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Effects of diagenesis on strontium, carbon, nitrogen and oxygen concentration and isotopic composition of bone

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Abstract—Paleodietary analysis based on variations in the trace element and stable isotopic composition of inorganic and organic phases in fossil bone depends on the assumption that measured values reflect *in vivo* values. To test for postmortem alteration, we measured $^{87}Sr/^{86}Sr$, $^{13}C/^{12}C$, $^{18}O/^{16}O$ and $^{15}N/^{14}N$ ratios and Sr concentrations in modern and prehistoric (610 to 5470 yr old) bones of animals with marine or terrestrial diets from Greenland.

Bones from modern terrestrial feeders have substantially lower Sr concentrations and more radiogenic $^{87}Sr/^{86}Sr$ ratios than those from modern marine feeders. This contrast was not preserved in the prehistoric samples, which showed almost complete overlap for both Sr concentration and isotopic composition in bones from the two types of animals. Leaching experiments, X-ray diffraction analysis and infrared spectroscopy indicate that alteration of the Sr concentration and isotopic composition in prehistoric bone probably results from nearly complete exchange with groundwater. Oxygen isotope ratios in fossil apatite carbonate also failed to preserve the original discrimination between modern terrestrial and marine feeders. The C isotope ratio of apatite carbonate did not discriminate between animals with marine or terrestrial diets in the modern samples. Even so, the ranges of apatite $\delta^{13}C$ values in prehistoric bone are more scattered than in modern samples for both groups, suggesting alteration had occurred. $\delta^{13}C$ and $\delta^{15}N$ values of collagen in modern bone are distinctly different for the two feeding types, and this distinction is preserved in most of the prehistoric samples.

Our results suggest that postmortem alteration of dietary tracers in the inorganic phases of bone may be a problem at all archaeological sites and must be evaluated in each case. While collagen analyzed in this study was resistant to alteration, evaluation of the possibility of diagenetic alteration of its isotopic composition in bones from other contexts is also warranted.

INTRODUCTION

RECONSTRUCTION OF prehistoric diet is a primary goal of many researchers attempting to unravel the history of human biological evolution and cultural development. Methods of studying paleodiets include analyses of skeletal morphology, tooth and stone-tool microwear, cultural artifacts and the abundances of excavated food items (WALKER, 1981). The relative utilization of dietary components has also been estimated from the trace element and stable isotopic composition of fossil bone, an approach based on studies that have linked feeding habits with variations in bone chemistry and isotopic composition in modern animals (for recent reviews, see VAN DER MERWE, 1982; SILLEN and KAVANAGH, 1982). In order for this approach to be valid for prehistoric bone, postmortem alteration of the bone chemical and isotopic compositions that are influenced by diet must not have occurred.

In this paper, we present the results of isotopic and trace element analyses of modern and prehistoric bones from species with known diets. The objective of the study was to test for diagenetic alteration of the most commonly used and potentially useful chemical and isotopic tracers of diet. We analyzed the $^{87}Sr/^{86}Sr$ ratios and strontium concentrations in total bone, $^{13}C/^{12}C$ and $^{18}O/^{16}O$ ratios in bone apatite carbonate, and $^{13}C/^{12}C$ and $^{15}N/^{14}N$ ratios in bone collagen. Each of these tracers in fresh bone reflects certain aspects of an animal's diet and therefore is potentially useful for dietary reconstruction.

The strontium concentration in fresh bone is determined by dietary intake and metabolic processes. The predominant use of strontium analysis of prehistoric bone has been the determination of the relative abundance of plants *versus* meat in the diet (SILLEN and KAVANAGH, 1982). TUREKIAN (1961) suggested that strontium concentration in bone would also discriminate between marine and terrestrial diets, although the only data of which we were aware (WESSEN *et al.*, 1978) did not support his proposal. Bone $^{87}Sr/^{86}Sr$ ratios should also reflect the relative consumption of marine and terrestrial foods if the isotopic composition of strontium in the environment where the terrestrial foods originated differed from that of seawater. Results reported here confirm that strontium concentration and isotopic composition in fresh bone can be used to determine the relative utilization of marine and terrestrial food sources in the appropriate geological settings.

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DENIRO and EPSTEIN (1978, 1981) demonstrated that the carbon and nitrogen isotope ratios in fresh bone collagen and the carbon isotope ratio of the carbonate occurring in the hydroxyapatite of fresh bone are determined by the isotopic composition of an animal's diet. For individuals subsisting primarily on terrestrial plants, the carbon isotope ratios reflect consumption of C_3 plants *versus* C_4 and CAM plants (VAN DER MERWE, 1982). Further, it has been suggested that the nitrogen isotopic composition of collagen should reflect legume *versus* non-legume consumption (DENIRO and EPSTEIN, 1981). Both carbon and nitrogen isotope ratios of collagen distinguish between the use of marine and terrestrial food sources (TAUBER, 1981; CHISHOLM *et al.*, 1982, 1983; SCHOENINGER *et al.*, 1983; HOBSON and COLLIER, 1984; SCHOENINGER and DENIRO, 1984).

