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Herbivore tooth oxygen isotope compositions: Effects of diet and physiology

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Abstract—The applicability of rapid and precise laser probe analysis of tooth enamel for $\delta^{18}O$ has been verified, and the method has been applied to different modern herbivores in East Africa. Sampling and pretreatment procedures involve initial bleaching and grinding of enamel to <75 μm, and elimination of adsorbed water and organic compounds with BrF_5. Typical analytical reproducibilities for 0.5–2 mg samples are ±0.08‰ (±1σ). Chemical and spectroscopic characterization of pretreated but unanalyzed samples show no alteration compared to fresh enamel. Solid reaction products are nearly pure CaF_2 with little evidence for residual O_2. Because laser probe fluorination extracts oxygen from all sites in the apatite structure (phosphate, structural carbonate, and hydroxyl), only unaltered tooth enamel (>95% apatite) can be analyzed reliably. Different East African herbivores exhibit previously unsuspected compositional differences. Average enamel $\delta^{18}O$ values (V-SMOW) are approximately: 25‰ (goat), 27‰ (oryx), 28‰ (dikdik and zebra), 29‰ (topi), 30‰ (gerenuk), and 32‰ (gazelle). These compositions differ from generalized theoretical models, but are broadly consistent with expected isotope effects associated with differences in how much each animal (a) drinks, (b) eats C3 vs. C4 plants, and (c) pants vs. sweats. Consideration of diet, water turnover, and animal physiology will allow the most accurate interpretation of ancient teeth and targeting of environmentally-sensitive animals in paleoclimate studies.

1. INTRODUCTION

The oxygen isotope ratio of tooth enamel has long been considered a potentially useful record of climate (Longinelli, 1984; Luz et al., 1984, 1990; Luz and Kolodny, 1985; Ayliffe and Chivas, 1990; D'Angela and Longinelli, 1990) because tooth enamel is well preserved in the fossil record, and tooth compositions depend on variables such as rain and food source compositions (Longinelli, 1984; Luz et al., 1984, 1990; Luz and Kolodny, 1985; Ayliffe and Chivas, 1990; D'Angela and Longinelli, 1990), which in turn depend on local and global climate (e.g., Gat, 1980; Dongmann et al., 1974; Epstein et al., 1977; Burk and Stuiver, 1981; Sternberg, 1989; Yakir, 1992). However, advances in using tooth oxygen isotope compositions for paleoclimate studies have been retarded both by the difficulties in reliably analyzing biogenic phosphates and in understanding the factors that influence animal isotope compositions. Some major factors include: body temperature, air oxygen composition, diet, drinking water composition, and the relative proportions of oxygen influxes and effluxes (e.g., Longinelli, 1984; Luz and Kolodny, 1985; Ayliffe and Chivas, 1990; Bryant and Froelich, 1995). Although body temperature for mammals and air oxygen isotope composition are nearly constant (Dole et al., 1954), drinking water and food compositions are climate-dependent (Gat, 1980; Dongmann, 1974; Epstein et al., 1977; Burk and Stuiver, 1981; Sternberg, 1989; Yakir, 1992), and the proportions of oxygen influxes and effluxes are genus dependent (Longinelli, 1984; Luz et al., 1984; D'Angela and Longinelli, 1990). Unless isotope effects are characterized at the genus level, paleoclimate studies cannot be fully quantitative. To improve the quality of biogenic phosphate analyses, we have developed a sampling technique that allows the use of rapid, highly precise laser fluorination methods. We then applied this approach in a study of modern herbivores from a small area in the Lake Turkana region of East Africa to investigate isotope compositional differences among different species, to evaluate the consistency of published theoretical models in predicting the isotope trends, and to determine the potential usefulness of these animals for paleoclimate studies.

2. LASER FLUORINATION OF TOOTH ENAMEL

Chemically, biogenic phosphates are hydroxyapatite [$Ca_5(PO_4)_3(OH)$], with substantial complex vacancy-balanced substitutions of $(CO_3)^{2-}$ and $(HPO_4)^{2-}$ for $(PO_4)^{3-}$, and smaller amounts of $(CO_3)^{2-}$ and $(F)^{1-}$ substitutions for $(OH)^{-1}$ (e.g., Elliot et al., 1985; Legros et al., 1987; Rey et al., 1991). Although the overall amounts of $(CO_3)^{2-}$ and $(HPO_4)^{2-}$ depend systematically on the degree of enamel maturity (e.g., Hiller et al., 1975; Deutsch and Gedalia, 1980, Rey et al., 1990, 1991), fully mature enamel has approximately the same chemical composition, independent of species (e.g., Rey et al., 1989; Quade et al., 1992; our unpubl. data). Some exceptions include the enamel of a few marine fish such as sharks and porgies which have anomalously high F-contents (LeGeros and Suga, 1980). In contrast to the chemical homogeneity of enamel, the timing of formation and maturation depends strongly on position within a tooth and within a jaw (e.g., Hillson, 1986).

