Carbon and Nitrogen Isotope Analysis of Human Bone Collagen from the Late Roman and Anglo-Saxon Cemetery of Wasperton, Warwickshire

by Gundula Müldner

Department of Archaeology, University of Reading, Whiteknights, P.O. Box 227, Reading RG6 6AB

g.h.mueldner@reading.ac.uk

Summary

This report presents the results of carbon and nitrogen stable isotope analysis of human bone collagen from the Late Roman to Anglo-Saxon cemetery of Wasperton in Warwickshire. Preservation of the material was very poor. A total of 27 samples from 22 individuals were processed, of these, 14 yielded a product for analysis by stable isotope ratio mass-spectrometry. However, in only seven of these samples, bone collagen was well enough preserved to give meaningful isotope data. These data fall within the range of other Late/Sub-Roman and Anglo-Saxon populations from Britain and suggest a predominantly terrestrial diet in which a large proportion of the dietary protein was derived from animal products (meat and/or dairy, possibly also freshwater resources). While the sample size is too small to comment on possible intra-population variation, the isotope data from Wasperton do not contradict the suggestion of a systematic difference in diet between the Late Roman and Anglo-Saxon periods.

Carbon and Nitrogen Stable Isotope Analysis for Dietary Reconstruction

Carbon and nitrogen stable isotope analysis of bone collagen is currently one of the best-established bone chemistry applications for reconstructing diet in archaeological populations. Isotopes are atoms of the same element with small but measurable differences in their atomic mass. Their abundance varies systematically in nature and also between certain types of food. As the body breaks down food to synthesise and renew body tissues, these differences are preserved, although somewhat altered, in the isotopic composition of consumers' tissues, including their bones. By extracting the collagen, which is the main bone protein and usually the best preserved constituent of archaeological bone, and analysing its isotopic composition it is therefore possible to reconstruct the chief components of the diet of an individual over the time of tissue formation – usually an average of the last ten years or more of a person's life (see Katzenberg, 2000; Sealy, 2001).

Carbon stable isotope ratios ($\delta^{13}C$) are particularly suited to track the consumption of plants of different photosynthetic pathways (C_3 and C_4) through the food web, or, in areas like temperate Europe where C_4-plants never played a significant role in human subsistence, to distinguish between terrestrial (C_3) and marine-based foods. Nitrogen stable isotope ratios ($\delta^{15}N$) increase by 3-5‰ with each trophic level (Bocherens & Drucker, 2003) and are therefore useful to assess the importance of plant versus animal protein in the diet. They can also help detect the consumption of aquatic resources which are usually more enriched in ^{15}N than terrestrial foods (Schoeninger et al., 1983). Because collagen is primarily synthesised from dietary protein, its stable isotope composition the main protein sources rather than diet as a whole (Ambrose & Norr, 1993; Tieszen & Fagre, 1993). Consequently, low protein foods, such as most fruit and vegetables, can be effectively invisible in the collagen isotope signal. Also, since there are no isotopic differences between different types of protein from the same animal, collagen stable isotope analysis cannot distinguish between the consumption of herbivore meat and dairy products (O'Connell *et al.*, 2001).

Previous Applications to Anglo-Saxon and Sub-Roman Populations

Stable isotope analyses for dietary reconstruction have been successfully applied to various archaeological populations from Britain (e.g. Richards *et al.*, 2000; Richards *et al.*, 1998; Richards *et al.*, 2003; Jay & Richards, in press). Applications to the Anglo-Saxon period are still relatively scarce. For the early Anglo-Saxon cemetery at Berinsfield (Oxfordshire), Privat *et al.* (Privat *et al.*, 2002) concluded that $\delta^{15}N$ ratios were correlated with wealth and rank categories inferred from the grave-goods. They proposed that low status individuals consumed more freshwater foods or possibly pork, while high status individuals derived most of their animal protein from herbivore meat and/or dairy. Fuller *et al.* (Fuller *et al.*, 2006) demonstrated isotopic variation between males and females in a late/sub-Roman (late 4th to mid-6th century AD) population from Queenford Farm in Oxfordshire and argued for differences in food distribution between the sexes that were based in their different roles in society. Most recently, Müldner & Richards (Müldner & Richards, submitted) observed significant differences between stable isotope data obtained from the Mid-Anglian burial ground of Belle Vue House in York and both Roman and Medieval populations from the same city, suggesting a significant decline in the consumption of marine foods in the Anglo-Saxon period. They proposed that the infrastructure, which regularly supplied foods from the Humber Estuary or the sea in the Roman and Medieval periods, had temporarily broken down.