The relationship between diet and the oxygen isotopic composition of bone apatite carbonate is not clear. The carbonate may be precipitated in isotopic equilibrium with water in the blood (LAND *et al.*, 1980). The isotopic composition of water in blood is determined primarily by the isotopic composition of drinking water, but a number of factors, including the oxygen isotopic composition of the diet, are also influential (LUZ *et al.*, 1984).

Only a few studies on postmortem alteration of the chemical and isotopic dietary tracers in bone have been published. Most of these concerned alteration of strontium concentration in interred bone, a topic reviewed recently by SILLEN and KAVANAGH (1982) and LAMBERT *et al.* (1984). Comparison of strontium levels in prehistoric bones that had different *in vivo* concentrations (*i.e.* bones of contemporaneous herbivores and carnivores) and analysis of the distribution of strontium within prehistoric bones and in the surrounding soil have been used to identify samples and sites in which diagenetic alteration has or has not occurred. No work has been published on the effects of diagenesis on bone strontium isotope ratios. Postmortem alteration of the carbon and nitrogen isotopic composition of bone collagen, accompanied by changes in the elemental composition and chemical nature of that organic fraction, has been demonstrated recently (DENIRO, 1985). LAND *et al.* (1980) and SCHOENINGER and DENIRO (1982) presented evidence that the carbonate in bone apatite can exchange with groundwater carbonate after burial. This interpretation has been disputed by SULLIVAN and KRUEGER (1983), who maintain that the carbon isotopic compositions of prehistoric bone apatite carbonate reflect the *in vivo* values (SULLIVAN and KRUEGER, 1981).

In order to test for postmortem alteration, we designed a study to determine if the chemical and isotopic dietary signatures in the organic and inorganic phases of bones of animals with known diets are preserved after burial. We analyzed bones from modern animals with purely marine or purely terrestrial diets from regions along the coast of Greenland. We chose marine and terrestrial feeders because the studies discussed above indicated that these feeding preferences should produce large differences in bone mineral strontium concentrations, bone collagen nitrogen and carbon isotope ratios, and bone apatite carbonate carbon isotope ratios. Because of the antiquity of the crust and the attendant high $^{87}Sr/^{86}Sr$ ratios (MOORBATH *et al.*, 1972), we expected that the $^{87}Sr/^{86}Sr$ ratios of bones of terrestrial feeders from Greenland should be easily distinguishable from those of marine feeders. Additionally, because of the very negative $\delta^{18}O$ values that characterize precipitation in Greenland (DANSGAARD *et al.*, 1969, 1975), we expected that terrestrial animals would have low $^{18}O/^{16}O$ ratios in their bone apatite carbonate relative to marine animals. The prehistoric bones we analyzed came from animals whose modern counterparts are either purely marine feeders or purely terrestrial feeders, and, in many cases, were members of the same genera or species as the modern animals we studied. On the assumption that the feeding habits of these species have not changed in the past 5500 years (the age of our oldest specimen), we interpreted significant departures of chemical and isotopic compositions of fossil bone from values we observed in modern bone as evidence for diagenetic alteration.

MATERIALS AND METHODS

Various types of bones from modern and/or prehistoric *Rangifer tarandus* (reindeer), *Ovibus moschatus* (musk ox), *Ovis/Capra* (sheep or goat; the excavated bones could not be assigned more specifically), *Phoca vitulina* (harbor seal), *Phagophilus groenlandicus* (harp seal), and *Phoca* sp. (seal) were used in this study. Modern bones are museum specimens from animals captured live along the southern coast of West Greenland. Most of the prehistoric specimens are from a midden excavated at the Norse farm Niaqussat at Godthåbs Fiord in the same area that the modern specimens were collected. One of the other prehistoric samples came from a midden at Solbakken, on the coast of Hall Land in western North Greenland. The rest of the prehistoric samples are surface finds of reindeer antlers from different locations in Peary Land, North Greenland, and are not associated with prehistoric settlements. The ground at Godthåbs Fiord thaws during the summer, while in North Greenland, all but a thin surface layer remains frozen. (All information in this paragraph from HENRIK TAUBER, pers. commun.)

Methods used to determine the concentration of collagen and apatite in bone and the elemental and/or isotopic composition of these phases have been described elsewhere (SCHOENINGER and DENIRO, 1982, 1984). Carbonate and collagen concentrations are given as dry weight percentages of the mineral fraction and total bone respectively. Collagen elemental compositions are given as atomic carbon-to-nitrogen ratios. Isotope ratios of carbon, nitrogen, and oxygen are reported in the δ notation, where

$$\delta^*X = \left[\frac{(^*X/X)_{\text{sample}}}{(^*X/X)_{\text{standard}}} - 1\right] \times 1000‰$$

and $^*X/X = {}^{13}C/{}^{12}C$, $^{15}N/^{14}N$ or $^{18}O/^{16}O$. The standards are the PDB carbonate for carbon and oxygen isotope ratios and AIR nitrogen for nitrogen isotope ratios. The precisions for the determinations of δ values, C/N ratios, collagen concentrations and carbonate concentrations are ±0.2‰, ±0.1, ±2% and ±2% respectively.

