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EA-IRMS: Tracking human and animal dietary habits using isotope fingerprints recorded in bone collagen

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Goal

Determine the dietary habits of humans and animals by detecting the nitrogen, carbon, sulfur isotope fingerprints recorded in bone collagen.

Introduction

Measurements of nitrogen and carbon stable isotopes (hereafter $\delta^{13}C$ and $\delta^{15}N$) in bone collagen have been interpreted reliably as an indicator of human and animal diets because they primarily reflect the protein fraction of the consumer's diet. It may also be possible to use $\delta^{13}C$ and $\delta^{15}N$ from mammal bone collagen records to track movement and past climate changes as vegetation sources (and hence food sources) change with temperature and aridity. More recently, sulfur stable isotopes (hereafter $\delta^{34}S$) have also been employed in dietary studies and may also be used to trace geographic origins of humans and animals because of their relationship to the underlying geology. Most research, however, has focused on $\delta^{13}C$ and $\delta^{15}N$, with far fewer studies incorporating $\delta^{34}S$.

A key challenge to simultaneous $\delta^{13}C$, $\delta^{15}N$ and $\delta^{34}S$ analysis in bone collagen was the accurate and precise analysis of small concentrations of sulfur (around 0.2-0.3%).

In this application note, we show $\delta^{13}C$, $\delta^{15}N$ and $\delta^{34}S$ data from bone collagen measured using the Thermo Scientific™ EA IsoLink™ IRMS System. The challenge of sulfur analysis is overcome and an insight on animal and human diet trends is observed.

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Analytical configuration

For simultaneous $\delta^{13}C$, $\delta^{15}N$ and $\delta^{34}S$ analysis, around 1 mg of dried, homogenized bone collagen material was weighed into tin capsules and introduced into the EA IsoLink IRMS System from the Thermo Scientific™ MAS Plus Autosampler. The combustion reactor was held at 1020 °C and was packed with an oxidizer (tungstic anhydride) and a reducer (copper wires). The produced N_2, CO_2 and SO_2 gases were separated using a temperature ramped GC. After separation, the gases were transferred to a Thermo Scientific™ DELTA V™ IRMS via the Thermo Scientific™ ConFlo IV™ Universal Interface. For our samples, the sample weights correspond to approximately 140 µg of N, 500 µg of C and 3 µg of S. $\delta^{13}C$, $\delta^{15}N$ and $\delta^{34}S$ were calibrated against USGS 40, USGS 42, IAEA S1, IAEA S2 and IAEA S3, respectively. Analysis time was less than 10 minutes, using less than 1.4 liters of helium per sample.

$\delta^{13}C$, $\delta^{15}N$ and $\delta^{34}S$ isotope fingerprints of bone

The $\delta^{13}C$ and $\delta^{15}N$ fingerprints in bone reflect the isotopic composition of the diet of the consumer, implying that it can be used as a way to reconstruct or trace diet. In addition, $\delta^{34}S$ fingerprints are used to account for the amount and origin of protein in the diet, which reflects the environment form which the plants/tissue was grown. Therefore, in conjunction with $\delta^{13}C$ and $\delta^{15}N$ fingerprints, $\delta^{34}S$ fingerprints can provide an overview of the consumer diet and the source location of the foods[1]. Here, our data suggest a diet strongly influenced by C_3 and C_4 plants derived from terrestrial habitats (cattle). However, the $\delta^{15}N$ fingerprints in the human bones indicate a diet influenced by marine based fauna, particularly fish.

$\delta^{13}C$, $\delta^{15}N$ and $\delta^{34}S$ data

The collagen data are presented in Table 1. The $\delta^{13}C$, $\delta^{15}N$ and $\delta^{34}S$ data are of very high precision (triplicate measurements), with most samples ≤0.1‰. In addition, Figure 1 illustrates the $\delta^{15}N$ and $\delta^{34}S$ data, which shows a differentiation of collagen type as a function of diet.

