}C_{collagen}$ and $\delta^{15}N_{collagen}$ data are reported with the error inherent in the international standards used for calibration (i.e. ±0.2‰, 1σ); internal reproducibility was <0.2‰ (1σ). Structural carbonate samples were linearly corrected to the international standards NBS-19 and LSVEC. All $\delta^{13}C_{structural\ carbonate}$ and $\delta^{18}O_{structural\ carbonate}$ data are reported with the error inherent in these standards (i.e. ±0.2‰, 1σ); internal reproducibility was <0.2‰ (1σ). Phosphate samples were linearly corrected to the international standards USGS-34 and USGS-35. The error inherent in these standards is ±0.2‰ (1σ); internal reproducibility was <0.4‰ (1σ). All $\delta^{18}O_{phosphate}$ data are therefore reported with an error of ±0.4‰ (1σ).

Statistical analyses

All regressions are parametric least squares regressions assuming normal distribution and constant variance. All correlations are Pearson product moment correlations assuming normal distribution and constant variance of the residuals.

Results

Diagenesis

Although the samples are relatively modern, their burial introduces potential for post-mortem diagenetic alteration. Data were selected for inclusion in analyses based upon the previously established C:N ratio range of 2.8–3.6 and a weight %N yield of ~11–16% for well-preserved collagen (DeNiro, 1985; Ambrose, 1990; Bocherens *et al.*, 1991, 1994, 1996, 1997; Drucker *et al.*, 2001, 2003; McNulty *et al.*, 2002; Coltrain *et al.*, 2004; Jorkov *et al.*, 2007). Approximately 80% (154 of 197) of analyzed samples satisfied these requirements (Figure 1). While criteria for the preservation of original phosphate and structural carbonate isotope values has been considered (Tuross *et al.*, 1989; Michel *et al.*, 1995; Person *et al.*, 1995, 1996; Iacumin *et al.*, 1996b; Kohn *et al.*, 1999; Zazzo *et al.*, 2004), this study includes only those samples yielding well-preserved organic collagen per the above stated criteria. It has been noted that mineral crystals in bones and teeth are nested within the organic matrix (Francillon-Vieillot *et al.*, 1990; Veis, 2003), such that the presence of well-preserved organic matter protects the mineral fraction of bones and teeth from recrystallization and subsequent isotope alteration (Nelson *et al.*, 1986; Person *et al.*, 1996; Tütken *et al.*, 2008).

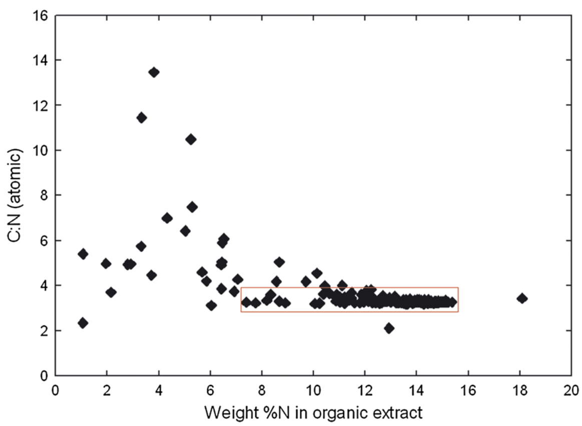

Figure 1. C:N ratios and weight %N yields from organic extracts. The rectangle identifies the samples that meet the criteria for good preservation.

Stable isotope ranges and correlations

The $\delta^{13}C_{collagen}$ ranges from −20.9 to −7.5‰; the $\delta^{13}C_{structural\ carbonate}$ ranges from −14.9 to −5.2‰ (Table 2). The $\delta^{13}C_{collagen}$ and $\delta^{13}C_{structural\ carbonate}$ are strongly correlated (Figure 2, R = 0.88). Both dentin and bone exhibit similar trends with strong correlations (R = 0.90, 0.87, respectively). Basic linear regression relationships are listed in Table 3. The average $\Delta^{13}C_{carb\text{-}coll}$ is 5.4 ± 1.1‰ (1σ) with little variation between dentin and bone. No significant correlation exists between $\Delta^{13}C_{carb\text{-}coll}$ and $\delta^{15}N_{collagen}$ (Figure 3, R = 0.26).

The $\delta^{18}O_{structural\ carbonate}$ ranges from +20.4 to +30.9‰; the $\delta^{18}O_{phosphate}$ ranges from +13.3 to +22.8‰ (Table 2). The correlation between $\delta^{18}O_{structural\ carbonate}$ and $\delta^{18}O_{phosphate}$ is moderate (Figure 4, R = 0.67). Tooth enamel shows the strongest correlation between values (R = 0.65), while dentin and bone show weaker correlations (R = 0.59, 0.35, respectively). Basic linear regression relationships are listed in Table 3. The average $\Delta^{18}O_{carb\text{-}phos}$ is 7.8 ± 1.5‰ (1σ) with little variation between dentin, enamel, and bone. Enamel shows a notably lower variability (±1.0, 1σ) compared to dentin (±1.5, 1σ) and bone (±1.6, 1σ).

