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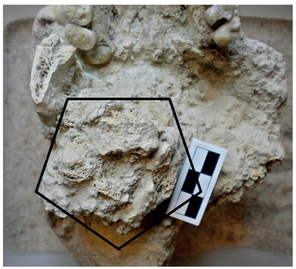

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12 DOSSIER: NEW PERSPECTIVES IN THE STUDY OF BIOARCHAEOLOGICAL REMAINS FROM THE 3RD MILLENNIUM BC IN SOUTHERN IBERIA

Coordinated by Marta Díaz-Zorita Bonilla

88 ESTUDIOS

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EL GENIL.

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PUNTOS DE SUSCRICION.

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LA CUEVA DE MENGA.

Rica en tradiciones y en recuerdos de gloria, aun de sus propios hijos, desconocidos ú olvidados, no lo es menos Antequera en célebres monumentos, brillantes páginas de su historia é indelebles huellas de las diversas civilizaciones, que desde las primeras edades del mundo tuvieron vida propia en su fecundo suelo.

Dejando, por hoy, á un lado la época romana, de la que tantas joyas atesora, vamos á penetrar en la nebulosa historia del pueblo

La Cueva de Menga se halla situada á mil y quinientos pasos de la ciudad de Antequera, sobre la derecha del camino que conducen á Granada. Es un *dolmen completo complicado* encerrado en un *tumulus semi-esférico*. Sus dimensiones son: longitud, veinticuatro metros y catorce centímetros; latitud, seis metros por el centro y cuatro por las extremidades; altura, dos metros y cincuenta á ochenta centímetros. Lo forman treinta y una piedras distribuidas de esta manera: veinte los muros, una el testero, cinco el techo, dos descubiertas á la entrada, y tres pilares

miento profano, practicado en tiempos modernos por manos ignorantes.

La mayor de las piedras que componen este magestuoso edificio, tiene, segun los cálculos del arquitecto D. Rafael Mitjina, veintitres piés de latitud, veintisiete de longitud, cuatro y medio de espesor, dos mil setecientos piés cúbicos de volúmen, y once mil ciento setenta y seis arrobas de peso. (1)

La historia guarda silencio acerca de los aparatos con que aquel pueblo semi-salvaje movia y trasportaba hace cuatro mil años esas

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Passage of La Pastora dolmen (Valencina de la Concepción, Sevilla).
Photo: Miguel A. Blanco de la Rubia.

MOBILITY PATTERNS AND PALEODIETARY INSIGHTS INTO HUMANS AND CATTLE AT THE COPPER AGE MEGA-SITE OF VALENCINA (SEVILLE, SPAIN) THROUGH δ^{18}O AND δ^{13}C ISOTOPE ANALYSES

Marta Díaz-Zorita Bonilla[1], K. J. Knudson[2], Javier Escudero Carrillo[1], Hervé Bocherens[3 4], and Leonardo García Sanjuán[5]

Abstract:

The mega-site of Valencina is currently a major focus of interest for the study of Copper Age Iberia. Remarkable megalithic monuments such as La Pastora, Montelirio or Structure 10.042-10.049 at PP4-Montelirio are found alongside hundreds of other features, including pits and large-sized ditches, some of which have yielded evidence of exotic material craftsmanship without parallels in Iberian Late Prehistory which also suggests long-distance contacts. Part of the flourishing experienced by Valencina in the 3rd millennium BC can be explained by its specific geographic location at the mouth of the Guadalquivir river, facing a marine gulf surrounded by lands of high agricultural potential. Other reasons, however, must have accounted for Valencina's growth as a mega-site, including particularly demographic and economic ones. In order to better understand the demographic and subsistence patterns of the communities that lived and/or frequented Valencina, we analysed 29 human and 7 faunal samples for δ^{18}O and δ^{13}C isotope analyses. The sampling strategy followed is based on a combination of bone and dental tissues. In addition an intra-tooth study was also carried out to observe intra-individual seasonal changes. Overall, this evidence contributes to the study of diet and mobility patterns, which can in turn provide insights of the demography and economy of the communities that used this mega-site.

Keywords: Stable isotopes, Copper Age, Mega-site, Mobility patterns, Palaeodiet, Oxygen, Carbon.

PAUTAS DE MOVILIDAD Y ESTRATEGIAS DE SUBSISTENCIA EN HUMANOS Y BÓVIDOS PROCEDENTES DEL MEGA-SITIO DE LA EDAD DEL COBRE DE VALENCINA (SEVILLA, ESPAÑA) A TRAVÉS DE LOS ISÓTOPOS ESTABLES DE δ^{18}O Y δ^{13}C

Resumen:

El mega-sitio de Valencina es actualmente un referente principal en el estudio de la Edad del Cobre en Iberia. Destacados monumentos megalíticos tales como La Pastora, Montelirio o la Estructura 10.042-10.049 del sector PP4-Montelirio aparecen junto a cientos de otras estructuras, incluyendo hoyos y zanjas de gran tamaño, algunos de los cuales han proporcionado evidencias de artesanía de materias primas exóticas sin paralelos en la Prehistoria Reciente ibérica, evidenciando también contactos de larga distancia. Parte del florecimiento de Valencina en el III milenio ANE puede ser explicado por su ubicación geográfica específica, en la desembocadura del río Guadalquivir y frente a un gran golfo marino rodeado de terrenos de alto potencial agrícola. Otras razones, sin embargo, deben explicar el crecimiento de Valencina como un mega-sitio, especialmente aquellas de carácter demográfico y económico. Para conocer mejor las pautas demográficas y subsistenciales de las comunidades que vivieron y/o frecuentaron Valencina, en este estudio hemos analizado 29 individuos humanos y 7 dientes de fauna según los análisis de isótopos estables de δ^{18}O y δ^{13}C. Se ha seguido una estrategia de muestreo combinada de tejidos óseos y dentales. También hemos seguido una estrategia de análisis intra-individuo basado en múltiples bandas en el diente para poder observar cambios estacionales. En conjunto, estos datos contribuyen al estudio de las pautas de dieta y movilidad, lo cual a su vez proporciona nuevas formas de analizar la demografía y la economía de las comunidades que usaron este mega-sitio.