Table 1. $\delta^{13}C$, $\delta^{15}N$ and $\delta^{34}S$ data from bone collagen.

Collagen type	$\delta^{13}C_{VPDB}$ (‰)	$\delta^{15}N_{AIR}$ (‰)	$\delta^{34}S_{VCDT}$ (‰)
Cattle	-16.0±0.01	6.12±0.03	12.71±0.07
Cattle	-17.8±0.02	6.69±0.01	10.30±0.05
Cattle	-17.6±0.01	5.45±0.01	7.20±0.09
Cattle	-17.7±0.01	5.73±0.01	2.21±0.01
Cattle	-18.8±0.01	8.96±0.02	5.71±0.03
Cattle	-19.8±0.10	3.14±0.07	4.28±0.10
Cattle	-20.3±0.02	5.06±0.06	4.96±0.02
Human	-18.7±0.01	8.50±0.03	3.58±0.04
Human	-18.9±0.01	9.38±0.01	2.90±0.04
Human	-18.7±0.01	9.45±0.02	4.69±0.02
Human	-17.9±0.01	10.72±0.01	3.09±0.05
Human	-18.5±0.01	9.46±0.01	4.72±0.02
Mammoth	-21.7±0.03	7.56±0.02	-25.43±0.21
Mammoth	-21.6±0.01	10.26±0.02	4.10±0.02

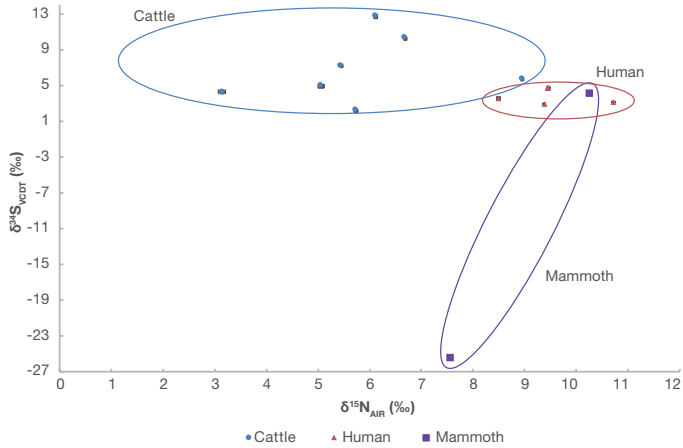

Figure 1. $\delta^{15}N$ and $\delta^{34}S$ showing separation of collagen type as a function of diet.

Summary: A new approach to NCS analysis

The $\delta^{13}C$, $\delta^{15}N$ and $\delta^{34}S$ fingerprints of bone collagen provide insights into current and past diet trends whilst also providing insight into the environmental origin of food sources. This is commonly applied in archaeology, palaeodietary research, forensic applications and studies investigating origin.

We have shown that the EA IsoLink IRMS System processes bone collagen samples for $\delta^{13}C$, $\delta^{15}N$ and $\delta^{34}S$ in a single sample drop. Traditionally, given the low concentration of sulfur in bone collagen of 0.2% to 0.3%, samples were measured for $\delta^{13}C$ and $\delta^{15}N$ using 0.5 mg to 1.0 mg of bone collagen, followed by a separate analysis for $\delta^{34}S$ using 10 mg to 15 mg of bone collagen: this meant, two separate analysis requiring different instrument configurations. However, the EA IsoLink IRMS System produces $\delta^{13}C$, $\delta^{15}N$ and $\delta^{34}S$ data from 1 mg of bone collagen, meaning overall cost per sample analysis is reduced, less maintenance is required and system throughput is significantly enhanced.

References

1. Nehlich, O., Oelze, V.M., Jay, M., Conrad, M., Stäuble, H., Teegen, W-R., Richards, M.P. (2014). Sulfur isotope ratios of multi-period archaeological skeletal remains from central Germany: A dietary study. Anthropologie. LII/1, 15-33.

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