Note that isotope data is not distinguished by site locality. While absolute values of $\delta^{13}C_{collagen}$, $\delta^{13}C_{structural\ carbonate}$, $\delta^{18}O_{structural\ carbonate}$, and $\delta^{18}O_{phosphate}$ differ by locality due to differing regional vegetation and meteoric water composition, there were no observable trends between $\Delta^{13}C_{carb\text{-}coll}$ or $\Delta^{18}O_{carb\text{-}phos}$ and locality. Likewise, $\delta^{15}N_{collagen}$ shows no correlation with individual sites. The implications of regional differences in isotope values will be discussed in a future manuscript.

Discussion

The correlation between $\delta^{13}C_{collagen}$ and $\delta^{13}C_{structural\ carbonate}$ is robust with fairly consistent $\Delta^{13}C_{carb\text{-}coll}$ values of ~4.2–6.4‰. Previous studies of humans show $\Delta^{13}C_{carb\text{-}coll}$ values of ~2.6–6.2‰, the lower range of which is found in individuals with a partially marine diet (Iacumin *et al.*, 1996a; Loftus & Sealy, 2012). Data from this study originates from individuals with strictly terrestrial diets and agrees with the larger spacing observed in similar individuals (Iacumin *et al.*, 1996a), as well as previous determinations of the offset for omnivorous non-human mammals (Krueger & Sullivan, 1984; Lee-Thorp *et al.*, 1989; Hedges, 2003). This observation suggests that carbon diet-tissue fractionations between different organic and mineral fractions of bone and dentin function similarly in humans and other large mammals. The strong correlation supports consistent fractionation of carbon isotopes between diet and carbon-containing fractions (i.e. collagen or hydroxyapatite) in bones and teeth. The $\Delta^{13}C_{carb\text{-}coll}$ does not correlate with $\delta^{15}N_{collagen}$ which suggests that diet in these humans is balanced such that the ultimate source of collagen carbon is consistently protein from animal flesh while the contribution to hydroxyapatite contains components from the whole diet. While the $\delta^{15}N_{collagen}$ serves as a rough indicator of the relative meat proportion in the diet, it appears that individuals with relatively lower animal protein intake are still allocating a similar proportion of animal derived carbon (compared to carbohydrate or lipid derived carbon) to their collagen.

The average $\Delta^{18}O_{carb\text{-}phos}$ of 7.8 ± 1.5‰ (1σ) is less than previously observed values of ~9–10‰ (Bryant *et al.*, 1996; Iacumin *et al.*, 1996a, 1996b; Martin *et al.*, 2008; Pellegrini *et al.*, 2011). While $\Delta^{18}O_{carb\text{-}phos}$ variation between dentin, enamel, and bone is minimal, enamel and dentin show notably lower intra-variability. Correlation between $\delta^{18}O_{structural\ carbonate}$ and $\delta^{18}O_{phosphate}$ values is only moderate at best with the strongest correlation noted for tooth enamel. All data included in these calculations demonstrated good collagen quality, thereby rendering diagenetic alteration an unlikely explanation for variability observed in these correlations or the $\Delta^{18}O_{carb\text{-}phos}$. This observation suggests that fractionation of oxygen when incorporated into $-CO_3$ or $-PO_4$ differs according to the mineral and body element. It is typically assumed that both the carbonate and phosphate precipitate from the same body water pool (Bryant & Froelich, 1995; Kohn, 1996); the resulting $\delta^{18}O$ values should therefore correlate. Data from human enamel in this study support this idea, but data from bone and dentin show support to a lesser degree.

Table 2. Data. Only data derived from samples determined to be well-preserved and therefore used in calculations are listed here