For strontium concentration and isotopic analyses, mechanically and ultrasonically cleaned non-cortical bone fragments were ashed at 800°C for 24 hours (TUREKIAN and

Table 1: Concentrations and isotopic compositions of bone strontium, apatite carbonate and collagen

SAMPLE	SPECIES & BONE[1]	SITE[2]	AGE[3]	STRONTIUM		APATITE CARBONATE[5]			COLLAGEN[5]			
				Conc.[4]	$^{87}Sr/^{86}Sr$	$\delta^{13}C$	$\delta^{18}O$	Conc.[4]	$\delta^{13}C$	$\delta^{15}N$	C/N	Conc.[4]
MODERN MARINE												
762	Phagophilus groenlandicus	1	14	877.9	0.70919	-11.5	- 6.9	4.0	-15.8	+15.4	3.3	21.2
763	Phoca vitulina	1	15	1074.8	0.70925	-13.2	- 5.2	3.4	-15.0	+16.5	3.3	28.4
764	Phagophilus groenlandicus	1	14			-11.9	- 6.7	3.4	-15.2	+15.9	3.3	23.7
765	Phoca vitulina	1	15	1073.5	0.70927	-11.3	- 5.6	4.2	-15.4	+18.8	3.3	22.7
774	Phoca vitulina	1	15			-12.7	- 6.0		-14.3	+17.7	3.2	20.3
775	Phoca vitulina	1	15			-12.8	- 5.5		-15.0	+15.2	3.3	20.7
777	Phagophilus groenlandicus	1	14			-12.2	- 5.6		-14.4	+15.8	3.2	22.1
773	Phagophilus groenlandicus	1	14			-10.6	- 6.4		-15.1	+15.6	3.3	25.0
MODERN TERRESTRIAL												
766	Rangifer tarandus	1	8	169.0	0.75421	-12.0	-10.5	4.8	-20.6	+ 3.4	3.2	23.5
767	Rangifer tarandus	1	8			-10.9	- 9.1	5.1	-20.1	+ 2.2	3.2	21.8
768	Rangifer tarandus	1	8	181.6	0.75828	-11.2	-10.3	5.0	-20.1	+ 2.4	3.2	24.2
769	Rangifer tarandus	1	8	200.8	0.73728	-11.1	-10.8	5.9	-20.6	+ 2.8	3.3	24.2
772	Rangifer tarandus	1	9			-12.2	-10.9	4.2	-20.4	+ 3.0	3.2	25.0
776	Rangifer tarandus	1	9			-13.5	-10.9	2.9	-20.5	+ 3.8	3.2	23.6
778	Rangifer tarandus	1	9			-12.0	- 9.5	5.1	-20.1	+ 2.3	3.2	23.2
779	Rangifer tarandus	1	9			-12.5	- 8.6	3.5	-20.6	+ 2.9	3.2	23.4
PREHISTORIC MARINE												
743	Phoca sp.	2	920±50	943.2	0.74585	-10.6	- 5.9	4.4	-13.1	+14.4	3.2	21.6
745	Phoca sp.	2	980±40	909.1	0.74570	-10.9	- 4.0		-12.9	+13.8	3.2	14.9
747	Phoca sp.	2	870±40	848.7	0.74793	-11.2	- 7.8	3.5	-12.9	+14.4	3.2	19.2
PREHISTORIC TERRESTRIAL												
744	Rangifer tarandus	2	990±40	907.3	0.74929							
742	Ovis/Capra	2	960±50	933.5	0.75376							
746	Ovis/Capra	2	640±50	1041.2	0.75037	-13.0	- 6.0	6.0	-19.4	+ 4.6	3.2	18.8
748	Ovis/Capra	2	610±50			-13.4	-11.3	4.9	-20.5	+11.6	3.2	23.2
750	Ovibos moschatus	3	3870±85			- 8.2	-12.7	5.0	-19.7	+ 4.8	3.2	24.9
751	Rangifer tarandus	4	5470±95			-10.8	- 3.8	5.9	-16.8	+12.7	3.2	17.1
752	Rangifer tarandus	5	2080±75			- 8.5	-12.1	4.8	-19.8	+ 2.3	3.1	13.0
753	Rangifer tarandus	6	1830±75			- 9.3	-11.4	3.7	-21.3	+ 1.0	3.2	15.4

[1]All samples for modern marine and terrestrial animals are mandibles. The prehistoric bones are: 743, sacrum; 745, humerus; 747, pelvis; 742 and 744, ulnas; 746, femur diaphysis; 748, metacarpal; 750, cranium; 751, 752 and 753, antlers.
[2]The key to the sites is given in Table 4.
[3]The ages of the bones indicate years since collection until the present for modern bones, and radiocarbon dates in years BP for the prehistoric specimens.
[4]Concentrations of strontium are in ppm for ashed bone; those of collagen and apatite carbonate are in dry weight percent for total bone and for the inorganic fractions of bone, respectively.
[5]The δ values are given in ‰.

KULP, 1956). Samples were then dissolved in 4 N HCl. Separation of Sr and mass spectrometry procedures are the same as described in DE PAOLO (1981). The Sr procedural blank was 2 ng and was insignificant relative to sample concentrations. Strontium concentrations are reported with respect to the weight of the ashed sample. The precisions for the determinations of the strontium isotope compositions are ±0.00005; those of the strontium concentrations are ±0.2 ppm.