Wasperton – Sample Selection

Samples were obtained from every adult individual with preserved bone that had been moved to the Department of Archaeology, University of York for skeletal assessment (22 in total). For five of these (Skeleton Nos. 26, 34, 41, 44 and 173) two samples from different anatomical elements were taken in order to increase the chances of success. Compact bone from a long bone shaft or, where these were not available, the cranial vault, was the sample material of choice, although the anatomical element could not always be determined with absolute certainty due to the fragmentation and generally very poor preservation of the human remains. In general, bone preservation was found to be even worse than expected, mostly owing to the adverse burial conditions on the site but aggravated by the fact that the remains were apparently not stored in a dry condition.

Because it became clear early on that collagen extraction would only be successful for few individuals and that the research potential of the assemblage was therefore very limited, it was decided not to expend any more financial resources on the analysis of animal bones to compare with the human data. This seemed most sensible also, because no faunal remains were available from Wasperton itself and analogues from other Anglo-Saxon sites in the Avon Valley would have to have been used instead, adding further uncertainty. In the following discussion of the results, herbivore isotope data from the early Anglo-Saxon cemetery of Berinsfield in Oxfordshire (Privat *et al.*, 2002) are used as the closest available analogue for environmental values at Wasperton.

Sample Preparation

Bone samples of typically 200-300 mg were prepared using the Longin Method (Longin, 1971) as outlined in Richards & Hedges (Richards & Hedges, 1999). Briefly, the bone was cleaned of adhering soil and the outer surfaces were removed with the aid of a drill. The samples were then demineralised over several days in 0.5 M HCl at temperatures around 5°C, after which the acid solution was discarded and the residue rinsed to neutrality with ultrapure de-ionised H_2O. The samples were then placed in sealed tubes in a pH 3 HCl solution and gelatinised for 48 h at 70°C, before the acid-insoluble residues were removed with the aid of a 5-8µm Ezee® filter. The remaining "collagen" was freeze-dried and weighed into tin capsules for isotopic analysis. $\delta^{13}C$ and $\delta^{15}N$ ratios were determined by continuous-flow isotope ratio mass spectrometry (CF-IRMS) using a Sercon elemental analyser coupled to a Europa 20-20 mass spectrometer in the School of Human and Environmental Sciences, University of Reading. Analytical error, calculated from repeat measurements of internal laboratory standards of known isotopic composition, was ±0.2‰ for both elements.

Results and Discussion

Collagen Preservation

Isotopic data and collagen quality indicators for the human bone samples from Wasperton are displayed in Table 1. As remarked before, sample preservation was rather poor. Of 27 samples, 14 yielded an extraction product for analysis by CF-IRMS, however, established criteria for the assessment of collagen preservation (see van Klinken, 1999) indicate that only seven of these were well enough preserved to give reliable palaeodietary data.

