

RADIOCARBON DATING BONE SAMPLES RECOVERED FROM GRAVEL SITES

SCIENTIFIC DATING REPORT

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HAD 84 1747 12569

Research Department Report Series 30/2007

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ISSN 1749-8775

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Summary

A range of pre-screening criteria which could be used on-site, in museums, or the laboratory were tested to determine which, if any, could be used to screen those suitable for radiocarbon dating prior to the time-consuming and costly collagen extraction pre-treatment chemistry.

Nearly 300 bones were analysed from 12 sites across southern England. The most successful screening method applied was the measurement of whole bone percent nitrogen. This method was consistently the most reliable predictor of suitability for radiocarbon dating. No other pre-screening criterion, or combination or criteria, showed a correlation with collagen preservation, either within the entire study or datasets from individual archaeological sites.

Keywords

Radiocarbon Dating
Animal Bone

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Introduction

Radiocarbon dating provides the backbone for most archaeological chronologies in the UK, particularly for the prehistoric period. The gravel terraces of our river valleys produce significant archaeological remains which often need to be dated. Bones, particularly those which articulate, are crucial to these chronologies as they provide substantial samples which relate directly to the context from which they were recovered.

However, many bones recovered from gravel sites in the UK have undergone severe chemical and physical degradation and subsequent microbial attack, resulting in greatly increased porosity and loss of up to 90% of the original collagen content. Such bones, therefore, have a relatively small possibility of containing sufficient collagen for reliable radiocarbon dating (ie more than 1% of the pristine amount) compared to bones from other contexts.

Radiocarbon pre-treatment chemistry is costly and time-consuming and so it is often not feasible to attempt to date bones, especially in large numbers, from gravel sites. However, sites which predominantly yield bone which contain insufficient surviving collagen to date do include a small fraction of bones in which collagen is better retained. The main aim of this project was to determine which, if any, analytical criteria could best be applied to gravel-deposited samples, and those from other sites known to yield poor bone preservation, either prior to their submission to the laboratory or during an initial screening process at the laboratory, to assess which bones might be successfully dated. Identification of any suitable diagnostic tests would make the dating of such sites more successful from a scientific standpoint, as well as faster and less costly.

Methods and Sampling

Samples of 298 human and animal bones (including *Sus scrofa*, *Bos primigenius*, *Bos taurus*, *Ovis aries*) were selected from 12 sites across southern England (Table 1). These were chosen to represent a range of contexts where bone preservation is thought to be poor or variable. A wide selection of bones, including femura, humeri, metacarpals, vertebrae, mandibula and scapulae, as well as antler, were sampled across a range of ages, from Neolithic to early medieval.

All of the bones were subjected to an initial screening process, which was designed to be used by archaeologists on-site or in museums. The criteria measured were:

- hardness
- colour (HSB: hue, saturation, brightness)
- dry bone bulk powder density
- microporosity.

Data from these bones was considered for scatter and range in the various measures. A total of 140 sub-samples were randomly selected and a further 60 selected judgements to cover specific cases missed in the random selection. These 200 bones were subjected to further diagnostic tests which could be carried out in the laboratory prior to pre-treatment for radiocarbon dating. These were:

- Fourier Transform Infra Red spectroscopy (FTIR)
- percent nitrogen and carbon analysis
- C:N atomic ratio of whole bone

The data produced for these bones was evaluated and half of the randomised samples (70) were selected again at random for full radiocarbon pre-treatment (using methods described

in Bronk Ramsey *et al.*, 2004). An additional 30 samples were chosen specifically to include those with as wide a range of values for the initial analyses as possible, although samples with very low nitrogen (<0.5%) were usually avoided as they were assumed to be unlikely to yield sufficient collagen for radiocarbon dating. The percentage yield of collagen, insoluble residue, and <30kD residue were calculated for each sample, and the stable isotopic values ($\delta^{13}\text{C}$, $\delta^{15}\text{N}$) and C:N atomic ratio of the purified collagen measured.

On the basis of the results observed throughout the study, 20 samples were selected for amino acid analysis, and 18 samples for small-angle X-ray scattering (SAXS) to represent samples with varying preservation states and from a range of sites.

Results

The results were studied to identify if any one, or any combination of, the diagnostic tests could be used to identify bones suitable for radiocarbon dating, and if any site specific trends could be observed. Details of the methods employed are provided in Appendix I; the data are provided in Appendices II and III.

Hardness: Hardness was measured using a modified Shore pencil durometer; the lower the value, the harder the bone. The hardness measurements ranged from 5 to 72 (arbitrary units) but failed to show any correlation with collagen preservation (Fig 1a). Some bones had very soft outer layers, and hence very high hardness values, but were actually much harder and well preserved under the surface. Other bones yielding low hardness values were simply brittle and poorly preserved. Several bones shattered on testing.

Colour: Neither hue, saturation, brightness, nor any combination of the three characteristics provided any indication of suitability for radiocarbon dating (Fig 1b). Bones tended to fall into several categories according to their colour following surface cleaning: those that became much darker (eg Fig 2a); those that were much lighter in colour beneath the surface (eg Fig 2b); and those that were unchanged (eg Figs 2c,d). Occasionally bones appeared to have several differently coloured layers beneath their surface. None of these observations provided any indication of collagen preservation.

Microporosity: Microporosity is defined as the volume of water uptake per gram of dried bone up to a relative humidity of 76%, which corresponds to all pores up to the theoretical diameter of 4nm being full of water. This has been suggested to be an indicator of bone diagenesis, as loss of protein has been observed to be reflected by both a loss in microporosity and an increase in crystallinity (Hedges *et al* 1995). Microporosity values in this study ranged from 0.07–0.19 cm^3g^{-1} , higher than the values of 0.025–0.105 cm^3g^{-1} reported from three archaeological sites by Hedges *et al.* (1995). This may be due to the small sample size used here, and by occasional inconsistencies in bone powder size (caused either by the use of different drill bits by different samplers or by fragmenting of the bone on sampling), but no correlation was observed between microporosity and the amount of collagen preserved in this study (Fig 1c).

Bulk powder density: These results may have been influenced by the same sample size issues as the microporosity samples, but again, no correlation between collagen preservation and bulk bone powder density was observed (Fig 1d). It should be noted that the bulk powder density values listed in this study of 0.24–0.52 gcm^{-3} are relative to each other, but that they may not be directly comparable to those in other published studies which analysed density of intact samples of bone. For example, Turner-Walker and Parry (1995) state that modern bone has a bulk density of $\sim 2.0 \text{ gcm}^{-3}$, and poorly preserved archaeological bone a bulk density of $\sim 0.8 \text{ gcm}^{-3}$.

FTIR: Modern bone normally contains 60–70% by weight of carbonated apatite crystals with a structure and composition similar to the mineral dahllite (Stiner *et al* 1995; Wright and Schwarz 1996). Infrared spectra of whole bone powder can provide both information on the crystallinity of the carbonate apatite crystals and a semi-quantitative estimate of the relative carbonate content of the mineral phase.

The crystallinity index or “splitting factor” (SF) of bone is calculated from the extent of splitting of the phosphate absorption peaks at $\sim 603\text{cm}^{-1}$ and 565cm^{-1} , as defined by Termine and Posner (1966) and Weiner and Bar-Yosef (1990). This represents a combination of both the relative sizes of the crystals and the extent to which the atoms are ordered within the lattice (Weiner and Bar-Yosef 1990).

During diagenesis, large crystals tend to grow at the expense of smaller ones, either rapidly over a period of a few months or years via weathering, or over many millennia during normal fossilization (Stiner *et al* 1995). Selective dissolution of more soluble, less ordered crystals may also occur in archaeological bones (Wright and Schwarz 1996). As recrystallization takes place, the two phosphate peaks become increasingly separated from each other, and this is reflected in the splitting factor: the higher the SF, the larger and/or the more ordered the crystals (Weiner and Bar-Yosef 1990).

Splitting factors of 2.5–2.9 have been recorded for fresh bones (Weiner and Bar-Yosef 1990; Stiner *et al* 1995; Sillen and Parkington 1996), and values of ~ 7 have been observed for highly fossilised or calcined bones (Weiner and Bar-Yosef 1990). All samples in this study had splitting factors of 3.6 and above, indicating that they were degraded to varying extents. There was no correlation in this study between splitting factor and percent collagen yield (Fig 3a).

The splitting factor is measured at the end of the spectrum, between ~ 500 and 750cm^{-1} and, in some cases, the spectrum was very poor and noisy, especially in the cases of very degraded bones. Despite baseline correction, this may have resulted in a slight elevation of the SF but not sufficiently to affect any correlation with collagen preservation. It should also be noted that SF has been observed to vary greatly throughout individual bones (Stiner *et al* 1995).

Carbonate content can be estimated from the ratio of the CO_3 and PO_4 absorbances at 1417cm^{-1} and 1035cm^{-1} respectively (Wright and Schwarz 1996). Loss of carbonate due to post-depositional recrystallization and hydroxyapatite alterations and/or burning results in lowering of the C:P ratio (Stiner *et al* 1995). The lower the C:P ratio, the more crystalline the bone.

Natural biogenic carbonate in bone has a C:P ratio of ~ 0.360 (Nielsen-Marsh, 1997 unpubl). In this study, C:P values ranged from 0.20 to 0.31, with the exception of 4 samples. These samples, 44 (from Eynesbury), 94, 97 (both from the Haddenham causewayed enclosure), and 175 (Etton causewayed enclosure) had C:P ratios of 0.61, 0.43, 0.34 and 0.33 respectively. Calcite peaks were observed at $\sim 710\text{cm}^{-1}$ in the FTIR spectra of all of these samples (but not in any other spectra) and may suggest growth of diagenetic calcite within the bone, especially for sample 44 (Eynesbury) The presence of calcite was reflected in the $\delta^{13}\text{C}$ of the whole bone, particularly for sample 44 which has the largest calcite peak and the most enriched $\delta^{13}\text{C}$ value of -11.7‰ (the majority of samples have $\delta^{13}\text{C}$ values which lie in the range -15 to -23‰). None of these samples were submitted for full radiocarbon pre-treatment, but they would all be expected to yield little collagen as none contained more than 0.2% nitrogen. All 5 samples with C:P ratios of 0.30 which were subjected to full pre-treatment yielded $>1\%$ collagen, but otherwise there was no correlation between C:P ratio and collagen preservation (Fig 3b).

Several studies have demonstrated that the relationship between C:P ratios and SF has reasonably good correlation with other pre-screening techniques (eg Petchey and Higham 2000). Sillen and Parkington (1996) also identified a very good correlation between SF and percent nitrogen in archaeological bones from one site in South Africa. In this study, there appears to be some correlation between samples with high SF (greater than ~ 4.8) and low C:P ratios (less than ~ 0.20) and preservation of collagen (Fig. 3c), but the relationship is not strong enough to use as a diagnostic test. Otherwise, no correlation within either the entire dataset or those of individual archaeological sites was found between C:P ratios and SF, or between either C:P or SF and any other pre-screening criteria.

Percent carbon and nitrogen analysis: Modern whole bone contains ~14% carbon (%C) (Sillen & Parkington, 1996). In this study, values ranged from 1.3 to 12.0% and all samples with 7.2% C and above yielded > 1% weight collagen and were therefore suitable for radiocarbon dating (Fig. 4a). However, there was no significant correlation between % weight collagen and %C, as the technique cannot distinguish between carbon present in collagen or contaminant.