Designation	Element	$\delta^{18}O_{phosphate}$ (‰, VSMOW)	$\delta^{18}O_{structural\ carbonate}$ (‰, VSMOW)	$\Delta^{18}O_{carb-phos}$ (‰, VSMOW)	$\delta^{13}C_{structural\ carbonate}$ (‰, VPDB)	$\delta^{13}C_{collagen}$ (‰, VPDB)	$\Delta^{13}C_{carb-coll}$ (‰, VPDB)	$\delta^{15}N_{collagen}$ (‰, air)	C:N
29LA1091-BOR-05	Metatarsal	17.9	26.5	8.5	−10.2	−15.9	5.7	10.4	3.3
29LA1091-BOR-06	Metatarsal	18.5	27.1	8.7	−11.2	−17.4	6.2	9.9	3.2
29LA1091-BOR-07	Metatarsal	17.4	27.1	9.7	−10.1	−17.2	7.1	12.3	3.3
29LA1091-BOR-11	Metatarsal	17.9	27.6	9.7	−7.0	−11.6	4.7	11.1	3.2
29LA1091-BOR-13	Metatarsal	16.4	25.1	8.7	−9.8	−16.5	6.7	11.3	3.2
29LA1091-BOR-14	Metatarsal	17.1	25.6	8.5	−10.9	−17.4	6.5	11.2	3.3
29LA1091-BOR-15	Metatarsal	17.4	27.5	10.1	−7.5	−14.2	6.7	11.1	3.2
29LA1091-BOR-18	Metatarsal	18.8	26.2	7.3	−6.7	−12.4	5.6	13.1	3.6
29LA1091-BOR-19	Metatarsal	17.0	27.1	10.1	−6.0	−12.0	6.0	10.3	3.2
29LA1091-BOR-23A	Talus	17.5	26.3	8.8	−9.0	−15.1	6.2	11.2	3.3
29LA1091-BOR-24	Metatarsal	13.3	23.2	9.9	−6.9	−12.3	5.4	10.5	3.2
29LA1091-BOR-26	Metatarsal	17.4	23.9	6.5	−5.7	−10.9	5.2	12.0	3.4
29LA1091-BOR-30	Metacarpal	15.8	24.5	8.7	−11.5	−18.1	6.6	11.3	3.3
29LA1091-BOR-31	Metatarsal	16.2	24.6	8.4	−5.2	−11.1	5.9	11.5	3.3
29LA1091-BOR-32	Metatarsal	18.1	24.4	6.3	−7.8	−13.3	5.5	12.0	3.4
29LA1091-BOR-33	Metatarsal	16.1	24.6	8.4	−8.4	−12.1	3.7	11.8	3.3
29LA1091-BOR-34	Metatarsal	16.5	25.8	9.3	−10.6	−18.2	7.6	10.7	3.2
29LA1091-BOR-35	Metatarsal	16.0	24.9	8.9	−6.1	−11.9	5.8	12.1	3.5
29LA1091-BOR-38	Metatarsal	17.2	29.7	12.5	−7.7	−14.0	6.3	9.9	3.2
29LA1091-BOR-39	Metatarsal	16.6	22.5	5.9	−10.3	−15.3	5.0	11.9	3.3
29LA1091-BOR-39	Metatarsal	16.6	20.8	4.2	−10.0	−15.0	5.0	12.3	3.3
29LA1091-BOR-39	Metatarsal	16.1	22.5	6.4	−10.3	−15.3	5.0	11.9	3.3
29LA1091-BOR-40	Metacarpal	15.1	24.6	9.5	−5.4	−11.6	6.1	10.7	3.4
29LA1091-BOR-41	Metatarsal	18.1	26.9	8.8	−8.0	−13.8	5.8	9.3	3.2
29LA1091-BOR-43	Metatarsal	18.0	23.0	5.1	−7.2	−12.9	5.7	11.4	3.4
29LA1091-BOR-44	Metatarsal	20.2	26.3	6.1	−9.8	−15.5	5.7	11.9	3.4
29LA1091-BOR-45	Metatarsal	18.5	24.1	5.6	−11.3	−17.6	6.3	11.9	3.4
29LA1091-BOR-47	Metatarsal	17.5	23.3	5.8	−8.9	−14.3	5.5	9.9	3.3
29LA1091-BOR-49	Metatarsal	17.0	25.9	8.9	−10.0	−16.7	6.7	11.4	3.2
29LA1091-BOR-50	Metatarsal	16.6	24.5	8.0	−10.0	−17.3	7.4	9.0	3.3
29LA1091-BOR-51	Metatarsal	18.7	26.0	7.3	−7.7	−13.5	5.8	11.0	3.3
29LA1091-BOR-54	Metatarsal	18.5	26.5	8.0	−7.6	−13.8	6.1	10.9	3.3
29LA1091-BOR-55	Metatarsal	17.7	24.7	7.0	−11.8	−18.0	6.3	12.7	3.3
29LA1091-BOR-57	Metatarsal	19.2	25.1	5.9	−5.6	−11.4	5.9	10.4	3.2
29LA1091-BOR-58	Metatarsal	17.2	23.7	6.4	−12.0	−18.6	6.6	11.3	3.4
29LA1091-BOR-62	Metatarsal	18.7	21.9	3.2	−7.0	−12.2	5.2	11.3	3.3
29LA1091-BOR-64	Metatarsal	16.2	25.1	8.9	−12.3	−19.2	7.0	11.6	3.2
29LA1091-BOR-65	Metatarsal	18.0	26.7	8.7	−12.7	−19.7	7.0	11.4	3.2
29LA1091-BOR-66	Metatarsal	14.9	25.9	10.9	−10.3	−17.5	7.2	10.8	3.2
51CAUSTEN-CC-07	Metacarpal	18.1	24.3	6.2	−9.4	−15.0	5.6	10.3	3.3
51CAUSTEN-CC-08	Metacarpal	15.9	21.7	5.8	−11.7	−17.0	5.3	11.3	3.3
51CAUSTEN-CC-11	Metacarpal	16.9	21.5	4.6	−12.2	−16.7	4.5	10.9	3.3
51CAUSTEN-CC-12	Metacarpal	17.3	24.0	6.6	−11.8	−16.2	4.4	12.1	3.2
51CAUSTEN-CC-13	Metacarpal	15.7	20.8	5.0	−10.9	−16.1	5.3	11.7	3.3
51CAUSTEN-CC-14	Metacarpal	17.4	23.4	6.0	−11.9	−17.2	5.3	11.1	3.3
51KEYWORTH-CC-01	Metatarsal	16.8	24.4	7.6	−10.8	−15.8	4.9	11.8	3.4
51KEYWORTH-CC-02	Metatarsal	15.6	24.3	8.7	−9.0	−15.0	6.0	10.7	3.3