Name	Anatomical Element	$\delta^{13}C$	$\delta^{15}N$	%C	%N	C/N	%Coll.
WASP-1	**?femur**	**-19.8**	**10.5**	**28.0**	**9.8**	**3.3**	**1.1**
WASP-3	?long bone						Nil
WASP-24	**?long bone**	**-20.6**	**10.0**	**38.4**	**12.8**	**3.5**	**9.0**
WASP-26A	long bone						Nil
WASP-26B	cranial vault						Nil
WASP-27	**temporal bone**	**-19.5**	**12.3**	**42.7**	**15.5**	**3.2**	**17.6**
WASP-28	cranial vault						Nil
WASP-30	?long bone						Nil
WASP-34A	tibia						Nil
WASP-34B	long bone						Nil
WASP-38	cranial vault						Nil
WASP-41A	cranial vault						Nil
WASP-41B	?femur						Nil
WASP-42	?long bone	-25.6	3.1	2.1	0.2	9.9	1.3
WASP-44A	?long bone	-25.7	2.4	2.1	0.2	10.4	0.9
WASP-44B	compact bone						Nil
WASP-46	?tibia						Nil
WASP-54	long bone	-20.5	11.9	34.0	11.1	3.6	0.9
WASP-55	**?compact bone**	**-20.4**	**11.6**	**40.0**	**13.9**	**3.4**	**6.2**
WASP-138	tibia	-25.8	-5.7	2.1	0.2	11.0	1.4
WASP-166	cranial vault	-22.8	10.9	6.2	1.5	4.9	2.5
WASP-169	**femur**	**-20.2**	**10.9**	**31.0**	**10.3**	**3.5**	**1.8**
WASP-173A	cranial vault	-21.2	11.7	28.4	8.7	3.8	1.2
WASP-173B	cranial vault						Nil
WASP-174	**cranial vault**	**-20.5**	**10.8**	**12.5**	**4.2**	**3.5**	**2.4**
WASP-190	cranial vault	-21.2	12.7	33.4	8.9	4.4	0.4
WASP-WNJ3*	**?rib**	**-19.8**	**12.2**	**39.7**	**13.8**	**3.3**	**5.3**

Table 1. Carbon and nitrogen stable isotope data and collagen quality indicators for human samples from Wasperton. Samples that yielded adequately preserved collagen for meaning isotopic data are highlighted in bold.
* Individual WASP-WNJ3 was available at the time of sampling but could since not be identified via the site records. It is possible that is does not belong to the Late-Roman/Anglo-Saxon assemblage.

Interpretation of the Dietary Signal

$\delta^{13}C$ ratios of these seven samples range from -20.6‰ to -19.5‰, with a mean of -20.1 ±0.4‰ (1σ). $\delta^{15}N$ ratios lie between 10.0‰ and 12.3‰ with a mean of 11.2 ±0.9‰. As Figure 1 illustrates, the Wasperton humans are therefore, on average, about 1‰ more enriched in ^{15}N than individuals from the Anglo-Saxon sites of Berinsfield (Privat *et al.*, 2002) and Belle Vue House (Müldner & Richards, submitted). While this may indicate a greater reliance on animal products at Wasperton, the great differences in preservation and sample size between the sites and the lack of environmental background data from Wasperton itself, make it impossible to confidently relate these differences to dietary variation between the three groups: As shown in Figure 2, all humans from Wasperton plot within the same range of individuals from the other two sites.