Palabras clave: Isótopos estables, Edad del Cobre, Mega-sitio, Pautas de Movilidad, Paleodieta, Oxígeno, Carbono.

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1. INTRODUCTION

Modern-day Valencina is located 6 km away from the city of Seville in the Lower Guadalquivir river basin. In the 3rd millennium BC, however, Valencina was at the mouth of the river, facing a marine gulf surrounded by lands of high agricultural potential (Fig. 1). The communities that inhabited this region in the 3rd millennium BC enjoyed a very favourable ecological setting (with arable land, pastures, marine resources and abiotic resources located in the nearby Sierra Morena mountains) as well as an advantageous geo-estrategic position, between the Mediterranean sea and the Atlantic ocean, and between Africa and Iberia. A systematic radiocarbon programme carried out recently has provided an entirely new base for the study of the occupation of this site between c. 3200 and 2300 cal BC (García Sanjuán *et al.*, Forthcoming a).

The largest 3rd millennium site in Iberia, Valencia hosts some remarkable megalithic monuments such as La Pastora, Montelirio or Structure 10.042-10.049, which are found alongside hundreds of other features, including pits and large-sized ditches. Some of these features have yielded abundant evidence of exotic material craftsmanship without parallels in Iberian Late Prehistory (Vargas Jiménez *et al.*, 2012; Murillo-Barroso and García Sanjuán, 2013; García Sanjuán *et al.*, 2013; Luciáñez Triviño *et al.*, 2014; Murillo-Barroso *et al.*, 2015; Morgado Rodríguez *et al.*, 2016; etc.). Key economic resources may have included pig farming under dehesa-type conditions as well as salt production, with exotic materials manufactured into sumptuous artefacts that played a key role in the expression of power (García Sanjuán, 2017).

Although major advances have been made in recent years, the study of the specific demographic and economic conditions that led to the flourishing of Valencina in the 3rd millennium BC still presents major gaps. To this end, in this paper we aim to investigate two problems of crucial importance: mobility patterns and subsistence strategies. Thus, the application of stable isotope analysis of oxygen and carbon to tooth enamel, cementum, dentine and bone hydroxyapatite from bone tissues of both human and cattle specimens provides the basis for a better understanding of the social and economic practices that led to the rise of Valencina as a mega-site, and also its specific environmental conditions.

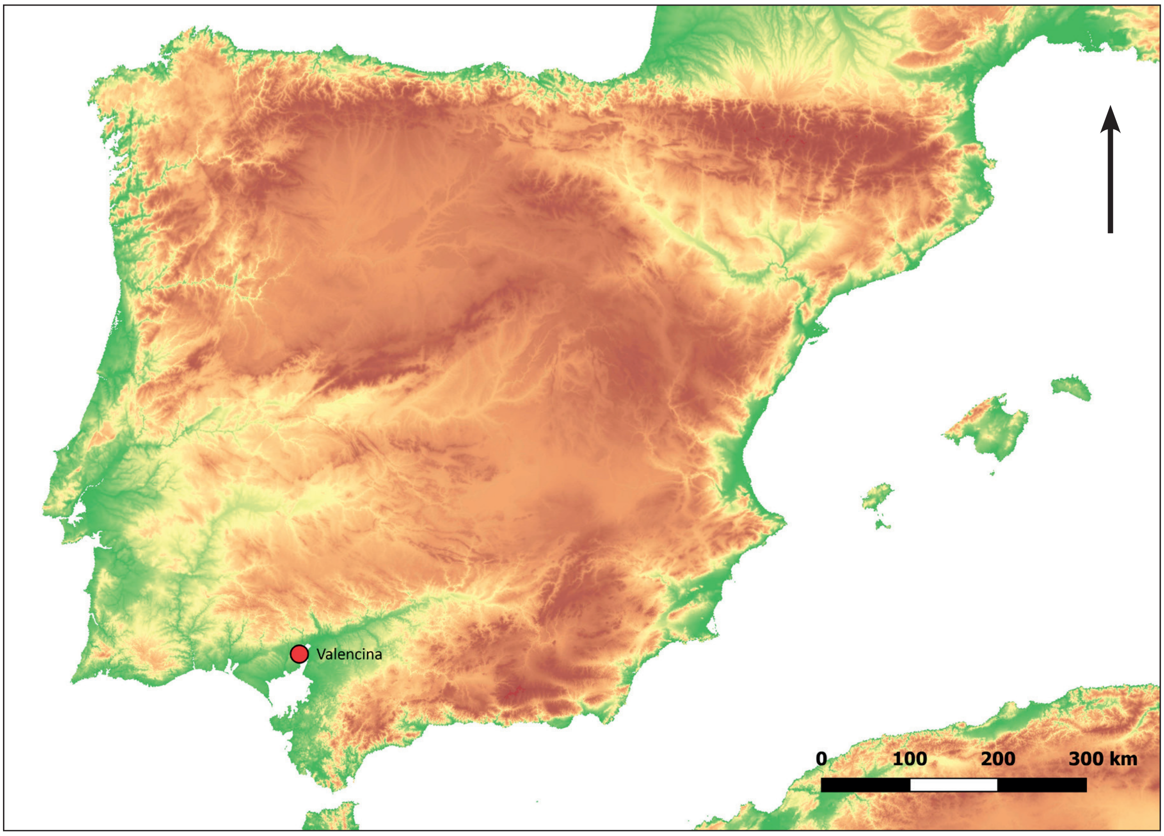

Fig. 1. Map with the location of Valencina (Seville, Spain) (Design: Manuel E. Costa Caramé).

