

Client: Your Company  
Contact(s): Your Name  
Analysis:  $\delta^{13}\text{C}$  and  $\delta^{15}\text{N}$  of collagen samples  
IA Ref. No.: Our LIMS Code  
From: Ian Begley  
Date: May 2, 2007

We have completed analysis of the collagen samples which arrived at our laboratory on the April 17, 2007. The results of analysis can be found attachment as an MS Excel worksheet.

### Method

The samples were measured with a duplication rate of 20 % (1 in 5 samples analysed in duplicate) with results for both replicates being reported.

The technique used for this analysis was EA-IRMS (elemental analyzer isotope ratio mass spectrometry). In this technique, samples and reference materials are weighed into tin capsules, sealed, and then loaded into an automatic sampler on a Europa Scientific Roboprep-CN sample preparation module. From there they were dropped into a furnace held at 1000 °C and combusted in the presence of oxygen. The tin capsules flash combust, raising the temperature in the region of the sample to ~ 1700 °C. The combusted gases are swept in a helium stream over a combustion catalyst ( $\text{Cr}_2\text{O}_3$ ), copper oxide wires (to oxidize hydrocarbons), and silver wool to remove sulphur and halides. The resultant gases ( $\text{N}_2$ ,  $\text{NO}_x$ ,  $\text{H}_2\text{O}$ ,  $\text{O}_2$ , and  $\text{CO}_2$ ) are swept through a reduction stage of pure copper wires held at 600 °C. This removes any oxygen and converts  $\text{NO}_x$  species to  $\text{N}_2$ . A magnesium perchlorate chemical trap removes water. Nitrogen and carbon dioxide are separated by a packed column gas chromatograph held at an isothermal temperature of 65 °C. The resultant chromatographic peaks enter the ion source of the Europa Scientific 20-20 IRMS where they are ionised and accelerated. Gas species of different mass are separated in a magnetic field then simultaneously measured on a Faraday cup universal collector array. For  $\text{N}_2$ , masses 28, 29, and 30 are monitored and for  $\text{CO}_2$ , masses 44, 45, and 46 are monitored.

Both references and samples are converted to gases and analysed in this manner. The analysis proceeds in a batch process, whereby a reference is analysed followed by a number of samples and then another reference.

## Reference Standards

The reference material used during analysis of all samples was IA-R042 (powdered bovine liver,  $\delta^{15}\text{N}_{\text{Air}} = 7.65 \text{ ‰}$ ,  $\delta^{13}\text{C}_{\text{V-PDB}} = -21.60 \text{ ‰}$ ). IA-R042 is traceable to IAEA-N-1 (ammonium sulphate,  $\delta^{15}\text{N}_{\text{Air}} = 0.40 \text{ ‰}$ ) and IAEA-CH-6 (sugar,  $\delta^{13}\text{C}_{\text{V-PDB}} = -10.43 \text{ ‰}$ ). IA-R042 was chosen as the reference material as, with a carbon content = 48.97 % and nitrogen content = 10.05 %, it closely matches the organic matrix of your samples.

Reference standards IA-R042, IAEA-N-1 (Ammonium Sulphate,  $\delta^{15}\text{N}_{\text{Air}} = 0.40 \text{ ‰}$ ), IAEA-N-2 (Ammonium Sulphate,  $\delta^{15}\text{N}_{\text{Air}} = 20.30 \text{ ‰}$ ), IAEA-CH-6 (Sucrose,  $\delta^{13}\text{C}_{\text{V-PDB}} = -10.43 \text{ ‰}$ ) and IA-R005 (Beet Sugar,  $\delta^{13}\text{C}_{\text{V-PDB}} = -26.03 \text{ ‰}$ , traceable to IAEA-CH-6) were measured as quality control check samples during analysis. Results for the quality control check samples are included in the results file.

The International Atomic Energy Agency, Vienna, distributes IAEA-CH-6 and IAEA-N-1 and IAEA-N-2 as inter-comparison standards.

The remaining sample material will be returned to you upon request. Otherwise it will remain in storage for a period of 3 months after which time it may be disposed of.

Please do not hesitate to contact us if you have any queries regarding the analysis of your samples.

