A Paleoenvironmental Reconstruction using Stable Isotopes of Red Deer (*Cervus elaphus*) Enamel at Lapa do Picareiro (Portugal)

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Background

Carbon and oxygen isotope compositions of bone collagen and tooth enamel from archaeological remains are tools frequently used in the reconstructions of the paleodiets and paleoenvironments of Neanderthals and anatomically modern humans (AMHs). Oxygen isotope values ($\delta^{18}O$) can mirror local rainfall and humidity while carbon isotope values ($\delta^{13}C$) can reflect C3/C4 plant cover distributions by using animal diet as a proxy (Schoeninger and Moore 1992; Schoeninger 1995). This poster presents preliminary stable carbon and oxygen isotope results from intra-tooth enamel samples from Lapa do Picareiro, a stratified cave site in Portuguese Estremadura, in an attempt to detect environmental changes during the Late Pleistocene and Early Holocene using red deer (*Cervus elaphus*). Picareiro is situated in Portuguese Estremadura (Fig. 2) and has evidence of human occupations spanning the Middle Paleolithic to the Bronze Age. It is one of three sites in southern Iberia to have Middle Paleolithic occupations dated post Heinrich Event 4. The teeth sampled here assess five different Middle and Upper Paleolithic levels from of the site: A, K, U, FF and JJ.

We then make a series of comparisons between our stable isotope results and: 1. red deer $\delta^{13}C$ data from Western European archaeological sites dating to the Magdalenian, 2. modern European temperate forest $\delta^{13}C$ values, and 3. geoarchaeological data from Picareiro (Benedetti et al. in review). In addition, we assess seasonality by way of intra-tooth sampling. These results will shed light on the environmental conditions in which Neanderthals and anatomically modern humans adapted, ultimately affecting their subsistence strategies, technology, mobility and settlement patterns.