Bone is composed by two different tissues; the organic part, e.g. collagen, and the inorganic part, e.g. the apatite or hydroxyapatite (DeNiro, 1985). In addition, teeth are made of three type of mineralized tissue composed by an inorganic phase, e.g. the tooth enamel (hydroxyapatite) and the organic phase, e.g. the dentine and the cementum (Hillson, 1997; Driessens and Verbeeck, 1990; Lowenstam and Weiner, 1989). In this research, we used bone hydroxyapatite, tooth enamel hydroxyapatite, dentine and cementum to investigate both diet and mobility patterns. Human samples were subject to two analysis, tooth enamel and maxillae or long bone (bone hydroxyapatite), whilst three different types of tissue were extracted from each animal for analysis: tooth enamel, cementum and dentine. Each of the different tissues holds different information about an individual´s lifetime; while tooth enamel is indicative of childhood stages, bone hydroxyapatite, cementum, and dentine reflect the later stages of adult life (Kohn *et al.*, 1997). A comparison of the four types of tissues allows the investigation of changes throughout the life of an individual. In addition, for those animal teeth (cattle) included in this study, a sampling strategy based on multiple bands was applied (Fig. 2). The purpose of this strategy was to investigate intra-individual changes in $\delta^{18}O_c$ and to observe seasonal changes, as previously undertaken in other archaeological contexts (Balasse *et al.*, 2001; Bendrey *et al.*, 2015).

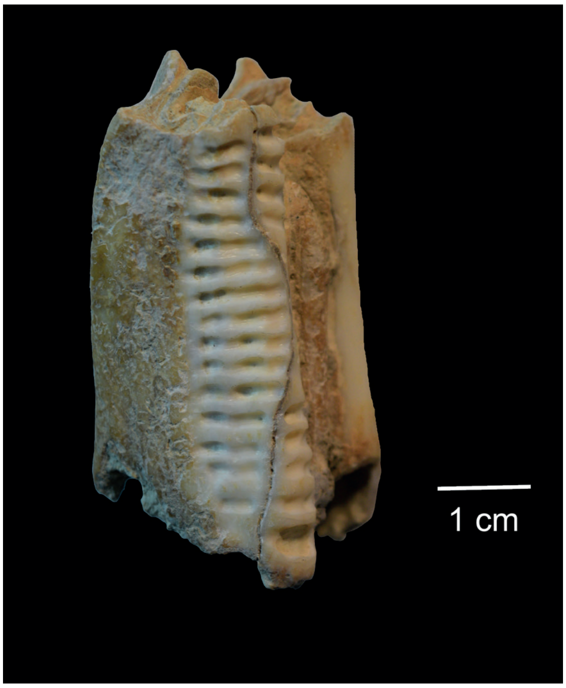

Fig. 2. Sample VCE-3, cattle upper third molar with multiple bands extracted for analysis (Photo: Marta Díaz-Zorita Bonilla).

Stable isotope analysis is widely used nowadays as a remarkably powerful tool to investigate past societies and to answer questions about topics such as subsistence strategies, climate and environmental conditions, and cultural and economic practices. In the past few years, this area of research has been quite prolific, particularly for the investigation of prehistoric contexts in Iberia (Lubell *et al.*, 1994; Arias, 2005;García-Guixé *et al.*, 2006a, 2006b; Roksandic, 2006; Umbelino, 2006; Fano, 2007; García García *et al.*, 2009; Salazar García, 2009, 2010, 2011; Fuller *et al.*, 2010; McClure *et al.*, 2011; Waterman, 2012; Fontanals-Colls *et al.*, 2012; Fontanals-Colls *et al.*, 2015; Díaz-Zorita Bonilla, 2017).

Although some research has been carried out during the past few years at Valencina (Fontanals-Coll *et al.*, 2015; Díaz-Zorita Bonilla, 2017), this is the first attempt to investigate mobility patterns and subsistence strategies using a multi-tissue approach through the application of $\delta^{18}O_c$ and $\delta^{13}C_c$ stable isotopes.

2. SAMPLING STRATEGY

Different sectors at the mega-Site of Valencina have been subject to analysis including El Algarrobillo, La Cima, La Gallega and La Alcazaba (Díaz-Zorita Bonilla, 2013, 2017), as well as the PP4-Montelirio sector (Robles Carrasco *et al.* 2017; García Sanjuán *et al.*, Forthcoming b) (Figure 3). The radiocarbon chronology of these sectors shows that they were not all used at the same time (García Sanjuán *et al.*, Forthcoming a). The human remains included in this study from El Algarrobillo (MNI=19) are coming from two different negative structures, a circular one and a second one consisting of two circular structures connected by a passage (Santana Falcón, 1993). The human remains from La Cima (MNI=2) were found at trench 6 mixed with material culture and large fragments of slate. Two individuals were documented, one of them in anatomical position with flexed lower limbs (Ruiz Moreno, 1991; Alcázar Godoy *et al.*, 1992). At La Gallega (MNI=2) several pit structures were documented and the human remains were found at Pit 10 and 11 along with material culture including copper artefacts and two idols (Martín Espinosa and Ruiz Moreno, 1992). Human remains from La Alcazaba (MNI=9) were found commingled at Structure 18 and 19. Both structures were pits where human remains where mixed with faunal and material culture (Cruz Auñón Briones and Arteaga Matute, 1996). The PP4-

