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Biogeochemical data from well preserved 200 ka collagen and skeletal remains

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Abstract

Data presented herein form part of a pilot study to reconstruct palaeoenvironment using isotopic data from 200 ka collagen and skeletal remains. To date such a study has only been possible on materials of up to 120 ka, due to poor preservation, but amino acid compositional analyses of mammal molar collagen herein indicate excellent preservation of the 200 ka collagen. Samples studied were found in association with faunal evidence for a temperate climate, and oxygen and carbon isotope data support this evidence. Collagen $\delta^{15}N$ values, however, are akin to those typically found in collagen from modern semi-arid to arid environments. Similar data have been reported for the Eemian interglacial, and it is likely that these data reflect specific environmental adaptations, as opposed to aridity. This pilot study emphasises the need for complementary isotopic and stratigraphical analyses in palaeoenvironmental studies.

Keywords: collagen; biogeochemical methods; climate mammals; enamel; isotopes; oxygen; carbon; nitrogen

1. Introduction

The isotopic composition of mammal skeletal and collagen remains has been used extensively to reconstruct palaeoenvironments and palaeodiet throughout the upper Pleistocene period (e.g. [1]). Such studies typically analyse carbon and nitrogen isotopes from collagen; and carbon and oxygen from skeletal material, i.e. teeth and bones. Carbon isotopes from collagen can be used to identify the animal's environment as $^{13}C/^{12}C$ ratios tend to be lower in closed-canopy forests than in open plant formations, such as steppes and grasslands. Nitrogen isotopes in collagen are used to infer aridity or water stress, and studies indicate that herbivores from arid, low rainfall environments exhibit higher N ratios than those from temperate environments [2,3]. In skeletal material, carbon isotopes largely reflect an animal's diet, i.e. the difference between C_3 and C_4 plant intake, whereas oxygen largely reflects in-

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gested water oxygen [4]. To date, palaeoenvironmental reconstruction using collagen and skeletal material has only been carried out on materials of up to 120 ka [1] because collagen is rarely well preserved beyond this timescale. Data presented herein form part of a pilot study on 200 ka collagen for palaeoenvironmental reconstruction.

Channel deposits at Stanton Harcourt in southern Britain are believed to correspond to marine oxygen isotope (MOI) Stage 7 (200 ka) [5], and floral and faunal remains are indicative of a fully interglacial temperate climate [6,7]. The site yields well preserved mammalian remains, including mammoth (*Mammuthus* sp.), straight tusked elephant (*Palaeoloxodon antiquus*), horse (*Equus ferus*), bison (*Bison priscus*), red deer (*Cervus elaphus*), brown bear (*Ursus arctos*), and lion (*Panthera leo*).

Mammuthus sp. remains far outnumber all other mammal remains at the site, which may indicate favourable conditions for this species. However, these mammoths had less evolved dentition and were about one third smaller in size than *Mammuthus primigenius* from cold stage deposits. Size reduction and 'primitive dentition' also characterise mammoths from other British sites attributed to MOI Stage 7 [8,9]. Reduction in mammalian body size has been attributed to insularity and/or an unfavourable environmental change [10,11].

Terrestrial biological data from MOI Stage 7 sites are rare; this site thus provides a unique opportunity to investigate terrestrial climatic conditions during this interglacial period. Herein we analyse oxygen, carbon and nitrogen isotopes extracted from collagen, carbonate and phosphate fractions of *Mammuthus* sp., *P. antiquus*, *E. ferus* and *B. priscus* molars. Other mammal samples had been chemically treated for preservation and were not suitable for analysis.

2. Materials and methods

2.1. Collagen extraction

Collagen from the dentine fraction was analysed following the method outlined in [12]. Collagen analyses were performed using a Carlo Erba automated carbon and nitrogen analyser coupled to a Europa Geo 20/20 isotope ratio-monitoring mass spectrometer. Typical replicate measurement errors are of the order of ± 0.2‰ for both $\delta^{13}C$ and $\delta^{15}N$, which are reported relative to PDB and AIR, respectively.