X-ray diffraction patterns and infrared spectra of powdered bone samples were obtained using standard methods (BONAR *et al.*, 1983; BLUMENTHAL *et al.*, 1972).

The amino acid compositions of collagen were determined as described by HARE (1980).

RESULTS

Strontium, carbon, nitrogen and oxygen isotopic compositions and concentrations are presented in Table 1, and sample locations in Table 4.

There is a clear distinction between modern marine and terrestrial feeding animals on the basis of bone strontium concentration and isotopic composition (Fig. 1). Seals (*Phagophilus* and *Phoca*) have bone strontium concentrations 700 to 900 ppm higher than reindeer (*Rangifer*), confirming the suggestion that such a separation should exist (TUREKIAN, 1961). Reindeer bones are isotopically radiogenic and heterogeneous. In contrast, the seal bones lie within our analytical uncertainty of the value measured for modern seawater ($^{87}Sr/^{86}Sr$ = 0.70923, DE PAOLO and INGRAM, 1985). This contrast in strontium concentrations and isotopic compositions between marine and terrestrial feeders is not preserved in the prehistoric bone samples, however (Fig. 1). Strontium concentrations for prehistoric seal

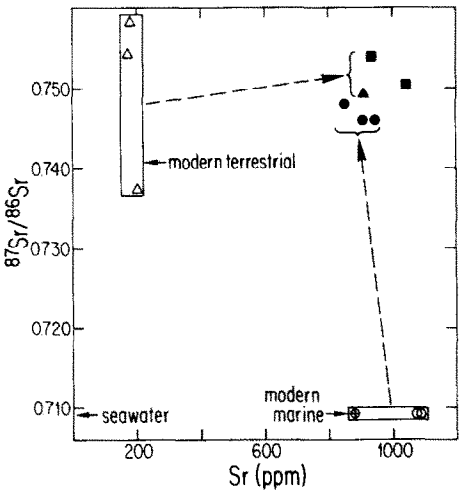

FIG. 1. Strontium isotope ratios and concentrations of bones from modern and prehistoric terrestrial and marine feeders. Open and solid symbols represent modern and prehistoric samples respectively. Circles indicate marine animals, while terrestrial animals are symbolized by various non-circular shapes. A detailed key to the symbols is given in Table 4. The boxes encompass the data for the indicated types of modern animals.

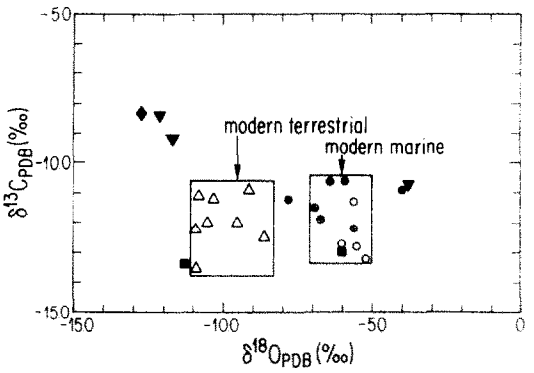

FIG. 2. Carbon and oxygen isotope ratios of the apatite carbonate of bones from modern and prehistoric terrestrial and marine feeders. Open and solid symbols represent modern and prehistoric samples respectively. Circles indicate marine animals, while terrestrial animals are symbolized by various non-circular shapes. A detailed key to the symbols is given in Table 4. The boxes encompass the data for the indicated types of modern animals.

overlap those for reindeer and sheep/goat. This overlap cannot be ascribed to *in vivo* differences in the strontium levels of the various bones we analyzed, because the range of strontium concentrations within the skeletons of modern seal and deer are only about 200 ppm (WESSEN *et al.*, 1978). Although $^{87}Sr/^{86}Sr$ ratios of prehistoric seal bones are still somewhat lower than those of bones from the prehistoric terrestrial feeders we measured, they are far removed from their original seawater composition, and even more radiogenic than that of one of the modern reindeer samples.

Oxygen isotope ratios in apatite carbonate of prehistoric bones also failed to preserve the contrast observed between modern marine and terrestrial animals (Fig. 2). Modern reindeer have $\delta^{18}O$ values 2–6‰ lower than those of marine animals, whereas the ranges of values from prehistoric terrestrial and marine animals overlap. There is no difference in the $\delta^{13}C$ values of bone carbonate apatite between modern marine and terrestrial feeders, which range from −10.6‰ to −13.5‰ (Fig. 2). Although most of the prehistoric specimens have $\delta^{13}C$ values in the same range, three terrestrial feeders have values substantially more positive than that of any modern animal we analyzed (Fig. 2).