Percent nitrogen (%N) values for whole bone provide an indication of protein survival as nitrogen in bone is derived solely from the proteinaceous component. %N can be used to indicate whether or not there is sufficient collagen in the bone for radiocarbon dating to be successful. The value will not, however, identify whether the nitrogen is present as collagen or as contaminants, nor will it specify the amount of non-nitrogenous soil-derived organic matter present (Hedges and van Klinken 1992).

Fresh bone contains ~ 4% nitrogen (Stafford *et al* 1988; Ambrose 1993). In this study, the maximum nitrogen detected was 3.7% (sample 210, Cleveland Farm). All samples with more than 2% nitrogen which underwent pre-treatment yielded sufficient collagen for AMS radiocarbon dating (2.4–6.5% wt collagen) but the majority of samples contained 1% nitrogen or less (Fig. 4b).

However, of the pre-screening techniques investigated in this study, %N of whole bone was the most reliable tool for identifying samples with sufficient collagen for radiocarbon dating. Linear regression analysis demonstrated that if 0.76% N is chosen as the threshold, 84% of those bones will be correctly identified as dateable or not dateable (Fig. 4b). Where C = % weight collagen and N = % nitrogen of whole bone:

$$C_{\text{pred}} = a + bN$$

(a = -0.02 ± 0.15, b = 1.36 ± 0.12 and R = 0.759).

Inclusion of the %C and/or the C:N atomic ratio of bone in the linear regression increased the prediction success to 85%, which is not significantly better.

C:N atomic ratio of whole bone: The C:N atomic ratio of whole bone can provide an indication of the general state of preservation of the collagen, the extent to which deamination has taken place and/or the extent of contamination by exogenous carbon-containing compounds such as humic acids (Tisnérat-Laborde *et al* 2003). A C:N ratio of greater than 5 demonstrates extensive diagenesis and/or the presence of a high proportion of humics (Tisnérat-Laborde *et al* 2003).

Samples within this study produced C:N atomic ratios of 3.6–98.1, although only 16% of the 200 samples analysed had ratios less than 5. In general, this study suggests that bones with C:N ratio of greater than ~6.5–7.0 are not suitable for radiocarbon dating, but one sample (number 126, Berinsfield) yielded 1.1% wt collagen on pre-treatment with a C:N ratio of 8.7 (Fig. 4c).

Full radiocarbon pre-treatment: Modern fresh bone typically contains ~ 22% wt collagen (van Klinken 1999). At the Oxford Radiocarbon Accelerator Unit, samples yielding less than 1% wt collagen after full pre-treatment are failed and not submitted for AMS dating. Of the 100 samples which underwent full radiocarbon pre-treatment, only 45 yielded greater than 1% wt collagen and were therefore deemed suitable for dating (Table 2).

The bones from Berinsfield generally demonstrated good preservation, as did those from Holloway Lane and Bestwall Quarry. It should be noted that the differing yields of collagen between samples from Berinsfield recorded in this study and those published by Privat *et al* (2002) are most likely due to differences in pre-treatment procedures: Privat *et al* used only an acid wash prior to gelatinization, and did not include the base wash and ultrafiltration utilized in this study and currently as standard for radiocarbon dating at the Oxford Radiocarbon Accelerator Unit (Bronk Ramsey *et al* 2004).

No samples from either Eynesbury or Haddenham Causewayed Enclosure contained sufficient collagen for dating, but this was unsurprising as all of the 40 samples analysed contained low levels of nitrogen.

C:N ratios of collagen are typically 2.9–3.6 (DeNiro 1985). Values out of this range are indicative of low collagen (Schoeninger *et al* 1989; Ambrose 1990), contamination (DeNiro 1985; Ambrose 1990) or diagenesis (Koch *et al* 1994). At the Oxford Radiocarbon Accelerator Unit, our cut-off for dating is 2.9–3.5.

All but four bones in this study had collagen with a C:N atomic ratio of 3.2 or 3.3, regardless of the amount of collagen preserved. One sample (number 35, Holloway Lane) had a C:N ratio of 2.9 and yielded 1.9% wt collagen, two samples (numbers 178 and 179, both from Etton causewayed enclosure) had C:N ratios of 3.4 and yielded 0.9% and 0.5% wt collagen respectively, and sample 182 (also from Etton) had a ratio of 86.4 due to very low nitrogen and yielded only 0.5% wt collagen.

Stable isotopic values ($\delta^{13}\text{C}$ and $\delta^{15}\text{N}$) of collagen are more likely to be linked to sample-specific variables (eg trophic-level in herbivores or carnivores in $\delta^{15}\text{N}$) and environmental influences (eg $\delta^{13}\text{C}$ canopy effects in forests) than collagen preservation (van Klinken 1999).

The amount of insoluble residue present after radiocarbon pre-treatment ranged from 0 to 42% (Fig. 4d) and varied greatly within archaeological sites. The percentage of material which passed through the ultrafilter during pre-treatment (ie < 30 kDa residue) reached a maximum of 1.6%, although the residue was often very sticky and difficult to weigh. Neither residue correlated with the percent weight collagen yielded for a given sample.

Amino acid analysis: allows the detailed composition of the proteins present to be determined and compared to those of collagen in modern bone (van Klinken 1999). However, for the majority of bones, there is little actual variation in the amino acid profiles themselves, it is only when the bones become very degraded that reduced levels of certain amino acids are present (van Klinken 1999). One of the limitations of amino acid analysis in this study was that there was insufficient collagen for the analysis to be carried out on many of the samples which failed pre-treatment.

Of the 20 collagen samples analysed, five were from bones which had yielded less than 1% wt collagen. It was not easy to distinguish these specific samples from those that were successful in pre-treatment using their amino acid profiles, except for sample number 182 (Etton) for which the pre-treatment product was a brown powder. This sample had reduced levels of hydroxyproline, proline, glycine, and alanine in comparison with the other collagen samples. Sample 280 (RMC Land), which also failed pre-treatment and which appeared brittle and glassy, was only differentiated from the other samples by slightly reduced alanine levels. Of the other samples with low collagen yields, sample 127 (Berinsfield) demonstrated

slightly reduced levels of alanine and aspartic acid, but samples 178 and 179 (Etton) were indistinguishable by their amino acid profiles from the samples with good collagen preservation.

Several studies have attempted to use ratios of various amino acids to categorise collagen preservation (eg Stafford *et al* 1988; DeNiro and Weiner 1988; Weiner and Bar-Yosef 1990). It was nearly impossible to assign any of the 20 bones sampled to the classes of preservation defined by Stafford *et al* (1988) on the basis of their amino acid profiles and whole bone percent nitrogen content. Sample 182 could clearly be assigned to class V (“extremely poorly preserved”), but most other bones could only be attributed to combined classes I–III (“modern”, “very well to well preserved”, and “moderately well preserved” respectively) using these criteria.

Weiner and Bar-Yosef (1990) used Gly/Asp ratios (glycine/aspartic acid ratios) to determine collagen purity: pure collagen has a Gly/Asp ratio of close to 7 (Weiner and Bar-Yosef 1990). They stated that collagen is not present in samples with a Gly/Asp ratio of less than ~ 6. However, in this study, sample 99 (Berinsfield) has a ratio of 5.6 but yielded 2.5% wt collagen with a C:N ratio of 3.2 and $\delta^{13}\text{C}$ of -20.2‰ , suggesting that it was not affected by contamination. Most of the other samples had Gly/Asp ratios of 6.4–7.5, except two with poor collagen preservation, samples 127 (Berinsfield) and 182 (Etton) with ratios of 8.2 and 30.9 respectively.

DeNiro and Weiner (1988) used several amino acid ratios - Asp/Pro (aspartic acid/proline), Asp/Gly (aspartic acid/glycine), and Asp+Thr+Ser+Glu/Pro+Gly+Hop ((aspartic acid + threonine + serine + glutamic acid)/(proline + glycine + hydroxyproline)) - to distinguish between modern, well-preserved, and poorly-preserved prehistoric bones. With the exception of sample 182, there is little variation between these ratios among the samples in this study, and all have ratios which agree with those of DeNiro and Weiner’s modern and well-preserved samples.

Small-angle X-ray scattering (SAXS): This technique allows for the accurate measurement of crystal shape, size, and orientation within bone and has been used to determine the degree to which the bone matrix has recrystallised (Wess *et al* 2001; Hiller *et al* 2004; Hiller and Wess 2006). Hiller *et al* (2004) and Hiller and Wess (2006) used the method to study the structural dimensions of crystallites in bones and demonstrated a link between alteration to crystal structure (in terms of thickness or shape) and other diagenetic changes such as loss of nitrogenous material. In this study, 18 samples were analysed using SAXS, but no correlation between crystal thickness and collagen preservation was observed (Fig 3d).

Interpretation and conclusions

Of the pre-screening criteria tested in this study, only %N (and, to a lesser extent, %C and C:N atomic ratio) of whole bone showed a good correlation with collagen preservation which could be applied to bone specimens to identify those suitable for radiocarbon dating. Linear regression analysis demonstrated that using a 0.76% N cut-off level would allow an 84% prediction rate for whether a bone was dateable or not. This prediction success was increased to 85% when including the %C and/or C:N atomic ratio data in the linear regression.

Percent nitrogen analysis of bone powder is a relatively quick and simple technique and could easily be implemented as a pre-screening technique prior to radiocarbon dating of bone samples from sites known to suffer from poor or variable preservation such as Brandon Staunch Meadow, Etton Causewayed Enclosure, and Kingsmead Quarry. This level of nitrogen is less than the 1% cut-off currently used at the Oxford Radiocarbon Accelerator

Unit to determine whether or not to proceed with pre-treatment of potentially poorly preserved bones.

No correlation between any of the pre-screening criteria and % weight collagen was observed within any of the individual archaeological sites studied.

Acknowledgements

The authors wish to thank everyone who provided material for this study and assisted with the laboratory work: Alex Bayliss, John Meadows, Derek Hamilton, and Wendy Hart (English Heritage); Angela Bowles, Christine Tompkins, Jane Davies, and Barbara Emery (Oxford Radiocarbon Accelerator Unit) for sampling the bones and full radiocarbon pre-treatment; Peter Ditchfield (Oxford Radiocarbon Accelerator Unit) for stable isotope analysis; Tony Willis (Department of Biochemistry, University of Oxford) for amino acid analysis; Colin Johnston and Alison Crossley for permission to use, and assistance with, FTIR at the Department of Materials, University of Oxford and BegbrokeNano (supported by the DTI MNT Capital Programme); Clerk Maxwell and Tim Wess (Cardiff University) for SAXS, and Jen Hiller (Diamond Synchrotron) for analysis of the data.

We are grateful to the following who kindly provided bone specimens for this study: Andrew Tester, Suffolk County Council (Brandon Staunch Meadow); Nick Elsdon, Museum of London Archaeology Service (Holloway Lane); Quinton Carroll, Cambridgeshire County Council (Eynesbury); Chris Evans, Cambridge Archaeological Unit, University of Cambridge (Haddenham); Dr Andrew Chamberlain, University of Sheffield (Berinsfield); Robin Jackson, Worcestershire County Council (Huntsman's Quarry); Lilian Ladle, Bestwall Quarry archaeology project; Dr Frances Healy, Cardiff University (Etton); Lorraine Mepham, Wessex Archaeology (Imperial College Sports Ground, Cleveland Farm, Kingsmead Quarry, RMC Land).

