Journal of Archaeological Science: Reports 5 (2016) 181–189

Contents lists available at ScienceDirect

Journal of Archaeological Science: Reports

journal homepage: www.elsevier.com/locate/jasrep

Environmental change and economic practices between the third and second millennia BC using isotope analyses of ovicaprid remains from the archeological site of Zambujal (Torres Vedras), Portugal

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ARTICLE INFO

Article history:
Received 11 August 2015
Received in revised form 10 November 2015
Accepted 12 November 2015
Available online xxxx

Keywords:
Portugal
Stable isotope analysis
Fauna
Diet
Environmental change
Neolithic
Copper Age

ABSTRACT

Widespread environmental changes are documented in the Iberian Peninsula between the third and second millennia BC. These include erosion, deforestation, and valley infilling, and are likely due to a combination of the 4.2 kya climate event and anthropogenic activities. Contemporary with these environmental changes are social and cultural transformations associated with the Copper to Bronze Age transition, such as political consolidation or fragmentation, shifts in burial practices, and changes in exchange patterns. The causal links between these environmental and cultural changes have never been tested, however. In this study, we evaluate whether the diet of animal domesticates, which directly respond to ecological changes, occurred. Specifically, we evaluate whether the diet of ovicaprids from the site of Zambujal, Portugal changed significantly over this period. Zambujal is an extensively excavated and comprehensively dated settlement in the Portuguese Estremadura, with faunal remains recovered from all five phases of the site's occupation, extending from the third to early second millennia BC. A sample of 29 ovicaprid mandibles and maxillae, grouped into two diachronically distinct periods of the site's occupation, suggests that changes did occur, as statistically significant differences were found for stable isotopic indicators of diet between the two groups. We suggest that these isotopic differences document the impact of environmental change on cultural changes related to animal husbandry in Portugal during this period.

1. Introduction

1.1. Environmental and cultural transformations in the Late Prehistory of the Iberian Peninsula

Widespread environmental changes are documented in the Iberian Peninsula between the third and second millennia BC. These include erosion, deforestation, and valley infilling. Most of these environmental transformations have been explained in terms of anthropic activities, such as forest clearance and cultivation, although climate change, including the 4.2 kya event, has also been proposed (García et al., 2006; López-Sáez et al., 2014). For example, following millennia of stability, soils at the Neolithic settlement of Encosta de Sant'Ana (Lisbon) were buried by sediments from the mass wasting of hillslopes bordering the site, around 3000 BC (Angelucci et al., 2007). Palynological data from the Segura Mountains of southern Spain demonstrate that mesophilous vegetation peaked between 6400 and 3200 BC, after which xerophytes, suggestive of fires, dominated (Carrión, 2002). At the Bronze Age settlement of Gatas, in southeast Spain, forests and soils degraded over the period of the site's use, between the third and second millennia BC (Castro et al., 1994; Castro et al., 1998:62–68; Castro et al., 1999a, 1999b; Castro et al., 2000; Gale, 1999). Palynological studies in central Spain show that Betulae/Pinus forests declined and heathlands expanded between 3300 and 2000 BC (López-Sáez et al., 2014).

Contemporary with these environmental transformations were the cultural shifts of the Late Neolithic/Copper Age to Bronze Age transition. Between 3500 and 2200 BC (the Late Neolithic and Copper Age), hierarchical societies first emerged in the Iberian Peninsula. These were associated with craft specialization (copper-based metallurgy, ceramics, flaked and groundstone lithics), long-distance trade (in copper, slate, variscite, amphibolite, North African ostrich eggshell and ivory), and sociopolitical differentiation (large fortified or ditched enclosed settlements with smaller satellite settlements) (Chapman, 2003; Gilman, 1987; Schuhmacher, 2012). These larger sites include Los Millares, Marroquíes Bajos, Perdigões, and Zambujal. Burials, which typically were collective, were housed mainly outside of settlements in caves, rockshelters, and megaliths. At the end of this period, between 2200 and 1500 BC, also known as the Early Bronze Age, another set of cultural

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http://dx.doi.org/10.1016/j.jasrep.2015.11.017
2352-409X/

changes occurred, although with significant regional variability. In the Iberian Southwest, most settlements were abandoned, and new ones, such as Agroal (Lillios, 1993), were established, although settlements, in general, are poorly known from this period in this region (Gamito, 2003). As in the earlier Copper Age, burials occurred outside settlement areas, but were housed in individualized stone cist tombs grouped into cemeteries. In the Southeast, most sites were abandoned, and new settlements on terraced and easily defended hilltops were established. These constitute what is known as the Argaric culture (Aranda Jiménez et al., 2015). During the Argaric, stratified societies emerged, and agriculture and craft production intensified. Mortuary practices changed dramatically, and the dead, in individual or double graves, were typically buried underneath houses. Throughout the Peninsula during the Early Bronze Age, long-distance exchange connections were either discontinued or concentrated in more local regional networks.

Despite the synchrony of cultural and ecological changes, Iberian archeologists have generally been reluctant to apply a causal link between the environmental and cultural changes of the Copper and Bronze Ages (though see Fábregas et al., 2003; López-Sáez et al., 2014). This caution is, of course, warranted, as correlation does not imply causation. In order to directly assess the causative role of environmental change on cultural changes for the third to second millennia BC, independently derived data are necessary.

This paper investigates this relationship by evaluating whether the diet of animal domesticates, which directly respond to environmental changes, occurred between the Late Neolithic/Copper Age and Early Bronze Age of the Iberian Peninsula. Specifically, we evaluate whether the diet of the ovicaprids from the settlement site of Zambujal (Fig. 1), in the Estremadura of Portugal, changed significantly over this period.