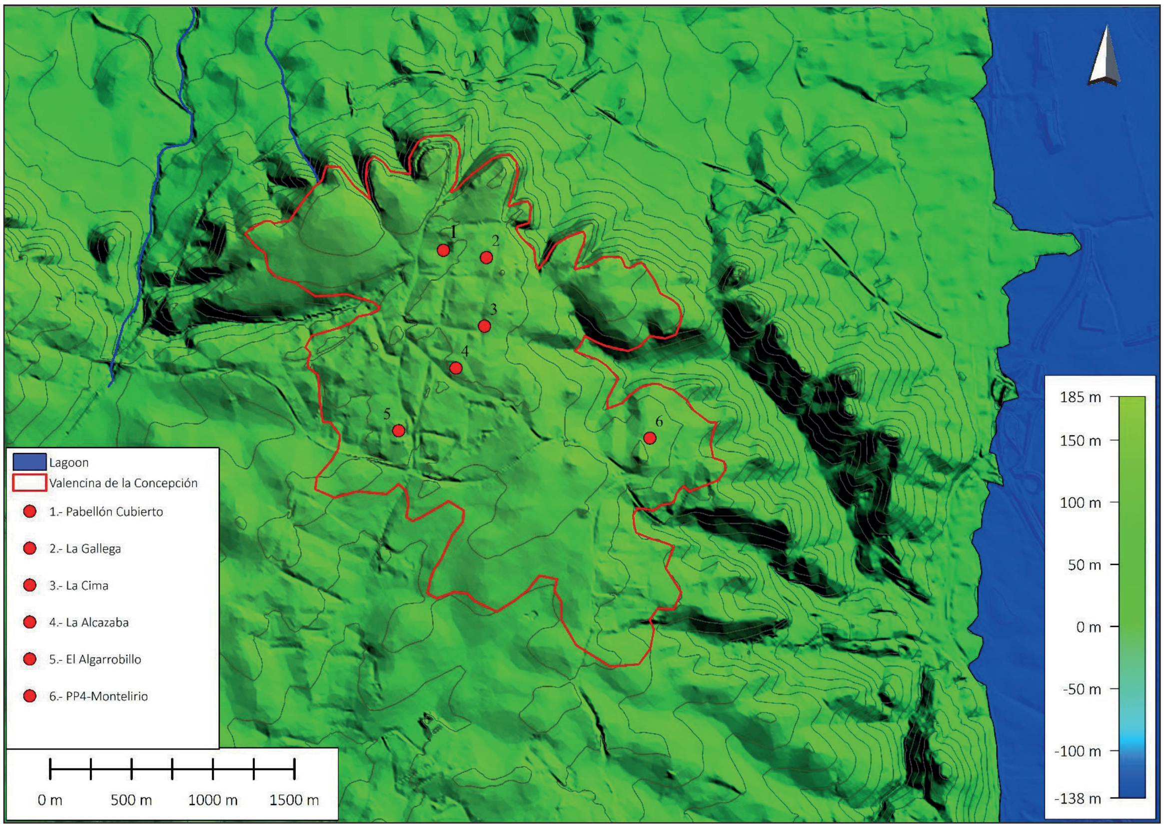

Fig. 3. Sectors at Valencina selected for analysis (Design: Javier Escudero Carrillo).

Montelirio sector includes several megalithic and non-megalithic structures including a large number of skeletal remains currently under analysis. For this study we have included only one individual from the megalithic Structure 10.042-10.049. This is a megalithic structure consisting of a 13 m corridor and two chambers. This individual was found in anatomical position at the lower depositional level at the second chamber (Structure 10.049) associated by a remarkable grave goods deposit (Díaz-Zorita Bonilla 2017; Robles Carrasco and Díaz-Zorita Bonilla, 2017; Mora Molina *et al.*, 2013; García Sanjuán *et al.*, Forthcoming b).

The faunal remains come from two different sectors, the PP4-Montelirio which was analysed in Liesau von Lettow-Vorbeck *et al.* (2014) and also from Pabellón Cubierto. The remains from the latter sector come from three different negative structures currently under study. More detailed information about this sector can be found in Ortega Gordillo (2013).

In total, 29 human samples (all adults of different sex and age categories) from 15 humans (MNI) and 7 faunal samples (all of them young adults) from 5 different individuals (MNI) were analysed (Tables 1 and 2). The criteria to sample the human material were based on the MNI (Díaz-Zorita Bonilla, 2017), preferably using molars. Priority was given to second molars, but depending on the availability of samples other teeth were also used for analysis if needed. For the bone material, maxillae or the skull from the same individual as the molars were always sampled. For humans, bulk enamel was sampled (sampling of a band of enamel from occlusal to the cemento-enamel junction or CEJ) and for the cattle, an intra-tooth sampling strategy was followed in the form of multiple parallel bands of about 1 mm each, extracting the sample from occlusal to CEJ (Fig. 2). Measurements of bands were taken in mm with sliding callipers from the occlusal part to the CEJ and are reported in Table 3.

Additionally, there is one animal for which three different teeth were analysed (M1, M2 and M3), potentially allowing us to detect if it moved during its lifetime or experienced any change in diet. Since there are differences among teeth formed *in utero* and those formed later in life (Table 4), variation amongst them can be used to investigate seasonality (Balasse *et al.*, 2003). Enamel mineralization in cattle teeth has been investigated by several authors (Suga

1982; Brown *et al.*, 1960; Hillson 2005). There are two different phases during enamel mineralization (amelogenesis); an initial one, when the matrix is in formation, and a subsequent maturation, when water and proteins are removed and a mature enamel is produced (Hillson 2005: 155). This process starts at the base of the cusps and finishes at the cervix. For cattle specifically, the enamel mineralization first starts in Molar 1 before birth and finishes 2-3 months after birth, whilst teeth eruption occurs between the 5th and 6th months of life. Molar 2 starts mineralization 1 month after birth and the process finishes around the 13th month. The eruption of M2 is between the 15th and the 18th month. The enamel mineralization in M3 starts around 9-10 months and lasts until the 24th month, whilst eruption does not take place until the 30th month, according to Silver (1969) and Hillson (2005: 232) (Table 4).