51KEYWORTH-CC-04	Radius	17.0	24.5	7.6	−10.2	−15.0	4.7	11.2	3.3
51KEYWORTH-CC-05	Fibula	16.8	25.0	8.2	−10.5	−15.5	5.0	10.4	3.4
51KEYWORTH-CC-06	Humerus	17.0	25.0	7.9	−10.7	−16.5	5.8	10.0	3.3
51KEYWORTH-CC-07	Metatarsal	16.1	23.4	7.4	−12.1	−17.3	5.2	9.8	3.3
51KEYWORTH-CC-08	Metatarsal	16.7	23.4	6.7	−11.9	−17.3	5.4	9.6	3.3
51KEYWORTH-CC-08	Tibia	19.6	24.1	4.5	−11.8	−17.4	5.6	9.8	3.3
51KEYWORTH-CC-10	Metatarsal	17.1	23.1	6.0	−10.9	−15.8	4.9	11.1	3.4
51KEYWORTH-CC-11	Temporal	16.9	24.5	7.6	−6.0	−16.4	10.4	11.3	3.5
51KEYWORTH-CC-12A	Femur	18.5	25.3	6.7	−10.9	−17.2	6.3	11.0	3.2
51KEYWORTH-CC-13	Metatarsal	18.3	25.3	6.9	−9.8	−15.8	6.0	10.8	3.3
51KEYWORTH-CC-14	Metacarpal	18.4	26.0	7.5	−12.9	−17.0	4.1	10.5	3.3
51MACOMB-CC-1	Metacarpal	16.6	25.9	9.4	−12.4	−17.2	4.8	12.2	3.3
51MACOMB-CC-2	Metacarpal	16.7	26.1	9.4	−12.4	−18.0	3.6	11.4	3.3
51WHITE-CC-01	Metacarpal	17.1	25.1	8.0	−10.5	−14.9	4.3	11.0	3.3
51WHITE-CC-02	Fibula	16.8	24.4	7.6	−9.2	−13.7	4.4	10.2	3.3
51WHITE-CC-03	Metatarsal	16.1	24.6	8.5	−10.2	−15.6	5.5	11.0	3.3
51WHITE-CC-07	Radius	17.2	24.5	7.3	−10.7	−17.1	6.4	10.9	3.3
51WHITE-CC-08	Fibula	17.1	24.8	7.7	−9.4	−13.0	3.6	11.7	3.3
51WHITE-CC-09A	Radius	17.7	24.4	6.7	−9.5	−15.5	5.9	10.1	3.3
51WHITE-CC-09B	Metacarpal	17.0	23.3	6.3	−7.9	−12.7	4.8	10.3	3.3
51WHITE-CC-10	Clavicle	16.3	25.1	8.8	−11.3	−16.0	4.6	12.1	3.3
51WHITE-CC-11	Fibula	16.6	20.4	3.8	−10.7	−15.5	4.8	10.5	3.3
51WHITE-CC-12B	Radius	16.1	20.4	4.3	−10.1	−16.0	6.0	10.2	3.3
51WHITE-CC-13	Radius	17.8	21.3	3.5	−11.8	−17.0	5.2	10.6	3.4
6CT58-5-AMM01	Femur	17.2	23.9	6.8	−8.9	−13.8	4.8	9.8	3.3
6CT58-5-AMM02	Femur	17.2	23.4	6.2	−9.2	−13.6	4.4	10.1	3.2
6CT58-5-AMM04A	Femur	16.0	24.9	8.9	−9.9	−15.1	5.2	10.3	3.2
6CT58-5-AMM05	Femur	17.4	24.7	7.3	−10.0	−15.9	5.9	9.8	3.2
6CT58-5-AMM07	Femur	15.8	25.3	9.5	−9.3	−15.0	5.7	9.6	3.3
6CT58-5-AMM08A	Femur	16.2	25.3	9.1	−8.0	−13.8	5.8	9.4	3.3
6CT58-5-AMM09	Humerus	16.6	23.9	7.3	−8.2	−13.9	5.7	9.7	3.4
6CT58-5-AMM11	Femur	16.4	23.0	6.6	−8.8	−13.8	5.0	9.5	3.2
6CT58-5-AMM19	Femur	17.8	25.6	7.8	−9.7	−15.2	5.5	9.7	3.4
6CT58-5-AMM20	Humerus	16.2	24.2	8.0	−8.6	−13.5	4.8	9.6	3.2
6CT58-5-AMM21	Femur	16.6	25.0	8.4	−9.5	−14.3	4.8	9.9	3.3
6CT58-5-AMM22	Femur	16.5	22.5	6.0	−9.5	−13.7	4.2	10.0	3.3
6CT58-5-AMM23	Femur	17.4	24.7	7.3	−7.3	−13.2	5.8	8.7	3.3
6CT58-5-AMM25	Temporal	16.8	25.9	9.1	−8.7	−13.7	5.0	9.4	3.3
6CT58-5-AMM26	Rib	17.5	25.6	8.1	−9.1	−13.8	4.7	9.4	3.3
6CT58-5-AMM27	Temporal	17.1	24.5	7.4	−9.0	−13.8	4.7	9.6	3.4
7NCE176-DHCA-V01	Petrous	17.9	24.9	7.0	−9.9	−14.3	4.4	10.0	3.3
7NCE176-DHCA-V02	Petrous	17.2	24.9	7.7	−7.1	−11.5	4.4	10.6	3.3
7NCE176-DHCA-V03	Mandible	17.8	24.8	7.0	−6.2	−11.5	5.3	9.7	3.4
7NCE176-DHCA-V04	Mandible	17.4	23.8	6.4	−9.4	−15.3	5.8	10.8	3.5
7NCE98A-WOODVILLE-04	Fibula	18.2	26.0	7.8	−6.9	−11.6	4.7	11.4	3.5
7NCE98A- WOODVILLE-SLOPEB	Ulna	16.7	24.2	7.5	−7.8	−12.1	4.3	10.4	3.3