1.2. Zambujal

Zambujal is a prehistoric fortified settlement, located approximately 3 km west of the town of Torres Vedras and 12 km from the Atlantic Ocean. It is situated on a promontory of a small hill, the Cabeço da Calvina, and near the Ribeira de Pedrulhos, a tributary of the larger Sizandro River (Fig. 2). The site was first settled in the beginning of the 3rd millennium BC and abandoned in the first half of the second millennium BC (radiocarbon dates range from 2900 to 1700 BC) (Kunst and Lutz, 2008). Culturally, it spans the Copper and Early Bronze Ages.

The remains of the fortifications of Zambujal were first discovered in 1930 by Leonel Trindade, who carried out the first excavations there in 1944. From 1959 to 1961, larger excavations were conducted jointly by Trindade and then-director of the museum of Torres Vedras, Aurélio Ricardo Belo (Hall et al., 1964). Between 1964 and 1973, Hermanfrid Schubart from the German Archeological Institute in collaboration with Edward Sangmeister (Institute of Prehistory of the University of Freiburg, Germany) carried out a new series of excavations at Zambujal. These excavations revealed five different phases of construction for the fortifications, with several sub-phases (Sangmeister and Schubart, 1981) (Fig. 3). The last series of excavations were started in 1994 (continued in 1995, 2001, 2002, 2004, 2007, and 2012). These were directed by Michael Kunst of the German Archeological Institute in collaboration with Hans-Peter and Margarethe Uerpmann from the University of Tübingen (Uerpmann and Uerpmann, 2003) and, in 2002, with Elena Morán and Rui Parreira (Kunst et al., 2002). During these campaigns a fourth fortification line was discovered. To this day, some of the remains of the fortification structures at the center of Zambujal stand four meters high and it has been estimated that some of these originally would have been nearly nine meters high, making them some of the most substantial in the Iberian Peninsula during this time period.

Zambujal appears to have had both defensive and domestic functions. Its defensive nature is suggested by its walls and towers, whose five different construction phases reveal the evolution of defensive strategies. However, within the site's walls archeologists have recovered the remains of round houses with hearths, a significant amount of faunal and botanical remains, as well as pottery, stone, bone, and copper artifacts, and personal ornaments, such as beads, (Jiménez Gómez, 1995; Kunst, 1987; Kunst, 1995; Sangmeister and Schubart, 1981; Uerpmann and Uerpmann, 2003). Features and objects associated with copper manufacture were also recovered within the site's walls (Müller et al., 2007; Sangmeister, 1995).

1.3. Paleoecology of the Estremadura, Neolithic through Bronze Age

The Estremadura region of west-central Portugal borders the Atlantic coast and encompasses both the Lisbon and Setúbal peninsulas. Bounded by the Mondego River in the North and harboring the large estuary systems of the Tagus and Sado to the South, the region has attracted human occupation since the Late Pleistocene. Today, the Estremadura occupies a climatic zone referred to as subtropical dry

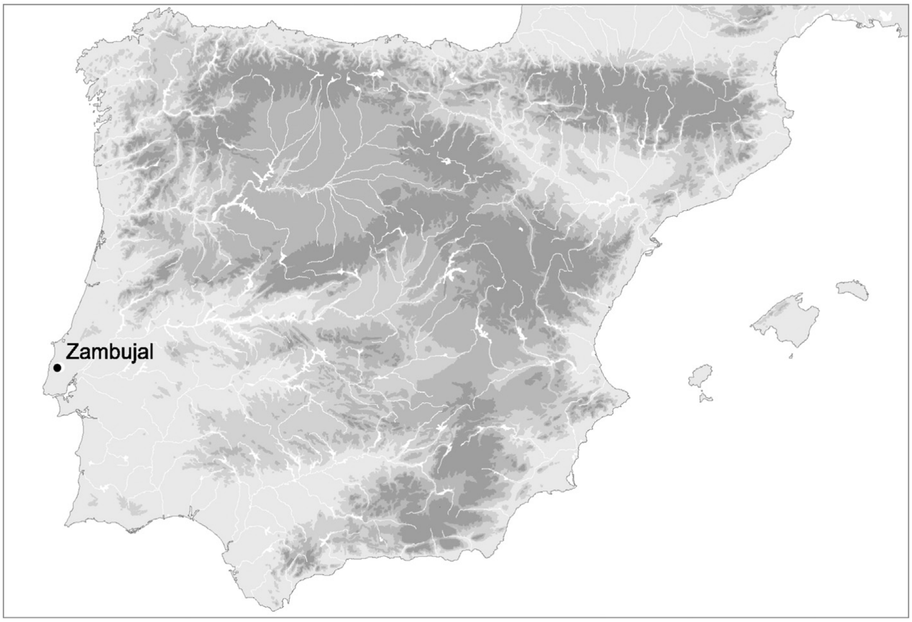

Fig. 1. Location of Zambujal in the Iberian Peninsula.

Fig. 2. Aerial view of Zambujal.

forest and exhibits a mixture of maritime conditions in the North and West and Mediterranean conditions in the South and Southeast (Albuquerque, 1984).