Both the $\delta^{13}C$ and $\delta^{15}N$ values for bone collagen clearly distinguish between modern seal and reindeer (Fig. 3). The difference between the two groups is approximately 5‰ in $\delta^{13}C$ values and 14‰ in $\delta^{15}N$ values, similar to the average difference previously reported for a large group of modern marine and terrestrial feeding animals (SCHOENINGER and DENIRO, 1984). With two exceptions (samples 748 and 751), collagen $\delta^{13}C$ and $\delta^{15}N$ values distinguish between prehistoric species feeding in the marine and terrestrial environments. Although the isotope ratios for the well behaved prehistoric samples do not coincide perfectly with those of the modern marine and terrestrial animals we studied, they do fall within the ranges reported for much larger groups of modern animals that feed in the two environments (SCHOENINGER and DENIRO, 1984).

DISCUSSION

Bone collagen carbon and nitrogen isotope ratios

All but two of the prehistoric bones we analyzed had collagen with $\delta^{13}C$ and $\delta^{15}N$ values similar to the values in bone collagen of modern animals with the same feeding habits (Fig. 3). The two exceptions, samples 748 and 751, are a goat/sheep and a reindeer. These two animals may have had diets that explain their pe-

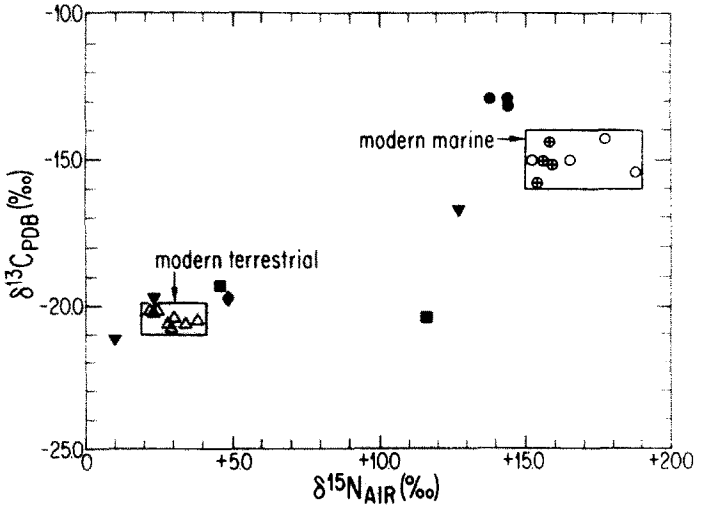

FIG. 3. Carbon and nitrogen isotope ratios of the collagen fraction of bones from modern and prehistoric terrestrial and marine feeders. Open and solid symbols represent modern and prehistoric samples respectively. Circles indicate marine animals, while terrestrial animals are symbolized by various non-circular shapes. A detailed key to the symbols is given in Table 4. The boxes encompass the data for the indicated types of modern animals.

Table 2: Amino acid compositions of bone collagen in mole %.

SAMPLE:	751	748	753	746	767	251[1]	253[2]	743	765
Hydroxyproline	9.7	9.7	9.6	10.0	10.3	9.8	9.5	10.0	9.8
Aspartic Acid	4.5	4.6	4.9	4.8	4.8	4.7	5.1	4.6	4.9
Threonine	2.0	1.9	1.8	2.1	1.9	1.9	2.0	2.0	2.0
Serine	3.7	3.3	3.5	3.4	3.5	3.5	3.6	4.2	4.3
Glutamic Acid	8.1	8.2	8.1	7.0	8.0	8.0	8.3	8.1	8.1
Proline	11.4	11.4	11.6	11.9	10.7	11.2	10.5	11.5	11.7
Glycine	33.6	32.8	32.8	33.1	32.8	33.1	33.8	32.2	32.5
Alanine	11.5	12.4	12.3	11.9	11.8	12.1	12.2	11.3	11.4
Valine	1.7	1.8	1.7	1.4	1.7	1.6	1.6	2.0	1.8
Methionine	0.6	0.6	0.5	0.5	0.5	0.4	0.2	0.7	0.5
Isoleucine	0.8	0.9	0.9	1.0	0.9	0.8	0.9	0.7	0.7
Leucine	2.4	2.6	2.6	2.7	2.6	2.6	2.6	2.6	2.6
Tyrosine	0.3	0.2	0.2	0.3	0.4	0.4	0.3	0.3	0.4
Phenylalanine	1.5	1.4	1.4	1.3	1.4	1.3	1.3	1.4	1.4
Histidine	0.6	0.5	0.5	0.7	0.5	0.6	0.4	0.5	0.7
Hydroxylysine	0.5	0.5	0.6	0.5	0.8	0.5	0.4	0.5	0.6
Lysine	2.5	2.6	2.7	2.5	2.7	2.7	2.6	2.6	2.4
Arginine	4.6	4.7	4.6	4.7	4.7	4.6	4.6	4.6	4.3

[1]Modern sheep (Ovis aries) and [2]modern goat (Capra sp.) from SCHOENINGER and DENIRO (1984).

culiar isotope ratios. The $\delta^{13}C$ and $\delta^{15}N$ values for the reindeer fall on a mixing line between pure marine and pure terrestrial feeders (Fig. 3), suggesting that this animal ate marine foods in addition to the terrestrial foods that comprised the diet of the other reindeer in this study. Under starvation conditions reindeer will eat plants refused by other animals (WESTERLING, 1970). In the case of sample 751, these may have included large quantities of seaweed or other marine plants. The isotope ratios for sample 748, the sheep/goat, do not fall on the mixing line. The $\delta^{13}C$ value for this sample is similar to those of other terrestrial feeders in this study, but its $\delta^{15}N$ value is about 10‰ higher than those of the terrestrial animals. It is possible that this animal subsisted on terrestrial plants growing along the coast. Such plants would have had normal $\delta^{13}C$ values relative to other terrestrial plants but higher $\delta^{15}N$ values because of the incorporation of nitrogen that came from seaspray (VIRGINIA and DELWICHE, 1982).