In most stable isotope studies of biogenic phosphates, carbon is preferentially extracted from the $(CO_3)^{2-}$ component

Table 1. Summary of Tooth Analyses Exclusive of East Africa:

Sample	$\delta^{18}O$ Enamel (‰)	$\pm 1\sigma$	n	Location	$\delta^{18}O$Water (‰)
Human					
Anonymous-A	15.97	0.11	8	Madison	-8
Anonymous-B	15.88	0.06	4	Madison	-8
Banfield, J.	18.64	0.05	5	S. E. Australia	-5
Barovich, K.	15.19	0.06	5	W. Pennsylvania	-8
Valley, D.	15.53	0.05	3	Madison, WI	-8
Valley, M.	17.52	0.09	7	Houston, TX	-4
Wilhite	16.00	0.14	4	Indianapolis, IA	-6
Non-Human					
Wolf	8.01	0.13	7	Arctic Circle, Canada	-18
Shark	21.83	0.14	3	North Carolina Coast (Atlantic)	0

through acid dissolution, and oxygen is preferentially extracted from the $(PO_4)^{3-}$ and $(HPO_4)^{2-}$ components (Tudge, 1960; Kolodny et al., 1983; O'Neil et al., 1994). Phosphate oxygen analysis involves initial acid dissolution of 10–100 mg of sample, reprecipitation as Bi- or Ag-phosphates, and reduction at high temperature with either fluorine compounds, Br_2, or graphite to produce either O_2 or CO_2, and has an analytical reproducibility of ± 0.2–0.5‰ (1σ; Luz et al., 1984; Tudge, 1960; Kolodny et al., 1983; Crowson et al., 1990; O'Neil et al., 1994, Stuart-Williams and Schwarcz, 1995).

Laser fluorination is an extremely rapid and precise method of analyzing oxygen isotope ratios in silicate and oxide minerals (e.g., Sharp, 1992; Mattey and Macpherson, 1993; Valley et al., 1995). Precisions can be routinely $\leq \pm 0.1$‰ (1σ), and typical sample sizes are 0.5–2.0 mg. We have explored the possibility of using laser fluorination for analyzing tooth enamel, and have found that if enamel is simply bleached, ground to a fine powder (<75 μm), and reacted with BrF_5 by heating with a high power ($\geq$5W) diffuse CO_2-laser beam (0.5–1 mm diameter), then precisions and sample sizes similar to those obtained for laser-fluorinated silicates and oxides can be achieved (Tables 1 and 2). Otherwise our analysis procedures follow those employed in previous laser studies (Valley et al., 1995). At the University of Wisconsin, these procedures include loading samples and standards individually into a 72-pit Ni block, which is then placed in a reaction chamber; evacuation followed by pretreatment overnight with $\sim$1000 μmoles of BrF_5 eliminates organic residues and adsorbed H_2O; samples are then individually heated with a laser in the presence of BrF_5 to produce free oxygen, the oxygen is converted to CO_2 with a heated graphite rod, and the CO_2 is analyzed in a mass spectrometer. Use of the laser approach significantly decreases the compositional uncertainty, sample preparation difficulty, amount of sample, and collection time required to analyze tooth enamel. Each enamel sample can be prepared in less than 15 min, and $\sim$4 samples/h can be reacted and analyzed in the UW laser extraction/mass spectrometer system.

We made special efforts to verify that no pre-lasing reaction or leaching of oxygen occurred, and that our sample processing did not bias compositions. Until heating, large enamel pieces retain natural surface polish, and powders remain pure white, suggesting no reaction. Backscattered electron imaging and quantitative electron probe analysis of enamel that was in the sample chamber during pretreatment, but not heated with the laser indicates no fluoride reaction products or changes of fluorine content compared with fresh enamel, and infra-red spectra on pretreated powders are indistinguishable from fresh enamel. These observations suggest that no isotope fractionation occurred due to preferential leaching of hydroxyl oxygen or structural carbonate during pretreatment. Yields and compositions are also unaffected by pretreatment times ranging from 1 to 42 h. Electron microprobe analyses of the post-reaction fluoride glasses reveal less than 2 mol% oxygen, which may be hydroscopic water introduced during polishing of glass beads rather than residual oxygen. Although enamel pieces are more difficult to analyze reliably by laser, analyses of apatite from a rock show no dependence of isotope composition on grinding, and the physical behavior of more finely ground igneous and metamorphic apatites during lasing is identical to that observed for enamel. Analysis of unbleached enamel has a poorer precision (± 0.20‰, 1σ) compared with enamel in which organic material was initially oxidized in NaOCl or H_2O_2, but average measured compositions differ by less than 0.05‰.