2.2. Amino acid analyses

The preservation of collagen can be tested through analysis of the amino acid profile [13]. For this purpose, the isolated collagen protein was dissolved in distilled water to create a solution containing approximately 6 μg of collagen in a 20 μl aliquot. In this study the collagen solutions were run on an ABI 420A derivatiser/analyser following hydrolysis for 24 h in 5.7 N hydrochloric acid at 110°C [14].

2.3. Enamel phosphate and carbonate analyses

Samples for phosphate $\delta^{18}O$ analyses were prepared using the silver phosphate method [15], which has an analytical reproducibility of ± 0.1–0.3‰ (1σ).

Samples for carbonate $\delta^{18}O$ and $\delta^{13}C$ analyses were pretreated with hydrogen peroxide before acidification with 100% H_3PO_4 at 90°C on an ISOCARB 2 preparation system. Analyses of the evolved CO_2 were carried out on a VG PRISM mass spectrometer, with reproducibility of ± 0.1‰ for $\delta^{13}C$ and ± 0.3‰ for $\delta^{18}O$.

3. Results

3.1. Collagen

Collagen $\delta^{13}C$ and $\delta^{15}N$ data from fossil samples SH6/152, SH6/244, SH1/353, SH4/45 and 339 are listed in Table 1. Four samples yielded C:N ratios of around 3.2, equivalent to that of fresh collagen [16], but samples SH7/126 and SH8/7 yielded insufficient collagen for analysis, and collagen from sample SH1/353 had a C:N value of 5, which may be suspect. Collagen $\delta^{13}C$ data for the four good samples are in the region of -21.6 to

Table 1
Isotopic data from collagen fractions from the Stanton Harcourt mammal molars

Animal	Sample	C:N ratio	C in collagen (%)	N in collagen (%)	Collagen $\delta^{13}C$	Collagen $\delta^{15}N$
B. priscus	339	3.3	23.2	8.5	−20.9	11.0
P. antiquus	SH6/244	3.4	23.9	8.2	−21.6	13.2
P. antiquus	SH4/45	3.0	37.9	14.5	−20.8	10.7
Mammuthus sp.	SH6/152	3.1	40.3	15.2	−21.1	10.9
Mammuthus sp.	*SH1/353*	*5.0*	*12.8*	*3*	*−23.6*	*11.2*

Carbon data are reported relative to PDB and nitrogen to AIR. Analytical precision is ±0.2‰ for C and N. $n=4$ for all samples, where n = number of samples from each specimen. SH1/353 is shown in italics as the C:N ratio was outside the acceptable range [16].

−20.8‰ (PDB). The $\delta^{15}N$ data are greater than 10‰ for all samples, with a range of 10.7–13.2‰ (AIR).

3.2. Amino acid profiles

Amino acid profiles were analysed in mammoth and elephant samples – SH4/45, SH6/244 and SH6/152 – and compared to profiles in modern elephants from Amboseli National Park, Kenya. Similar quantities of amino acids were extracted. These data (Fig. 1) show the modern and ancient collagen profiles to be remarkably similar, which suggests that the 200 ka collagen is well preserved.

3.3. Enamel

Oxygen and carbon isotope data from molar enamel carbonate and phosphate are listed in Table 2. The mammalian skeleton consists of both phosphate and carbonate, and phosphate oxygen ($\delta^{18}O_{PO4}$) is related to carbonate oxygen ($\delta^{18}O_{CO3}$) by $\Delta_{(CO3-PO4)}=9.2$‰ [15]. From Table 2, we see that for samples SH6/152, SH1/353 and SH4/45 $\Delta_{(CO3-PO4)}$ has a mean value of 9.3‰, consistent with modern values and therefore indicative of well preserved material. Carbonate $\delta^{13}C$ ranges between −12.1 and −10.1‰ and carbonate $\delta^{18}O$ ranges between 24.3 and 26.5‰. Phos-

