Paleodietary reconstruction

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Introduction

Bones are living structures and require nutrients throughout an individual's life. Due to a number of metabolic processes occurring in all living organisms, dietary signals from food and nutrients are incorporated into an organism's tissues – including bone - and can be identified through the analysis of carbon and nitrogen isotopic concentrations (Ambrose & Norr,1993, Schwarcz & Schoeninger, 1991). Stable isotope analysis of carbon ($\delta^{13}C$) and nitrogen ($\delta^{15}N$) ratios in bulk collagen has proven to be a resourceful technique in the study of past populations diet and lifestyle. This technique can discriminate between broad categories of foods, distinguishing between the consumption of marine and terrestrial protein owing to the enriched carbon ($\delta^{13}C$) values of marine organisms; as well as differentiating between two groups of plants that use different photosynthetic pathways, C_4 plants (i.e. millet, sorghum, sugar cane, maize) and C_3 plants (i.e. the majority of crops, fruits and vegetables growing in temperate climates). In addition, nitrogen ($\delta^{15}N$) values provide an indication on the position of an individual within the food chain, where organisms become increasingly enriched in ^{15}N with each trophic level (Katzenberg,2008).

Materials and method

Four individuals (575-680 CE) from the archaeological site Casa dos Mosaicos Romanos in Rua Antonio Joaquim Granjo, Setubal were sampled and analysed for carbon ($\delta^{13}C$) and nitrogen ($\delta^{15}N$). The methodology followed a standard collagen extraction method (Longin, 1971) with an additional ultrafiltration step (Brown *et al*. 1988). Collagen samples were analysed in duplicate using isotope ratio mass spectrometry (IRMS) with a Sercon 20-22 at the BioArCh facilities, University of York. Isotopic values are reported following standard practice as the ratio of the heavier isotope to the lighter one (δ values in parts per mille ‰) relative to internationally defined standards for carbon $^{13}C/^{12}C$ (VPDB: Vienna Pee Dee Belemnite) and nitrogen $^{15}N/^{14}N$ (AIR). The analytical error, calculated from repeated measurements of each sample, a bovine control, and international standards, was <0.2‰ (1σ) for both $\delta^{13}C$ and $\delta^{15}N$. Sufficient collagen was extracted in all samples (>1%) and collagen quality met published criteria (De Niro, 1985; van Klinken, 1999).

Results and discussion

The results are presented in Table 1 and illustrated in Figure 1 in relation to other published isotopic data from the same geographical area.

Three out of four individuals show relatively enriched nitrogen and carbon values, while one individual (AJG 8) has lower values. The associated enrichment in both ^{13}C and ^{15}N suggests a significant reliance on marine resources for this group of individuals. Consumption of C_4 crops is unlikely due to both the extremely high $\delta^{15}N$ values and lack of C_4 fodder in contemporary herbivores from nearby Monte da Cegonha (Saragoça *et al*., 2016, Figure 1) The estimated contribution of marine resources to the diet has been calculated

Table 1 - List of analysed individuals and the relative carbon and nitrogen ratios. The percentage of marine resources contribution to diet is calculated as per the equation Marine2 (Arneborg *et al.,* 1999; Richards & Hedges, 1999)

Sample ID	Sex	Age	C:N	%C	%N	$\delta^{13}C_{coll}$	$\delta^{15}C_{coll}$	% of marine resources
AJG 10	M	Adult	3.23	40.63	14.74	-16.29	12.91	52.4
AJG 7	I	Juvenile	3.23	41.44	14.95	-16.12	16.09	54.2
AJG 9	I	Juvenile	3.18	40.82	15.01	-16.89	12.87	45.7
AJG 8	I	Adolescent	3.15	38.56	14.30	-17.21	11.30	42.1

based on the $\delta^{13}C$ values as per the equation Marine2 (Arneborg *et al*., 1999; Richards & Hedges 1999) which indicated a diet where marine protein makes up about 42% for all individuals with a highest value of 54%. These estimations take into account the $\delta^{13}C$ ratio in bulk collagen, which represents solely the protein fraction of the diet, leaving aside the contribution of lipids and carbohydrates. Therefore, although almost half of their protein intake was based on marine resources, it is expected that a large component of the diet were carbohydrates and lipids, whose contribution is not detectable in collagen when protein consumption is adequate (Ambrose & Norr, 1993).

