The transition to agriculture in south-western Europe: new isotopic insights from Portugal's Atlantic coast

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For the past fifteen years a succession of stable isotope studies have documented the remarkable abrupt dietary transition from the Mesolithic to the Neolithic in Western and Northern Europe. The key region of Portugal, with Late Mesolithic shell middens and burials apparently coexisting with the earliest Neolithic, provides further illustration of the nature of that transition. Individuals from Neolithic contexts there had significantly different diets from their Mesolithic counterparts. No evidence was found for a transitional phase between the marine-oriented Mesolithic subsistence regimes and the domesticated, terrestrial Neolithic diet. Two later Neolithic individuals, however, showed evidence for partial reliance on marine or aquatic foods.

This raises questions about the possible persistence of marine dietary regimes beyond the Mesolithic period. This article is followed by a brief note by Mary Jackes and David Lubell.

Keywords: Portugal, Mesolithic, Neolithic transition, stable isotope analysis, carbon, nitrogen, diet

Methods

Sampling protocols were aimed at capturing a large number of individuals from a variety of sites representing different geographical regions, time periods and funerary contexts (Table S1). Bone materials from 284 individuals excavated from 26 Neolithic sites were selected for sampling. An additional 68 individuals from five Mesolithic sites in the Sado Valley were also sampled as part of this project and are considered in detail elsewhere (Guiry *et al*. 2015). With the exception of Carcavelos (with 35 left femora and 6 mandibles) and Carrascal (with 3 right femurs and 2 mandibles), sampling was performed on the same element within each site to prevent duplication of data. A literature review was also undertaken to source $\delta^{13}C$ and $\delta^{15}N$ as well as radiocarbon data from additional individuals. Note that some $\delta^{13}C$ data been sourced from radiocarbon measurements and may be less accurate than data produced explicitly for palaeodietary work. A variety of wild and domestic faunal remains (n=43) from five sites were also analysed to provide a stable isotope baseline for human data. Generally, Neolithic faunal and human remains are found in mutually exclusive (i.e. settlement *versus* funerary) archaeological contexts. For that reason, most of the faunal materials analysed here cannot be directly attributed to particular groups of humans.

Samples weighing between 150mg and 1g were cut from specimens and cleaned of visible surface contamination. Collagen was extracted from bone samples following well established methods outlined by Richards and Hedges (1999) as well as Brown and colleagues (1988). In brief, chunks of bone were demineralised in 0.5M hydrochloric acid and collagen pseudomorphs were gelatinised in water adjusted to a pH value of 3 and heated to 75°C for 48 hours. Gelatins were then filtered sequentially using Ezee filters (5–8μm mesh) and ultrafilters (30kd). Filtered gelatins were frozen and lyophilised in a freeze-dryer for 48 hours.

Stable carbon and nitrogen isotope measurements were performed on 0.5mg of collagen. Where collagen yields allowed, samples were run in duplicate. Measurements were made using a Thermo-Finnigan Delta Plus XL isotope ratio mass spectrometer coupled via continuous flow to a Calro Erba elemental analyser at the Department of Human Evolution, Max Planck Institute for Evolutionary Anthropology in Leipzig, Germany. Instrumental error for $\delta^{13}C$ and $\delta^{15}N$ measurements was ±0.1‰ and ±0.2‰, respectively. Statistical analyses (a One Way ANOVA followed by a post hoc Bonferroni test [significance level set at 0.5]) were performed using SPSS 17.0. Stable isotope measurements are considered acceptable if derived from a sample with carbon to nitrogen (C:N) ratios between 2.9 and 3.6 and carbon and nitrogen concentrations above 16% and 8%, respectively (DeNiro 1985; Van Klinken 1999).

Calibration of new and previously published radiocarbon dates was undertaken in OxCal v4.2 using the Intcal13 and Marine13 calibration curves for atmospheric and marine carbon (Reimer *et al.* 2013). All dates were calibrated to an uncertainty of 2σ and are presented as median values to facilitate concise visual representation in figures. Where relevant, marine dietary protein intake was calculated using conservative $\delta^{13}C$ endpoints of -20‰ and -12‰ to represent pure terrestrial (C_3) and marine consumers, respectively. Marine dietary contributions were calculated to the nearest percent using a linear mixing equation ([|-20‰| - |Consumer|]/8 = % marine dietary contribution). Radiocarbon dates for individuals with ≥25% marine dietary protein contribution were calibrated to offset marine reservoir effects. Affected dates were corrected using the 'marine/mixed curve' option in OxCal v4.2, employing the Portugal marine mean reservoir offset (ΔR) of 256±29 years and a dietary uncertainty of ±10% marine carbon (Meiklejohn *et al.* 2009). Individuals with a marine dietary contribution of <25% were calibrated strictly using the Intcal13 terrestrial curve (Reimer *et al.* 2013). Freshwater reservoir effects were not taken into account due to the lack of a suitable freshwater ΔR value for the region.

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