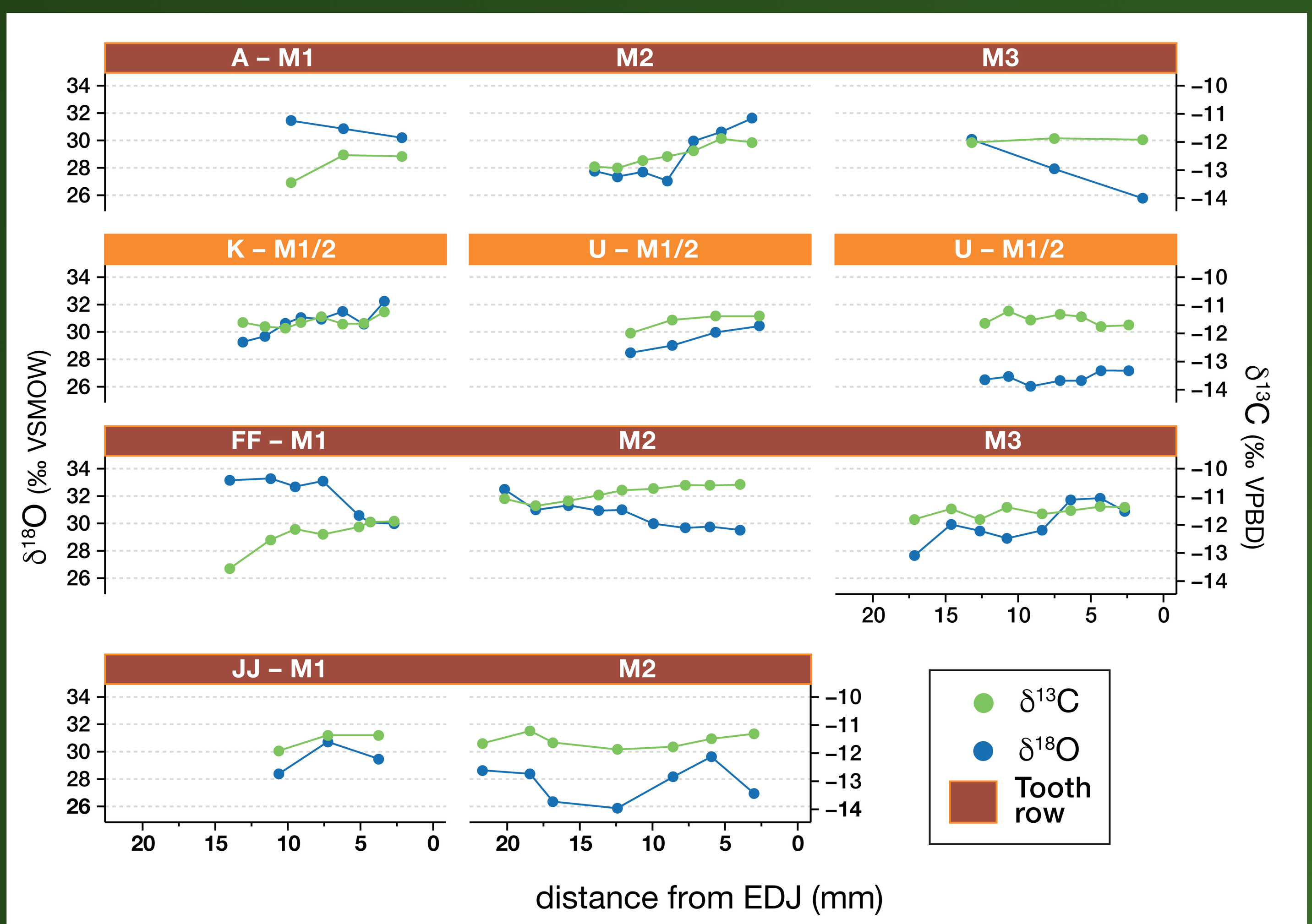

Fig 1. Tooth profiles showing carbon and oxygen results. Orange headers indicate isolated teeth while maroon headers indicate teeth from same tooth row. Note lack of strong sinusoidal patterning.

Methods

Stable Isotopes: Both RIT and UNM laboratories followed the same protocol. We sampled the right mandibles from five individuals (see table 1). Using a high-speed drill with a diamond bit, we sampled each tooth at equal intervals from the root to the crown of the tooth to create a homogenized powder. Pretreatment protocol followed Koch et al. 1997. Each enamel sample was sonicated for 15 minutes in a 2-3% NaOCl solution and then 10 minutes in 0.1M acetic acid. Samples were rinsed three times with DI H_2O after each treatment and air-dried. About 3-10mg of enamel apatite was weighed into glass extainer vials and were subjected to a phosphoric acid reaction at 50°C degrees for six hours. The CO_2 product of this reaction was measured on a Thermo-Scientific GasBench from Bremen, Germany, coupled to a Delta V isotope ratio mass spectrometer at the University of New Mexico Center for Stable Isotopes. Samples analyzed in the Stable Isotope Ratios in the Environment Laboratory (SIREAL) at the University of Rochester were reacted with phosphoric acid at 70°C for one hour.

Comparison of ancient and modern regional carbon values: Using data synthesized in Stevens et al. (2014), we compared our red deer $\delta^{13}C_{bioapatite}$ values to red deer $\delta^{13}C_{collagen}$ values obtained from various archaeological sites in different regions of Europe that dated to the Magdalenian, the only chronologically compatible time period between data sets. In order to make all $\delta^{13}C_{bioapatite}$ and $\delta^{13}C_{collagen}$ data comparable, all $\delta^{13}C$ values were converted to reflect $\delta^{13}C_{diet}$, which in the case of the red deer, is a proxy for the local vegetation. The fractionation factor between $\delta^{13}C_{tissue}$ -$\delta^{13}C_{diet}$ used here is 5‰ for collagen (Ambrose and Norr 1993) and 14‰ for bioapatite (Cerling and Harris 1999). For bioapatite, an additional correction accounting for the offset caused by differences in atmospheric CO_2 through time was made (Keeling et al. 2010).