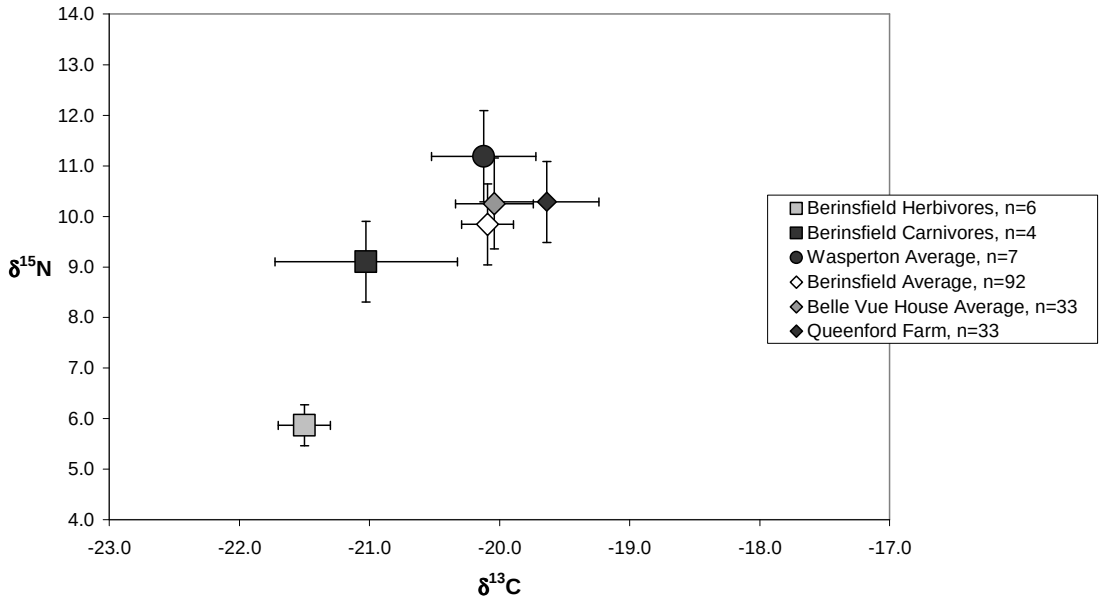

Figure 1. Mean carbon and nitrogen isotope values for humans from Wasperton, Berinsfield (Privat *et al.*, 2002), Queenford Farm (Fuller *et al.*, 2006) and Belle Vue House (Müldner & Richards, submitted) in comparison with animal bone data for herbivores and carnivores from Berinsfield. Error bars indicate ± 1σ.

While the $\delta^{13}C$ ratios indicate that the diet of the Wasperton humans was very predominantly terrestrial in origin (but see below), the $\delta^{15}N$ data suggest that they derived a large proportion of their dietary protein from animal products such as meat and/or dairy. Using herbivore isotope data from early Anglo-Saxon Berinsfield in Oxfordshire as an analogue for environmental values at Wasperton, the relatively large difference between mean herbivore and human $\delta^{15}N$ (5.3‰) could also suggest smaller contributions from omnivore meat (pork) or freshwater resources to the diet (see Privat *et al.*, 2002). However, as the complexity of

nitrogen isotope ecology and metabolism is becoming increasingly apparent, the interpretation of unusually high $\delta^{15}N$ ratios in humans is currently a subject of considerable debate in palaeodietary studies (see Müldner & Richards, submitted for more detailed discussion). Alternative factors such as elevated plant $\delta^{15}N$ due to manuring (Bogaard *et al.*, in press) and a possible variation in diet-tissue spacing dependent on the amount of protein in the diet (Sponheimer *et al.*, 2003), may have to be taken into account.