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Table 1. Details of the 12 sites from southern England from which bones were sampled for this study

Site Name	Location	Age of Bones
Brandon Staunch Meadow	Suffolk	Anglo-Saxon, early medieval
Holloway Lane, Harmondsworth	London Borough of Hillingdon	Pre-historic multi-period site, especially late Neolithic-early Bronze Age
Eynesbury	Barford Road, St Neots, Cambs.	Neolithic, from Neolithic to Anglo-Saxon site
Haddenham Causewayed Enclosure	Haddenham, Cambs.	Early Neolithic features, from multi-period site
Wally Corner, Berinsfield	Dorchester-on-Thames, Oxon	Anglo-Saxon cemetery
Huntsman's Quarry	Kemerton, Worcestershire	Late Bronze Age features, from multi-period site
Bestwall Quarry	Wareham, Dorset	Bronze Age, Romano-British, Anglo-Saxon
Etton Causewayed Enclosure	Maxey, nr Peterborough, Cambs.	Early Neolithic
Imperial College Sports Ground, Harlington	London Borough of Hillingdon	Romano-British
Cleveland Farm	Ashton Keynes, Wiltshire	Iron Age or Romano-British
Kingsmead Quarry, Horton	Windsor & Maidenhead, Berks.	Bronze Age
RMC Land, Harlington	London Borough of Hillingdon	Bronze Age, Romano-British, Anglo-Saxon

Table 2. Collagen yields for samples which underwent full radiocarbon pre-treatment

Site	No. bones sampled	No. bones pre-treated	No. bones >1% collagen	% bones >1% collagen
Brandon Staunch Meadow	30	8	4	50
Holloway Lane	12	4	3	75
Eynesbury	18	4	0	0
Haddenham Causewayed Encl.	43	10	0	0
Wally Corner, Berinsfield	31	16	13	81
Huntsman's Quarry	25	7	2	29
Bestwall Quarry	13	9	9	100
Etton Causewayed Encl.	18	7	3	43
Imperial College Sports Ground	20	5	1	20
Cleveland Farm	28	10	4	40
Kingsmead Quarry	30	7	1	14
RMC Land, Harlington	30	13	5	38
Total	298	100	45	45

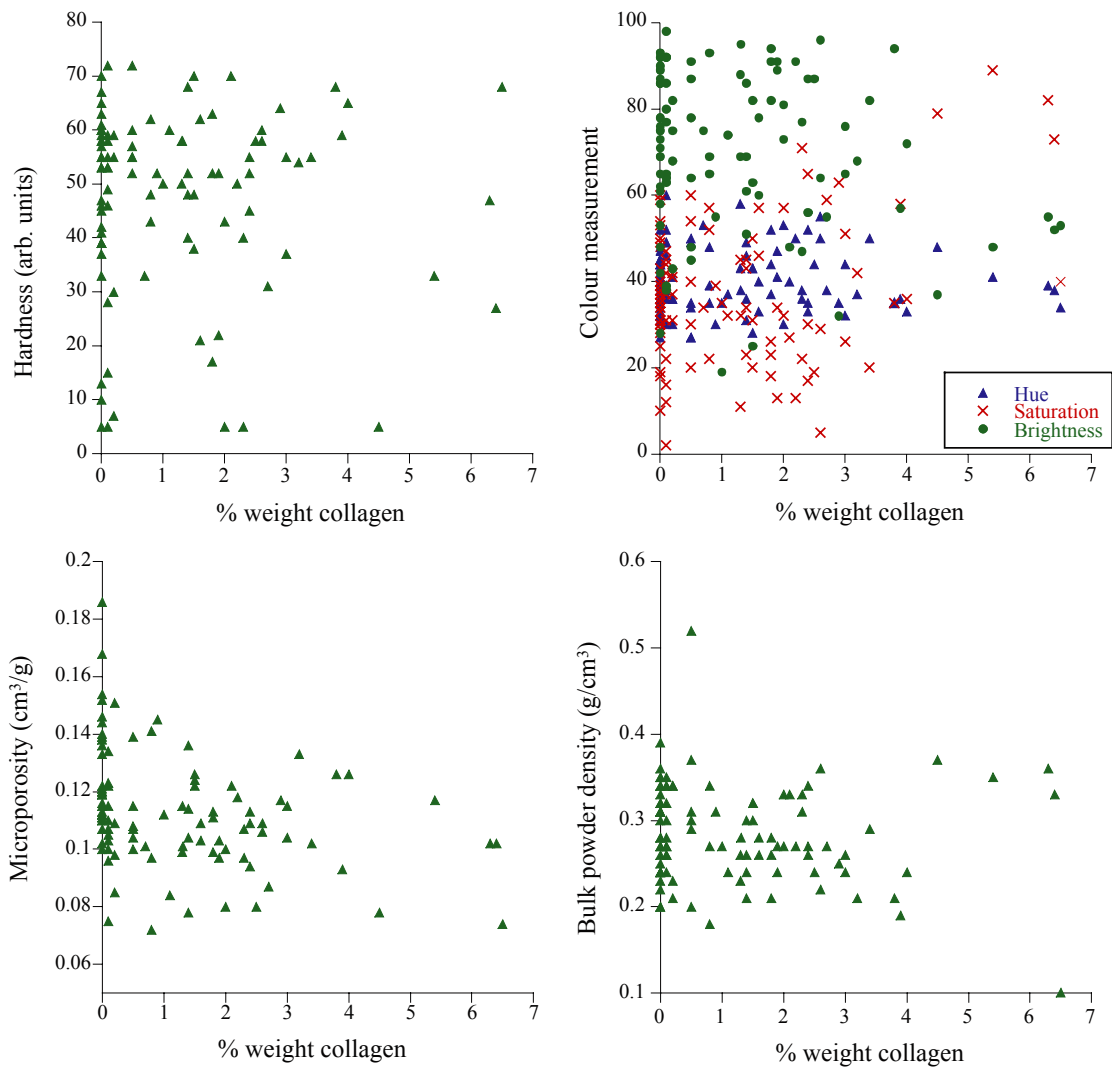


Figure 1. Plots of a) hardness, b) colour measurement (hue, saturation, and brightness), c) microporosity, and d) bulk powder density against % weight collagen

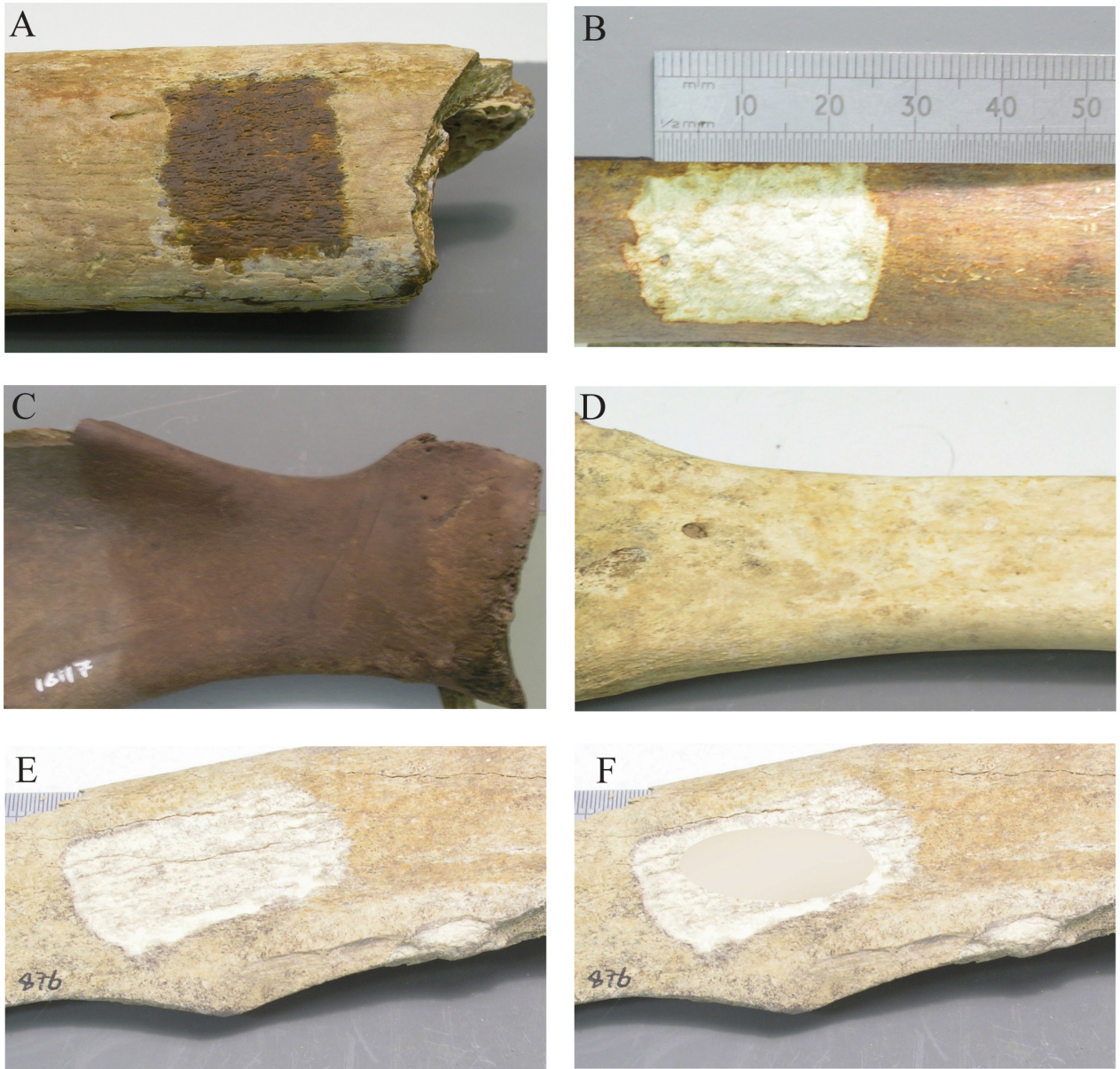


Figure 2. Examples of the variation in appearance of bone and the effect of cleaning the bone surface by shotblasting. Hue (H), saturation (S), and brightness (B) values varied accordingly: A) Sample 60 (Eynesbury) H: 31, S: 37, B: 41; B) Sample 123 (Berinsfield) H: 58, S: 11, B: 91; C) Sample 175 (Etton) H: 25, S: 34, B: 45; D) Sample 282 (RMC Land, Harlington) H: 46, S: 26, B: 90; E) Sample 265 (RMC Land, Harlington) H: 47, S: 8, B: 93; F) Sample 265 showing area of Gaussian blur averaging used to measure HSB