Lvl.	Lab Number	Material	Cal 2σ Range (cal yr BP)	Tooth	# of Intra-tooth samp.	Cultural Affiliation
A	<9,475 cal yr BP			M2	7	Iron age
K	Wk-31354	bone	18,512-18,006	M1/2	8	Magdalenian
U	Beta-234373	charcoal	27,220-26,525	M1/2	7	Gravettian
	Beta-208222	charcoal	27,461-26,395	M1/2	8	
	Beta-234374	charcoal	27,246-26,550			
FF	UGAMS-20479	charcoal	36,355-35,800	M1	7	Middle Paleolithic
	Wk-41259	charcoal	38,524-36,381	M2	9	
	Beta-247964	charcoal	33,485-31,705			
	Wk-32219	bone	38,086-36,355	M3	7	
	Wk-28843	bone	32,291-31,087			
JJ4	Wk-28844	bone	46,381-42,094	M1	4	
	UGAMS	charcoal	45,420-44,451	M2	7	

Table 1. Picareiro radiocarbon dates for the levels targeted in this study and sample information

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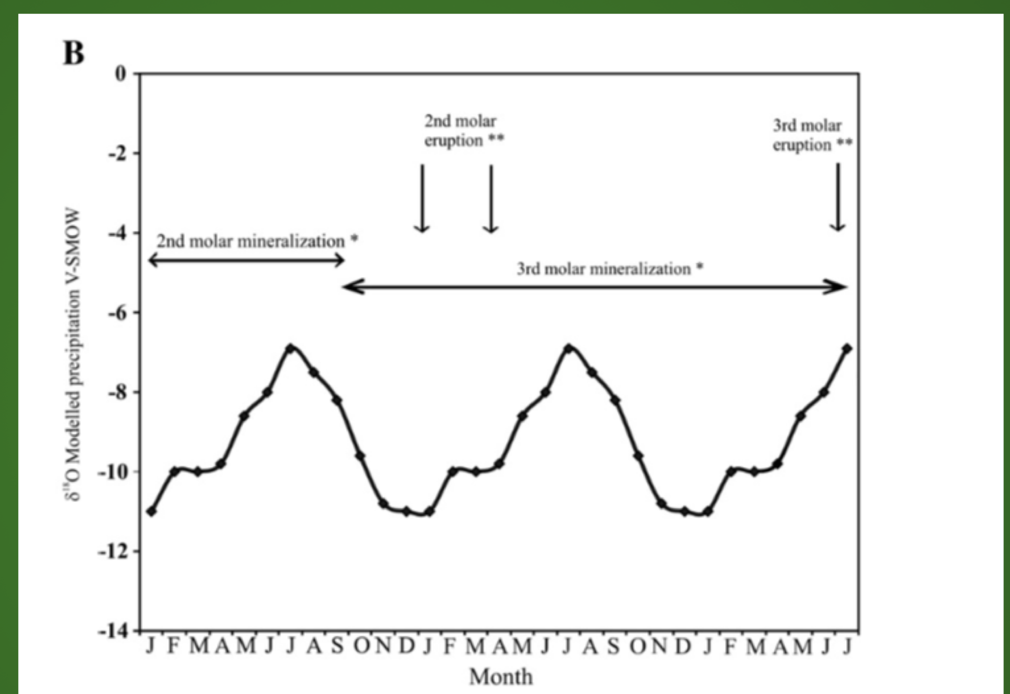
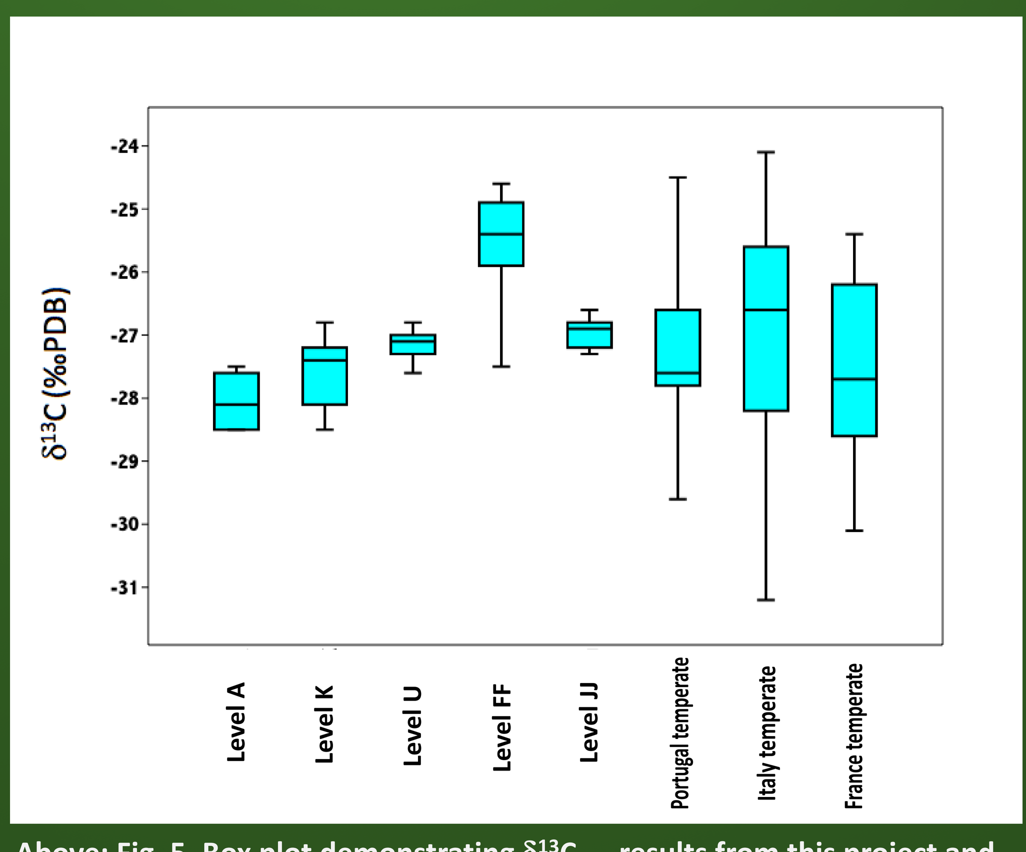
Above: Fig. 2. Location of Lapa do Picareiro.