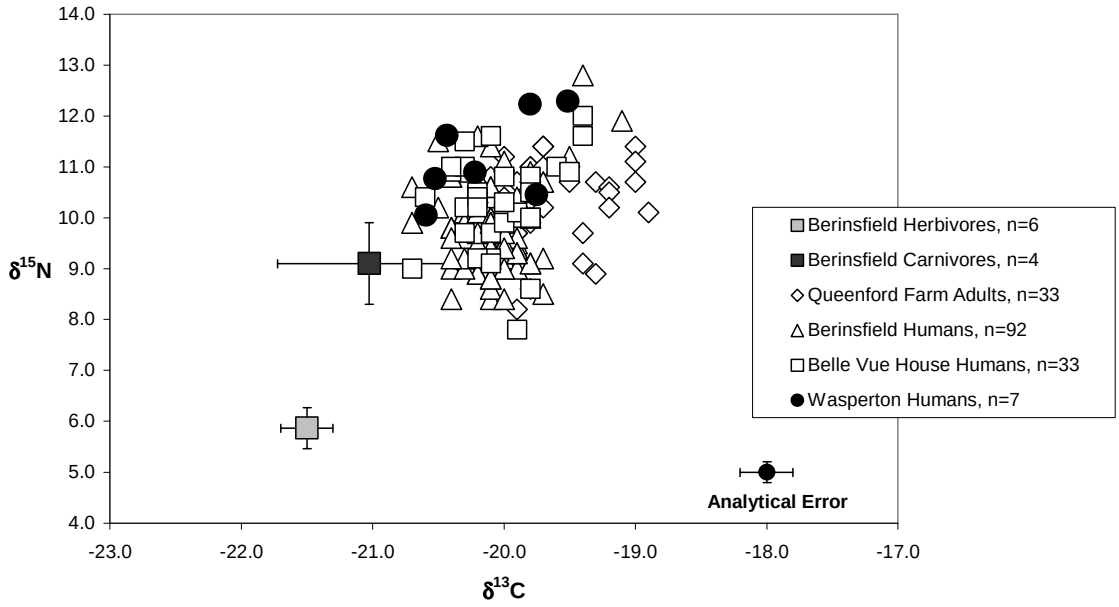

Figure 2. Carbon and nitrogen stable isotope data for individuals from Wasperton in comparison to humans from Berinsfield (Privat *et al.*, 2002), Queenford Farm (Fuller *et al.*, 2006)and Belle Vue House (Müldner & Richards, submitted). Also shown are mean values for herbivores and carnivores from Berinsfield (±1σ).

Intra-Population Variation

The small number of successful samples from Wasperton does not really allow for commenting on possible intra-population differences in diet and the following comments must therefore be regarded with extreme caution.

Sex could be assessed for only three of the individuals (WASP-24, WASP-27, WASP-55), while for five of them age-at-death was estimated (WASP-1, WASP-24, WASP-27, WASP-55, WASP-174). Neither of these biological categories appears to be correlated with diet as reflected in the stable isotope data.

A plot of isotopic values against chronological phases (Figure 3) reveals that the two Late Roman burials (WASP-1, WASP-27) exhibit slightly higher $\delta^{13}C$ ratios (< -20.0‰) than the rest, which fit in well with the data available from other Late/Sub-Roman sites in England: Poundbury in Dorset (Richards *et al.*, 1998), Queenford Farm in Oxfordshire and the city of

York (Müldner & Richards, submitted). Environmental records indicate climatic change to cooler and wetter conditions between the Late Roman and the Anglo-Saxon period (Dark, 2000), and as biosphere $\delta^{13}C$ ratios are linked to both temperature and humidity (see Heaton, 1999), these values could theoretically be explained by environmental rather than dietary change. However, no trend towards more negative $\delta^{13}C$ ratios can be observed in animal bone isotope data from Roman and Anglo-Saxon York (the only one of these sites for which complementary faunal data are available). For the case of York at least, the elevated $\delta^{13}C$ ratios of the Mid- to Late Roman population from Trentholme Drive were therefore attributed to a small marine component in the diet of most individuals (Müldner & Richards, submitted). This interpretation is based on a good faunal base-line and a large human sample and cannot simply be transferred to the case of Wasperton (or indeed Queenford Farm and Poundbury which also lack animal bone data), especially since the identification of minor (≤ 20% of dietary protein) consumption of marine protein by stable isotope analysis is notoriously difficult (see Hedges, 2004; Richards & Schulting, 2006). Nevertheless, it is interesting to note that the data from these Southern English sites (including Wasperton) do not contradict the suggestion of a systematic difference in diet between the Late Roman and Anglo-Saxon periods, nor the possibility that food was sourced from further afield while the infrastructure of the Roman Empire still existed (see Müldner & Richards, submitted).