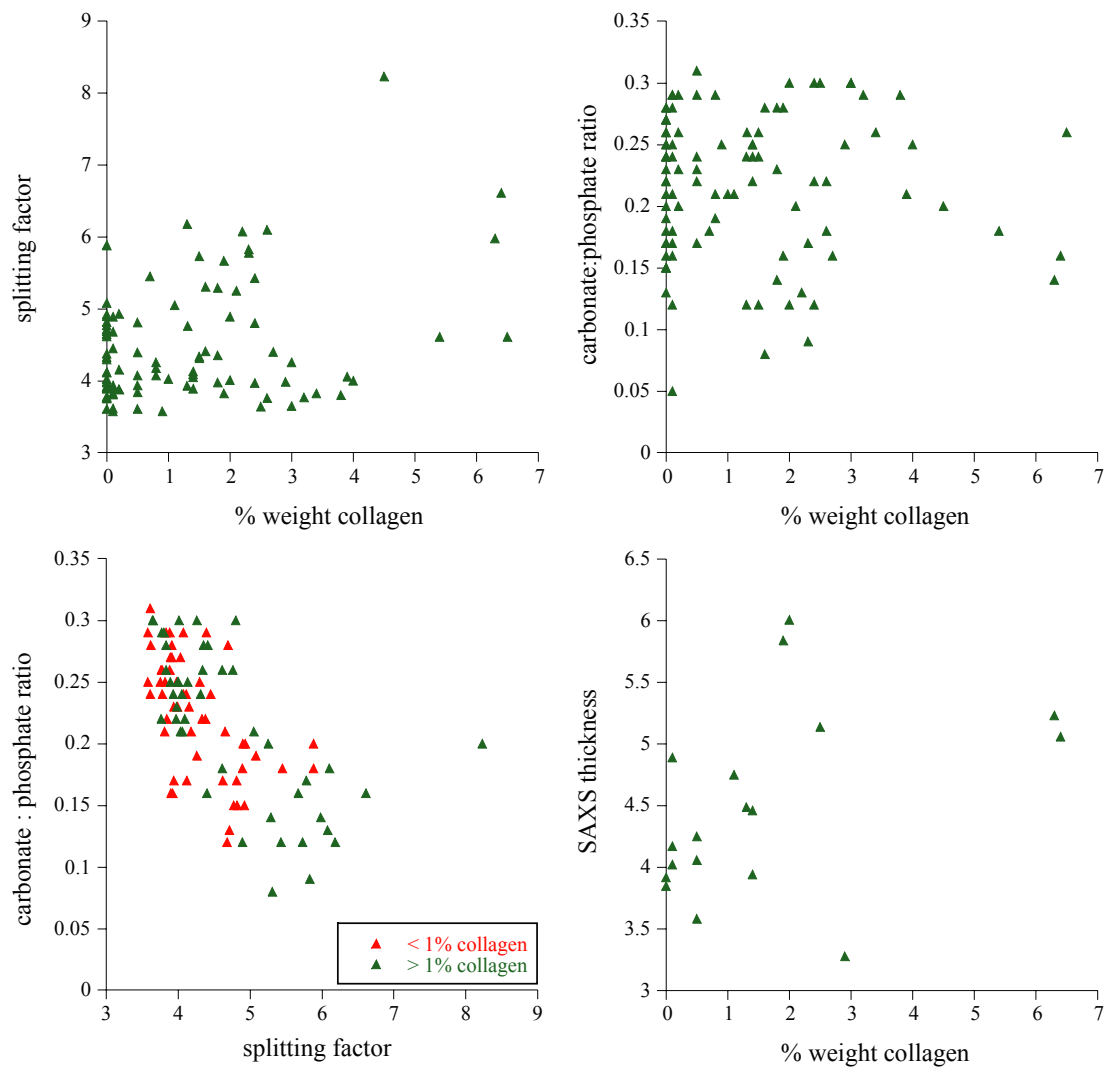


Figure 3. Plots of a) splitting factor (SF) and b) carbonate : phosphate ratio (C:P) against % weight collagen; c) splitting factor vs carbonate : phosphate ratio; d) SAXS crystallinity thickness against % weight thickness

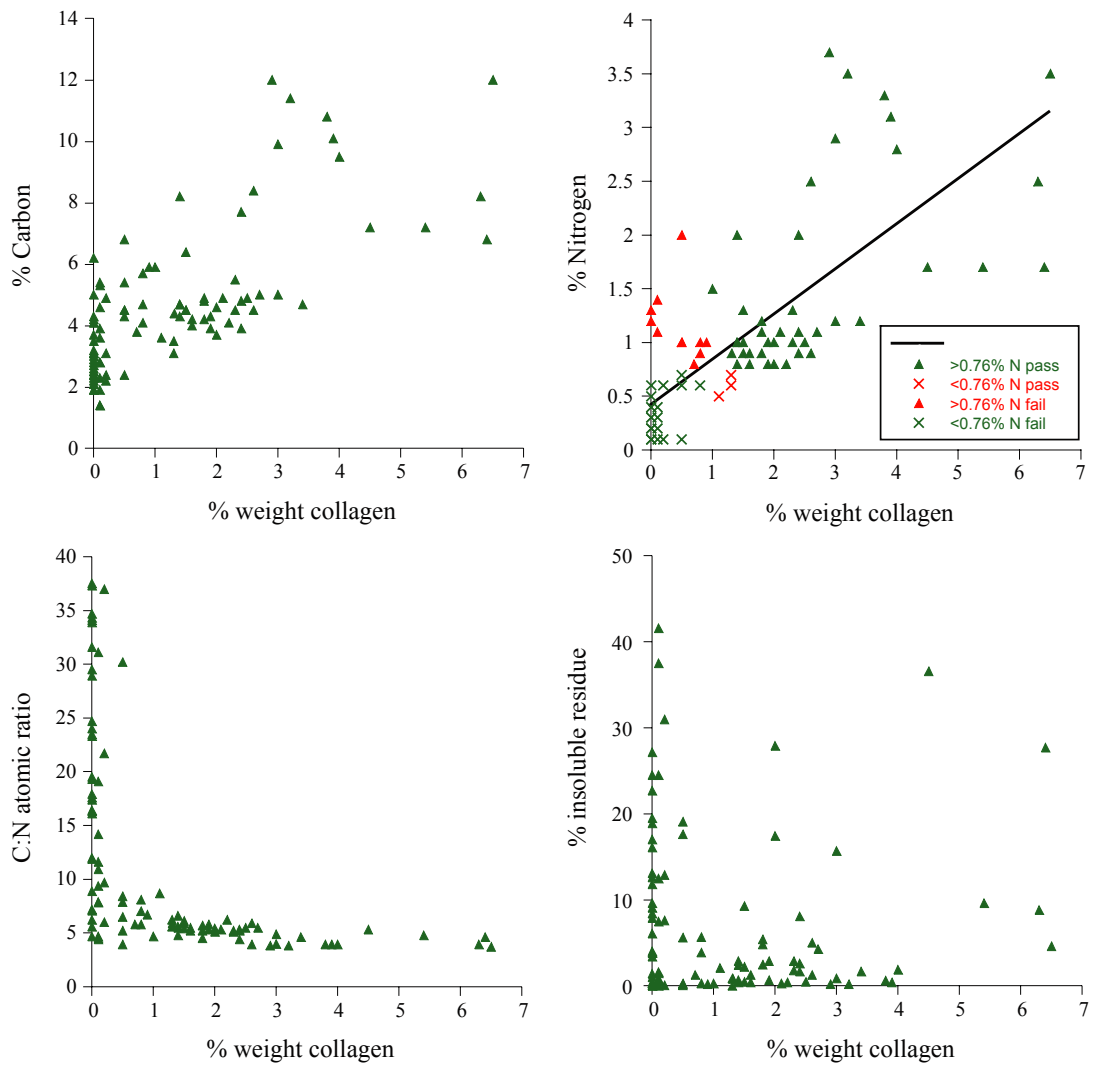


Figure 4. Plots of a) % carbon, b) % nitrogen, showing the line of linear regression and indicating those which “passed” and “failed” radiocarbon treatment (ie those with yields of >1% and <1% weight collagen respectively). Samples which are correctly predicted to pass or fail radiocarbon pre-treatment are in green, and those which are incorrectly predicted in red; c) C:N atomic ratio of whole bone, and d) % insoluble residue against % weight collagen

Appendix I: Methods

Hardness: An Intron Shore Pencil Durometer was used to measure hardness of a bone by determining the depth of penetration of an indenter into the bone. The durometer was modified to require 5kg to fully compress the spring, and the origin blunt indenter was sharpened to a 60° angle. Three closely-spaced measurements were taken from a flat portion of the bone (where possible), and the average of the three values calculated. The units are arbitrary.

Colour: Each bone was shot-blasted with fine aluminium oxide powder (Swan-Blaster, Crystal Mark Inc, Glendale CA, USA; Airbrasive powder No. 1, REG Abrasives Ltd, Dartford, UK) to remove surficial contamination before being digitally photographed against a Kodak grey card background. The image was adjusted for grey-point and HSB (hue, saturation, brightness) were measured over a sample area using Gaussian blur averaging in Adobe Photoshop software.

Sampling: The cleaned section of the bone was drilled using a tungsten carbide spherical burr drill bit (3mm diameter) at a low speed, and ~1g bone powder collected. For consistency the same bit and speed was used where possible within the Oxford Radiocarbon Accelerator Unit, although this was not always possible for bones sampled elsewhere.

Microporosity: 20–30mg bone powder was transferred to a 4mL Wheaton vial of known weight. Samples were oven-dried at 100°C for a minimum of three days prior to weighing. The samples were then transferred to a thermostatically-controlled humidity chamber at 25°C with at 75% relative humidity (RH). Humidity was controlled using sulphuric acid and measured with a hygrometer (Pike 1993 unpubl). Samples were left for 3 days prior to weighing. Each weight measurement was repeated 3 times and an average calculated. Microporosity was calculated as follows:

$$\text{Microporosity} = \frac{75\% \text{ RH weight} - \text{oven dry weight}}{\text{oven dry weight}}$$

according to Nielsen-Marsh (1997 unpubl).

Bulk powder density: The samples which had been weighed out for microporosity measurements were dried in an oven at 100°C for at least three days and then allowed to cool over silica gel in a dessicator. A 1.5mm micro-curette with an approximate volume of 1.77mm³ was used to weigh out three aliquots of dried bone powder. For each aliquot, the micro-curette was loaded up with bone powder which was pressed down against the side of the vial. The micro-curette was then tapped three times to remove excess or loose powder, before the surface of the bone powder was levelled off using a clean scalpel. The average of the three weights was calculated and divided by 1.77 to calculate the bulk powder density. Note that such a small volume was required due to the limited amount of bone powder available, and that the values for should be taken as relative to each other, but not absolute due to uncertainty of the volume of the micro-curette. As for the microporosity measurements, bulk powder density could not be calculated for samples which had fragmented on drilling due to heterogeneous grain size.

FTIR: Infra-red spectra of bone powder was obtained using Varian Excalibur series FTIR with a Specac Golden Gate ATR at BegbrokeNano, part of the Department of Materials, Oxford University. Data was manipulated and measured using Digilab Resolutions Pro 4.0 software. Each sample was run in triplicate for 20 scans and each spectrum subject to background subtraction and baseline correction. The splitting factor (SF), or crystallinity index, was calculated for each spectrum according to Weiner and Bar-Yosef (1990) as follows:

$$\text{SF} = \frac{\text{height of peak at } 603\text{cm}^{-1} + \text{height of peak at } 565\text{cm}^{-1}}{\text{distance from the baseline to the trough between the 2 peaks}}$$

when the baseline was drawn between 500 and 750 cm^{-1} .

The carbonate : phosphate ratio (C:P) was calculated according to Wright and Schwarcz (1996) as:

$$\text{C:P} = \frac{\text{height of carbonate peak at } 1417 \text{ cm}^{-1}}{\text{height of phosphate peak at } 1022 \text{ cm}^{-1}}$$

Both SF and C:P were calculated for all three spectra for each sample and averaged.

Elemental analysis: %C, %N, and C:N atomic ratio.

Whole bone and collagen samples were analysed using an automated carbon and nitrogen elemental analyser (Carlo Erba EA1108) coupled with a continuous-flow isotope ratio-monitoring mass spectrometer (Europa Geo 20/20).

Full radiocarbon pre-treatment: The method is detailed by Bronk Ramsey *et al* (2004).