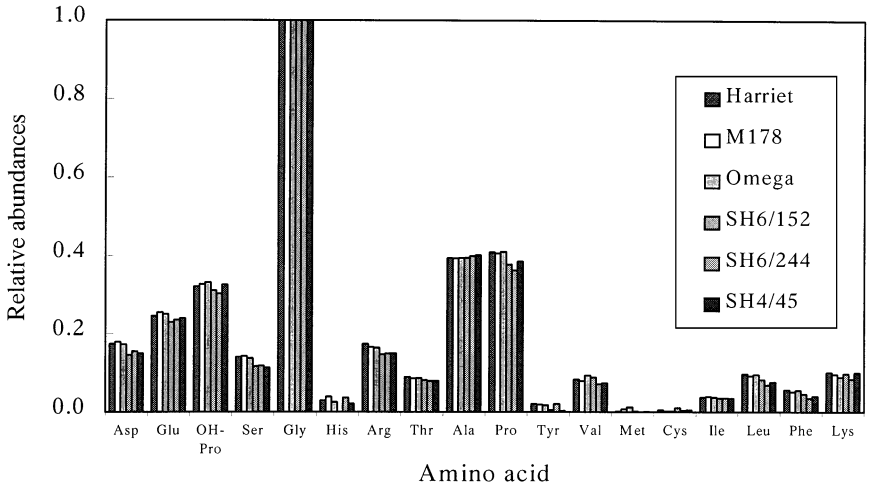

Fig. 1. Amino acid data from modern elephant (Harriet, Omega and M178) and fossil elephant and mammoth (SH6/152, SH6/244, SH4/45) samples. Similar amino acid compositions were observed in both modern and fossil collagen. Abbreviations: aspartic acid – Asp, glutamic acid – Glu, hydroxyproline – OH-Pro, serine – Ser, glycine – Gly, histidine – His, arginine – Arg, threonine – Thr, alanine – Ala, proline – Pro, tyrosine – Tyr, valine – Val, methionine – Met, cysteine – Cys, isoleucine – Ile, leucine – Leu, phenylalanine – Phe, lysine – Lys.

Table 2
Isotopic data from carbonate and phosphate fractions from the Stanton Harcourt mammal molars

Animal	Sample	CO_3 $\delta^{18}O$	CO_3 $\delta^{13}C$	PO_4 $\delta^{18}O$	Water $\delta^{18}O$[a]
Mammuthus sp.	SH6/152	25.6	−11.4	16.2	−8.7
P. antiquus	SH4/45	24.3	−11.3	15.0	−10.0
Mammuthus sp.	SH1/353	26.5	−11.3	17.1	−7.6
P. antiquus	SH6/244	24.9	−12.1	–	−9.3
B. priscus	339	24.6	−11.3	–	−8.5
B. priscus	SH7/126	24.8	−10.1	–	−8.1
E. ferus	SH8/7	25.4	−12.1	–	−9.0

Water $\delta^{18}O$ data are calculated using animal specific relationships. Oxygen data are reported relative to SMOW, carbon to PDB. Analytical precision is ±0.1‰ for carbonate $\delta^{13}C$, ±0.3‰ for carbonate and phosphate $\delta^{18}O$. $n=4$ for all samples, where n = number of samples from each specimen.

[a] Denotes values calculated from PO_4 or from CO_3 assuming $\Delta^{18}O_{PO4-CO3} = 9.2‰$.

phate $\delta^{18}O$, for the three samples analysed, ranges between 15.0 and 17.1‰.

4. Calculating palaeowater $\delta^{18}O$

There is a general relationship between precipitation $\delta^{18}O$ and ambient temperature [17–19], where lower water $\delta^{18}O$ values indicate lower ambient temperatures. This relationship can be complicated, however, by local environmental parameters. Precipitation in present-day southern Britain has a mean $\delta^{18}O$ value of around −7‰. If similar environmental conditions prevailed at 200 ka, then similar precipitation $\delta^{18}O$ would be expected. A means of obtaining palaeoprecipitation $\delta^{18}O$ data is through interpretation of mammal phosphate and carbonate $\delta^{18}O$.