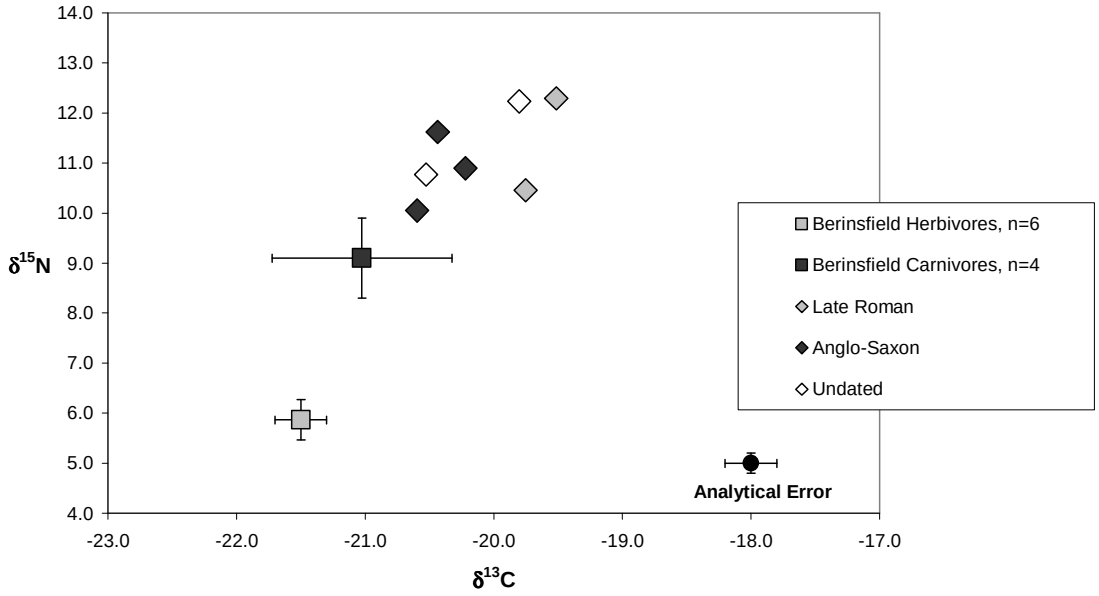

Figure 3. Carbon and nitrogen stable isotope data for humans from Wasperton dated to the Late Roman and Anglo-Saxon periods. Also shown are mean values for herbivores and carnivores from Berinsfield (± 1σ) (Privat *et al.*, 2002).

Correlation with Oxygen and Strontium Isotope Data

Comparison was also made between the carbon and nitrogen stable isotope data and the results of the oxygen and strontium isotope analyses (Montgomery, Evans & Chenery, separate report). Due to differential preservation of the skeletal remains, there are only 5 individuals for which measurements of all four elements are available (WASP-1; WASP-24, WASP-27, WASP-55, WASP-174). Of these WASP-24 and WASP-27 belong to the group that can be tentatively regarded as "local" based on their strontium and oxygen isotope signatures, while the others fall outside of the "local" range, either on grounds of their tooth enamel $^{87}Sr/^{86}Sr$ ratio alone (WASP-1, WASP-55) or because both strontium and oxygen isotope signature are not consistent with "local" values (WASP-174).