The interpretations presented in this study depend principally on compositional differences, and analytical reproducibility is the most important parameter. The excellent reproducibility obtained for laser analyses of bleached enamel (± 0.08‰, 1σ) allows robust interpretations regarding compositional variations. Nonetheless, we made additional effort to evaluate analytical accuracy. Multiple laser analyses of an igneous apatite that has been previously analyzed by fluorination in Ni reaction vessels (Farquhar et al., 1993) suggests that a correction to the raw laser data of $+1.7 \pm 0.1$‰ is required, and we have routinely applied a 1.7‰ upward correction to all apatite data. Some enamel samples were additionally analyzed for $\delta^{18}O$ of PO_4^{3-} alone using the approach of O'Neil et al. (1994), and corrected laser analyses are the same as the isotope composition of the PO_4^{3-} groups to $\sim \pm 0.2$‰ (M. Schoeninger and J. R. O'Neil, unpubl. data).

It is important to note that unlike other phosphate analytical techniques, laser fluorination does not extract one single oxygen component. Instead, three oxygen components (phosphate, structural carbonate, and hydroxyl) contribute significantly to the overall isotope analysis. The similarity

Table 2. Summary of Analyses from the Lake Turkana Area, Kenya:

Sample	$\delta^{18}O$(‰, V-SMOW)	$\delta^{13}C$(‰)	Ave	Replication Difference
Zebra (bleached)				
BZ2142 #1	28.07, 28.22	-8.8,-9.3	28.14	0.15
BZ2142 #2	27.62, 27.72		27.67	0.10
BZ2144	27.42, 27.44	-9.3	27.43	0.02
BZ2306 #1	28.13, 28.13	-8.7	28.13	0.00
BZ2306 #2	28.04, 28.41		28.22	0.37
Average Zebra		-9.1	27.84	
Dikdik (bleached)				
DD2251 (M2)	27.42, 27.64	--	27.53	0.22
DD2284 #1 (M3)	27.42, 27.52	--	27.47	0.10
DD2284 #2 (M3)	27.73, 27.56		27.64	0.17
DD2278	28.04, 28.03	-18.9	28.04	0.01
DD2256 (M3+M1)	28.17, 27.91, 27.78	-14.0	27.95	±0.20
DD2347#1 (M1+M2)	28.58, 28.62	-16.2	28.60	0.04
DD2347#2 (M1+M2)	29.60, 29.41		29.50	0.19
DD2283 (M2+M3)	30.42, 30.57	-19.8	30.50	0.15
Average Dikdik		-18.0	28.44	
Gazelle (bleached)				
GG2180	32.36, 32.60, 32.45	-14.8	32.47	±0.12
GG2109	32.16, 32.16, 32.13	-14.0	32.15	±0.02
GG2090 #1 (M3)	30.71, 30.66	-15.6	30.68	0.05
GG2090 #2 (M3)	30.52, 30.40	____	30.46	0.12
Average Gazelle		-15.0	31.73	
Goat (bleached)				
Goat2188	26.46, 26.54	-10.6	26.50	0.08
Goat2181	23.84, 23.85	-12.3	23.84	0.01
Average Goat		-11.5	25.17	
Gerenuk (unbleached)				
GN2245 (M3)	29.82, 30.06	-20.3	29.94	0.24
Oryx (unbleached)				
OR2120 (M2)	26.08	-9.0	26.08	
OR2119 (M1)	27.53, 27.29	-9.5	27.41	0.24
OR2122 (M1)	27.34	-9.7	27.34	
Average Oryx		-8.8	26.94	
Topi (unbleached)				
TP2484 (M3)	27.09, 27.33	-7.3	27.21	0.24
TP2484 (P4)	29.88, 30.16	-7.3	30.02	0.28
TP2172	29.61, 29.66	-8.4	29.64	0.05
Average Topi		-7.8	28.96	

$\delta^{18}O$ from laser analyses of tooth enamel, $\delta^{13}C$ from collagen analysis of bone. Values of $\delta^{18}O$ normalized to UW standard UWG-2=5.8‰ (Valley et al., 1995) and SP3-3 apatite (Farquhar et al., 1993). Average $\delta^{13}C$ based on additional collagen analyses of specimens not analyzed for $\delta^{18}O$. Average duplication difference of 0.11 for $\delta^{18}O$ of bleached samples (n=32), implies a $\pm 1\sigma$ reproducibility of ~ ±0.08‰. "M1"-"M3" refer to molars; "P4" refer to premolar; "bleached" indicates initial oxidation of organic material in NaOCl; "unbleached" indicates untreated enamel. All dentition was permanent.