The present landscape of the Estremadura is, in part, a product of climatic and anthropogenic events that occurred throughout the Holocene. At the beginning of the Holocene, the Atlantic coast experienced eustatic sea rise, which reached its maximum around 7000 cal BC (Zazo et al., 1996; Dabrio et al., 2000; Dias et al., 2000; Vis et al., 2008). This coincided with a climatic shift toward warmer and wetter conditions throughout the Iberian Peninsula (Pérez-Obiol et al., 2011), and other regions in the world, marking the Holocene Climate Optimum. The sea rise resulted in the flooding of river valleys throughout the Iberian Peninsula. Most likely the result of anthropogenic activities, including forest clearance and farming, these estuaries filled with alluvium over the subsequent 2000 years (Vis et al., 2008). Similar processes can be documented near the site of Zambujal. There, in the early Holocene, a large estuary along the Sizandro valley was formed, which extended 18 km inland from the Atlantic coast (Hoffman, 1990; Dambeck et al., 2010). By ca. 4500 cal BC, the Sizandro estuary had become smaller and brackish (Lord et al., 2010).

By the time that site of Zambujal was established, in the Late Neolithic/Copper Age (ca. 2800 BC), the estuary extended only 10 km from the coast, and soil erosion from intensive agriculture had filled much of the remaining waterways (Dambeck et al., 2010). The river valleys as well as the estuary also rapidly aggraded. These processes are evidenced by the brackish water sediments studied by Lord et al. (2010), which were buried under 17 m of alluvium, and a soil boring on the Ribeira de Pedrulhos, some hundred meters from Zambujal, which encountered a bone beneath 12 m of alluvium dated to 2910–2755 BC (Dambeck et al., 2010). Settlement expansion in the area also led to increased deforestation (Van Leeuwaarden and Janssen, 1985; Castro et al., 2000), which likely contributed to the estuary decline.

Another possible contributor to the changes in the landscape of the Estremadura during the Holocene was the 4.2 kya event, also known as the Bond event 3 (Bond et al., 2001). Associated with cooler and dryer conditions, this event has also been linked to episodes of cultural fragmentation (or collapse) in diverse regions of the world. (Jalut et al., 1997). The contemporaneity of these events (i.e., the complete filling of the estuary and the abandonment of Zambujal and other Late Neolithic settlements in the Sizandro around 1800 BC) points to the possibility that environmental factors, exacerbated by the 4.2 kya event, played a role in transforming the social and political landscape, particularly in impacting trade and communication routes. Kunst and Trindade (1990) have suggested that the Sizandro River, which flows to the Atlantic, provided strategic access to marine resources and trade networks in southern Portugal and that population aggregation in this area during the Late Neolithic was related to trade and mobility. The infilling of the Sizandro would have reduced access to important marine and trade resources in this region and may have triggered the dissolution of its communities economic and social networks.

1.4. Stable isotope analysis in archeological research

Stable isotope analysis is an important part of archeological research because it can provide a record of dietary patterns from prehistoric humans and animals and, thus, can provide data about climate, environment, and culturally practices in past populations (Hedges et al., 2004) The published work related to this methodology continues to grow each year (cf. Le Bras-Goude et al., 2013 or Fontanals-Coll et al., 2015; see Ambrose and Krigbaum, 2003; Katzenberg and Saunders, 2008; Schoeninger and Moore, 1992, and Tykot, 2014 for general reviews). Bone has two components. One is an organic protein composed of collagen fibers, and the other is composed of inorganic minerals called apatite or hydroxyapatite. Experimental studies have shown that that stable isotope ratios in bone collagen are formed from dietary protein (Ambrose and Norr, 1993; Richards and Hedges, 1999; Tykot, 2004), while ratios from bone apatite reflect the whole diet (proteins, carbohydrates, and lipids) (Ambrose and Norr, 1993; Schwarcz, 2000; Tykot, 2002, 2004). In humans and animals the values of δ^{13}C and δ^{15}N from bone collagen are commonly used to calculate the dietary protein input of C_3, C_4 and CAM plants, and, for carnivores and omnivores, the consumption of marine and terrestrial animal proteins (Schoeninger and DeNiro, 1984; Ambrose and Norr, 1993; Chisholm et al., 1982; Norr, 1995; Smith and Epstein, 1971; Tykot, 2002:216).

In bone apatite, δ^{13}C values are used to evaluate patterns of C_3, C_4 and CAM plant consumption and marine protein input within the whole diet. When both $\delta^{13}C$ data on both collagen and apatite are available, investigations of $\delta^{13}C$ collagen-apatite spacing can provide more information about of dietary attributes. In particular, comparing of $\delta^{13}C$ in collagen and apatite can help to quantify the dietary intake of marine versus terrestrial protein and C_3 versus C_4 plants (Kellner and Schoeninger, 2007).

In bone apatite, oxygen isotope values ($\delta^{18}O$) can vary according to water sources and, therefore, can be used to recognize plants and animals from different geographic locations Stuart-Williams et al., 1996; White et al., 2004). Additionally, $\delta^{18}O$ values can be enriched via breastmilk consumption due to the oxygen isotope fractionation between mother and offspring, thus, these values can be used to distinguish suckling young and weaning time periods (Fricke and O'Neil, 1996).

Climatic, geologic and biodiversity differences in ecological zones lead to variations in the available isotopeswhich are incorporated into hard tissues. Therefore, the isotopic values in human and animal tissues can reveal information about precipitation (Bradley, 1999; Celle-Jeanton et al., 2001), soil salinity (van Groenigen and van Kessel, 2002), forest density (van der Merwe and Medina, 1991) and geologic formations and substrates (Bentley, 2006) in the local landscape. This allows diachronic studies of isotopic patterns in plants and the hard tissues of animals to be used to elucidate environmental changes over time (cf. Lohse et al., 2014, Drucker et al., 2003, and Aires et al., 2008). In particular isotopic enrichment in animal tissues have been linked to post-Neolithic, anthropogenic deforestation — based upon the "canopy effect" in which dense forest canopies produce more carbon depleted plants near the forest floor where many larger herbivore graze and deforestation leads to more carbon enrichment in many terrestrial herbivores who are forced to forage in more open landscapes and fallow fields (van der Merwe and Medina, 1991).