Mammal phosphate and carbonate $\delta^{18}O$ are directly influenced by body water $\delta^{18}O$, which is largely controlled by ingested (surface water) $\delta^{18}O$. This relationship, however, can also depend upon additional factors including animal species and dietary intake [20,21].

Different species can exhibit different relationships between phosphate (and carbonate) $\delta^{18}O_{PO4}$ and ingested water $\delta^{18}O_{sw}$, i.e. the influence of ingested water on skeletal oxygen is variable due to variations in metabolism and water intake, but in general the relationship is approximately 0.75 [21]. Herein we analyse elephant, mammoth, horse and bison $\delta^{18}O$. The relevant relationships between $\delta^{18}O_{PO4}$ and $\delta^{18}O_{sw}$ for these mammals are discussed below.

The investigation of [22] showed that the relationship between phosphate oxygen and ingested water oxygen ($\delta^{18}O_{PO4}$–$\delta^{18}O_{sw}$) for elephants and mammoths had an unusually high slope. A more recent study [23] verified this relationship, on the basis of faunal and dietary data, to be:

$$\delta^{18}O_{PO4} = 0.88 \times \delta^{18}O_{sw} + 23.9 \quad (1)$$

The relationship between *Bison* sp. enamel and ingested water $\delta^{18}O$ has not been established. This can be developed, however, from data presented by [24]:

$$\delta^{18}O_{PO4} = 0.77 \times \delta^{18}O_{sw} + 21.9 \quad (2)$$

For *Equus* sp., $\delta^{18}O_{PO4}$–$\delta^{18}O_{sw}$ has been extensively studied [25–27], and according to [27] it is:

$$\delta^{18}O_{PO4} = 0.71 \times \delta^{18}O_{sw} + 22.6 \quad (3)$$

Palaeowater $\delta^{18}O$ data for the Stanton Harcourt site, calculated using these species specific relationships, are listed in Table 2. Palaeowater $\delta^{18}O$ ranges from −7.6‰ to −10.0‰.

5. Discussion

The amino acid data show that the fossil and modern collagen profiles are very similar thus implying that the Stanton Harcourt collagen is well preserved, despite the antiquity of the samples. These are the first samples older than ~120 ka

to produce reliable collagen data, providing promise of further successful study of such ancient samples. That channel deposits have produced such high quality samples may be surprising, but similar collagen yields have previously been extracted from Seine River deposits [28].

Carbon isotope data obtained from both the carbonate and collagen fractions of the Stanton Harcourt mammal molars are consistent with an open environment consisting of C_3 trees and plants [4], as expected from faunal and floral evidence. The oxygen isotope data from the carbonate and phosphate fractions can be used to infer that mean surface water $\delta^{18}O$ at Stanton Harcourt was generally similar 200 ka to that of present-day Britain, thus suggesting that the Stanton Harcourt mammals experienced similar temperate conditions.

The collagen $\delta^{15}N$ data, however, are higher than expected for a temperate climate. Animal collagen $\delta^{15}N$ is influenced by dietary intake and because plants from arid water-stressed regions exhibit higher $\delta^{15}N$ than normal, higher $\delta^{15}N$ values are seen in animals consuming these plants [29,30]. Field data also show that mammal $\delta^{15}N$ may be further elevated relative to dietary $\delta^{15}N$ under conditions of physiological stress, such as water stress associated with aridity [2,3].

Without baseline floral $\delta^{15}N$, it is impossible to interpret precisely the elevated $\delta^{15}N$ measured in the Stanton Harcourt mammals, but it can be said that these values are similar to those of modern elephants from semi-arid environments (Fig. 2). The faunal and floral evidence from Stanton Harcourt, however, does not support a hypothesis of aridity, and as the empirical relationship between bison and mammoth collagen $\delta^{15}N$ and precipitation has not been established, inferring aridity from these data may be premature. One may also expect $\delta^{18}O$ values to be elevated if aridity was indeed a factor. It is of note that similar high N values have been recorded in Eemian herbivores from a forested environment [1], perhaps reflecting environmental parameters specific to certain parts of an interglacial period.