Briefly:

- coarsely ground bone powder (~600mg) was sequentially treated with hydrochloric acid (0.5M), sodium hydroxide (0.1M), and hydrochloric acid (0.5M) with thorough rinsing with ultrapure (MilliQ™) water between each reagent;
- crude collagen was then gelatinized in pH3 solution at 75°C for 20 hours;
- the gelatin solution was filtered using a 9 μm polyethylene Eezi-filter™ which had been cleaned by ultrasonication in ultrapure water for 20 minutes;
- the filtered gelatin was transferred into a pre-cleaned ultrafilter (Vivaspin™ 15 30 kD MWCO) and centrifuged at 2500–3000 rpm until 0.5–1.0mL of the > 30 kD gelatin fraction remained (typically 20–40 minutes);
- this fraction was freeze-dried and the resulting purified collagen weighed.

During this procedure, both the insoluble residue left after Eezi-filtering and the gelatin which passed through the ultrafilter were freeze-dried and weighed, and the percentage insoluble residue and < 30 kD component calculated respectively. The percent collagen yield was also calculated in relation to the starting weight of the bone powder.

The stable isotopic values ($\delta^{13}\text{C}$, $\delta^{15}\text{N}$) and C:N ratio of the collagen was measured as for the whole bone powder.

Amino acid analysis: Analysis was carried out by Tony Willis at the MRC Immunochemistry Unit in the Department of Biochemistry, University of Oxford. 2.5 nanomoles of an internal standard mixture of nor-valine and sarcosine was added to each sample of purified collagen (~1 mg) and samples were dried in pyrolysed Pyrex tubes (Corning 9820 culture tubes) before being hydrolysed in vapour-phase for 22 hours at 110°C. The hydrolysing medium was 5.7N hydrochloric acid (constant boiling) with a trace of phenol added. Amino acid analysis was performed using an Agilent 1100 series HPLC with a G1327A autosampler and G1321A fluorescence detector. Samples were derivitised with ortho-phthalaldehyde (OPA) for the primary amino acids and 9-fluorenylmethyl chloroformate (FMOC) for the secondary amino acids. 1 μl of redissolved hydrolysate was injected onto a Hypersil AA-ODS HPLC column (Agilent) to resolve the derivitised amino acids. Results were integrated using Agilent ChemStation software with three point calibration in each batch of samples using standard runs of 10, 25, and 100 picomoles, and are given as mole %.

SAXS: Analysis was carried out by Clerk Maxwell in the School of Optometry and Vision Sciences, Cardiff University. Bone powder (~ 15mg) was loaded into a specially designed sample carriage between two mica sheets. This was attached to a sample stage which was mounted into the vacuum chamber of the NanoSTAR (Bruker AXS, Karlsruhe) X-ray facility.

The SAXS configuration uses a sample-to-detector distance of 1.25m. Bone scattering profiles typically require 3-hour exposures. Data is collected according to Wess *et al* (2001), corrected for camera distortions, subjected to ultralene background subtraction, and images analysed using in-house software. The two-dimensional detector output is converted into spherically averaged one-dimensional profiles, and values for crystal thickness determined.

Appendix II: Data

ID	Site	Site ID	Hardness	Colour			Micro-porosity	Bulk powder density	Whole bone powder				Splitting factor	Carbonate: phosphate	Radiocarbon pre-treatment				Collagen				SAXS Ave thickness nm
				Hue	Saturation	Brightness			$\delta^{13}C$	%N	%C	C/N			% Insol	% <30kD	% wt coll	OK/fail	No reps	$\delta^{13}C$	$\delta^{15}N$	C/N	
			arb. unit				cm ³ /g	g/cm ³	‰										‰	‰			
1	Brandon Staunch Meadow	1708	55	36	48	68	0.083	0.31	-24.32	0.2	3.2	17.6	5.06	0.16									
2	Brandon Staunch Meadow	1779	53	36	52	67	0.088	0.19															
3	Brandon Staunch Meadow	1803	59	36	58	57	0.093	0.19	-19.95	3.1	10.1	3.9	4.06	0.21	0.4	0.1	3.9	OK	3	-19.63	10.85	3.2	
4	Brandon Staunch Meadow	1816	59	33	52	67	0.086	0.22	-23.46	1.2	5.7	5.7	4.31	0.18									
5	Brandon Staunch Meadow	1830	47	33	42	67	0.080	0.18															
6	Brandon Staunch Meadow		27	39	52	78																	
7	Brandon Staunch Meadow	1838	58	35	54	66	0.071	0.23	-20.10	1.0	4.8	5.7	4.32	0.18									
8	Brandon Staunch Meadow	1850	29	38	56	66	0.071	0.28	-27.48	0.3	4.2	17.6	4.51	0.19									
9	Brandon Staunch Meadow	1861	62	35	52	65	0.072	0.18	-21.98	1.0	5.7	7.0	4.18	0.21	5.7	0.1	0.8	fail	2	-19.76	10.32	3.3	
10	Brandon Staunch Meadow	1863	68	34	40	53	0.074	0.10	-18.91	3.5	12.0	3.7	4.61	0.26	4.6	0.0	6.5	OK	3	-19.50	10.10	3.2	
11	Brandon Staunch Meadow	1882	61	35	53	66	0.114	0.24	-19.44	2.1	7.7	4.2	4.58	0.22									
12	Brandon Staunch Meadow	1898	55	39	47	78	0.113	0.29															
13	Brandon Staunch Meadow	1900	55	35	54	64	0.107	0.37	-22.36	0.6	4.3	8.4	4.81	0.17	5.6	0.1	0.5	fail	1	-19.63	9.94	3.3	
14	Brandon Staunch Meadow	1917	51	34	53	58	0.097	0.43															
15	Brandon Staunch Meadow	1919	33	42	44	82	0.108	0.27	-21.61	0.7	4.4	7.5	4.79	0.17									
16	Brandon Staunch Meadow	1923	52	37	50	72	0.081	0.35															
17	Brandon Staunch Meadow	3067	48	39	57	69	0.097	0.34	-20.77	0.9	4.7	5.8	4.26	0.19	3.9	0.0	0.8	fail	3	-19.32	10.55	3.3	
18	Brandon Staunch Meadow	3081	28	35	54	61	0.092	0.22	-19.69	3.5	11.5	3.9	5.06	0.26									
19	Brandon Staunch Meadow	3082	55	35	54	65	0.096	0.30	-20.90	1.3	5.7	5.2	4.23	0.18									
20	Brandon Staunch Meadow	3090	57	32	45	47	0.090	0.32															
21	Brandon Staunch Meadow	3103	55	32	51	65	0.115	0.24	-19.32	2.9	9.9	4.0	4.26	0.30	0.9	0.1	3.0	OK	3	-19.26	9.33	3.2	
22	Brandon Staunch Meadow	3112	60	33	51	67	0.110	0.14															
23	Brandon Staunch Meadow	3113	58	37	56	66	0.109	0.20	-19.08	3.7	11.3	3.6	4.29	0.24									
24	Brandon Staunch Meadow	3127	62	38	51	65	0.100	0.20															
25	Brandon Staunch Meadow	3136	68	36	45	61	0.104	0.21	-20.67	2.0	8.2	4.8	4.05	0.24	2.9	0.1	1.4	OK	3	-20.19	9.69	3.3	
26	Brandon Staunch Meadow	4009	58	36	56	64	0.101	0.24															
27	Brandon Staunch Meadow	4035	58	31	45	48			-19.41	3.2	10.4	3.8	4.48	0.26									
28	Brandon Staunch Meadow	4042	50	38	51	78	0.093	0.18															
29	Brandon Staunch Meadow	4054	49	36	47	65	0.096	0.26	-23.22	0.3	3.9	14.2	4.89	0.18	1.6	0.0	0.1	fail					
30	Brandon Staunch Meadow	8007	60	34	55	57	0.084	0.21															
31	Holloway Lane	HL 87 B1	47	36	16	89	0.100	0.18	-19.87	0.4	2.7	8.3	4.68	0.14									
32	Holloway Lane	HL 87 B14	17	37	18	82	0.111	0.21	-22.38	1.2	4.8	4.5	5.29	0.14	4.8	0.4	1.8	OK	3	-24.00	7.91	3.2	
33a	Holloway Lane	HL 87 B62		36	28	78	0.111	0.18															
33b	Holloway Lane	HL 87 B62b		34	26	78																	
34	Holloway Lane	HL 87 B65	47	34	32	75	0.107	0.30	-25.08	1.5	5.1	4.0	5.64	0.16									
35	Holloway Lane	HL 87 B66	22	41	13	91	0.097	0.27	-22.09	1.0	4.3	5.3	5.67	0.16	2.9	0.4	1.9	OK	3	-24.31	7.99	2.9	5.84
36	Holloway Lane	HL 87 B17	67	39	34	76	0.100	0.26															
37	Holloway Lane	HL 87 B24	5	39	37	63	0.075	0.34	-22.81	0.2	1.4	7.9		0.05	41.6	0.1	0.1	fail					
38	Holloway Lane	HL 87 B45	7	36	29	78	0.088	0.21															
39	Holloway Lane	HL 87 B67	25	53	4	94	0.085	0.23	-21.32	1.0	4.3	4.9	4.95	0.18									
40	Holloway Lane	HL 82 III 42	10	30	8	76	0.084	0.21	-21.66	1.1	5.0	5.2	4.30	0.19									
41	Holloway Lane	HL 82 III 61	40	38	22	77	0.107	0.31	-20.30	1.0	4.5	5.2	5.78	0.17	1.8	0.1	2.3	OK	3	-21.72	7.50	3.2	
42	Holloway Lane	WGF 79 41	32	38	11	91	0.090	0.29															
43	Eynesbury	3249 2365	34	47	26	87	0.105	0.21	-16.85	0.1	2.2	35.8	4.44	0.21									
44	Eynesbury	3249 2366	6	43	43	58	0.067	0.30	-11.69	0.0	7.3	0.0	3.97	0.61									
45	Eynesbury	6087 2329	59	30	42	43	0.151	0.34	-19.84	0.1	2.4	21.7	3.88	0.29	7.6	0.0	0.2	fail					