between our whole enamel analyses and independently extracted phosphate $\delta^{18}O$ suggests that compositional compensation of the carbonate and hydroxyl components fortuitously yields a $\delta^{18}O$ similar to that of the phosphate component alone. Because the fractionations between carbonate and hydroxyl components relative to phosphate are unknown, this inference cannot be assessed independently. These issues are not likely to be significant for modern mature enamel, as long as the carbonate and hydroxyl components are precipitated in equilibrium with body water, because the proportions of the components and their relative internal fractionations are then constant. That is, the whole enamel $\delta^{18}O$ should simply be offset by a constant amount from the $\delta^{18}O$ of any one component. Fossil material may be more problematic, because it is possible to diagenetically change the composition of one component independently of the others (e.g., Barrick and Showers, 1994). Thus, future laser analyses of fossil enamel will require verification that all three components of the fossil material are unaltered.

Laser analytical yields for coarse geologic apatite are ~85%, and for bleached powdered enamel are ~80%. The low yield as well as the 1.7‰ compositional correction could

be due to the production of a small quantity of high $\delta^{18}O$ P-O(-F) compounds in the sample chamber during lasing. Formation of fractionated C-O-F compounds from the structural carbonate component seems less likely, as the presence of graphite inclusions in minerals does not affect oxygen yields or compositions in our laser extraction system (M. J. Kohn and J. W. Valley, unpubl. data). Formation of phosphorus compounds might be enhanced by the thermal gradient between the reaction site and the walls of the reaction chamber. Externally heated Ni reaction vessels have a negligible thermal gradient and apatite analyses have better yields (Farquhar et al., 1993). One likely compound, POF_3, has a melting point of −68°C and would be frozen in liquid nitrogen traps and so lost prior to oxygen conversion and analysis. A similar effect involving low $\delta^{18}O$ S-O(-F) compounds has been proposed for laser probe analyses of mixed silicate/sulfide samples (Kohn and Valley, 1994). Because the P:O ratio in biogenic apatites is essentially constant, we assume that the 1.7‰ correction is constant. The difference between the yields for enamel vs. geologic apatite is probably due to differences in adsorbed water. Mature enamel has 2–5% water by weight, and pre-drying enamel in an oven at 85°C for 10 min improves yields by 2–3%.

One reason we focused on tooth enamel rather than other phosphates is because enamel is more coarsely crystalline (crystallite length is >1600 nm) and is nearly free of organic impurities (>95% crystalline; Hillson, 1986). In contrast, other biogenic phosphates have a lower apatite content (e.g., bone, dentine and cementum are only 70–75% crystalline) and the crystallites are smaller (20–100 nm; Hillson, 1986). Our attempts to analyze dentine and cementum resulted in higher analytical blanks, lower yields, and poorer reproducibility, and consequently we restricted our application to enamel. We have not tested the technique on either bone or the phosphate components of marine shells.

Enamel analyses by laser probe have yields, corrections, reproducibilities and sample sizes that compare favorably with alternative methods of analyzing phosphates. Enamel sample sizes by most other methods are typically 10–20 mg, yields are between 25% (using graphite: O'Neil et al., 1994) and 100% (using BrF_5: Crowson et al., 1990), reproducibility is ±0.2–0.3‰ (1σ), and use of graphite may involve a correction of a few tenths of a permil (O'Neil et al., 1994). An alternative method involves reacting Ag_3PO_4 with Br_2 (Stuart-Williams and Schwarcz, 1995), which improves the reproducibility (~ ±0.15‰, 1σ) and speeds reaction time to ~25 min, but has yields of less than 20% and requires a correction of 10.9‰. A third approach is simply to melt the enamel with a laser to produce a small amount of CO_2 gas (Sharp and Cerling, 1995). In this method, the carbon is likely derived from structural carbonate, but the oxygen reservoir tapped by this method is unclear. Thermal decomposition studies of human tooth enamel (e.g., Holcomb and Young, 1980) suggest that the released oxygen is most likely a mixture of the carbonate and hydroxyl reservoirs, and might not include significant phosphate oxygen. Thus, in comparison with other approaches laser fluorination is a viable analytical approach for tooth enamel.