(*Continues*)

Table 2. (Continued)

Designation	Element	$\delta^{18}O_{phosphate}$ (‰, VSMOW)	$\delta^{18}O_{structural\ carbonate}$ (‰, VSMOW)	$\Delta^{18}O_{carb\text{-}phos}$ (‰, VSMOW)	$\delta^{13}C_{structural\ carbonate}$ (‰, VPDB)	$\delta^{13}C_{collagen}$ (‰, VPDB)	$\Delta^{13}C_{carb\text{-}coll}$ (‰, VPDB)	$\delta^{15}N_{collagen}$ (‰, air)	C:N
7NCE98A-DHCA-01	Metacarpal	17.8	25.6	7.8	−6.1	−11.3	5.2	11.2	3.4
7NCE98A-DHCA-08	Metatarsal	17.9	24.3	6.4	−6.6	−10.9	4.2	10.9	3.2
FABC-08-107a	Metacarpal	16.2	24.8	8.6	−10.6	−16.2	5.6	10.7	3.3
FABC-08-3300	Metatarsal	16.7	24.2	7.4	−11.5	−17.0	5.5	8.8	3.4
FABC-08-63b	Metatarsal	16.8	23.8	7.1	−11.1	−16.6	5.5	10.5	3.4
FABC-08-8000	Metacarpal	16.2	23.5	7.3	−7.8	−12.4	4.6	10.7	3.4
FABC-08-9100	Phalanx	17.0	24.1	7.1	−5.8	−12.6	6.8	11.1	3.4
GLO-099-1A	Femur	18.5	27.0	8.5	−7.5	−13.5	5.9	11.5	3.3
GLO-099-2A	Femur	17.6	26.7	9.2	−8.9	−15.2	6.3	10.8	3.3
GLO-099-2C	Femur	18.6	26.0	7.4	−8.1	−12.3	4.2	10.4	3.3
GLO-099-2E	Femur	19.2	26.5	7.3	−7.0	−12.9	5.9	10.4	3.3
GLO-099-2H	Femur	19.4	27.2	7.8	−7.5	−12.5	5.0	10.9	3.3
GLO-099-2R	Metatarsal	18.9	23.6	4.7	−8.9	−16.0	7.1	10.7	3.3
GLO-099-2 V	Femur	18.9	26.8	7.9	−8.7	−12.5	3.8	10.3	3.3
GLO-099-2X	Femur	16.9	25.1	8.2	−10.6	−12.7	2.1	10.1	3.3
TRINITY-EAST-10	Metacarpal	17.5	24.5	7.0	−7.5	−12.2	4.6	11.8	3.2
TRINITY-EAST-11	Temporal	–	26.2	–	−7.6	−13.8	6.2	12.5	3.4
TRINITY-EAST-13	Metacarpal	18.2	26.2	8.0	−6.8	−11.5	4.7	11.4	3.5
TRINITY-EAST-14	Temporal	–	25.8	–	−7.9	−12.3	4.5	10.4	3.4
TRINITY-EAST-26	Temporal	18.2	25.6	7.4	−8.4	−12.9	4.5	11.3	3.3
TRINITY-WEST-01	Temporal	16.6	24.6	8.0	−8.2	−10.8	2.5	9.7	3.6
29LA1091-BOR-06	Premolar - dentin	18.5	25.3	6.8	−14.2	−20.6	6.4	6.2	3.2
29LA1091-BOR-06	Premolar - enamel	19.6	26.5	6.9	−13.9	–	–	–	–
29LA1091-BOR-07	Premolar - dentin	17.8	22.4	4.6	−12.1	−19.2	7.1	12.1	3.5
29LA1091-BOR-07	Premolar - enamel	20.0	25.4	5.4	−12.3	–	–	–	–
29LA1091-BOR-13	Premolar - dentin	17.9	26.8	8.9	−10.9	−16.6	5.7	11.1	3.3
29LA1091-BOR-14	Premolar - enamel	17.9	25.6	7.7	−13.0	–	–	–	–
29LA1091-BOR-15	Premolar - enamel	21.1	28.3	7.2	−1.4	–	–	–	–
29LA1091-BOR-47	Premolar - dentin	18.1	26.7	8.7	−6.9	−13.0	6.1	10.3	3.3
29LA1091-BOR-47	Premolar - enamel	17.6	26.3	8.7	−6.4	–	–	–	–
29LA1091-BOR-49	Molar - dentin	18.1	24.8	6.7	−13.0	−20.7	7.8	11.4	3.6
29LA1091-BOR-49	Molar - enamel	17.7	26.6	8.9	−14.2	–	–	–	–
51KEYWORTH-CC-05	Premolar - enamel	19.6	26.2	6.6	−9.9	–	–	–	–
51KEYWORTH-CC-12A	Premolar - dentin	18.5	24.8	6.3	−13.3	−17.8	4.5	12.2	3.3
51KEYWORTH-CC-12A	Premolar - enamel	19.9	26.3	6.4	−13.3	–	–	–	–
7NCE176-DHCA-X01L	Molar - dentin	17.9	24.5	6.6	−7.7	−12.8	5.1	10.6	3.4
GLO-099-2A	Molar - dentin	16.7	27.4	10.7	−9.2	−15.5	6.3	10.8	3.3
GLO-099-2A	Molar - enamel	16.9	26.4	9.5	−9.5	–	–	–	–
GLO-099-2AA	Canine - dentin	20.0	28.4	8.4	−6.8	−12.2	5.4	10.2	3.3
GLO-099-2AA	Canine - enamel	20.4	28.3	7.9	−6.0	–	–	–	–
GLO-099-2B	Canine - dentin	–	28.7	–	−6.8	−11.9	5.1	10.1	3.3
GLO-099-2BB	Molar - dentin	21.1	28.3	7.3	−8.1	−13.7	5.6	12.1	3.3
GLO-099-2BB	Molar - enamel	22.8	28.9	6.2	−7.4	–	–	–	–
GLO-099-2C	Molar - dentin	19.6	30.5	10.9	−7.9	−12.3	4.4	10.0	3.2
GLO-099-2C	Molar - enamel	20.4	27.4	7.0	−8.1	–	–	–	–
GLO-099-2CC	Molar - dentin	16.8	26.3	9.6	−12.9	−18.2	5.4	8.1	3.1
GLO-099-2CC	Molar - enamel	16.5	25.7	9.2	−13.0	–	–	–	–
GLO-099-2D	Canine - dentin	20.2	30.8	10.5	−6.4	−12.7	6.3	10.3	3.3