Analysed and Reported by:

Verified by:

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## Iso-Analytical Laboratory Report

### Client Details

Name: Your Company  
Contact: Your Name

### Sample Details

Number: 15  
Material: Collagen

### Sample Tracking

IA Reference No.: Our LIMS Code  
Date of Arrival: April 17, 2007

### Analysis Details

Isotope(s):  $\delta^{13}\text{C}$  &  $\delta^{15}\text{N}$   
Method: EA-IRMS  
Report Date: May 2, 2007

Duplicate Results shown in **BOLD**

Sample Number	Date	Bone	Nitrogen (%)	$\delta^{15}\text{N}_{\text{Air}}$ (‰)	Mean $\delta^{15}\text{N}_{\text{Air}}$ (‰)	Carbon (%)	$\delta^{13}\text{C}_{\text{V-PDB}}$ (‰)	Mean $\delta^{13}\text{C}_{\text{V-PDB}}$ (‰)	C:N Ratio
<b>T29</b>	<b>Late Middle Ages</b>	<b>phalanx</b>	<b>15.29</b>	<b>8.32</b>		<b>42.63</b>	<b>-19.61</b>		<b>2.79</b>
<b>T29</b>	<b>Late Middle Ages</b>	<b>phalanx</b>	<b>14.39</b>	<b>8.38</b>	<b>8.35</b>	<b>40.21</b>	<b>-19.64</b>	<b>-19.62</b>	<b>2.79</b>
<b>T62</b>	<b>Late Middle Ages</b>	<b>phalanx</b>	<b>15.93</b>	<b>10.73</b>		<b>44.21</b>	<b>-19.51</b>		<b>2.78</b>
<b>T62</b>	<b>Late Middle Ages</b>	<b>phalanx</b>	<b>14.10</b>	<b>10.75</b>	<b>10.74</b>	<b>39.05</b>	<b>-19.56</b>	<b>-19.53</b>	<b>2.77</b>
<b>T114</b>	<b>Late Middle Ages</b>	<b>phalanx</b>	<b>14.24</b>	<b>8.04</b>		<b>39.98</b>	<b>-20.28</b>		<b>2.81</b>
<b>T114</b>	<b>Late Middle Ages</b>	<b>phalanx</b>	<b>13.31</b>	<b>8.09</b>	<b>8.07</b>	<b>37.43</b>	<b>-20.23</b>	<b>-20.26</b>	<b>2.81</b>
M04	Early Middle Ages	phalanx	16.19	9.48		44.9	-18.88		2.77
M37	Early Middle Ages	phalanx	11.10	10.54		31.04	-18.73		2.80
M41	Early Middle Ages	phalanx	14.19	10.80		39.41	-18.57		2.78
M59	Early Middle Ages	phalanx	12.90	9.36		36.09	-18.88		2.80
M64	Early Middle Ages	phalanx	13.21	8.60		36.95	-17.80		2.80
M68	Early Middle Ages	phalanx	14.15	9.82		39.5	-18.87		2.79
M108	Early Middle Ages	phalanx	8.02	8.63		22.93	-19.72		2.86
M121	Early Middle Ages	phalanx	9.42	12.78		26.71	-18.74		2.84
M126	Early Middle Ages	phalanx	13.61	10.74		38.14	-18.09		2.80
M150	Early Middle Ages	phalanx	13.20	10.13		36.94	-18.81		2.80
M181	Early Middle Ages	phalanx	13.79	10.22		38.8	-19.06		2.81
M197	Early Middle Ages	phalanx	12.79	11.10		35.43	-19.10		2.77

### Quality Control Check Samples

	IA-R042 (Bovine Liver)		IAEA-N-1 (Ammonium Sulphate)	IAEA-N-2 (Ammonium Sulphate)	IAEA-CH-6 (Sucrose)	IA-R005 (Beet Sugar)
	$\delta^{15}\text{N}_{\text{Air}}$ (‰)	$\delta^{13}\text{C}_{\text{V-PDB}}$ (‰)	$\delta^{15}\text{N}_{\text{Air}}$ (‰)	$\delta^{15}\text{N}_{\text{Air}}$ (‰)	$\delta^{13}\text{C}_{\text{V-PDB}}$ (‰)	$\delta^{13}\text{C}_{\text{V-PDB}}$ (‰)
	7.56	-21.53	0.55	20.42	-10.43	-26.12
	7.66	-21.66	0.65	20.32	-10.49	-26.08
	7.60	-21.41				
Mean	7.61	-21.53	0.60	20.37	-10.46	-26.10
St. Dev.	0.05	0.12	0.07	0.07	0.04	0.03
N	3	3	2	2	2	2
Accepted Value	7.65	-21.60	0.40	20.30	-10.43	-26.03

end of results table