We have evaluated the applicability of laser fluorination by analyzing tooth enamel from samples obtained in different areas of the world. Biogenic phosphate oxygen isotope composition correlates strongly with local rainwater composition (Longinelli, 1984; Luz et al., 1984, 1990; D'Angela and Longinelli, 1990), which varies geographically (Gat, 1980). Figure 1 demonstrates that our data follow closely the trends delineated in previous studies, supporting the use of our laser fluorination procedure for analyzing teeth.

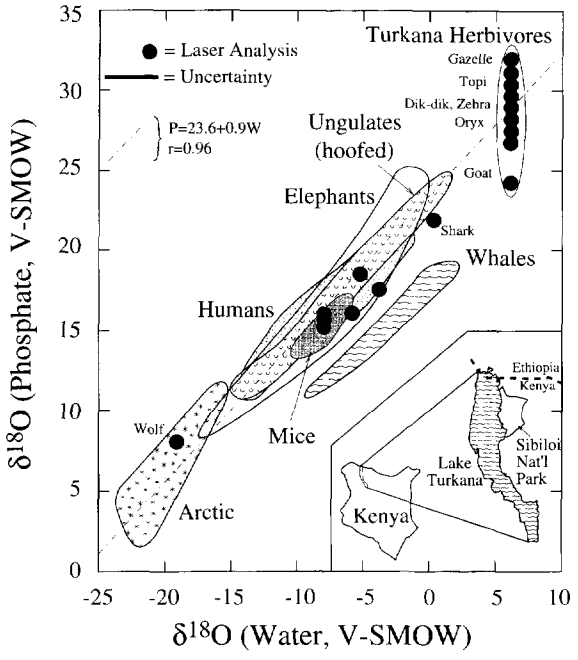

FIG. 1. Values of tooth $\delta^{18}O$ by laser fluorination vs. average $\delta^{18}O$ of local water for six humans, one shark, and one wolf. Where not directly measured, water compositions are based on the worldwide summary of Gat (1980). Compositional fields for humans, arctic animals (musk ox and dog), ungulates (pig, deer, cattle, and sheep), mice, elephants, and whales (including dolphins and porpoises) from published conventional phosphate analyses (Longinelli, 1984; Luz et al., 1984; Ayliffe and Chivas, 1990; D'Angela and Longinelli, 1990; Yoshida and Miyazaki, 1991; Huertas et al., 1995) shown for comparison. Height and width of black box shows ±2σ analytical error in laser isotope analyses and estimated uncertainty in surface water compositions, respectively. Regressed dash-dot line does not include data for whales or laser analyses. The good correlation between our tooth measurements and water compositions, and the consistency with previous studies supports the use of the laser fluorination technique for determining tooth $\delta^{18}O$ compositions. Inset: Location of Sibiloi National Park in Kenya.

3. INTER-SPECIES ISOTOPE VARIATIONS

Having demonstrated the validity of tooth analyses by laser fluorination, we chose to investigate the variability of tooth compositions for animals from the Sibiloi National Park on the east side of Lake Turkana in East Africa (Fig. 1). This park is well suited for study because the Lake Turkana area has been the subject of much climate and paleoclimate research (e.g., Cerling et al., 1988; Johnson et al., 1991), and it is well characterized with respect to rainfall, weather patterns, food and drinking water sources, and drinking water compositions (Cerling et al., 1988). Average yearly rainfall is extremely low (≤ 200 mm/y), and drinking

water is derived almost exclusively from long-term surface water sources with a nearly constant composition (Cerling et al., 1988; Johnson et al., 1991). Because the park is surrounded by human pastoral groups, animals likely obtained their food and water solely from within the park boundaries and were nonmigratory. This limits the number of plant species available as food, and restricts drinking water almost exclusively to Lake Turkana. Extensive mixing in Lake Turkana makes its water composition almost constant. Three reported $\delta^{18}O$ measurements (Cerling et al., 1988) range from 5.6 to 6.1‰ (V-SMOW), and the minimum and maximum oxygen isotope compositions throughout the year are thought to be 5 and 7‰ respectively (Johnson et al., 1991). A 2‰ range in drinking water $\delta^{18}O$ is also consistent with the range of $\delta^{18}O$ values for rainfall observed during the year at Khartoum, Sudan (IAEA, 1992), the nearest climatically similar IAEA station.