GLO-099-2DD	Canine - enamel	20.4	27.4	7.0	−6.7	–	–	–	–
GLO-099-2EE	Canine - dentin	21.1	29.2	8.1	−5.9	−10.3	4.4	11.0	3.2
GLO-099-2EE	Canine - enamel	22.1	29.5	7.4	−4.4	–	–	–	–
GLO-099-2 G	Canine - dentin	17.7	26.6	8.9	−13.1	−19.7	6.6	10.9	3.3
GLO-099-2 G	Canine - enamel	19.2	27.6	8.5	−14.0	–	–	–	–
GLO-099-2H	Molar - dentin	20.3	28.7	8.4	−6.7	−12.6	5.9	11.0	3.3
GLO-099-2H	Molar - enamel	19.7	28.4	8.7	−6.8	–	–	–	–
GLO-099-2I	Canine - dentin	20.0	28.4	8.5	−7.4	−12.2	4.7	10.0	3.2
GLO-099-2I	Canine - enamel	19.6	28.4	8.8	−7.4	–	–	–	–
GLO-099-2 J	Canine - dentin	19.2	28.8	9.7	−7.4	−12.5	5.0	10.9	3.3
GLO-099-2 J	Canine - enamel	20.2	28.3	8.1	−8.5	–	–	–	–
GLO-099-2 K	Molar - dentin	20.5	29.1	8.6	−6.2	−11.6	5.4	10.8	3.3
GLO-099-2 K	Molar - enamel	21.4	28.8	7.5	−6.4	–	–	–	–
GLO-099-2 L	Canine - dentin	17.9	26.8	8.9	−11.8	−15.8	4.0	11.8	3.2
GLO-099-2 L	Canine - enamel	19.2	26.4	7.2	−12.3	–	–	–	–
GLO-099-2 M	Molar - dentin	19.0	28.9	9.9	−5.7	−12.1	6.4	9.9	3.3
GLO-099-2 M	Molar - enamel	19.6	28.1	8.5	−5.7	–	–	–	–
GLO-099-2 N	Molar - dentin	19.9	28.1	8.2	−8.1	−13.5	5.5	11.7	3.3
GLO-099-2 N	Molar - enamel	19.5	27.6	8.1	−9.1	–	–	–	–
GLO-099-2O	Molar - dentin	20.9	29.2	8.3	−6.6	−11.6	5.0	11.9	3.3
GLO-099-2P	Canine - dentin	20.3	28.9	8.6	−7.2	−9.2	2.0	10.1	3.3
GLO-099-2P	Canine - enamel	19.5	28.1	8.7	−7.4	–	–	–	–
GLO-099-2Q	Molar - dentin	18.7	28.5	9.8	−8.8	−13.3	4.5	11.5	3.3
GLO-099-2R	Molar - dentin	18.1	28.1	10.0	−6.1	−10.9	4.9	10.6	3.2
GLO-099-2S	Molar - dentin	18.1	28.6	10.5	−9.5	−14.0	4.5	10.9	3.3
GLO-099-2S	Molar - enamel	21.6	30.1	8.4	−9.6	–	–	–	–
GLO-099-2 T	Canine - dentin	19.9	29.6	9.7	−7.5	−15.0	7.4	10.3	3.5
GLO-099-2 T	Canine - enamel	20.1	29.6	9.6	−6.7	–	–	–	–
GLO-099-2U	Canine - dentin	16.7	26.8	10.2	−12.0	−18.8	6.8	9.2	3.2
GLO-099-2U	Canine - enamel	18.6	26.3	7.7	−12.4	–	–	–	–
GLO-099-2 V	Molar - dentin	19.0	30.1	11.1	−6.7	−12.7	6.0	10.7	3.3
GLO-099-2 V	Molar - enamel	21.1	28.8	7.6	−5.8	–	–	–	–
GLO-099-2X	Incisor - dentin	–	28.1	–	−7.8	−13.9	6.2	10.4	3.5
GLO-099-2Y	Molar - dentin	17.1	26.5	9.5	−10.7	−10.7	0.0	11.7	3.3
GLO-099-2Y	Molar - enamel	18.2	25.3	7.1	−13.2	–	–	–	–
GLO-099-2Z	Canine - dentin	–	28.5	–	−6.3	−12.1	5.8	9.6	3.2
TRINITY-EAST-22A	Molar - dentin	19.6	27.0	7.4	−14.6	−20.9	6.3	10.5	3.3
TRINITY-EAST-22A	Molar - enamel	21.0	28.5	7.5	−15.0	–	–	–	–
TRINITY-EAST-25	Premolar - dentin	–	26.9	–	−14.9	−20.8	5.9	11.3	3.5