Below: Fig. 3. Intra-tooth sampling of red deer teeth.

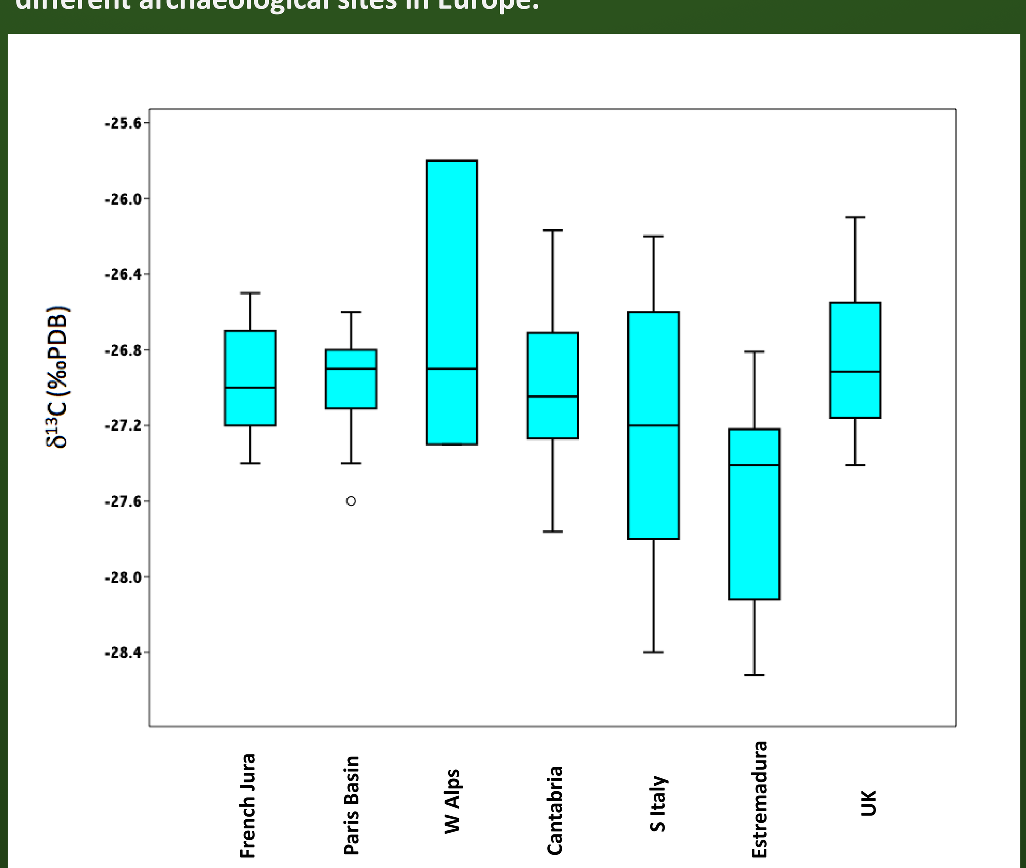


Above: Figure 4. Tooth development and mineralization rates for a modern red deer population over time. (Figure: Stevens et al. 2011).



Above: Fig. 5. Box plot demonstrating $\delta^{13}C_{diet}$ results from this project and $\delta^{13}C$ values of modern temperate forests (Hemming et al. 2005).

Below: Fig 6. Box plot demonstrating $\delta^{13}C_{diet}$ comparisons with data from different archaeological sites in Europe.