Sector	Individual	Bone/Tooth	Sex	Age	$\delta^{18}O_{VPDB}$ (‰)	VSMOW (‰)	$\delta^{18}OP$ (‰)	$\delta^{18}Odw$ (‰)	$\delta^{13}C$ (‰)
La Cima	C-1	URM2	F	18-25	-3.14	27.63	18.57	-5.12	-12.72
La Cima	C-1	R Tibia	F	18-25	-5.14	25.56	16.55	-8.24	-11.97
La Gallega	GAL-1	URI1	F	>45	-4.30	26.43	17.40	-6.93	-13.47
La Gallega	GAL-1	L Scapula	F	>45	-5.06	25.64	16.63	-8.11	-11.21
La Alcazaba	ALC-1	LRM1	F	25-35	-3.59	27.16	18.12	-5.82	-12.70
La Alcazaba	ALC-1	Mandible	F	25-35	-4.99	25.72	16.70	-7.99	-11.71
La Alcazaba	ALC-2	URM3	F	30-34	-2.93	27.84	18.78	-4.80	-12.91
La Alcazaba	ALC-2	Skull	F	30-34	-4.24	26.49	17.46	-6.83	-10.86
El Algarrobillo	ALG-1	URM3	M	18-25	-3.71	27.04	18.00	-6.01	-12.15
El Algarrobillo	ALG-1	Skull	M	18-25	-4.82	25.89	16.87	-7.73	-12.19
El Algarrobillo	ALG-2	URM1	M	18-25	-3.21	27.55	18.50	-5.22	-13.49
El Algarrobillo	ALG-2	Maxilla	M	18-25	-8.02	22.60	13.65	-12.71	-11.15
El Algarrobillo	ALG-7	URM3	F	25-35	-4.41	26.31	17.28	-7.10	-12.35
El Algarrobillo	ALG-7	Skull	F	25-35	-4.53	26.20	17.17	-7.28	-10.01
El Algarrobillo	ALG-9	Maxilla	U	A	-7.34	23.30	14.33	-11.65	-12.36
El Algarrobillo	ALG-10	URM2	M	25-35	-3.74	27.00	17.96	-6.06	-12.31
El Algarrobillo	ALG-10	Maxilla	M	25-35	-5.88	24.80	15.80	-9.38	-11.32
El Algarrobillo	ALG-11	URM3	M	18-25	-5.23	25.47	16.46	-8.38	-12.96
El Algarrobillo	ALG-11	Skull	M	18-25	-4.40	26.33	17.30	-7.08	-12.32
El Algarrobillo	ALG-12	ULM2	F	25-35	-3.91	26.83	17.79	-6.33	-12.95
El Algarrobillo	ALG-12	Skull	F	25-35	-5.38	25.31	16.31	-8.61	-10.79
El Algarrobillo	ALG-13	URM3	F	25-35	-3.27	27.49	18.44	-5.32	-12.67
El Algarrobillo	ALG-13	Skull	F	25-35	-5.00	25.71	16.70	-8.01	-10.35
El Algarrobillo	ALG-16	URP1	U	A	-5.08	25.62	16.61	-8.14	-12.49
El Algarrobillo	ALG-16	Skull	U	A	-4.63	26.09	17.06	-7.44	-11.14
El Algarrobillo	ALG-17	URM2	U	A	-3.69	27.06	18.02	-5.97	-11.59
El Algarrobillo	ALG-17	Maxilla	U	A	-5.50	25.19	16.18	-8.80	-11.87
PP4-Montelirio	PP4-M-1	URM1	M	18-25	-2.87	27.90	18.84	-4.71	-13.33
PP4-Montelirio	PP4-M-1	Long bone	M	18-25	-5.34	25.36	16.35	-8.54	-11.59

Table 1. List of human isotope analysis results (Legend: U= upper; R= right; M= molar; L= left; P= premolar; I= incisor; F= female; M= male; U= undetermined; A= adult).

Sample	Structure	Teeth	Age
VCE-1	18	M3Right	Adult
VCE-2		M1Right	Young adult (30-36 months)
VCE-3		M2Right	
VCE-4		M3Right	
VCE-6	195	M1Right	>14 months
VCE-7	115	M3Right	Young adult (30-36 months)

Table 2. List of cattle samples from Pabellón Cubierto sector.