46	Eynesbury	6087 2389	60	27	45	33	0.133	0.39	-22.45	0.2	2.1	15.7	4.15	0.24								
47	Eynesbury	6301 2346	33	39	45	30	0.099	0.40	-19.35	0.1	1.3	26.9	4.29	0.24								
48	Eynesbury	6354 2348	29	35	51	33	0.110	0.36														
49	Eynesbury	6355	60	43	37	76	0.111	0.34	-19.51	0.1	2.7	31.6	4.38	0.22	9.6	0.0	0.0	fail				
50	Eynesbury	6355 2385	48	42	26	39	0.110	0.29	-19.11	0.2	3.2	26.2	4.23	0.26								
51	Eynesbury	6355 2385	40	40	35	52		0.00														
52	Eynesbury	6360	50	44	61	38	0.113	0.28	-16.68	0.1	2.2	21.3	4.32	0.28								
53	Eynesbury	6360 2368	46	34	42	50	0.099	0.46														
54	Eynesbury	6360 2369		40	50	35	0.109	0.40														
55	Eynesbury	6361	55	36	31	75	0.109	0.21	-18.37	0.1	2.2	37.0	3.88	0.26	12.9	0.1	0.2	fail				
56	Eynesbury	6361 2375		32	28	47	0.099	0.32														
57	Eynesbury	6363 2355	56	40	17	21	0.143	0.28	-19.94	0.1	2.0	19.6	4.52	0.24								
58	Eynesbury	6363 2357	49	47	32	28	0.124	0.24	-17.41	0.0	2.3	67.5	5.04	0.19								
59	Eynesbury	6379	46	30	31	38	0.103	0.34	-19.63	0.1	1.4	19.1	3.58	0.29	37.5	0.2	0.1	fail				
60	Eynesbury	6394 2384	50	31	37	41	0.104	0.26														
61	Haddenham CE	HAD 82 380 2234	47	43	24	57	0.125	0.28	-21.30	0.1	2.3	24.6	4.19	0.21								
62	Haddenham CE	HAD 82 403 2413	49	44	28	58	0.107	0.31														
63	Haddenham CE	HAD 82 403 2422	46	39	53	42	0.116	0.33	-18.69	0.1	2.2	24.7	4.33	0.22	17.0	0.0	0.0	fail				
64	Haddenham CE	HAD 82 403 2423	50	42	39	64	0.132	0.22														
65	Haddenham CE	HAD 82 471 2566	51	39	36	63	0.093	0.22	-21.23	0.2	5.4	27.8	4.25	0.21								
66	Haddenham CE	HAD 82 471 2567	55	38	38	38	0.090	0.27	-21.02	0.1	1.3	24.1	5.98	0.14								
67	Haddenham CE	HAD 82 476 2832	13	42	43	69	0.112	0.24	-18.70	0.1	2.6	23.5	4.65	0.21	6.1	0.0	0.0	fail				
68	Haddenham CE	HAD 84 1747 12527	44	41	35	59	0.117	0.25														
69	Haddenham CE	HAD 84 1747 12556	56	53	45	38	0.083	0.32														
70	Haddenham CE	HAD 84 1747 12569	48	43	41	60	0.080	0.39	-23.36	0.6	3.1	6.3	3.87	0.23								
71	Haddenham CE	HAD 84 1749 12599	32	45	32	76	0.139	0.23	-19.40	0.2	3.7	26.7	4.15	0.31								
72	Haddenham CE	HAD 84 1801/1809 11580	7	41	37	82	0.085	0.34	-19.81	0.6	3.1	6.0	4.93	0.20	31.0	0.2	0.2	fail	1	-21.51	5.24	3.2
73	Haddenham CE	HAD 84 1818 12548	67	54	26	89	0.108	0.26														
74	Haddenham CE	HAD 84 1837 14610	58	48	32	73	0.106	0.22														
75	Haddenham CE	HAD 87 3801 27641	47	26	41	48	0.111	0.18	-18.44	0.1	2.7	26.4	4.84	0.20								
76	Haddenham CE	HAD 87 3812 27628	58	37	47	57	0.124	0.21	-25.27	0.1	2.7	35.2	4.99	0.17								
77	Haddenham CE	HAD 87 3813 27968	60	49	30	87	0.108	0.20	-19.91	0.7	4.5	7.9	3.94	0.23	0.1	0.0	0.5	fail	1	-21.66	6.61	3.3
78	Haddenham CE	HAD 87 3813 28048	45	49	47	79	0.129	0.18														
79	Haddenham CE	HAD 87 3813 28050		50	39	76	0.118	0.27														
80	Haddenham CE	HAD 87 3813 28051	10	47	39	73	0.122	0.24	-22.10	0.3	4.1	17.9	5.88	0.20	0.4	0.0	0.0	fail				
81	Haddenham CE	HAD 87 3822 27735	40	48	50	55		0.30	-19.49	0.3	3.9	13.3	4.77	0.23								
82	Haddenham CE	HAD 87 3840 27675	20	53	27	88	0.096	0.32														
83	Haddenham CE	HAD 87 3857 27992	33	40	48	62	0.113	0.22	-22.33	0.1	1.9	16.1	4.90	0.20	0.4	0.0	0.0	fail				
84	Haddenham CE	HAD 87 3890 28104	28	40	50	56	0.103	0.30														
85	Haddenham CE	HAD 87 3898 28161	5	53	35	75	0.129	0.30	-22.15	0.4	3.6	11.4	5.54	0.21								
86	Haddenham CE	HAD 87 3898 28199	58	36	41	64	0.123	0.25	-21.76	0.8	4.7	7.2	4.30	0.23								
87	Haddenham CE	HAD 87 3931 28244	33	53	34	75	0.101	0.00	-21.87	0.8	3.8	5.8	5.45	0.18	1.3	0.3	0.7	fail	2	-23.07	7.14	3.2
88	Haddenham CE	HAD 87 3931 27962	40	42	43	58	0.138	0.24														
89	Haddenham CE	HAD 87 3938 27971	5	40	41	76	0.151	0.29	-23.58	0.2	3.6	18.2	4.40	0.22								
90	Haddenham CE	HAD 87 3939 27961	34	38	38	62	0.122	0.29														
91	Haddenham CE	HAD 87 3947 27959	55	48	35	86	0.120	0.20	-20.33	0.2	4.3	28.9	3.91	0.27	1.4	0.0	0.0	fail				
92	Haddenham CE	HAD 87 3952 28056	32	39	42	75	0.104	0.29														
93	Haddenham CE	HAD 87 3952 28268	16	41	37	68	0.099	0.32	-20.26	0.1	3.0	24.8	5.18	0.21								
94	Haddenham CE	HAD 87 3953 27963	38	40	45	58	0.090	0.22	-15.64	0.1	5.6	98.1	4.58	0.43								
95	Haddenham CE	HAD 87 3953 27976	30	36	41	68	0.098	0.23	-20.23	0.6	4.9	9.7	4.15	0.23	0.1	0.1	0.2	fail	1	-20.87	9.38	3.2
96	Haddenham CE	HAD 87 3965 27986	52	40	51	54	0.098	0.35														

97	Haddenham CE	HAD 87 3969 27960	32	49	46	83	0.086	0.34	-16.31	0.2	4.5	24.0	4.72	0.34										
98	Haddenham CE	HAD 87 3973 27985	37	53	32	78	0.082	0.24																
99	Berinsfield	BERWC74 Grave 1	58	44	19	87	0.080	0.24	-17.20	1.0	4.9	5.5	3.64	0.30	0.5	0.0	2.5	OK	3	-20.21	9.26	3.2	5.14	
100	Berinsfield	BERWC74 Grave 5	57	47	29	73	0.083	0.31																
101	Berinsfield	BERWC74 Grave 6	46	28	37	95	0.114	0.24	-18.53	1.2	5.3	5.2	3.87	0.24										
102	Berinsfield	BERWC74 Grave 8	58	38	32	95	0.101	0.28	-20.44	0.9	4.4	5.9	4.76	0.26	0.8	0.1	1.31	OK	3	-20.15	10.44	3.2		
103	Berinsfield	BERWC74 Grave 14	63	44	26	94	0.099	0.28	-17.73	1.1	4.9	5.2	3.98	0.23	5.4	0.2	1.8	OK	3	-19.78	10.00	3.2		
104	Berinsfield	BERWC74 Grave 18	68	39	31	93	0.105	0.23																
105	Berinsfield	BERWC74 Grave 20	60	45	20	85	0.103	0.25	-17.83	1.4	5.6	4.5	3.70	0.27										
106	Berinsfield	BERWC74 Grave 22	58	48	40	78	0.101	0.23																
107	Berinsfield	BERWC74 Grave 28	52	47	34	89	0.103	0.24	-17.59	0.8	3.9	5.8	3.83	0.28	0.7		1.9	OK	3	-20.29	9.75	3.2		
108	Berinsfield	BERWC74 Grave 35	55	43	33	80	0.101	0.32	-21.58	1.0	4.5	5.1	4.04	0.24										
109	Berinsfield	BERWC74 Grave 49	28	49	38	80	0.100	0.32	-17.18	0.3	2.8	11.6	4.45	0.24	24.5	0.0	0.1	fail						
110	Berinsfield	BERWC74 Grave 61	47	45	40	87	0.094	0.31																
111	Berinsfield	BERWC74 Grave 63	55	50	20	82	0.102	0.29	-18.08	1.2	4.7	4.6	3.83	0.26	1.7	0.3	3.4	OK	3	-20.29	9.32	3.2		
112	Berinsfield	BERWC74 Grave 72	48	45	42	85	0.116	0.21	-18.49	0.7	4.4	7.8	4.37	0.23										
113	Berinsfield	BERWC74 Grave 73	58	43	45	69	0.099	0.26	-17.85	0.7	3.5	5.6	3.93	0.24	0.0	0.0	1.3	OK	3	-20.03	9.83	3.2	4.49	
114	Berinsfield	BERWC74 Grave 82	48	44	40	86	0.104	0.28																
115	Berinsfield	BERWC74 Grave 83	55	52	17	87	0.109	0.26	-18.13	1.1	4.8	5.2	4.80	0.30	2.6	0.4	2.4	OK	3	-20.41	9.62	3.2		
116	Berinsfield	BERWC74 Grave 86	49	40	41	80	0.129	0.21																
117	Berinsfield	BERWC74 Grave 92	37	44	26	76	0.104	0.26	-19.04	1.2	5.0	4.9	3.65	0.30	15.7		3.0	OK	3	-19.97	8.41	3.2		
118	Berinsfield	BERWC74 Grave 102	54	45	22	85	0.102	0.24																
119	Berinsfield	BERWC74 Grave 107	43	53	32	81	0.100	0.27	-18.69	1.0	4.6	5.4	4.01	0.30	17.4	0.3	2.0	OK	3	-20.23	9.22	3.2		
120	Berinsfield	BERWC74 Grave 110	52	49	43	69	0.078	0.30	-20.91	1.0	4.7	5.6	4.13	0.25	2.4	1.6	1.4	OK	3	-19.83	9.70	3.2		
121	Berinsfield	BERWC74 Grave 121	52	52	23	91	0.113	0.26	-17.59	0.9	4.2	5.7	4.35	0.28	2.5		1.8	OK	3	-19.86	9.99	3.2		
122	Berinsfield	BERWC74 Grave 128	65	48	15	90	0.096	0.29																
123	Berinsfield	BERWC74 Grave 133	62	58	11	91	0.096	0.29	-18.11	0.8	3.9	5.5	4.83	0.24										
124	Berinsfield	BERWC74 Grave 134	48	43	20	89	0.111	0.22																
125	Berinsfield	BERWC74 Gr 141/2	62	40	46	78	0.109	0.26	-18.20	0.8	4.0	5.5	4.41	0.28	0.4	0.0	1.6	OK	3	-19.70	10.06	3.2		
126	Berinsfield	BERWC74 Gr 148	60	37	32	74	0.084	0.24	-18.11	0.5	3.6	8.7	5.05	0.21	2.1	0.0	1.1	OK	3	-20.13	10.84	3.2	4.75	
127	Berinsfield	BERWC74 Gr 149	55	27	60	78	0.100	0.29	-18.35	1.0	4.5	5.2	4.39	0.29	17.6	0.8	0.5	fail	1	-19.72	9.01	3.2	4.25	
128	Berinsfield	BERWC74 Gr 150/1	42	37	59	58	0.112	0.23																
129	Berinsfield	BERWC74 Gr 150/2	42	36	60	61	0.136	0.24	-18.44	0.2	2.8	16.3	4.69	0.28	3.4	0.0	0.0	fail						
130	Huntsman's Quarry	HWCM 21698 1103 Bag 1	36	43	45	72	0.134	0.29																
131	Huntsman's Quarry	HWCM 21698 1103 Bag 2	48	46	23	86	0.136	0.24	-19.39	0.8	4.3	6.6	3.89	0.25	0.7	0.1	1.4	OK	3	-21.71	7.16	3.2	3.94	
132	Huntsman's Quarry	HWCM 21698 1104A	42	48	25	93	0.160	0.29	-22.21	0.6	4.2	8.4	4.18	0.26										
133	Huntsman's Quarry	HWCM 21698 1104B	58	52	22	92	0.122	0.24	-16.84	0.4	2.8	7.8	3.83	0.29	0.0	0.0	0.1	fail	0					
134	Huntsman's Quarry	HWCM 21698 1111A	58	34	54	50	0.160	0.23																
135	Huntsman's Quarry	HWCM 21698 1111B	55	46	18	93	0.118	0.27	-18.45	0.7	3.7	5.8	3.94	0.26										
136	Huntsman's Quarry	HWCM 21698 1111C	52	47	20	79	0.116	0.28																
137	Huntsman's Quarry	HWCM 21698 1111D	53	44	50	69	0.154	0.28	-16.43	0.1	2.5	37.5	3.91	0.28	0.3	0.0	0.0	fail	0					
138	Huntsman's Quarry	HWCM 21698 1242	53	64	6	96	0.124	0.25	-18.64	0.5	4.2	9.8	4.28	0.27										
139	Huntsman's Quarry	HWCM 21698 1517	52	44	48	63	0.148	0.22	-15.07	0.1	2.8	40.0	4.02	0.23										
140	Huntsman's Quarry	HWCM 21698 1517	46	55	5	95	0.122	0.23																
141	Huntsman's Quarry	HWCM 21698 1830A	48	43	20	82	0.124	0.32	-19.48	0.9	4.5	6.1	4.34	0.26	2.2	0.2	1.5	OK	3	-21.52	5.90	3.2		
142	Huntsman's Quarry	HWCM 21698 1830B	57	40	35	86	0.146	0.25																
143	Huntsman's Quarry	HWCM 21698 1830C	57	40	38	85	0.122	0.28	-19.13	1.2	5.2	5.3	3.87	0.26										
144	Huntsman's Quarry	HWCM 21698 1830D	53	41	36	76	0.162	0.29																
145	Huntsman's Quarry	HWCM 21698 1834	43	48	22	93	0.141	0.27	-17.98	0.6	4.1	8.1	4.07	0.29	0.3	0.0	0.8	fail	2	-22.15	7.15	3.2		
146	Huntsman's Quarry	HWCM 21698 1836	60	49	19	92	0.131	0.26																