To date, collagen analyses of contemporaneous mammoth, horse and bison have shown mammoth $\delta^{15}N$ to be notably higher than that of bison and horse [35–37], a feature that has been attributed to distinctive dietary adaptations [35]. However, at Stanton Harcourt the bison and elephant display similar $\delta^{15}N$ values, which may be indicative of an environment that did not permit differentiation of feeding habitats. This hypothesis assumes, however, that these mammals would exhibit the same isotopic symptoms under the same environmental conditions. Such a scenario may have arisen from overcrowding or a reduction in food resources, but it has not been possible, as yet, to determine from stratigraphic data whether such conditions existed.

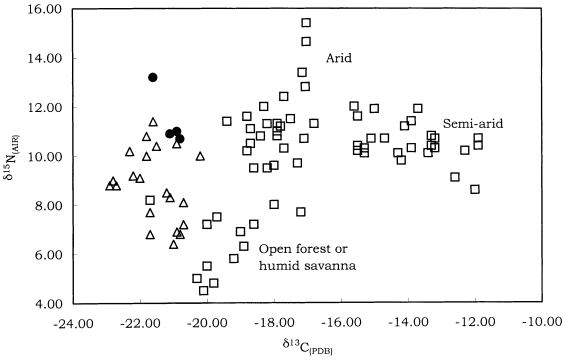

Fig. 2. Collagen $\delta^{13}C$ versus $\delta^{15}N$ for Stanton Harcourt mammoth, elephant and bison (SH6/152, SH4/45, 339) (closed circles) shown alongside *L. africana* collagen isotopic data taken from [31–34] (open squares) and cold climate mammoth data [35–37] (open triangles). These data show how these isotopes can be used to differentiate between arid, semi-arid, open forested and humid savanna environments. The Stanton Harcourt data lie within values typical of semi-arid environments.

6. Conclusions

The collagen data are the first of this age to yield reliable amino acid profiles, indicative of unaltered collagen. The similarity in isotopic values between the mammoth, elephant and bison suggests that they shared a similar environment. However, the type of environment cannot be easily defined. The nitrogen data are similar to those from modern semi-arid environments but a hypothesis that such an environment existed at Stanton Harcourt does not tally with the faunal

and floral nor the carbon and oxygen data. It is possible that certain parts of interglacials provide conditions that give rise to elevated $\delta^{15}N$ amidst temperate conditions but our current understanding of the factors affecting collagen $\delta^{15}N$ in large mammals cannot provide a definitive explanation.

This pilot study emphasises the need for the combination of several techniques for a more comprehensive understanding of palaeoenvironments. Work is underway to further investigate this site.

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References

[1] H. Bocherens, D. Patou-Mathis, M. Otte, D. Bonjean, M. Toussaint, A. Mariotti, Palaeoenvironmental and palaeodietary implications of isotopic biogeochemistry of later interglacial and Neandertal and mammal bones in Scladina Cave (Belgium), J. Archaeol. Sci. 26 (6) (1999) 599–607.

[2] J.C. Sealy, N.J. van der Merwe, J.A. Lee-Thorp, J.L. Lanham, Nitrogen isotopic ecology in southern Africa: implications for environmental and dietary tracing, Geochim. Cosmochim. Acta 51 (1987) 2707–2717.

[3] D.R. Gröcke, H. Bocherens, A. Mariotti, Annual rainfall and nitrogen-isotope correlation in macropod collagen: application as a palaeoprecipitation indicator, Earth Planet. Sci. Lett. 153 (1997) 279–285.

[4] P.L. Koch, M.L. Fogel, N. Tuross, Tracing the diets of fossil animals using stable isotopes, in: K. Lajtha, R.H. Michener (Eds), Stable Isotopes in Ecology and Environmental Science, Blackwell Scientific Publications, 1994.