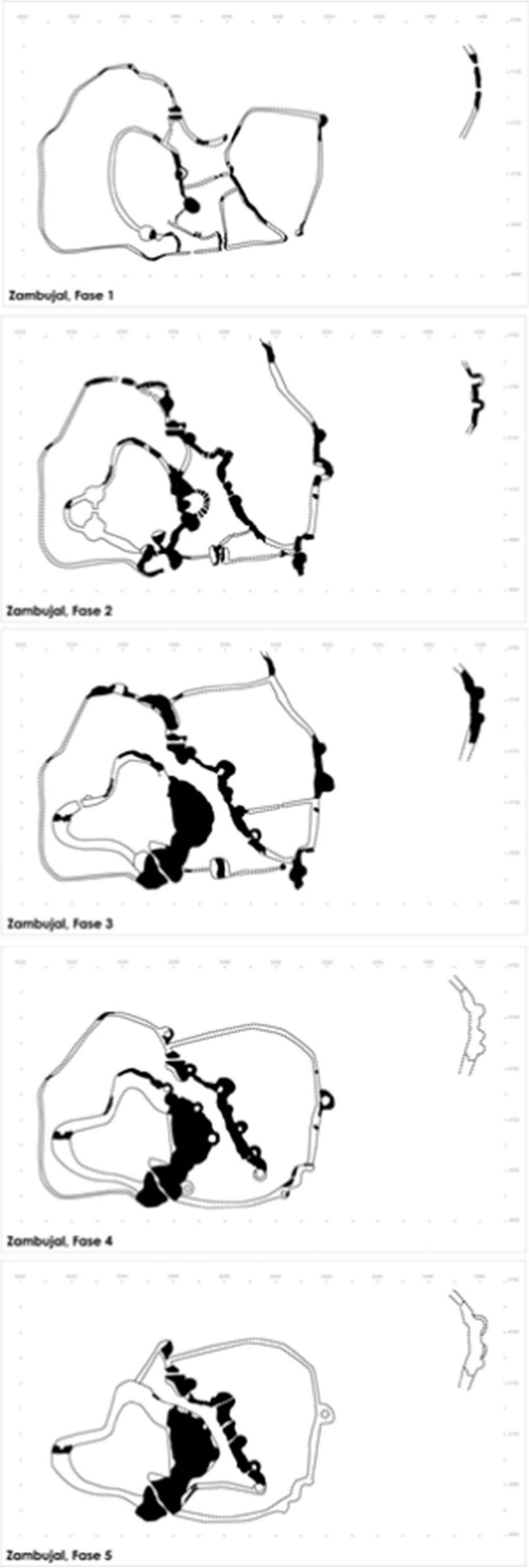

Fig. 3. Construction phases at Zambujal.

2. Materials and methods

In order to evaluate if faunal diets exhibit changes over time that would provide evidence of environmental changes that affected food webs, stable isotope analyses were completed on bone and tooth samples from 29 distinct adult ovicaprids recovered from Zambujal. Ovicaprids were selected for two reasons: 1) ovicaprids are more likely than other domesticated animals to graze and browse on wild plants rather than human-produced fodder. Thus, ecological changes which result in shifts in wild plant life are more likely recorded in the isotope record of the hard tissues of these animals, and 2) ovicaprids were available in well-dated contexts. All the 29 samples for stable isotope analysis come from the excavations of E. Sangmeister and H. Schubart. The chronology of Zambujal is based on a division in construction phases (with several sub-phases), established by Sangmeister and Schubart (1981, 226–255). The sampled faunal remains are classified into two groups: the older phase (Phases 1–3b, $n = 22$) and the younger phase (Phases 3c–5, $n = 7$) (Table 1). Chronologically the older phase spans a time period encompassing the first half of the 3rd millennium BC and the younger phase dates from the end of the 3rd millennium to the first half of the 2nd millennium BC (Kunst and Lutz, 2008).

The ovicaprid bone and tooth samples were processed at the Laboratory for Archaeological Science at the University of South Florida where collagen and apatite samples were obtained using the techniques described in Tykot (2006). Samples were pretreated with a buffered acetic acid (CH_3COOH) solution in order to remove nonbiogenic carbonates as such treatments have been shown to be effective in removing postmortem contamination from the burial environment while retaining the in vivo isotopic signature (Koch et al., 1997; Tykot, 2002). Both collagen yields and C:N ratios in collagen were measured, as yields of less than 1% have been found to be too degraded for reliable analysis, and C:N ratios between 2.9 and 3.7 are generally found to indicate good preservation (DeNiro, 1985; Tykot, 2002). The samples were analyzed for nitrogen, carbon, and oxygen isotopes using a CHN analyzer coupled with a

Table 1
Results of stable isotope analysis.