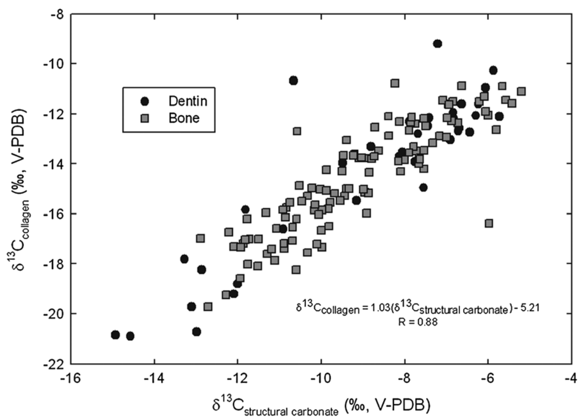

Figure 2. Carbon isotope values from structural carbonate and collagen. The displayed formula is a linear regression on all data.

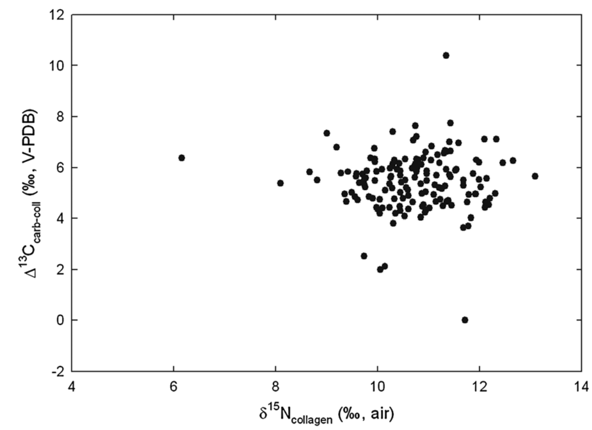

Figure 3. Nitrogen isotope data from collagen and the spacing between carbon isotopes in structural carbonates and collagen.

It is however known that the carbonate fraction of hydroxyapatite also incorporates an isotope contribution from blood- DIC. This same pool of blood-DIC is tapped during collagen formation, albeit to a much lesser degree (Jim *et al.*, 2004; Froehle *et al.*, 2010). It is possible that the pool of blood-DIC is therefore fractionated during collagen formation, which occurs before mineralization of hydroxyapatite (Francillon-Vieillot *et al.*, 1990; Veis, 2003), thus diminishing the correlation between the $\delta^{18}O_{structural\ carbonate}$ and $\delta^{18}O_{phosphate}$ values in bone and dentin. The noted difference between the $\Delta^{18}O_{carb\text{-}phos}$ value of 7.8‰ in humans from this study and values of ~9–10‰ observed in other research focusing on non-human mammals could also be due to a species-specific metabolic or physiological mechanism.