Samples of Burchell's zebra (*Equus burchelli*), goat (*Capra hircus*), dikdik (*Rhynchotragus guentheri*), Grant's gazelle (*Gazella granti*), topi (*Damaliscus corrigum*), gerenuk (*Litocranius walleri*), and oryx (*Oryx beisa*) were obtained from surface-exposed modern skeletons. Zebra is from the order *Perissodactyla*, family *Equidae*, whereas the other animals are from the order *Artiodactyla*, family *Bovidae*. Goat is from the subfamily *Caprinae*, whereas the others belong to different tribes within the subfamily *Antelopinae*. These animals were chosen for analysis based on known differences in dietary and drinking habits, which could be used to evaluate theoretical models, as well the potential of future paleoclimate studies using zebra and gazelle teeth that commonly occur in local sedimentary rocks. Skeleton weathering ranged up to stage 3 (0–7 years exposure; Behrensmeyer, 1978), and should not have affected tooth isotope compositions (Koch et al., 1990). Oxygen isotope compositions were measured at the Department of Geology and Geophysics, University of Wisconsin, by using the laser fluorination technique, and carbon isotope compositions were determined on bone collagen from the same animals (method of Schoeninger and DeNiro, 1984) to facilitate interpretations of diet (Table 2).

Measured $\delta^{18}O$ compositions are reported in Table 2, and plotted in Fig. 2 against $\delta^{13}C$ to allow comparison of $\delta^{18}O$ with diet. High $\delta^{13}C$ grazers exhibit a positive correlation between $\delta^{18}O$ and $\delta^{13}C$, with $\delta^{18}O$ and $\delta^{13}C$ increasing in the order goat, oryx, zebra, and topi. Low $\delta^{13}C$ browsers and mixed feeders show a similar trend, with $\delta^{18}O$ increasing in the order gerenuk + dikdik to gazelle, but the trend is offset to higher $\delta^{18}O$ from that of the grazers.

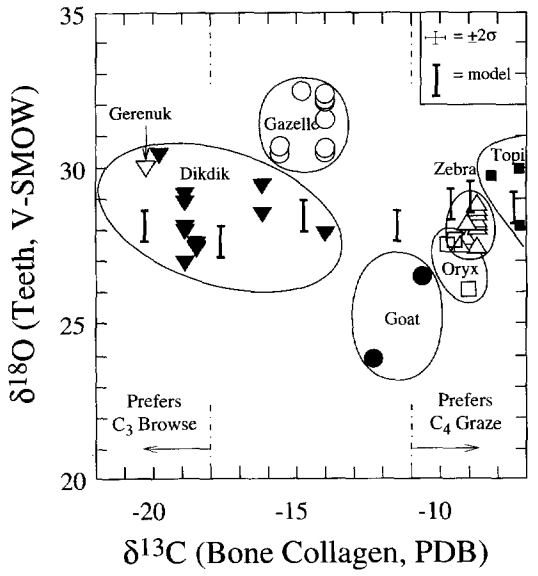

FIG. 2. $\delta^{18}O$ of teeth vs. $\delta^{13}C$ of bone collagen for animals from the Turkana area analyzed in this study. Analyses by Kohn et al. (1996) of a single gazelle, dikdik, and zebra from the same area are also included. Large differences in oxygen isotope composition among species most likely reflect physiological and dietary differences, including drinking vs. nondrinking, consumption of grasses (C4) vs. shrubs (C3), and panting vs. sweating. Thick error bars indicate theoretical predictions of average tooth $\delta^{18}O$ using the model of Bryant and Froelich (1995), which is based on generalized physiological equations; the ±0.5‰ $\delta^{18}O$ range reflects their stated uncertainties in intrinisic parameters. Each error bar is centered on the average $\delta^{13}C$ of each species. The differences between measured data and theoretical models suggests that physiological and dietary deviations from averages are quantitatively important.

4. EXPLAINING THE ISOTOPE COMPOSITIONS

There are several possible explanations for the isotope trends and offset. Firstly, if the animals inhabit different areas of the park, then compositional differences may simply reflect differences in local water sources. This is particularly important for evaluating the observed isotope offset between the grazers and the browsers/mixed feeders. Relatively high $\delta^{18}O$ gazelle, gerenuk, and dikdik are drought-tolerant and all live inland from the lake, whereas relatively low $\delta^{18}O$ zebra, goat, and topi are water dependent and stay close to Lake Turkana. Although this might suggest a compositional difference between inland vs. lake water sources, oryx are water independent and inhabit the same areas as gazelle, gerenuk, and dikdik. If inland water had an elevated $\delta^{18}O$, we would expect oryx to have a $\delta^{18}O$ significantly higher than zebra, goat, and topi. Because oryx compositions are similar to the near-lake species, we conclude that local water source compositions are not significantly different.