Sample #	Phase	Time period	Collagen			C:N	Apatite			Enamel		
			USF lab #	$\delta^{13}C$	$\delta^{15}N$		USF lab #	$\delta^{13}C$	$\delta^{18}O$	USF lab #	$\delta^{13}C$	$\delta^{18}O$
1614	1	Early phase	19942	−20.9	6.8	3.3	19971	−10.2	−1.7	20000	−11.1	0.1
197	1	Early phase	19943	−20.8	4.8	3.2	19972	−10.3	−1.2	20001	−10.7	0.6
1593	(1c/)2	Early phase	19944	−19.5	7.2	3.3	19973	−10.0	−1.9	20002	−10.3	−1.8
882	(1c/)2	Early phase	19945	−20.3	5.8	3.2	19974	−10.4	−1.3	20003	−11.1	−1.3
884	(1c/)2	Early phase	19946	−20.8	5.1	3.3	19975	−10.0	−1.3	20004	−10.5	−0.7
1539	(1c/)2/3	Early phase	19947	−20.5	5.9	3.2	19976	−11.3	0.0	20005	−10.7	0.4
1538A	(1c/)2a	Early phase	19948	−20.3	5.3	3.2	19977	−10.1	−0.4	20006	−11.8	−1.0
1538B	(1c/)2a	Early phase	19949	−20.3	5.4	3.1	19978	−10.6	−0.2	20007	−11.4	−0.6
175	1a	Early phase	19950	−19.9	5.0	3.2	19979	−10.6	−1.3	20008	−11.4	−1.3
266	1b	Early phase	19951	−20.5	6.0	3.1	19980	−11.4	−0.4	20009	−11.1	1.1
173	1b-3a	Early phase	19952	−20.5	5.5	3.2	19981	−10.1	−0.9	20010	−12.6	−1.3
154	1c-3a	Early phase	19953	−20.5	5.8	3.2	19982	−10.5	−0.9	20011	−12.2	−1.2
184	1c-3a	Early phase	19954	−20.2	5.5	3.2	19983	−10.5	−1.4	20012	−12.2	−0.8
134	1c-3a	Early phase	19955	−20.9	4.9	3.3	19984	−10.1	−1.4	20013	−12.6	−0.5
828	2a	Early phase	19956	−20.8	4.4	3.1	19985	−10.5	−0.5	20014	−12.0	0.2
68080A	2b/c	Early phase	19957	−20.6	5.6	3.1	19986	−11.5	−0.7	20015	−11.7	0.3
68080B	2b/c	Early phase	19958	−20.1	5.8	3.2	19987	−10.2	0.0	20016	−12.5	0.3
68079	3a	Early phase	19959	−21.0	5.0	3.3	19988	−9.7	−0.8	20017	−11.9	−1.2
68073A	3a	Early phase	19960	−20.1	5.7	3.1	19989	−10.5	−0.6	20018	−10.6	−0.6
68073B	3a	Early phase	19961	−20.4	5.8	3.2	19990	−11.0	−0.7	20019	−11.4	−2.5
1599	3a	Early phase	19962	−21.2	6.0	3.1	19991	−12.0	−0.5	20020	−12.7	−0.9
321	3b	Early phase	19963	−20.3	6.1	3.2	19992	−9.7	0.2	20021	−12.3	0.2
596	3c/4a	Late phase	19964	−21.0	5.2	3.2	19993	−10.8	−1.1	20022	−12.5	−0.3
492A	3c/4a	Late phase	19965	−20.7	4.0	3.2	19994	−10.8	−2.0	20023	−12.3	−0.3
492B	3c/4a	Late phase	19966	−20.8	4.7	3.1	19995	−12.3	−1.0	20024	−12.6	−1.2
170	4a	Late phase	19967	−20.6	5.1	3.2	19996	−9.9	−1.2	20025	−11.4	−1.3
327	4b/5	Late phase	19968	−20.3	3.8	3.2	19997	−8.2	−1.0	20026	−12.8	0.7
1482	4c	Late phase	19969	−21.1	4.8	3.2	19998	−10.3	−0.5	20027	−13.1	−1.7
328	4/5	Late phase	19970	−20.4	5.8	3.2	19999	−7.7	−3.6	20028	−11.4	−0.9
			Avg	−20.5	5.4			−10.4	−1.0		−11.8	−0.6
			Min	−21.2	3.8			−12.3	−3.6		−13.1	−2.5
			Max	−19.5	7.2			−7.7	0.2		−10.3	1.1
			Std	0.4	0.7			0.9	0.7		0.8	0.9

Finnigan MAT Delta Plus stable isotope ratio mass spectrometer using continuous flow for the bone collagen, and a Finnigan MAT Delta Plus instrument using a Kiel III device with 100% phosphoric acid (H_3PO_4) at 90 °C for the bone apatite.

All isotope ratios are reported using the delta (δ) notation and calibrated to an international standard using the following standard formula: $\delta = [(R_{sample} / R_{standard}) - 1) \times 1000]$, where R_{sample} is the ratio of the heavy isotope to light isotope in the sample (e.g. $^{13}C/^{12}C$ or $^{15}N/^{14}N$), and $R_{standard}$ is the ratio of the heavy isotope to light isotope in the reference standard and δ is the difference in isotopic composition of the sample relative to that of the reference, expressed in per thousands (‰) (see Tykot, 2006). In this study the $\delta^{13}C$, $\delta^{15}N$ and $\delta^{18}O$ notations are calculated using Ambient Inhalable Reservoir (AIR) and the Pee Dee Belemnite (PDB) standards. The analytical precision of the employed instrument was ±0.1‰ for the $\delta^{13}C$ and $\delta^{18}O$ data and ±0.2‰ for the $\delta^{15}N$ data. Student t-tests were used to ascertain if statistically significant ($P < 0.05$) differences in the dietary data existed between sampled groups. All statistical calculations were completed using NCSS 2003 software.