Specimen	Tooth	Distance mm	d 13C -corr (‰ VPDB)	d 18O-corr (‰ VPDB)	d 18O-corr (‰ VSMOW)	d18OP	d1O8dw
VCE-1	M3 R	cement	-7.75	-3.85	26.89	17.85	-4.57
VCE-1	M3 R	36	-10.44	-2.07	28.73	19.65	-3.66
VCE-1	M3 R	33	-10.87	-1.26	29.56	20.46	-2.94
VCE-1	M3 R	30.5	-10.01	-0.62	30.22	21.11	-3.02
VCE-1	M3 R	28	-9.40	-0.69	30.15	21.04	-3.00
VCE-1	M3 R	25.5	-8.57	-0.68	30.16	21.05	-2.78
VCE-1	M3 R	22.5	-8.00	-0.48	30.37	21.25	-2.55
VCE-1	M3 R	18	-7.29	-0.27	30.58	21.46	-2.91
VCE-1	M3 R	15.5	-7.20	-0.60	30.24	21.13	-3.50
VCE-1	M3 R	12	-7.41	-1.12	29.71	20.61	-3.26
VCE-1	M3 R	9	-8.03	-0.91	29.92	20.82	-3.96
VCE-1	M3 R	7	-8.62	-1.53	29.29	20.20	-6.57
VCE-1	M3 R	dentine	-9.45	-2.72	28.06	18.99	-5.30
VCE-2	M1 R	34.0	-6.47	-1.06	29.76	20.66	-3.44
VCE-2	M1 R	30.0	-4.86	-1.04	29.78	20.68	-3.41
VCE-2	M1 R	27.0	-4.79	-1.56	29.25	20.16	-4.00
VCE-2	M1 R	23.5	-4.86	-1.76	29.05	19.96	-4.21
VCE-2	M1 R	20.0	-5.34	-2.40	28.39	19.31	-4.94
VCE-2	M1 R	17.0	-6.35	-2.78	28.00	18.93	-5.36
VCE-2	M1 R	14.0	-7.54	-3.32	27.44	18.38	-5.98
VCE-2	M1 R	11.0	-9.30	-3.64	27.11	18.06	-6.34
VCE-2	M1 R	7.5	-10.33	-3.95	26.78	17.74	-6.69
VCE-2	M1 R	4.0	-11.29	-3.72	27.02	17.98	-6.43
VCE-2	M1 R	cementum	-8.68	-4.72	25.99	16.97	-7.55
VCE-2	M1 R	dentine	-10.93	-3.81	26.93	17.89	-6.52
VCE-3	M2 R	43	-10.89	-2.65	28.13	19.06	-5.22
VCE-3	M2 R	40	-11.19	-2.75	28.03	18.96	-5.33
VCE-3	M2 R	37	-11.37	-2.13	28.67	19.59	-4.63
VCE-3	M2 R	35	-11.72	-1.62	29.18	20.10	-4.07
VCE-3	M2 R	33	-11.40	-0.91	29.92	20.82	-3.26
VCE-3	M2 R	31	-10.98	-1.21	29.61	20.51	-3.60
VCE-3	M2 R	29	-10.59	-0.28	30.57	21.46	-2.55
VCE-3	M2 R	27	-10.17	-0.95	29.88	20.78	-3.31
VCE-3	M2 R	25	-10.05	-0.41	30.43	21.32	-2.71
VCE-3	M2 R	23	-9.75	-0.12	30.74	21.62	-2.37
VCE-3	M2 R	21	-9.54	-0.93	29.90	20.79	-3.29
VCE-3	M2 R	19	-9.49	-1.60	29.21	20.12	-4.04
VCE-3	M2 R	16	-9.54	-1.86	28.94	19.86	-4.33
VCE-3	M2 R	14	-9.66	-3.00	27.77	18.71	-5.6
VCE-3	M2 R	11	-9.61	-3.74	27.01	17.96	-6.44
VCE-3	M2 R	6	-10.01	-4.28	26.45	17.42	-7.05
VCE-3	M2 R	dentine	-10.30	-3.35	27.40	18.35	-7.32
VCE-3	M2 R	cement	-9.03	-4.52	26.20	17.17	-6.01
VCE-4	M3 R	dentine	-9.77	-4.23	26.50	17.47	-6.06
VCE-4	M3 R	cement	-8.54	-4.48	26.24	17.21	-6.91
VCE-4	M3 R	38	-9.68	-3.40	27.35	18.30	-6.26
VCE-4	M3 R	34	-9.90	-4.16	26.57	17.54	-5.68
VCE-4	M3 R	30	-10.49	-3.58	27.17	18.12	-4.05
VCE-4	M3 R	25	-10.99	-3.06	27.70	18.64	-2.36
VCE-4	M3 R	20	-11.25	-1.61	29.20	20.11	-2.09
VCE-4	M3 R	16	-10.54	-0.11	30.75	21.63	-2.09
VCE-4	M3 R	11	-10.02	0.13	30.99	21.87	-2.43

Specimen	Tooth	Distance mm	d 13C -corr (‰ VPDB)	d 18O-corr (‰ VPDB)	d 18O-corr (‰ VSMOW)	d18OP	d1O8dw
VCE-4	M3 R	7	-9.62	0.14	31.00	21.88	-4.59
VCE-4	M3 R	2	-9.18	-0.17	30.69	21.57	-7.28
VCE-4	M3 R	-4	-9.29	-2.09	28.71	19.63	-6.99
VCE-6	M1 R	cement	-8.47	-4.38	26.35	17.31	-7.18
VCE-6	M1 R	32	-11.23	-4.39	26.33	17.30	-5.66
VCE-6	M1 R	28	-12.71	-3.04	27.72	18.66	-5.99
VCE-6	M1 R	25	-12.58	-3.33	27.42	18.37	-5.96
VCE-6	M1 R	22	-12.49	-3.31	27.44	18.39	-6.13
VCE-6	M1 R	19	-12.47	-3.46	27.30	18.25	-6.49
VCE-6	M1 R	15	-12.34	-3.78	26.96	17.92	-7.37
VCE-6	M1 R	12	-12.60	-4.56	26.16	17.13	-7.89
VCE-6	M1 R	9	-12.39	-5.02	25.68	16.66	-8.15
VCE-6	M1 R	6	-12.65	-5.26	25.44	16.42	-8.63
VCE-6	M1 R	3	-12.67	-5.68	25.00	16.00	-7.16
VCE-6	M1 R	dentine	-9.59	-5.00	25.70	16.68	-7.86
VCE-7	M3 R	cementum	-8.76	-4.52	26.20	17.18	-0.50
VCE-7	M3 R	31	-9.08	1.55	32.46	23.30	-1.79
VCE-7	M3 R	27	-9.14	0.40	31.27	22.14	-3.75
VCE-7	M3 R	23	-9.68	-1.34	29.47	20.38	-4.86
VCE-7	M3 R	19	-11.01	-2.33	28.45	19.38	-3.93
VCE-7	M3 R	14	-11.53	-1.50	29.31	20.22	-2.59
VCE-7	M3 R	10	-10.32	-0.31	30.54	21.42	-2.95
VCE-7	M3 R	5	-9.14	-0.63	30.21	21.10	-7.32
VCE-7	M3 R	dentine	-10.57	-4.37	26.35	17.32	-7.16
VCE-8	Molar	Enamel	-0.40	30.45	21.34	-0.86	-10.16

Table 3. Cattle isotope analysis results.