It is also possible that samples 748 and 751 had their collagen isotope ratios altered in the postmortem environment. However, two observations suggest that diagenetic alteration does not account for the anomalous isotopic compositions of collagen in these two samples. First, both collagen samples have C/N ratios of 3.2, which is the same (within analytical precision) as the C/N ratios for all other collagen samples we analyzed (Table 1). Analysis of over one hundred bone collagen samples from prehistoric marine and terrestrial feeders has shown that significant diagenetic alteration of collagen $\delta^{13}C$ and/or $\delta^{15}N$ values (on the order of 5‰ or more) is always accompanied by shifts in the collagen C/N ratios to values outside the range observed for fresh collagen samples, which is 2.9–3.6 (DENIRO, 1985). Second, collagen samples from 751 and 748 have amino acid compositions similar to those of collagens from modern and other prehistoric reindeer or sheep/goat (Table 2, Fig. 4), indicating that diagenetic processes have not altered their amino acid compositions. We conclude that the isotope ratios of collagen from samples 748 and 751 have not undergone postmortem alteration because diagenetic processes that alter collagen isotopic composition would also affect the elemental and amino acid composition of the collagen.

Most of the prehistoric bones analyzed in this study had collagen concentrations of 10–20 weight percent, substantially less than the 20–30 weight percent that characterizes modern bone (Table 1; HARE, 1980). The low temperature in the depositional environments in which these bones were interred presumably is responsible for the relatively good preservation of collagen in our samples (TURROS *et al.*, 1980). For comparison, bones analyzed by DENIRO and EPSTEIN (1981), ranging in age from 8000 years BP to 1000 years BP, that were excavated from dry caves in the Tehuacan Valley of Mexico had collagen concentrations ranging from 12 to 3 weight percent. Whatever processes were responsible for removal of collagen from the prehistoric bones we studied did not cause changes in the isotopic, elemental, or amino acid compositions of the collagen. Further study will be necessary to identify these processes, and to determine how they differ from diagenetic processes that do cause changes in collagen chemistry and isotopic and elemental compositions (DENIRO, 1985).

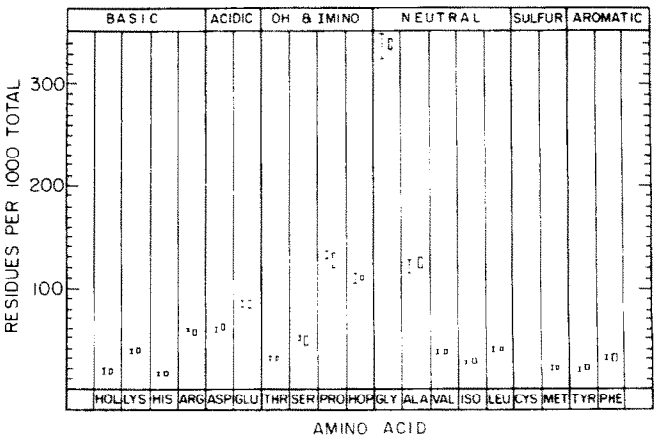

FIG. 4. Ranges of amino acid concentrations in all collagen samples listed in Table 2, indicated by boxes, compared with ranges for collagen in fresh bone from various species (HARE, 1980), indicated by vertical bars.

Bone apatite carbon and oxygen isotope ratios

The carbon isotopic composition of carbonate in the hydroxyapatite of the modern bone we analyzed did not distinguish between terrestrial and marine feeders (Fig. 2). This observation can be explained by the fact that the modern marine animals are carnivores, while the terrestrial animals are herbivores. KRUEGER and SULLIVAN (1984) observed that in herbivores, the $\delta^{13}C$ values of bone apatite carbonate are approximately 7‰ more positive than those of collagen, while for carnivores the difference is only 3‰ in the same direction. In light of this observation, the 5–6‰ difference in $\delta^{13}C$ values of bone collagen between seals and reindeer should not be reflected in $\delta^{13}C$ values of their bone apatite carbonate (Table 1). Thus, the lack of contrast between the carbonate $\delta^{13}C$ values of prehistoric marine and terrestrial feeders is not proof of diagenesis. Even so, the larger scatter in the $\delta^{13}C$ values for the prehistoric samples relative to those for modern samples (Fig. 2) does suggest some diagenetic alteration of isotopic compositions.