3. Results

3.1. Collagen data

The results of the isotopic tests are presented in Table 1 and Table 2.

The $\delta^{13}C$ values from the ovicaprid bone collagen from both time periods suggest a C_3 plant-based diet (Fig. 4) and no statistically significantly differences in the $\delta^{13}C$ values between the younger and later time periods were found ($p = 0.1732$). In contrast, the $\delta^{15}N$ values of the two time periods showed a marked decline over time. This difference was found to be highly statistically significantly different at the >.05 level (t-test $p = 0.0062$). For the bone apatite, $\delta^{13}C_{ap}$ values do not exhibit any statistically significantly differences between the two temporal samples ($p = 0.2112$), but both samples show some carbon isotope enrichment ($\delta^{13}C_{ap}$ values <11‰) which suggests C_4 plant consumption, a finding that is not evidenced by the bone collagen data (Fig. 5).

For the $\delta^{18}O$ values from bone apatite a statistically significantly difference was found between the two time periods ($p = 0.0359$), with a decrease in $\delta^{18}O$ enrichment in the sampled animals over time. This suggests a change in the isotopic composition of water over time or a change in drinking water source. One animal with a strongly divergent $\delta^{18}O$ value is likely nonlocal. When the $\delta^{18}O$ from tooth enamel is examined, no statistically significant differences between the two groups are found ($p = 0.6999$), but the temporally later group does continue to exhibit a trend toward oxygen isotope depletion. In contrast to the $\delta^{13}C$ values from the bone apatite, the $\delta^{13}C$ values from dental enamel show a statistically significant difference between the two groups ($p = 0.0325$) (Fig. 6). Animals from the later time period show less carbon enrichment, suggesting less C_4 plant input.

4. Discussion

We expected isotopic signatures animal diets in the region near Zambujal during the Neolithic through Early Bronze Age to reflect a mainly C_3 food web as the main cultivated crops for human and animal consumption were wheat, barley, legumes, and olives (all C_3 plants) (Castro, 1995; Cardoso, 2007) and few native C_4 plants are known to exist during this time in this region. This expectation was met by the collagen data as the sampled fauna from both time periods appear to have obtained the majority of their protein from a food web based upon C_3 plants.

However, the C_4 signature in the bone apatite is inconsistent with the collagen data and suggests that some C_4 or CAM plants were being

Table 2
Results of statistical tests.

Phase	Sample size	Mean	SD	SE	95% LCL of mean	95% UCL of mean	DF	t-Value	p-Value
δ^{13}Cco									
Early	22	−20.5	0.4	0.08	−20.7	−20.3	27	1.3991	0.1732
Late	7	−20.7	0.3	0.11	−21.0	−20.4			
Total	29								
δ^{15}N									
Early	22	5.6	0.6	0.14	5.3	5.9	27	2.9691	0.0062
Late	7	4.8	0.7	0.26	4.1	5.4			
Total	29								
δ^{13}Cap									
Early	22	−10.5	0.6	0.13	−10.8	−10.2	27	−1.2806	0.2112
Late	7	−10.0	1.6	0.60	−11.5	−8.5			
Total	29								
δ^{18}Oap									
Early	22	−0.8	0.6	0.12	−1.1	−0.6	27	2.2008	0.0358
Late	7	−1.5	1.0	0.39	−2.4	−0.5			
Total	29								
δ^{13}Cen									
Early	22	−11.6	0.8	0.16	−11.9	−11.3	27	2.2543	0.0324
Late	7	−12.3	0.7	0.25	−12.9	−11.7			
Total	29								
δ^{13}Oen									
Early	22	−0.1	0.9	0.19	−1.0	−0.2	27	0.3895	0.6999
Late	7	−0.7	0.8	0.31	−1.5	0.0			
Total	29								

consumed. Millet, the main C_4 crop cultivated during prehistory in Europe, was first domesticated in China and spread into Eastern Europe (Hungry) by 6000 BC (Lightfoot et al., 2013). However, although some seeds of millet have been recovered from Chalcolithic sites in central Spain (Rios and Liesau, 2011) evidence for the widespread cultivation of millet is not found in the Iberian Peninsula until the Middle Bronze Age (Rovira Buendía, 2007). Other possible sources of the carbon enrichment could include seaweed and sea grasses that are both C_4 plants and would have been available in the large estuarine environment that existed until the Bronze Age (Kunst, 1995). Such crops may even have been harvested by humans and used for crop fertilization (Waterman et al., 2015) and, as such, made more readily available to sheep and goats. The use of seaweed as a fertilizer can also lead to δ^{13}C enrichment of cultivated C_3 plants by increasing the δ^{13}C values and the salinity of the soil (Farquhar et al., 1989) — an enrichment which can be passed on to the consumers.

As the carbon enrichment is only exhibited in the apatite it suggests that the C_4 is mainly providing nutrients toward the carbohydrate and/or fat component of the diet (Ambrose et al., 1997). When the δ^{13}C collagen and apatite are compared using the Kellner and Schoeninger (2007) model of isotopic indicators of dietary inputs, C_3 plants are indicated for protein sources, but some portion of dietary energy (fats and sugars) are coming from C_4 plants (Fig. 7). Similar to the sheep and goats in this study, isotopic studies from human remains recovered from some burials in the region also found that C_4 plants contributions to dietary energy (Waterman et al., 2015).