The diet of Portuguese archaeological populations is largely unknown for the historical period. While a series of studies have been performed on Mesolithic and Neolithic individuals, only one recent paper presented the first data on an historical population's diet. Saragoça and colleagues (2016) analysed both faunal and human remains from the Late Antique site of Monte da Cegonha (predominantly 7th CE). The results showed this population had a mainly terrestrial and rather homogeneous diet, based on terrestrial animal and C_3 plants (fig. 1). The distribution of the human individuals reflects the diet of the herbivores from the same site and chronology, indicating that this population relied mostly on terrestrial protein. The difference between the values of Setubal AJG individuals and Monte da Cegonha is statistically significant for both carbon and nitrogen (Mood's median test $\delta^{13}C$ values $p=0.02$, $\delta^{15}N$ values $p=0.04$). This striking difference between the two assemblages is further appreciated when the Portuguese Mesolithic individuals from Muge and Sado are considered (Lubell *et al*., 1994;, Umbelino *et al*., 2007). Regardless of the big chronology gap of these population, the Mesolithic isotopic values and their relative reliance on marine food is closer to the AJG individuals rather than the inland individuals from Monte da Cegonha, contemporaneous with the Setubal population. When the AJG individuals are further compared to other European contemporaneous populations, the high reliance on marine resources remains a peculiar element, since the majority of the studies reported a rather uniform terrestrial diet for this period (Fuller *et al*., 2006; Sandias & Müldner, 2015; Mion *et al*., 2016; Rissech *et al*., 2016; Lubritto *et al*., 2017). The only other populations showing similar values for both nitrogen and carbon are the Vikings from Orkney (550-1066 CE) and two Galician populations (2nd-7th C CE), whose economy and subsistence was mainly based on fishing (Richards *et al*., 2006; Kaal *et al*., 2016; López-Costas & Müldner, 2016). This first comparison might suggests that the marine vocation of Setubal was preserved during the Late Antique/Visigoth period and fishing was still widely practiced, supplying the markets of the town with abundant marine fish. On the other hand, caution is necessary when dealing with such a small sample size and the values showed by these individuals might not be representative of the dietary trends

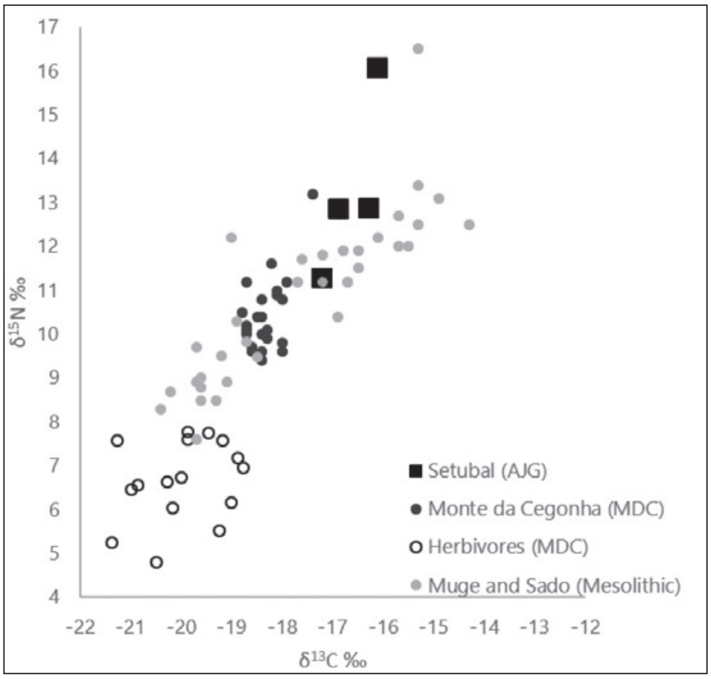

Figure 1 - Plot showing the values of $\delta^{13}C$ and $\delta^{15}N$ for the AJG-Setubal individuals (red squares), compared to the Late Antique individuals from Monte da Cegonha (Saragoça *et al.,* 2016) and the Mesolthic individuals from Muge and Sado (Lubell *et al.,* 1994, Umbelino *et al.,* 2007).

of Setubal during the Visigoth period, but rather portraying a specific dietary choice or occupation (fishing) of these particular individuals.

Conclusion

Although the scarcity of isotopic studies on Portuguese historical populations and the small sample size of this assemblage from Setubal, it is remarkable that two contemporaneous populations showed a substantial difference in their diet. This difference in consumption might reflect differences in subsistence strategies because of economic, political and social dynamics as well as geographical and environmental factors, with Setubal being on the coast and Monte da Cegonha (7th C) located inland. Even these few individuals add significant data to aid in our understanding of the diversity of diet and economy in Western Iberia during this period. Further exploration is necessary in order to ascertain whether the consumption of marine resources was prominent in Setubal for the vast majority of the population or if these values are an expression of single individuals' dietary preferences. Dietary results from a wider chronological period, and especially the Roman time, would be particularly beneficial to determine the impact of the fish processing industry, taking place in Setubal, on the level of marine resources consumed by the local population.

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