The observation that the 2–6‰ difference in the $\delta^{18}O$ values of apatite carbonate from the bones of modern seal and reindeer is not preserved in prehistoric marine and terrestrial animals (Fig. 2) is also reasonably explained as a consequence of diagenesis. There is an alternative explanation, however. The $\delta^{18}O$ values for the prehistoric terrestrial animals could have been shifted towards the more positive values that characterize marine mammals because of climatically induced increases in the ^{18}O concentration in the water sources they utilized. Contrary to such an interpretation is the observation that the $\delta^{18}O$ values of ice in cores from Greenland covering the same ages as our prehistoric specimens (Table 1) lie within ±1‰ of the values for ice forming at present on top of the cores (DANSGAARD *et al.*, 1969, 1975). For this reason, diagenetic effects, rather than climatically induced variations in the isotopic composition of water, are likely to be responsible for the overlap in the $\delta^{18}O$ values of carbonate apatite in the bones of prehistoric marine and terrestrial mammals.

The method we used to treat bone samples before isotopic analysis of their apatite carbonate removes authigenic carbonate minerals introduced during diagenesis (SULLIVAN and KRUEGER, 1981; SCHOENINGER and DENIRO, 1982). In this regard, note the similar concentrations of carbonate in the modern and prehistoric bones listed in Table 1. Thus, the isotopic shifts we observed result from apatite carbonate interactions with air or groundwater carbon and oxygen, and are not caused by postmortem addition of carbonate phases.

Bone mineral strontium concentrations and isotope ratios

The strontium isotope ratios and concentrations of the samples we analyzed show clear evidence for postmortem alteration (Fig. 1). Because the differences in strontium concentration and isotopic composition of bone caused by marine *versus* terrestrial feeding are at least three orders of magnitude greater than our analytical precision, diagenetic changes in these parameters are unambiguous. All of the prehistoric samples included in the strontium analyses came from a single excavation at Godthåbs Fiord. The shift of both marine and terrestrial bones from their inferred widely disparate original strontium concentrations and isotopic compositions toward the common values they share as fossils (Fig. 1) suggests that these bones interacted with a common source of strontium, most likely in groundwater. Other investigators have observed overlaps in strontium and barium concentrations in prehistoric bones of different species whose *in vivo* levels should have differed significantly (BOAZ and HAMPEL, 1978; WESSEN *et al.*, 1977; SILLEN, 1981). Such observations have also been interpreted as evidence for postmortem interaction with the depositional environment (SILLEN, 1981; SILLEN and KAVANAGH, 1982).

Strontium from soil moisture or groundwater may be introduced into fossil bone in any of four ways. First, the strontium may be associated with authigenic minerals such as calcium carbonate that were added during diagenesis. Second, the strontium may be incorporated if bone hydroxyapatite becomes more crystalline during interment (WYCKOFF, 1972). Third, the strontium may replace calcium and strontium in the original hydroxyapatite crystal structure. Finally, the strontium may not be bound within any crystalline structure but may instead reside in microcracks, dislocations or vacancies, or adsorbed onto the surface of the apatite crystals. In order to try to discriminate among these possibilities, we conducted X-ray diffraction and infrared spectroscopic analyses on selected samples.

The X-ray powder diffraction patterns for the modern (samples 763, 765, 774, 775) and prehistoric (743, 747) marine animals and modern (251, 253, 766–769) and prehistoric (746, 748) terrestrial animals show only two broad peaks at the diffraction angles that characterize poorly crystalline hydroxyapatite (BONAR *et al.*, 1983). Infrared spectroscopy of the same samples indicated that there were no differences in the degrees of crystallinity of the modern and prehistoric samples. Specifically, the magnitudes of the splitting of the antisymmetric bending mode of phosphate between 550 and 600 cm^{-1}, which is a measure of the degree of hydroxyapatite crystal size/perfection (BLUMENTHAL *et al.*, 1972), were similar for prehistoric marine and terrestrial animals compared to those for their modern counterparts. The absence of evidence for secondary mineralization and for changes in crystallinity in the prehistoric bone indicates that the drastic changes in strontium concentration and isotopic ratios we observed must be caused, in large part, by the adsorption of groundwater strontium, or by its exchange with the original strontium or calcium in the bone apatite. In order to differentiate between these two possibilities,

we conducted a leaching experiment on modern and prehistoric *Phoca* (seal) bone.

For the leaching experiment, samples 743 and 765 were ashed as described above and then powdered in an agate mortar. An aliquot of each sample was weighed and then leached in a solution consisting of equal volumes of glacial acetic acid and water for three hours while being treated with ultrasound. After centrifugation, the leachate was decanted, and combined with three ultra-pure H_2O rinses of the residue. The residue was dried, then weighed to determine the weight loss by leaching. The prehistoric sample (743) was more susceptible to leaching, losing 59% of its weight, compared to 24% for the modern sample (765). Further results of the leaching experiment are shown in Table 3. The $^{87}Sr/^{86}Sr$ ratios of the leachate and residue from the prehistoric sample are the same within analytical uncertainty. The difference with respect to the value for untreated bone is probably a result of heterogeneous alteration of the sample. Compared with the modern sample, which showed essentially no shift in strontium concentration after leaching, the prehistoric bone has strontium that is more labile in that its residue has approximately half the concentration of the unleached sample (Table 3).