The δ^{13}C values from the dental enamel exhibit similar carbon enrichment that can be attributable to C_4 plants. For the enamel, the later time period shows a statistically significant decrease in the carbon enrichment suggesting less C_4 plant input in the diet of the animals for the later period. Although not statistically significant, a slight decrease in carbon enrichment is also seen in the apatite data over time. This finding suggests that the C_4 signature is likely related to a local plant, rather than the early importation of millet.

However, since teeth represent a more narrow formation window than bone, this difference may represent aspects of biological growth and development, including breastmilk consumption and seasonal dietary attributes. Alternatively, as these detected differences are from small samples sizes, are not strongly statistically significantly different, and are from a mixture of sheep and goat, we may be detecting a difference in sheep versus goat diet rather than a real change in plant availability or selection over time.

However, the difference in the δ^{15}N values from bone collagen in the two sampled groups was found to be highly statistically significant suggesting that the later animals were consuming less nitrogen-enriched plants. This may indicate either a change in the types of plants in the environment or possibly a change in the soil salinity, as increases in soil salinity have been documented to enrich the carbon and nitrogen isotope values in plants (van Groenigen and van Kessel, 2002). Elevated δ^{15}N values have been documented in other prehistoric settings for animals that grazed in saltwater marsh environments (Britton et al., 2008). Droughts can increase soil salinity, and drought resistant animals, like caprines, may exhibit enriched δ^{15}N values in hard tissues formed during times of water stress due to physiological changes related to water conservation (Ambrose and DeNiro, 1986). Additionally, middening and manuring are anthropogenic activities that can lead to increases in δ^{15}N values (Bogaard et al., 2007; Bogaard, 2012). Thus, in this study, in addition to possible shifts in available plants or used pasture lands, a decrease in soil salinity related to an increase in rainfall and/or a shift in human agricultural activities could explain this decrease in nitrogen enrichment over time. These same changes may also be tied to the decrease in carbon enrichment in the later time period.

Depletions of the δ^{18}O values are also found in the animals recovered from the later phase. These changes were found to be statistically significant in the bone apatite but not in the dental enamel, the opposite of the δ^{13}C values. Since the δ^{18}O values are tied to water source and intake, this shift toward oxygen depletion may indicate environmental changes. In particular, oxygen isotope depletion is associated with increased precipitation and/or lower temperature (Bradley, 1999; Price and Burton, 2011:91–92). Thus, this change in stable isotope values may suggest that the climate had become wetter and cooler during the later occupation phase. Alternatively, as with the other isotopic shifts, this may indicate a change in animal foraging and drinking locations.

As discussed in the introduction, the Late Neolithic through Early Bronze Age was a time of population increase and social and political development in the southwestern Iberia. However, at the end of this time period, many settlements and burials were abandoned. The reasons for these abandonments are still not clear but anthropic activities are thought to have resulted in widespread environmental transformations. Climate change, including the 4.2 kya event, has also been proposed as a possible factor (Lillios et al., under review). The 4.2 kya event resulted in more arid conditions throughout the Mediterranean

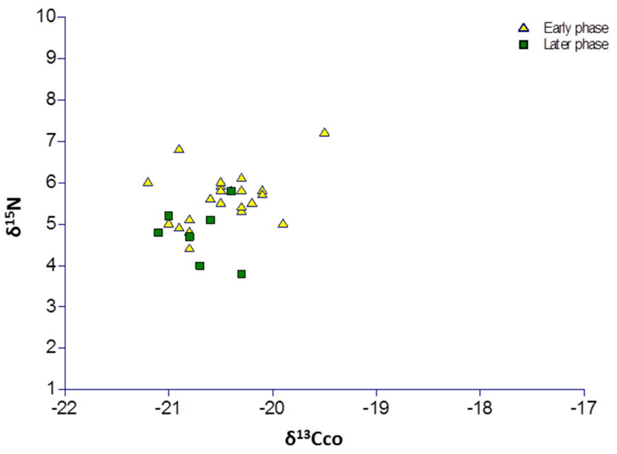

Fig. 4. Scatterplot of δ^{15}N and δ^{13}Cco.

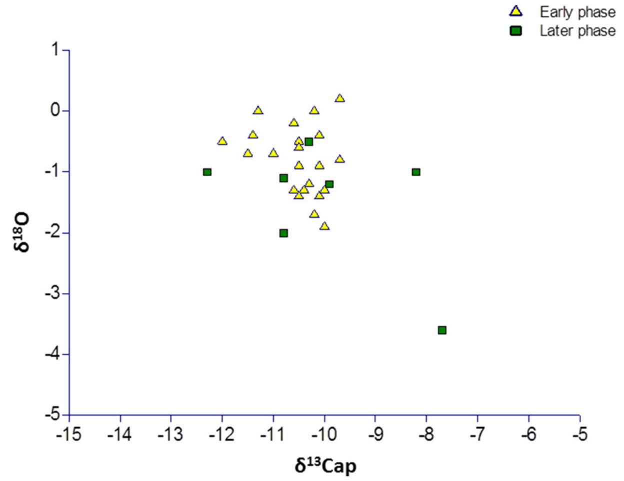

Fig. 5. Scatterplot of δ^{18}Oap and δ^{13}Cap.