Teeth	Start	Finish	Eruption
M1	In utero	2-3 months	6 months
M2	1 months	13 months	18 months
M3	9-10 months	24 months	30 months

Table 4. Mineralization and tooth eruption in cattle following Hillson (2005).

3. CARBON AND OXYGEN ISOTOPE ANALYSIS METHODOLOGY

Oxygen and carbon isotope analysis recorded in the carbonate fraction (hydroxyapatite) of tooth enamel and bone depend both on the environment and the individual's physiology (Ambrose and DeNiro, 1986; Kohn *et al.*, 1996). Oxygen isotope signatures in water in normal conditions reflect the temperature and precipitations, although this varies depending on some environmental factors, such as altitude and latitude (e.g. Bryant and Froelich, 1996; Craig, 1961, Luz *et al.*, 1984; Sponheimer and Lee-Thorp, 1999, Luz and Kolodny, 1985). In addition, if any environmental and climatic change has occurred during the tooth mineralization, it is also possible to detect this through variations in oxygen isotopes (Balasse, 2002).

Carbon isotope signatures in bone and tooth apatite are meant to reflect the carbon sources in the whole diet, which includes proteins, carbohydrates and lipids (Ambrose and Norr, 1993; Lee-Thorp and Merwe, 1991; Lee-Thorp *et al.*, 1989). Depending on their photosynthetic pathway, different values are given for C_3 or C_4 plants (Tieszen and Chapman, 1992).

The comparison among tooth enamel (first years of life) versus cementum, dentine or bone hydroxyapatite (last stage of life) would allow us to observe changes through time. In addition, intra-tooth sampling performed in the animal´s teeth would enable us to investigate seasonality.

Oxygen and carbon isotope analysis of archaeological hydroxyapatite (bone and enamel) carbonate

($\delta^{18}O_c$ and $\delta^{13}C_c$) were performed at two different institutions, the Department of Geosciences at the University of Tübingen (Germany) and the W.M. Keck Foundation Laboratory for Environmental Biogeochemistry at Arizona State University (United States of America). The former was done using the Gasbench II Finnigan MAT 252 in the Isotope Geochemistry Group at the Department of Geosciences, University of Tübingen (samples were calibrated using NBS-18 ($\delta^{18}O$ = -22.9‰, $\delta^{13}C$ = -5.0‰ V-PDB) and the NBS-19 ($\delta^{18}O$ = -2.2‰, $\delta^{13}C$ = 1.9‰ V-PDB) with a reproducibility of ±0.1‰ for both $\delta^{18}O$ and $\delta^{13}C$ values) and the latter using a Thermo-Finnigan MAT 253 stable isotope ratio mass spectrometer (the reproducibility for NBS-19 is ±0.2‰ for $\delta^{18}O$ and ±0.2‰ for $\delta^{13}C$).

Samples were extracted using three different devices; at Arizona State University, a Dremel Minimite-750 cordless drill and a Dremel 3956-02 Variable Speed MultiPro drill equipped with an engraving cutter were used, and a portable dental drill (model Thumb) was employed at the University of Tübingen. Before extracting the samples, the outer part of the teeth and bones were mechanically removed to avoid any organic material which could cause contamination (Budd *et al.*, 2000; Montgomery *et al.*, 1999; Waldron 1981, 1983; Waldron *et al.*, 1979). Sample preparation followed standard methodologies (Koch *et al.*, 1997) during which about 10 mg of human tooth enamel and bone powder were treated with 0.24 mL of 2% NaOCl and then 0.24 mL of 0.1M CH3COOH. Oxygen and carbon isotope ratios ($\delta^{18}O_c$, $\delta^{13}C_c$) are expressed relative to the VPDB (Vienna PeeDee Belemnite) carbonate standard and are expressed in per mil (‰) using the formula: $\delta^{18}O = (((^{18}O/^{16}O_{sample})/(^{18}O/^{16}O_{standard})) - 1) \times 1000$ (Coplen 1994; Craig 1961). Some other formulae were applied to convert values into drinking water ($\delta^{18}O_{dw}$) $\delta^{18}O_{VSMOW} = (1.03091 \times (\delta^{18}O_{VPDB})) + 30.91$, $\delta^{18}O_{VPDB} = (0.97002 \times \delta^{18}O_{VSMOW}) - 29.98$, $\delta^{18}O_{c(VSMOW)} = (8.5 + (\delta^{18}Op)) / 0.98$, and $\delta^{18}O_{p(VSMOW)} = (0.64 \times (\delta^{18}O_{dw})) + 22.37$ (Coplen *et al.* 1983; Lacumin *et al.* 1996; Muller *et al.* 2003; Wolfe *et al.* 2001).

4. RESULTS

The mean human enamel (n=14) $\delta^{18}O_c$ is -6.1 ±1.1 ‰ (1σ) and the values range from -8.3‰ to -4.7‰ (Table 1). The mean $\delta^{18}O_c$ of the human bone hydroxyapatite (n=15) is -8.5 ±1.6 ‰ (1σ) and the values range from -12.7 ‰ to -6.8‰ (Table 1). The mean $\delta^{18}O_c$ faunal tooth enamel (n=7) is -4.0 ±1.9‰ (1σ); the faunal bone $\delta^{18}O_c$ values range from $\delta^{18}O_c$= -0.4‰ to -6.9‰. The faunal $\delta^{18}O_c$ dentine (n=6) is -5.5 ±2.6‰ (1σ) and the faunal $\delta^{18}O_c$ cementum is -6.5 ±1.1‰ (1σ) (Fig. 4; Table 3). In addition, we proceeded to perform the intra-tooth individual analysis of six teeth, three of them belonging to the same individual. Results are shown in Tables 2 and 3.