By analogy with experiments that employ acid leaching of minerals to remove ions adsorbed onto crystal surfaces or in microcracks (GANCARZ and WASSERBURG, 1977; NELSON and BICKFORD, 1980; GOPEL *et al.*, 1985), we believe that the leaching of the prehistoric bone was so severe that similarly adsorbed ions should have been removed from the residue. The absence of a shift in isotopic composition caused by leaching argues for the exchange of strontium in groundwater with original calcium or strontium atoms in the bone apatite as the major mechanism of diagenetic alteration in the samples analyzed in this study. We estimate that original strontium must constitute less than one percent of the total strontium in the prehistoric bone used in the leaching experiment. If more than this amount of original strontium were present, the isotopic composition of the residue would differ from that of the leachate. It may be possible to analyze the $^{87}Sr/^{86}Sr$ ratios of small volumes of a prehistoric bone such as sample 743 using an ion microprobe in order to determine whether or not diagenetic replacement of *in vivo* strontium is complete.

Table 3: Strontium concentrations and isotopic compositions of untreated modern and prehistoric seal bone and of their leached fractions and residues[1].

Sample	Sr(ppm)	$^{87}Sr/^{86}Sr$
Modern seal (765. *Phoca vitulina*)		
untreated	1073.5	0.70927
leachate[2]	843.6	0.70958
residue	1086.5	0.70923
Prehistoric seal (743. *Phoca* sp.)		
untreated	943.2	0.74585
leachate[2]	1049.0	0.74559
residue	467.0	0.74554

[1]Leached fractions and residues were produced using a 1:1 mixture (by volume) of glacial acetic acid and water.
[2]Expressed as concentration in bone removed by leaching.

Table 4: Symbol key for Figs. 1-3.

SYMBOL	SITE	SPECIES	AGE
⊕	1	*Phagophilus groenlandicus*	modern
△	1	*Rangifer tarandus*	modern
○	1	*Phoca vitulina*	modern
▲	2	*Rangifer tarandus*	prehistoric
●	2	*Phoca* sp.	prehistoric
■	2	*Ovis/Capra*	prehistoric
◆	3	*Ovibus moschatus*	prehistoric
▼	4,5,6	*Rangifer tarandus*	prehistoric

The sites are: 1, unspecified locations in southern West Greenland; 2, Godthåbs Fiord, southern West Greenland; 3, Solbakken, Hall Land, western North Greenland; 4, 5, and 6, Kap Moltke, Paralleldalen, and Kap Morris Jessup, respectively, all in Peary Land, North Greenland.

CONCLUSIONS

The results of this study have specific implications for the nature of the diagenetic processes that affected the chemical and isotopic compositions of the prehistoric bones we analyzed, and more general consequences for the use of chemical and isotopic tracers in bone as paleodietary indicators.

Our data show no persuasive evidence for alteration of carbon and nitrogen isotopic composition of bone collagen during diagenesis. In contrast, the isotopic and chemical tracers that reside in the inorganic phases of bone did not preserve their dietary signatures. The strontium concentrations and isotopic compositions give the most clearcut evidence of this, in that all prehistoric samples excavated from the same site, regardless of their original composition, were altered to similar ranges of values. In the case of the seal bones, strontium concentrations remained approximately constant, so that the isotopic data are the only indication that the original bone strontium had been replaced. The differences in strontium concentrations between modern and prehistoric reindeer bones demonstrate that the similarity of concentrations in modern and prehistoric seal bones was only coincidental. An obvious problem with interpreting dietary tracers in the inorganic phases of bone is that these phases may be contaminated by minerals deposited from groundwater. But the leaching experiment reported here demonstrates that nearly total replacement of structurally bound ions is also possible. By inference, similar effects may occur for other alkaline earths useful for dietary analysis, such as barium (WESSEN *et al.*, 1978; ELIAS *et al.*, 1982; LAMBERT *et al.*, 1984).

In extending the observations reported here to applications of isotopic and chemical analysis of bone

from other locations, we emphasize that the samples we analyzed are not representative of the usual types of samples and sites of interest to most prehistorians. They were chosen to maximize the possibility of identifying diagenetic alteration in bone strontium and apatite carbonate. In this regard, we note that bones of marine animals are less dense than those of terrestrial ones, so that the latter, as well as dense skeletal elements such as enamel, may be more resistant with regard to strontium alteration than the seal bones analyzed here. However, the convergence of bone strontium concentration and isotopic composition for the prehistoric terrestrial and marine feeders suggests similar susceptibility to alteration. The stability of collagen and the absence of diagenetic alteration of its $\delta^{13}C$ and $\delta^{15}N$ values in the samples we analyzed probably are due, in part, to the low temperature at which the bones resided prior to being excavated. Bones excavated from sites where higher temperatures prevailed are more likely to be affected by postmortem processes that can, in some cases, lead to alteration of collagen elemental and isotopic ratios (DENIRO, 1985). Our results should be taken as a warning that diagenetic alteration of bone chemistry and isotopic composition is a potential problem at every site. Diagenetic effects must be evaluated by application of the approach used here or by other means prior to attempting to reconstruct prehistoric diet from the chemical or isotopic composition of fossil bone.

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