and the Middle East (Burjachs et al., 1997; Dalfes et al., 1997; Magny et al., 2002; Peiser et al., 1998; Zielhofer et al., 2008). While we would think that these events would also result in drier conditions throughout the Iberian peninsula, as has been documented in eastern Spain (cf., Araus et al., 1997), based on the isotopic indicators of diet for the ovicaprids examined in this study, conditions were potentially wetter with this increased precipitation resulting in less enriched oxygen isotopes and decreased soil salinity which, in turn, led to less enrichments of δ^{13}C and δ^{15}N values in local plants. Alternatively, these changes in isotopic indicators of ovicaprid diets could signal a change in foraging locations. The estuaries near Zambujal are documented to have rapidly filled in during the time of occupation. It may have been that in the later phases of the site's occupation foraging and grazing conditions near the estuaries became unsustainable or were reserved for other agricultural activities, and animals were moved to pasture elsewhere where water sources and plant life were less isotopically enriched. While it is possible that the sheep and goats in this study consumed some human-grown fodder, it is expected that these animals were obtaining most of their sustenance from wild plants. Organized transhumance related to ovicaprid husbandry is documented during the Roman occupation of the Iberian Peninsula (Defourneaux, 1971). However, how, and if, animals were moved systematically from winter and summer pasture locations in Neolithic and Copper Age Portugal is unclear and likely animal husbandry practices were variable and flexible according to community needs (Harrison, 1985). Animals may also have been commonly gazed in fallow and reaped agricultural fields as has been noted in other locations (Halstead, 2006). Thus, we must consider that the variation in isotopic values may be reflective of changes in domesticate production and/or variations in regional rather than only local environmental patterns.

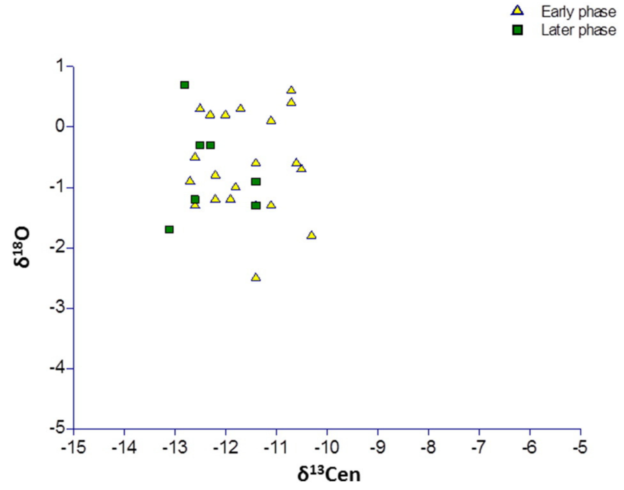

Fig. 6. Scatterplot of δ^{18}Oen and δ^{13}Cen.

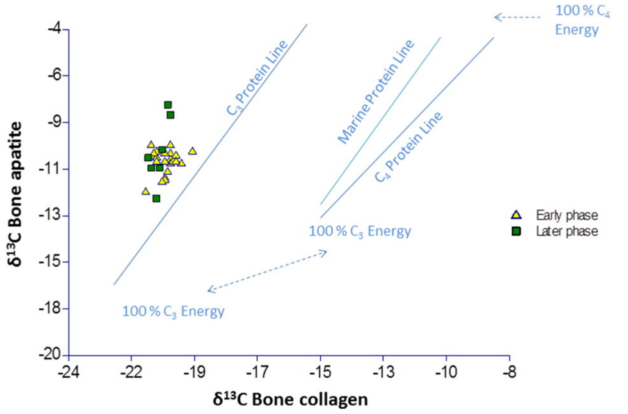

Fig. 7. Scatterplot of δ^{13}C collagen and apatite using model proposed by Kellner and Schoeninger (2007).

A final consideration is that the sheep/goats sampled in this study originated from different locations and were brought into Zambujal for consumption, possibly related to feasting events. Waterman et al. (2014) found that some ovicaprids remains from Zambujal exhibited non-local strontium isotope signatures. As the findings in this paper are based upon small samples sizes (in particular for the later samples) further investigation is needed to clarify these results.

5. Conclusion

In this paper we compare stable isotopic data gathered from bone and tooth samples from 29 ovicaprids recovered from two distinct time periods from the settlement of Zambujal (Torres Vedras, Portugal). The goal of this work was to identify changes in ovicaprids diets over time that could provide insights into how environmental changes of the third millennium BC may have affected local farming populations and triggered the eventual settlement abandonments. Our findings demonstrate that ovicaprids from earlier and later occupation phases exhibit statistically significant differences in stable isotope values, with the animals from the later occupation phases exhibiting less carbon, nitrogen, and oxygen isotope enrichment. In both time periods there is evidence of C_4 plant consumption but only in the bone apatite and dental enamel, which suggests that these plants largely contributed to the carbohydrate or fat component of the diet. The contribution of C_4 plants in these animals' diets appears to decrease over time. The finding of more isotope depletion in the later time period was unexpected in relation to the known environmental changes and may indicate either an increase in precipitation, a shift in available plants for consumption or that the animals from the later occupations were commonly being pastured in different locations than the animals from the earlier occupations. Both possibilities likely reflect changing local ecological conditions related to anthropogenic activities and climatic fluctuations. Thus, the results of this study document the biological impacts of these changes in animal diets at Zambujal, which in turn can be used to add to our understanding of the changing local resources and food procurement strategies of the local human populations.

Acknowledgments

We are grateful for support provided by the National Science Foundation (# 1153568). We would like to thank Isabel Luna and other staff members of the Museu Municipial de Torres Vedras for help and support obtaining samples for this project. We would also like to thank Simon Davis and Matthew Hill for their advice and input on fauna selection and sampling procedures and Robert D. Bowers and Ethan Goddard for laboratory preparation and isotopic analysis of the bone and tooth samples.

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