A second possible reason for the isotope differences is that different teeth or parts of teeth are compositionally different, and that vagaries of sampling fortuitously biased apparent average compositions. Isotope heterogeneity among teeth is supported by some of the data in Table 2, which show intertooth isotope differences of up to 3‰. Furthermore, zoning studies of individual teeth from a Turkana zebra and from two Nairobi gazelle (Kohn et al., 1996) indicate that these variations likely reflect seasonal differences in water and food source compositions. However, these more detailed studies in addition to theoretical modeling show that the expected seasonal variation in Kenya is at most 2–3‰, and so it is unlikely that the 6–8‰ differences observed between some species could be caused solely by sampling differences.

The only remaining likely explanation for the isotope differences is that they are the result of differences in animal physiology and diet. Enamel $\delta^{18}O$ parallels body water $\delta^{18}O$ (Longinelli, 1984; Luz and Kolodny, 1985; D'Angela and

Longinelli, 1990), and body water composition is quantitatively related to three major oxygen sources: air oxygen, lake (drinking) water, and food, as well as to three major oxygen losses: CO_2, liquid water (sweat, urine, fecal water), and water vapor (Luz and Kolodny, 1985). Although the oxygen isotope composition of air is the same worldwide (Dole et al., 1954) and local water compositions and teeth from a single animal show limited variability (Cerling et al., 1988; Johnson et al., 1991; Kohn et al., 1996), diet and physiology are quite different among the animals analyzed.

Bryant and Froelich (1995) proposed that herbivore oxygen isotope composition depends principally on body size. Based on fractionation, energy, and water turnover studies of humans and of captive animals, they derived equations to relate phosphate composition of wild herbivores to source water composition and body size. These equations were derived from average physiological parameters, and one question we wished to examine was how closely their generalized model predicted compositions for specific animals. Results of their predicted compositions are shown in Fig. 2 (error bars) using as input parameters the $\delta^{18}O$ value of Lake Turkana (6‰) and the known body sizes of the animals (5 kg for dikdik, 30 kg for gerenuk and goat, 50 kg for gazelle, 120 kg for topi, 200 kg for oryx, and 250 kg for zebra; e.g., Sachs, 1967). The predicted compositions using the Bryant and Froelich (1995) model are nearly the same for all the animals studied because the body-size dependence of $\delta^{18}O$ is rather weak in this size range. These predictions substantially underestimate the measured $\delta^{18}O$ values for the browsers and mixed feeders (dikdik, gerenuk, and gazelle), and although the match for the other animals is significantly better, the correlation between $\delta^{18}O$ and $\delta^{13}C$ remains unexplained. Consequently, we assume that specific physiological and dietary differences not originally considered by Bryant and Froelich (1995) are likely responsible for the observed variations.

One of the most important parameters in understanding animal $\delta^{18}O$ is the ratio of daily water turnover to energy expenditure (the water economy index; Nagy and Peterson, 1988). Although larger animals might on average be somewhat less capable of conserving water compared to smaller animals (as suggested by Bryant and Froelich, 1995), the scatter among different animals is extreme (e.g., Nagy and Peterson, 1988). At the most fundamental level, some animals do not drink water (e.g., 200 kg oryx), other animals drink every few days (e.g., 30 kg goat), and others must drink every day (e.g., 70 kg human). These behavioral differences reflect differences in water conservation capabilities that are not strictly mass-dependent. Additionally, physiological factors such as diet, the amounts of fecal and urinary water loss, and heat loss mechanisms (sweating vs. panting) vary significantly among genera. Because liquid water and water vapor outputs have different compositions, and because drinking water has a different composition from other oxygen sources, a detailed assessment of physiology is probably important for understanding animal $\delta^{18}O$. This is especially true in semi-arid settings like the Lake Turkana region, where a mix of animals with varying water conservation capabilities is expected.

Because wild herbivores also obtain 35–50% of their oxygen from plants, and because different plants respond differently to climate, specific plant physiology and fractionation processes should also affect the $\delta^{18}O$ of animals through diet and food selection. Plant cellulose is uniformly enriched over plant leaf water by ~27‰, but the ^{18}O enrichment of plant water over surface water is quite variable (Dongmann et al., 1974; Epstein et al., 1977; Burk and Stuiver, 1981; Sternberg, 1989; Yakir, 1992). The fractionation between stem and surface water is ~0‰, whereas preferential evaporative loss of $H_2{}^{16}O$ from leaves typically elevates the $\delta^{18}O$ value of their water by 10‰ or more (Dongmann et al., 1974; Sternberg, 1989; Yakir, 1992). Furthermore, preferential evapotranspiration by C4 plants (warm climate grasses) late in the day (when it is dryer) further elevates $\delta^{18}O$ of C4 over C3 plants (cool climate grasses, shrubs, and trees; Sternberg, 1989; Yakir, 1992). In arid settings, the C4-C3 composition difference is as large as 10‰ (Sternberg et al., 1984), but in cooler, moister settings it is ≤1‰ (e.g., Epstein et al., 1977). Although due to different causes, this C3-C4 $\delta^{18}O$ trend parallels the well-known enrichment of ^{13}C in C4 plants relative to C3 (e.g., O'Leary, 1988; Farquhar et al., 1989).