147	Huntsman's Quarry	HWCM 21698 1838	50	48	30	85	0.183	0.24	-15.65	0.1	2.6	37.2	3.80	0.24								
148	Huntsman's Quarry	HWCM 21698 1839	45	47	22	91	0.162	0.22	-18.01	0.1	3.4	47.5	4.30	0.25								
149	Huntsman's Quarry	HWCM 21698 1855	60	43	31	86	0.152	0.26	-18.52	0.1	2.9	33.9	3.89	0.27	9.1	0.1	0.0	fail	0			
150	Huntsman's Quarry	HWCM 21698 2010	52	46	28	89	0.117	0.28														
151	Huntsman's Quarry	HWCM 21698 2032A	58	44	40	85	0.141	0.34	-17.88	0.1	2.4	35.9	4.25	0.25								
152	Huntsman's Quarry	HWCM 21698 2032B	50	53	12	89	0.111	0.33														
153	Huntsman's Quarry	HWCM 21698 2032C	59	38	45	77	0.134	0.28	-18.13	0.4	3.6	9.4	3.62	0.28	0.8	0.0	0.1	fail	0			
154	Huntsman's Quarry	HWCM 21698 2050	45	37	42	75	0.126	0.24														
155	Bestwall Quarry	BQ01J (951)	5	48	79	37	0.078	0.37	-22.17	1.7	7.2	5.3	8.23	0.20	36.6	0.0	4.5	OK	3	-21.45	6.45	3.2
156	Bestwall Quarry	BQ92A (17)	52	33	65	56	0.094	0.27	-21.20	0.9	3.9	5.3	5.43	0.12	1.7	0.0	2.4	OK	3	-21.86	6.09	3.2
157	Bestwall Quarry	BQ92A (26)	30	29	52	65	0.074	0.37														
158	Bestwall Quarry	BQ95C (803)	5	36	71	47	0.097	0.33	-22.73	1.3	5.5	5.1	5.83	0.09	2.9	0.2	2.3	OK	3	-22.16	6.72	3.3
159	Bestwall Quarry	BQ98G (55)	47	39	82	55	0.102	0.36	-24.51	2.5	8.2	3.9	5.98	0.14	8.8	0.1	6.3	OK	3	-22.14	6.89	3.2
160	Bestwall Quarry	BQ99G (456)	31	38	59	55	0.087	0.27	-21.15	1.1	5.0	5.5	4.40	0.16	4.3	0.2	2.7	OK	3	-21.43	7.41	3.2
161	Bestwall Quarry	BQ99G (494)	33	41	89	48	0.117	0.35	-21.53	1.7	7.2	4.8	4.61	0.18	9.6	0.2	5.4	OK	3	-21.99	5.31	3.2
162	Bestwall Quarry	BQ99G (525)	5	30	57	73	0.080	0.33	-22.33	0.8	3.7	5.1	4.89	0.12	27.9	0.1	2.0	OK	3	-22.48	5.96	3.3
163	Bestwall Quarry	BQ99G (526)	50	32	56	61	0.101	0.24														
164	Bestwall Quarry	BQ99H (71)	47	31	49	61	0.095	0.32	-20.41	0.8	4.1	5.6	4.06	0.16								
165	Bestwall Quarry	BQ99H (88)	27	38	73	52	0.102	0.33	-22.73	1.7	6.8	4.6	6.61	0.16	27.7	0.5	6.4	OK	3	-22.58	7.32	3.3
166	Bestwall Quarry	BQ99H (427)	21	33	57	60	0.103	0.28	-22.10	0.9	4.2	5.2	5.31	0.08	1.3	0.2	1.6	OK	3	-22.58	7.19	3.2
167	Bestwall Quarry	BQ2KS (278)	22	34	68	55	0.098	0.29														
168	Etton CE	B9943	68	35	35	94	0.126	0.21	-20.55	3.3	10.8	3.9	3.80	0.29	0.6	0.7	3.8	OK	3	-22.08	6.72	3.2
169	Etton CE	B15663		41	33	85	0.158	0.24	-18.58	0.2	3.3	24.5	4.33	0.25								
170	Etton CE	B14807	72	37	42	39	0.115	0.27	-15.46	0.1	2.3	31.1	3.82	0.25	7.5	0.0	0.1	fail				4.17
171	Etton CE	B15724	50	32	51	53	0.154	0.33														
172	Etton CE	B15726	65	33	36	72	0.126	0.24	-20.91	2.8	9.5	3.9	4.00	0.25	1.9	0.9	4.0	OK	3	-22.37	6.17	3.2
173	Etton CE	B15733	52	34	27	91	0.136	0.24														
174	Etton CE	B15735	54	37	42	68	0.133	0.21	-20.40	3.5	11.4	3.8	3.77	0.29	0.2	0.8	3.2	OK	3	-21.95	4.75	3.2
175	Etton CE	B16117	52	25	34	45	0.135	0.48	-15.00	0.1	3.7	45.0	4.08	0.33								
176	Etton CE	B16121	55	34	34	64	0.128	0.35	-19.31	0.1	3.2	28.6	3.82	0.28								
177	Etton CE	B16290	65	51	27	40	0.132	0.31	-13.13	0.1	1.9	34.5	4.14	0.25								
178	Etton CE	B16299	52	30	39	55	0.145	0.31	-20.36	1.0	5.9	6.7	3.58	0.25	0.2	0.0	0.9	fail	3	-20.85	6.03	3.4
179	Etton CE	B16300	52	27	40	48	0.139	0.30	-20.22	1.0	5.4	6.5	4.07	0.24	0.3	0.0	0.5	fail	1	-21.50	4.84	3.4
180	Etton CE	B14818	53	31	37	45	0.094	0.40	-15.20	0.1	1.6	25.2	3.81	0.27								
181	Etton CE	B14822	52	38	37	80	0.099	0.41														
182	Etton CE	B14826	72	34	40	45	0.104	0.52	-18.02	0.1	2.4	30.2	3.61	0.31	19.1	0.0	0.5	fail	1	-29.37		86.4
183	Etton CE	B14547	48	34	43	47	0.098	0.32														
184	Etton CE	B15477	58	37	54	49	0.119	0.28	-17.94	0.1	2.8	30.6	3.86	0.25								
185	Etton CE	B16302	35	32	36	55	0.110	0.41														
186	Imperial Coll Sports Grd	IMC96 1283	59	34	40	78	0.110	0.32	-21.69	0.2	2.4	17.6	3.92	0.16	3.9	0.0	0.0	fail	0			
187	Imperial Coll Sports Grd	IMC96 1285A	60	39	39	68	0.102	0.19														
188	Imperial Coll Sports Grd	IMC96 1285B	50	36	35	80	0.100	0.20	-19.44	0.1	2.1	17.1	4.41	0.16								
189	Imperial Coll Sports Grd	IMC96 1286A	34	43	14	93	0.103	0.25	-21.00	0.3	2.6	11.4	6.02	0.15								
190	Imperial Coll Sports Grd	IMC96 1286B	41	35	33	87	0.121	0.39	-21.93	0.2	2.1	12.0	4.77	0.15	11.8	0.2	0.0	fail	0			
191	Imperial Coll Sports Grd	IMC96 1873A	38	37	36	87	0.117	0.22														
192	Imperial Coll Sports Grd	IMC96 1873B	49	37	40	67	0.122	0.39	-20.75	0.2	2.0	13.6	4.79	0.14								
193	Imperial Coll Sports Grd	IMC96 1873C	58	39	35	74	0.123	0.25														
194	Imperial Coll Sports Grd	IMC96 4844A	47	40	32	83	0.122	0.40	-22.85	0.3	1.8	7.2		0.09								
195	Imperial Coll Sports Grd	IMC96 4844B	70	40	40	82	0.091	0.28	-22.09	0.2	2.3	11.7	4.99	0.14								
196	Imperial Coll Sports Grd	IMC96 4844C	61	38	35	78	0.100	0.27	-20.39	0.5	3.0	7.1	4.71	0.13	8.4	0.5	0.0	fail	0			
197	Imperial Coll Sports Grd	IMC96 4847A	48	47	38	69	0.101	0.20														