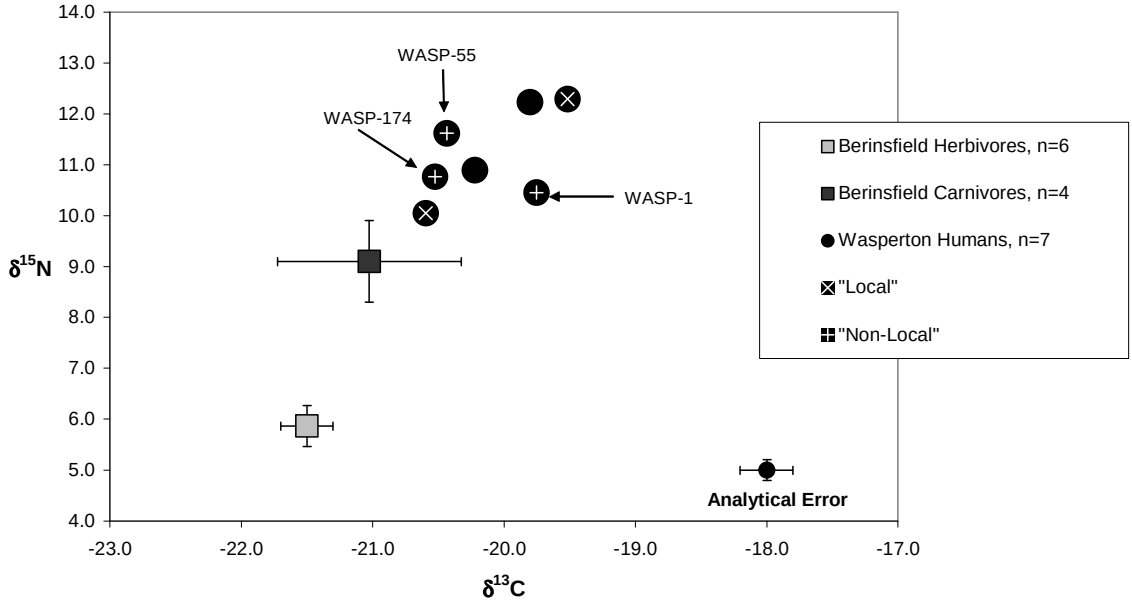

Fig. 4. Carbon and nitrogen stable isotope ratios for Wasperton humans shown in relation to the results of the strontium and oxygen isotope analyses (Montgomery, Evans & Chenery, separate report). Further explanations, see text.

As is apparent from Figure 4, the results of the dietary isotope analyses bear no meaningful correlation with the oxygen and strontium isotope data: WASP-24 and WASP-27, both characterised as "local", exhibit between them the largest differences in $\delta^{13}C$ and $\delta^{15}N$ ratios observed between any two individuals in the group. If diet at Wasperton indeed changed between the Late Roman and Anglo-Saxon period (see above), the difference may of course be explained by the fact that the two skeletons are from different chronological phases (WASP-24 is dated to Phase 2a, WASP-27 to Phase 1a).

WASP-1, characterised by more radiogenic $^{87}Sr/^{86}Sr$ than the "local" group, also exhibits a slightly elevated $\delta^{13}C$ ratio, which could be taken to suggest that the individual consumed terrestrial foods from a region warmer than the UK (see Richards *et al.*, 1998). However, this interpretation is not supported by the $\delta^{18}O$ data which is consistent with English drinking water and in the same range as the majority of the other samples (see Montgomery, Evans & Chenery, separate report). The opposite is true for WASP-174, whose elevated tooth enamel $\delta^{18}O$ value suggests a childhood in a warmer climate, while the bone collagen $\delta^{13}C$ ratio is the most negative but one of all samples from Wasperton. At last, WASP-55, who displays less radiogenic $^{87}Sr/^{86}Sr$ than the "local" group, also does not stand out in any way by their carbon and nitrogen data.

The lack of correlation between "dietary isotopes" and those indicative of residential mobility may not be so surprising when one considers that, although all of them are ultimately derived from the food and drink ingested by an individual, they are tracking very different things: For one, tooth enamel, the material of choice for strontium and oxygen isotope analyses, is laid down in early childhood and not remodelled later on, while bone collagen reflects an average of diet over the last 10 years or more of an individual's life (Sealy *et al.*, 1995). In addition, strontium in consumer tissues is predominantly derived from plants (Burton & Wright, 1995) while carbon in bone collagen is biased towards protein-rich foods (i.e. mostly animal products) (Ambrose & Norr, 1993; Tieszen & Fagre, 1993). If - as tentatively suggested for Wasperton in analogy with other English sites – the carbon and nitrogen isotope data indeed reflect dietary differences between the Late Roman and Anglo-Saxon period, these could have further served obscure potential links between the results of the stable and radiogenic isotope analyses.

Conclusions

Poor bone preservation has unfortunately greatly limited the analytical potential of the Wasperton assemblage. The small data-set that could be obtained shows great similarities with other Late/Sub-Roman and Anglo-Saxon stable isotope data available from Britain and could be tentatively interpreted as contributing to an emerging picture that Anglo-Saxon subsistence was much more reliant on terrestrial and possibly local resources than in the preceding Roman and subsequent Medieval periods (see Müldner & Richards, submitted).

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