[5] D.Q. Bowen, S. Hughes, G.A. Sykes, G.H. Miller, Land–sea correlations in the Pleistocene based on isoleucine epimerisation in non-marine molluscs, Nature 340 (1989) 49–51.

[6] K. Scott, C. Buckingham, Quaternary fluvial deposits and palaeontology at Stanton Harcourt, Oxfordshire, in: S.G. Lewis, D. Maddy (Eds.), The Quaternary of the South Midlands and the Welsh Marches, Quaternary Research Association, 1997.

[7] C.M. Buckingham, D.A. Roe, K. Scott, A preliminary report on the Stanton Harcourt Channel Deposits (Oxfordshire, England); geological context, vertebrate remains and Palaeolithic stone artefacts, J. Quat. Sci. 11 (1996) 397–415.

[8] A.M. Lister, Evolution and taxonomy of Eurasian mammoths, in: J. Shoshani, P. Tassy (Eds.), The Proboscidea: Trends, Evolution and Palaeoecology, Oxford University Press, 1996, pp. 203–218.

[9] D.C. Schreve, Mammalian Biostratigraphy of the Later Middle Pleistocene in Britain, University College of London, London, 1997.

[10] A.M. Lister, Palaeontology; mammoths in miniature, Nature 362 (1993) 288–289.

[11] S.L. Vartanyan, V.E. Garutt, A.V. Sher, Holocene dwarf mammoths from Wrangel island in the Siberian Arctic, Nature 362 (1993) 337–339.

[12] M.P. Richards, R.E.M. Hedges, Stable isotope evidence for similarities in the types of marine foods used by late Mesolithic humans at sites along the Atlantic Coast of Europe, J. Archaeol. Sci. 26 (1999) 717–722.

[13] S. Turban-Just, S. Schramm, Stable carbon and nitrogen isotope ratios of individual amino acids give new insights into bone collagen degradation, Bull. Soc. Geol. Fr. 169 (1998) 109–114.

[14] R.L. Heinrikson, S.C. Meredith, Amino acid analysis by reverse phase high performance liquid chromatography: precolumn derivatization with phenylisothiocyanate, Anal. Biochem. 136 (1984) 65–74.

[15] P. Iacumin, H. Bocherens, A. Mariotti, A. Longinelli, Oxygen isotope analyses of co-existing carbonate and phosphate in biogenic apatite: a way to monitor diagenetic alteration of bone phosphate, Earth Planet. Sci. Lett. 142 (1996) 1–6.

[16] M.J. DeNiro, Postmortem preservation and alteration of in vivo bone collagen isotope ratios in relation to palaeodietary reconstruction, Nature 317 (1985) 806–809.

[17] H.C. Fricke, J.R. O'Neil, The correlation between $^{18}O/^{16}O$ ratios of meteoric water and surface temperature: its use in investigating terrestrial climate change over geologic time, Earth Planet. Sci. Lett. 170 (1999) 181–196.

[18] K. Rozanski, L. Araguás-Araguás, R. Gonfiiantini, Isotopic patterns in modern global precipitation, in: P.K. Swart, K.C. Lohmann, J. McKenzie, S. Savin (Eds.), Climate Change in Continental Isotopic Records, Geophysical Monographs 78, 1993, pp. 1–36.

[19] W. Dansgaard, Stable isotopes in precipitation, Tellus 16 (1964) 436–468.

[20] J.D. Bryant, P.N. Froelich, W.J. Showers, B.J. Genna, A tale of two quarries: Biologic and taphonomic signatures on the oxygen isotope composition of tooth enamel phosphate from Modern and Miocene equids, Palaios 11 (1996) 397–408.

[21] M.J. Kohn, Predicting animal $\delta^{18}O$: Accounting for diet and physiological adaptation, Geochim. Cosmochim. Acta 60 (1996) 4811–4829.

[22] L.K. Ayliffe, A.M. Lister, A.R. Chivas, The preservation of glacial–interglacial climatic signatures in the oxygen isotopes of elephant skeletal phosphate, Palaeogeogr. Palaeoclimatol. Palaeoecol. 99 (1992) 179–191.