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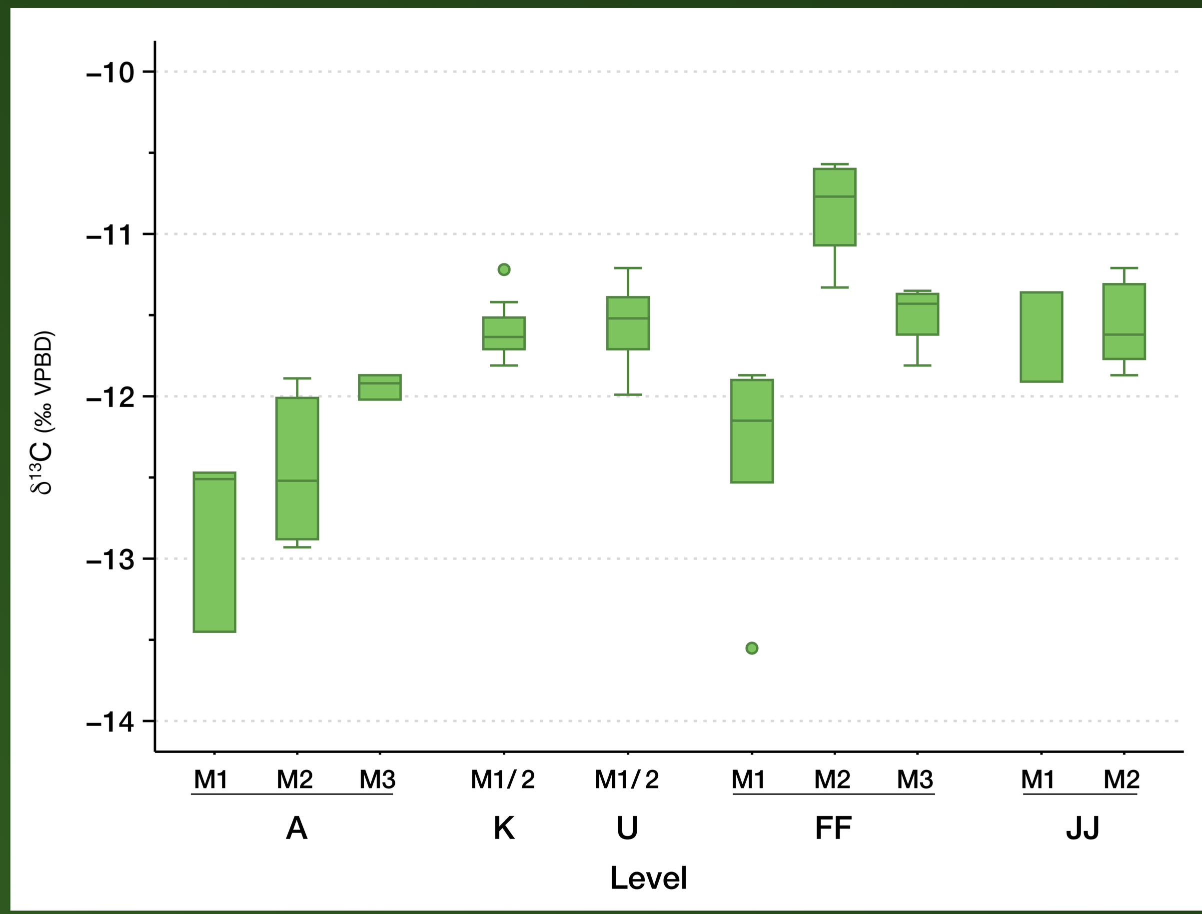
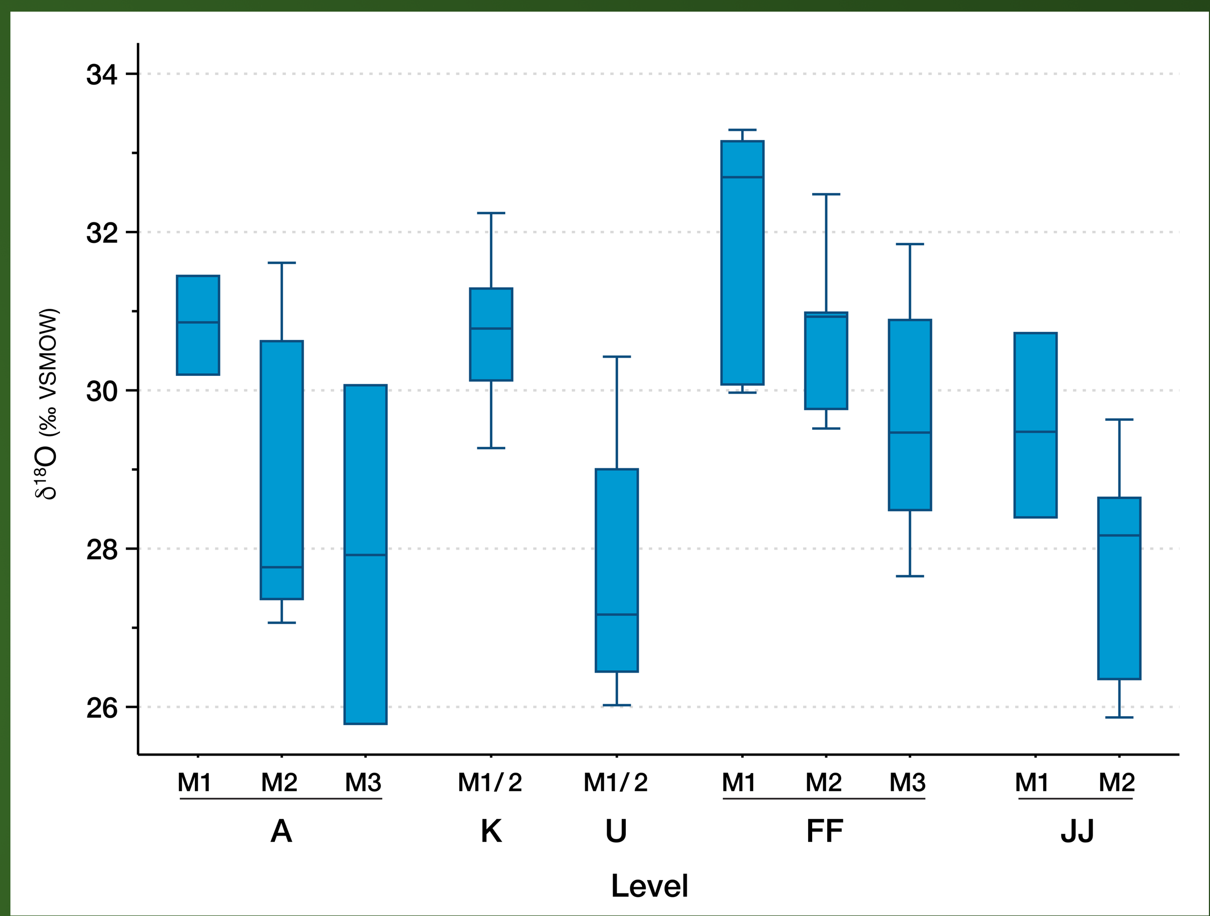