Based on plant and animal physiological characteristics, we expect animals with low water turnover to have a higher $\delta^{18}O$, because they derive more body water from isotopically enriched food sources and less from drinking isotopically depleted lake waters. Furthermore, we expect consumers of C4 leaves to have a higher $\delta^{18}O$ (and higher $\delta^{13}C$) than consumers of C3 stems, because of the $\delta^{18}O$ offset between C4 and C3 plants, and because of the compositional difference between stem and leaf water. Finally, because sweat, urine, and fecal water have a higher $\delta^{18}O$ than water vapor, animals that pant to lose heat, have high urinary salt concentrations, and have low fecal water contents, should have a higher $\delta^{18}O$ than animals that lose more of their water as liquid.

Although topi and gerenuk are not as well studied, the physiologies and dietary patterns of goat, zebra, dikdik, gazelle, and oryx are well known. As demonstrated in several dietary and physiological studies in arid or semi-arid settings, there are significant differences in what each of these animals eats, how it dissipates heat, and in its water turnover rate (e.g., Taylor, 1968; Shkolnik et al., 1972; Maloiy, 1973; Dawson et al., 1975; Nge'the and Box, 1976; Hoppe, 1977; Hoppe et al., 1977; King et al., 1978; Spinage et al., 1980; Maloiy et al., 1988; Hossaini-Hilali et al., 1994). Qualitatively, the observed isotope trends are consistent with the physiological and dietary differences among these animals. For example, the general trends of increasing $\delta^{18}O$ with increasing $\delta^{13}C$ observed for the grazers (goat, oryx, zebra, and topi) and for the browsers and mixed feeders (gerenuk, dikdik, and gazelle) are consistent with the elevation of C4 $\delta^{18}O$ over C3, as was also inferred by Koch et al. (1990). The offset between grazers and browsers/mixed feeders may reflect several factors. Gazelle, gerenuk, and dikdik are drought tolerant, employ panting as a principal means of thermal regulation, and select leaves preferentially over stems for food. In contrast, the other animals drink every day (except oryx), sweat, and tend to consume more stems. That is, the materials that gazelle, gerenuk, and dikdik ingest have higher than average $\delta^{18}O$ (plants rather than water, and

leaves rather than stems), and the water they output has lower than average $\delta^{18}O$ (vapor rather than liquid) compared to goat, oryx, zebra, and topi. This will tend to elevate gazelle, gerenuk, and dikdik $\delta^{18}O$ over the other animals, and may cause of the observed isotope offset.

5. DISCUSSION AND CONCLUSIONS

The compositional differences exhibited by different herbivores in East Africa indicate that diet and physiology have a strong control on animal isotope compositions. Conversely, an accurate knowledge of animal physiology and diet allows the sensitivity of each genus to environmental variables like humidity and temperature to be assessed. For example, a drought-tolerant animal that obtains all its water from food (e.g., antelope) will have a composition that is sensitive to humidity (Ayliffe and Chivas, 1990; Luz et al., 1990) because of the strong dependence of plant composition on humidity. In contrast, an animal that drinks daily and that has high water turnover (e.g., goat or zebra) will have a composition that is more sensitive to surface water compositions. Once the genus-specific dependencies of oxygen isotope compositions on present-day climate are determined through detailed studies in different settings, then analysis of the isotope composition of fossils within the geologic record should allow quantitative determination of past climate changes. For example, because zebra are water-dependent and gazelle are not, the compositions of these two animals might discriminate water composition changes from humidity effects.

The area in East Africa chosen for study, Lake Turkana, is especially important for future paleoclimate research for two reasons. Firstly, East Africa is climatically sensitive to global changes of temperatures, as illustrated by the Pliocene global climate models of Chandler et al. (1994) and Sloan et al. (1996). These models suggest that global warming causes greater seasonality and surprisingly cooler temperatures in East Africa. Secondly, significant hominid radiation occurred in the Lake Turkana area during the last few million years. The present-day sub-Saharan climate barely supports human life, and the Plio-Pleistocene climate must have been different for such a radiation to have occurred (e.g., Vrba et al., 1995). A quantitative determination of past climate at Lake Turkana from fossil tooth enamel of physiologically different species would therefore allow a detailed assessment of how climate change may have affected the rise of the human species.

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