249	Kingsmead Quarry	H54635 3824A	65	34	41	78	0.139	0.21															
250	Kingsmead Quarry	H54635 3824B	65	36	37	86	0.144	0.23	-20.74	0.1	3.2	34.7	3.78	0.26	3.8	0.0	0.0	fail	0				
251	Kingsmead Quarry	H54635 3824C	50	36	44	80	0.163	0.41	-23.98	0.1	2.9	27.1	4.70	0.23									
252	Kingsmead Quarry	H54635 3824D	65	33	38	81	0.125	0.31	-19.66	0.2	4.7	37.0	3.95	0.24									
253	Kingsmead Quarry	H54635 3824E	60	34	38	76	0.171	0.37	-22.50	0.1	2.7	32.4	5.52	0.25									
254	Kingsmead Quarry	H54635 3850A	63	33	36	89	0.168	0.31	-20.98	0.1	3.2	37.3	4.03	0.27	4.0	0.0	0.0	fail	0				
255	Kingsmead Quarry	H54635 3850B	60	37	23	90	0.161	0.43															
256	Kingsmead Quarry	H54635 3850C	60	41	17	93	0.143	0.23	-17.55	0.1	3.0	35.4	3.61	0.24									
257	Kingsmead Quarry	H54635 3850D	60	36	42	73	0.131	0.27	-18.73	0.0	2.2	51.6	3.85	0.26									
258	Kingsmead Quarry	H54635 3850E	60	39	30	90	0.146	0.24	-19.69	0.1	3.1	34.1	3.61	0.24	1.0	0.0	0.0	fail	0				
259	Kingsmead Quarry	H54635 7255A	54	46	37	65	0.145	0.29															
260	Kingsmead Quarry	H54635 7255B	19	50	28	74	0.161	0.31	-20.68	0.1	2.6	27.5	4.34	0.19									
261	Kingsmead Quarry	H54635 7255C	50	48	42	54	0.162	0.28															
262	Kingsmead Quarry	H54635 7255D	55	44	49	53	0.186	0.32	-24.26	0.2	2.1	16.4	5.88	0.18	16.1	0.0	0.0	fail	0			3.92	
263	Kingsmead Quarry	H54635 7255E	5	42	54	41	0.148	0.23	-19.38	0.1	2.5	29.4	4.59	0.19									
264	RMC Land, Harlington	SIE00 876A	53	49	12	92	0.107	0.27	-21.10	1.1	4.6	4.7	3.94	0.17	0.7	0.0	0.1	fail					
265	RMC Land, Harlington	SIE00 876B	48	47	8	93	0.118	0.27	-22.79	1.2	4.7	4.6	5.66	0.17									
266	RMC Land, Harlington	SIE00 876C	38	43	31	63	0.122	0.30	-20.58	1.0	4.5	5.4	5.73	0.12	9.3	0.2	1.5	OK	3	-21.22	6.96	3.2	
267	RMC Land, Harlington	SIE00 876D	48	45	28	89	0.131	0.25															
268	RMC Land, Harlington	SIE00 876E	63	39	43	75	0.116	0.27	-20.49	1.0	4.4	5.3	4.60	0.14									
269	RMC Land, Harlington	SIE00 877A	47	49	16	85	0.114	0.28															
270	RMC Land, Harlington	SIE00 877B	39	52	10	93	0.110	0.31	-21.53	1.2	5.0	4.7	4.12	0.17	0.3	0.0	0.0	fail	0				
271	RMC Land, Harlington	SIE00 877C	68	40	23	75	0.124	0.33	-23.76	1.5	5.2	4.2	4.95	0.18									
272	RMC Land, Harlington	SIE00 877D	43	56	6	95	0.119	0.24	-19.73	0.9	4.8	6.0	4.91	0.16									
273	RMC Land, Harlington	SIE00 877E	35	50	14	86	0.118	0.33															
274	RMC Land, Harlington	SIE00 1301A	58	45	18	92	0.102	0.25	-21.38	0.6	3.1	6.2	4.82	0.15	0.2	0.0	0.0	fail	0				
275	RMC Land, Harlington	SIE00 1301B	50	58	11	88	0.115	0.23	-22.83	0.6	3.1	6.2	6.18	0.12	0.9	0.0	1.3	OK	3	-21.31	6.91	3.2	
276	RMC Land, Harlington	SIE00 1301C	59	54	8	90	0.113	0.23	-20.33	0.4	3.0	9.8	5.65	0.13									
277	RMC Land, Harlington	SIE00 1301D	38	63	10	75	0.093	0.23	-19.65	0.3	2.7	10.5	5.47	0.12									
278	RMC Land, Harlington	SIE00 1301E	50	50	13	91	0.118	0.27	-20.69	0.8	4.1	6.2	6.08	0.13	0.4	0.0	2.2	OK	3	-21.58	8.10	3.2	
279	RMC Land, Harlington	SIE00 2741A	58	45	27	92	0.120	0.29															
280	RMC Land, Harlington	SIE00 2741B	57	50	20	91	0.115	0.31	-21.30	2.0	6.8	3.9	3.84	0.22	0.2	0.0	0.5	fail	1	-21.48	6.95	3.2	4.06
281	RMC Land, Harlington	SIE00 2741C	70	40	27	48	0.122	0.33	-20.07	1.1	4.9	5.3	5.25	0.20	0.3	0.0	2.1	OK	3	-21.57	7.65	3.3	
282	RMC Land, Harlington	SIE00 2741D	58	46	26	90	0.104	0.28	-18.91	0.7	3.9	6.7	4.67	0.19									
283	RMC Land, Harlington	SIE00 2741E	60	44	28	85	0.109	0.24															
284	RMC Land, Harlington	SIE00 2742A	55	60	2	98	0.110	0.30	-21.14	1.4	5.4	4.4	3.81	0.21	1.5	0.0	0.1	fail	0				
285	RMC Land, Harlington	SIE00 2742B	67	42	27	73	0.108	0.34	-22.87	2.0	6.4	3.7	4.49	0.19									
286	RMC Land, Harlington	SIE00 2742C	58	55	5	96	0.109	0.22	-19.36	0.9	4.5	5.9	6.10	0.18	1.3	0.0	2.6	OK	3	-21.58	8.27	3.2	
287	RMC Land, Harlington	SIE00 2742D	31	45	7	96	0.098	0.23	-19.25	0.6	3.3	6.6	5.34	0.14									
288	RMC Land, Harlington	SIE00 2742E	53	39	16	86	0.105	0.26	-21.30	1.4	5.3	4.5	3.90	0.16	0.5	0.0	0.1	fail					4.89
289	RMC Land, Harlington	SIE00 3921A	18	41	16	92	0.080	0.30															
290	RMC Land, Harlington	SIE00 3921B	5	44	28	93	0.119	0.31	-21.75	0.4	2.5	7.2	-	-	27.2	0.1	0.0	fail	0				
291	RMC Land, Harlington	SIE00 3921C	38	67	4	98	0.111	0.15	-19.07	0.2	2.3	16.1	6.39	0.13									
292	RMC Land, Harlington	SIE00 3921D	58	41	34	90	0.138	0.34	-19.91	0.3	2.4	8.9	4.92	0.15	13.1	0.0	0.0	fail	0				
293	RMC Land, Harlington	SIE00 3921E	43	40	33	86	0.116	0.23															
294	Haddenham CE	C540.839.60	40	40	29	89	0.114	0.26	-18.61	0.1	2.0	20.7	6.24	0.16									
295	Haddenham CE	C391D	49	38	26	79	0.145	0.24	-20.75	0.1	2.5	20.4											
296	Haddenham CE	C474D	37	40	31	58	0.133	0.24	-22.41	0.1	2.3	24.0	5.08	0.19	18.9	0.0	0.0	fail	0				
297	Haddenham CE	C475D	50	56	36	46	0.100	0.28	-19.90	0.1	1.6	13.4	5.26	0.17									
298	Haddenham CE	C3965K	67	37	36	89	0.126	0.24	-21.79	0.1	3.6	29.2	4.36	0.21									

Appendix III: Data (amino-acid ratios)

ID	Site	Site ID	AMINO ACID ANALYSIS															
			Asp	Glu	Ser	Gly	Thr	Ala	Arg	Tyr	Val	Met	Phe	Ile	Leu	Lys	ohP	Pro
			Mole %															
35	Holloway Lane	HL 87 B66	4.2	8.8	3.6	31.6	1.9	11.9	5.5	0.1	2.5	0.3	1.4	1.3	2.6	2.8	9.7	11.7
99	Berinsfield	BERWC74 Grave 1	4.6	8.5	3.7	25.7	2.2	12.5	6.3	0.1	3.2	0.6	1.6	1.3	3.0	3.0	10.8	12.9
107	Berinsfield	BERWC74 Grave 28	4.4	9.0	3.8	28.1	2.1	11.9	6.0	0.2	3.1	0.5	1.5	1.3	2.9	3.1	9.9	12.4
117	Berinsfield	BERWC74 Grave 92	4.3	9.2	3.4	32.1	1.9	11.8	5.7	0.0	2.8	0.3	1.3	1.1	2.6	2.7	9.3	11.5
126	Berinsfield	BERWC74 Grave 148	4.1	8.8	3.4	30.4	1.9	11.7	5.9	0.1	2.9	0.4	1.3	1.1	2.6	2.9	10.1	12.4
127	Berinsfield	BERWC74 Grave 149	3.9	8.7	3.4	32.0	1.8	12.3	5.6	0.0	2.9	0.3	1.2	1.0	2.3	3.3	9.6	11.8
131	Huntsman's Quarry	HWCM 21698 1103 Bag 2	4.3	9.0	4.0	31.9	1.8	10.9	5.7	0.1	2.4	0.1	1.4	1.3	2.7	3.1	9.5	11.8
159	Bestwall Quarry	BQ98G (55)	4.3	8.8	3.5	27.9	1.9	11.5	6.2	0.2	2.5	0.1	1.5	1.4	2.9	3.6	10.6	13.0
162	Bestwall Quarry	BQ99G (525)	4.4	9.1	3.4	28.8	1.9	11.8	5.9	0.2	2.7	0.0	1.4	1.3	2.7	3.2	10.5	12.6
165	Bestwall Quarry	BQ99H (88)	4.3	8.9	4.1	30.8	2.0	11.1	5.8	0.2	2.3	0.4	1.4	1.2	2.5	2.2	10.3	12.3
166	Bestwall Quarry	BQ99H (427)	4.1	9.0	3.5	29.5	2.1	11.1	5.9	0.2	2.7	0.5	1.5	1.2	2.8	3.1	10.3	12.5
178	Etton CE	B16299	4.2	9.4	3.3	30.2	1.9	11.2	5.8	0.2	2.7	0.2	1.5	1.1	2.7	2.8	10.1	12.8
179	Etton CE	B16300	4.2	9.5	3.5	31.7	1.8	11.2	5.5	0.2	2.6	0.1	1.4	1.1	2.6	2.6	9.6	12.3
182	Etton CE	B14826	0.8	0.0	6.7	24.7	1.7	6.9	3.0	2.0	1.9	7.8	1.1	5.1	7.3	15.2	5.6	10.3
210	Cleveland Farm	W257 1149E	4.3	9.1	4.0	28.9	1.9	11.1	6.1	0.3	2.5	0.2	1.5	1.4	2.9	3.0	10.2	12.6
234	Kingsmead Quarry	H54635 1980A	4.3	9.7	3.7	29.1	1.9	11.1	6.2	0.1	2.5	0.1	1.3	1.3	2.8	3.4	10.7	11.9
266	RMC Land, Harlington	SIE00 876C	4.2	9.2	3.3	28.8	1.9	11.9	6.0	0.1	2.6	0.0	1.4	1.3	2.5	3.3	11.0	12.7
280	RMC Land, Harlington	SIE00 2741B	4.4	9.3	4.1	30.5	1.9	10.9	6.1	0.2	2.4	0.1	1.4	1.2	2.6	2.2	10.1	12.7
281	RMC Land, Harlington	SIE00 2741C	4.6	9.4	3.4	28.6	1.9	11.1	6.1	0.1	2.8	0.0	1.6	1.4	3.0	3.0	10.5	12.6
286	RMC Land, Harlington	SIE00 2742C	4.3	9.2	3.7	28.6	1.9	11.2	6.1	0.2	2.7	0.0	1.5	1.4	2.8	3.0	10.8	12.8