Above: Fig. 7. Boxplot of $\delta^{13}C$ results per tooth.

Below: Fig. 8. Boxplot of $\delta^{18}O$ results per tooth. Note the enrichment of M1 and to a lesser extent M2 values due to suckling maternal milk (Stevens et al. 2011).



Results/Discussion

Previous analyses indicated that $\delta^{13}C$ values obtained were consistent between UNM and RIT labs and that $\delta^{18}O$ values were interpretively similar (Carvalho et al. 2017). Although there are fractionation factors in $\delta^{15}N$ and $\delta^{18}O$ isotopes between a mother's tissue and suckling young, $\delta^{13}C$ values from M1 do not seem to be depleted due suckling maternal milk and if these effects exist they are negligible (Fig 7; Balasse 2002). The effect of suckling, however, is clearly visible in the enrichment of $\delta^{18}O$ values in M1 (mineralizes in utero/during lactation) and M2 (mineralizes during partial lactation; Fig. 8; Brown and Chapman 1991; Stevens et al. 2011).

In terms of seasonality, neither the $\delta^{13}C$ nor the $\delta^{18}O$ intra-tooth results demonstrate a strong sinusoidal patterning that would be indicative of seasonal climate variation (See Figs. 1 & 4). This could be due to sampling intervals being shorter than the enamel maturation time and isotopic shifts recorded in an individual's tooth, resulting in isotope ratios that don't reflect the full breadth of seasonal change (Passey and Cerling 2002).

The $\delta^{13}C_{diet}$ values for all levels of Picareiro fall within the range of $\delta^{13}C$ values for modern temperate forests in Portugal, Italy and France, suggesting that during time periods represented by targeted levels, the local landscape is composed of a temperate forested landscape in which red deer foraged (Fig. 5; Hemming et al. 2005). This does not suggest that the environmental and/or climate did not change during the time periods targeted here but that the temperate forest environment was present in this region during warmer phases of the Late Pleistocene. More data is needed to conclusively test this.

Red deer $\delta^{13}C_{diet}$ values obtained from archaeological remains in southern Italy and Estremadura are slightly lower than those of the other regions (See Fig. 6). However, all values are consistent with those of modern European temperate forests (Hemming et al. 2005). The slightly depleted $\delta^{13}C_{diet}$ values from these two southern regions may reflect an environment with more canopy cover and shaded forest floors where the evaporative stress is slightly less in comparison to more northern regions (Quade et al. 1995).

There were no significant correlations between the $\delta^{13}C$ and $\delta^{18}O$ values presented here and the geoarchaeological data from Picareiro. The $\delta^{13}C$ values suggest the presence of a temperate forest in targeted levels and reflect only vegetation, not other factors affecting sedimentation in the cave. It is also important to consider that $\delta^{18}O$ ratios of an animal can be affected (to varying degrees) by a suite of environmental factors such as shifting precipitation regimes, geology, altitude, and proximity to the coast, necessitating comparisons between archeological remains and an established modern baseline (Hamilton et al. 2018). Thus, in order to make accurate statements about our $\delta^{18}O$ values and their relationship to climate, we need to create our own modern baselines for Estremadura.

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