[23] A.M. Jones, P.L. Koch, E.D. Young, Modeling $\delta^{18}O$ in proboscidean phosphate, in preparation.

[24] H.C. Fricke, W.C. Clyde, J.R. O'Neil, Intra-tooth variations in $\delta^{18}O_{PO4}$ of mammalian tooth enamel as a record of seasonal variations in continental climate variables, Geochim. Cosmochim. Acta 62 (1998) 1839–1851.

[25] B. Sánchez Chillón, M.T. Alberdi, G. Leone, F.P. Bonadonna, B. Stenni, A. Longinelli, Oxygen isotopic composition of fossil equid tooth and bone phosphate: an archive of difficult interpretation, Palaeogeogr. Palaeoclimatol. Palaeoecol. 107 (1994) 317–328.

[26] J.D. Bryant, B. Luz, P.N. Froelich, Oxygen isotopic composition of fossil horse tooth phosphate as a record of continental paleoclimate, Palaeogeogr. Palaeoclimatol. Palaeoecol. 107 (1994) 303–316.

[27] A.D. Huertas, P. Iacumin, B. Stenni, B.S. Chillón, A. Longinelli, Oxygen isotope variations of phosphate in mammalian bone and tooth enamel, Geochim. Cosmochim. Acta 59 (1995) 4299–4305.

[28] H. Bocherens, A. Tresset, F. Wiedemann, F. Giligny, F. Lafage, Y. Lanchon, A. Mariotti, Bone diagenetic evolution in two French Neolithic sites, Bull. Soc. Geol. Fr. 168 (5) (1997) 555–564.

[29] T.H.E. Heaton, J.C. Vogel, G. von la Chevallerie, G. Collett, Climatic influence on the isotopic composition of bone nitrogen, Nature 332 (1986) 822–823.

[30] T.H.E. Heaton, The $^{15}N/^{14}N$ ratio of plants in South Africa and Namibia: relationship to climate and coastal/saline environments, Oecologia 74 (1987) 236–246.

[31] H.P. Schwarcz, T.L. Dupra, S.I. Fairgrieve, ^{15}N enrichment in the Sahara: In search of a global relationship, J. Archaeol. Sci. 26 (1999) 629–636.

[32] J.C. Vogel, B. Eglinton, J.M. Auret, Isotope fingerprints in elephant bone and ivory, Nature 346 (1990) 747–748.

[33] N.J. van der Merwe, J.A. Lee-Thorp, J.F. Thackeray, A. Hall-Martin, F.J. Kruger, H. Coetzee, R.H.V. Bell, M. Lindeque, Source-area determination of elephant ivory by isotopic analysis, Nature 346 (1990) 744–746.

[34] P.L. Koch, J. Heisinger, C. Moss, R.W. Carlson, M.L. Fogel, A.K. Behrensmeyer, Isotopic tracking of change in diet and habitat use in African elephants, Science 267 (1995) 1340–1343.

[35] H. Bocherens, M. Patou-Mathis, D. Bonjean, M. Otte, A. Mariotti, Paleobiological implications of the stable isotopic signatures (^{13}C, ^{15}N) of fossil mammal collagen in Scladina Cave (Sclayn, Belgium), Quat. Res. (1997) 370–380.

[36] H. Bocherens, M. Fizet, A. Mariotti, R.A. Gangloff, J.A. Burns, Contribution of isotopic biogeochemistry (^{13}C, ^{15}N, ^{18}O) to the paleoecology of mammoths (*Mammuthus primigenius*), Hist. Biol. 7 (1994) 187–202.

[37] H. Bocherens, G. Pacaud, P.A. Lazarev, A. Mariotti, Stable isotope abundances ^{13}C, ^{15}N in collagen and soft tissues from Pleistocene mammals from Yakutia: implications for the palaeobiology of the Mammoth Steppe, Palaeogeogr. Palaeoclimatol. Palaeoecol. 126 (1996) 31–44.