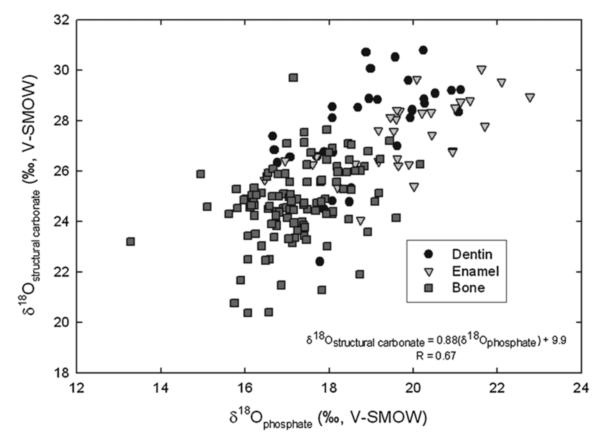

Figure 4. Oxygen isotope values of structural carbonates and phosphates. The displayed formula is a linear regression on all data.

Table 3. Linear regression equations

Body Element	Regression Formula	R	Average $\Delta^{13}C_{carb\text{-}coll}$
Carbon isotope comparisons. All formulas based on $\delta^{13}C$ referenced to V-PDB.			
Bone and dentin combined	$\delta^{13}C_{collagen} = 1.03(\delta^{13}C_{structural\ carbonate}) - 5.21$	0.88	
Bone	$\delta^{13}C_{collagen} = 1.00(\delta^{13}C_{structural\ carbonate}) - 5.48$	0.87	
Dentin	$\delta^{13}C_{collagen} = 1.07(\delta^{13}C_{structural\ carbonate}) - 4.78$	0.9	
			5.4 ± 1.1‰ (1σ)
Oxygen isotope comparisons. All formulas based on $\delta^{18}O$ referenced to V-SMOW.			
Body Element	**Regression Formula**	**R**	**Average $\Delta^{18}O_{carb\text{-}phos}$**
Bone, dentin, enamel combined	$\delta^{18}O_{structural\ carbonate} = 0.88(\delta^{18}O_{phosphate}) + 9.9$	0.67	
Bone	$\delta^{18}O_{structural\ carbonate} = 0.51(\delta^{18}O_{phosphate}) + 15.9$	0.35	
Dentin	$\delta^{18}O_{structural\ carbonate} = 0.87(\delta^{18}O_{phosphate}) + 11.3$	0.59	
Enamel	$\delta^{18}O_{structural\ carbonate} = 0.63(\delta^{18}O_{phosphate}) + 14.9$	0.65	
			7.8 ± 1.5‰ (1σ)

With only moderate correlation between $\delta^{18}O_{structural\ carbonate}$ and $\delta^{18}O_{phosphate}$, the use of one to predict the other is unfeasible. Various contrasting studies have suggested that an observed correlation indicates good preservation (Bryant *et al.*, 1996; Iacumin *et al.*, 1996a, 1996b), while divergence from a strict correlation is still observed in some well-preserved samples (Martin *et al.*, 2008; Kirsanow & Tuross, 2011; Pellegrini *et al.*, 2011). Given that this study incorporated external controls for diagenesis and included only well-preserved samples in all subsequent correlation calculations, we hereby contend that presence/absence of correlation between $\delta^{18}O_{structural\ carbonate}$ and $\delta^{18}O_{phosphate}$ is not indicative of diagenetic state. Given that this study produced different correlations in different body elements (i.e. enamel, dentin, or bone), it appears that this method of examining diagenesis is unreliable, and we recommend using it with caution. Consideration must be given to the body element tested if such a correlation is used as a diagenetic indicator. Rather it is hereby recommended that an alternate method of examining diagenesis be used, such as collagen quality indicators or examination of recrystallization (Person *et al.*, 1995; Kohn *et al.*, 1999; Thompson *et al.*, 2011). All such methods have their own inherent level of error, but with proper consideration of the data are likely to provide a better indication of diagenesis than isotope comparisons alone.

Conclusion

This study determined human-specific isotope spacing values and regression equations for the relationships between $\delta^{13}C_{collagen}$, $\delta^{13}C_{structural\ carbonate}$, $\delta^{18}O_{structural\ carbonate}$, and $\delta^{18}O_{phosphate}$ in individuals with a largely terrestrial diet. The average $\Delta^{13}C_{carb\text{-}coll}$ of 5.4‰ is fairly consistent between bone and dentin with a strong correlation between $\delta^{13}C_{collagen}$ and $\delta^{13}C_{structural\ carbonate}$. The lack of correlation between $\Delta^{13}C_{carb\text{-}coll}$ and $\delta^{15}N$ suggests that these individuals allocate a similar proportion of animal protein to their carbon input regardless of absolute meat intake. The average $\Delta^{18}O_{carb\text{-}phos}$ of 7.8‰ is lower than previous studies, but is consistent between bone, dentin, and enamel. Enamel shows the strongest correlation between $\delta^{18}O_{structural\ carbonate}$ and $\delta^{18}O_{phosphate}$ values compared to dentin and bone. This has potential implications concerning isotopic fractionation of blood-DIC during collagen formation, as well as potential implications for species-specific fractionations of oxygen during bone/tooth mineralization. Using the $\delta^{18}O_{structural\ carbonate}$ and $\delta^{18}O_{phosphate}$ correlation for determination of diagenetic state is tenuous and should be used with caution and attention to the particular body element (i.e. bone, dentin, enamel) being considered. This robust data set can now provide a basis of comparison for human isotope studies which typically involve small sample numbers and an attempt to limit destructive analyses of valuable and rare specimens.

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