In the case of the cattle, an intra-tooth sampling was performed (Tables 2 and 3). Results are plotted from the occlusal part of the tooth crown to the cemento-enamel junction (CEJ) following a time series (Figs. 5 and 6). VCE-1 represents a young adult specimen. It should be taken into account that VCE-2, VCE-3 and VCE-4 belong to the same individual, aged as a young individual. VCE-6 is an individual older than 14 months and VCE-7 is a young adult. Results for sample VCE-1 showed that $\delta^{18}O_c$ on tooth enamel vary from -2.9‰ to -6.9‰, VCE-2 results of $\delta^{18}O_c$ tooth enamel vary from -3.4‰ to 6.4‰, sample VCE-3 results of $\delta^{18}O_c$ tooth enamel vary from -2.3‰ to -7.0‰, VCE-4 results of $\delta^{18}O_c$ tooth enamel vary from -2.0‰ to -7.2‰, VCE-6 results of $\delta^{18}O_c$ tooth enamel vary from -5.6‰ to -8.6‰ and VCE-7 results of $\delta^{18}O_c$ tooth enamel vary from -1.7‰ to -7.3‰ (Fig. 5).

The $\delta^{13}C$ and $\delta^{18}O$ values of dentine and cement of cattle teeth are essentially in the same range as the isotopic values of enamel of their respective teeth, suggesting no significant deviation of the isotopic values from the biogenic ones. Thus, it would appear that $\delta^{13}C$ and $\delta^{18}O$ values in human bone did not suffer significant diagenetic change.

The mean $\delta^{13}O_c$ human tooth enamel (n=14) is -12.7 ±0.5‰ (1σ); the human enamel $\delta^{13}O_c$ values range from – 7.1‰ to -12.4‰. The observed mean $\delta^{13}O_c$ for the faunal tooth enamel (n=7) is -9.8 ±1.5‰ (1σ) (Fig. 6); the faunal enamel $\delta^{13}O_c$ values range from – 7.1‰ to -12.4‰. The faunal $\delta^{13}O_c$ dentine (n=6) is -9.8 ±0.7‰ (1σ) and the faunal $\delta^{13}O_c$ cementum is -8.8 ±1.5‰ (1σ).

The $\delta^{13}O_c$ values showed a difference of 3‰ among humans and faunal remains which might be in relation to the different processes of carbon assimilation between humans and cattle.

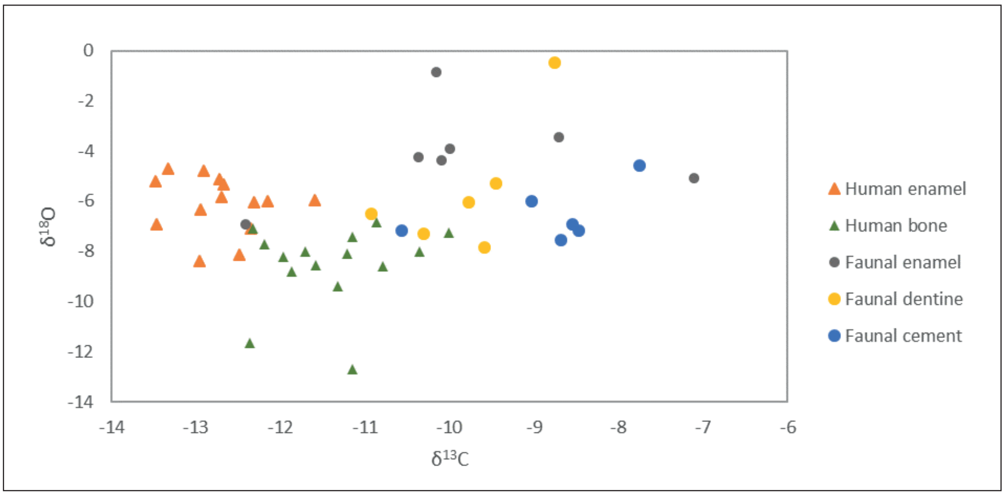

Fig. 4. Results of the δ18O and δ13C for the multi-tissue sampling.

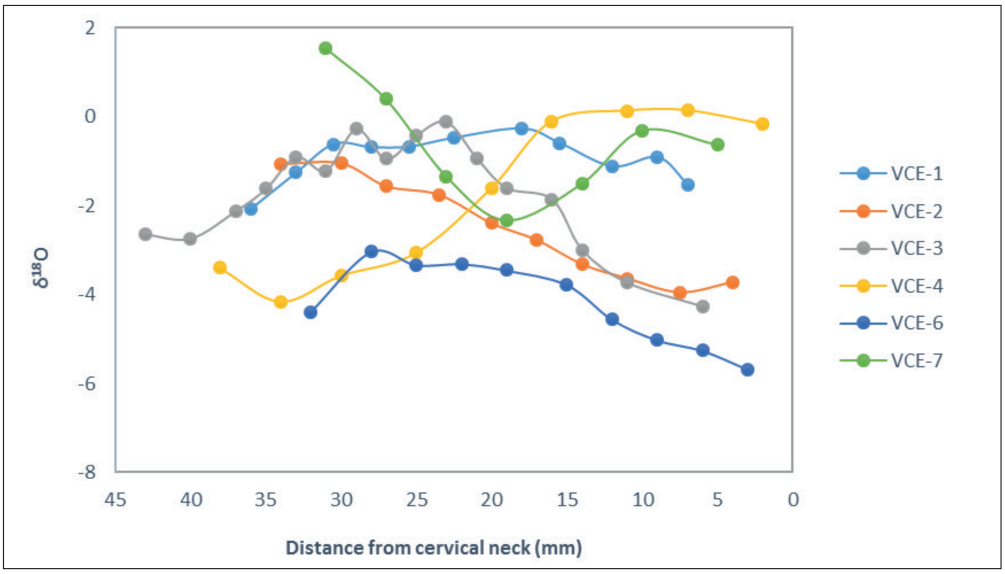

Fig. 5. Cattle tooth enamel δ18O results.

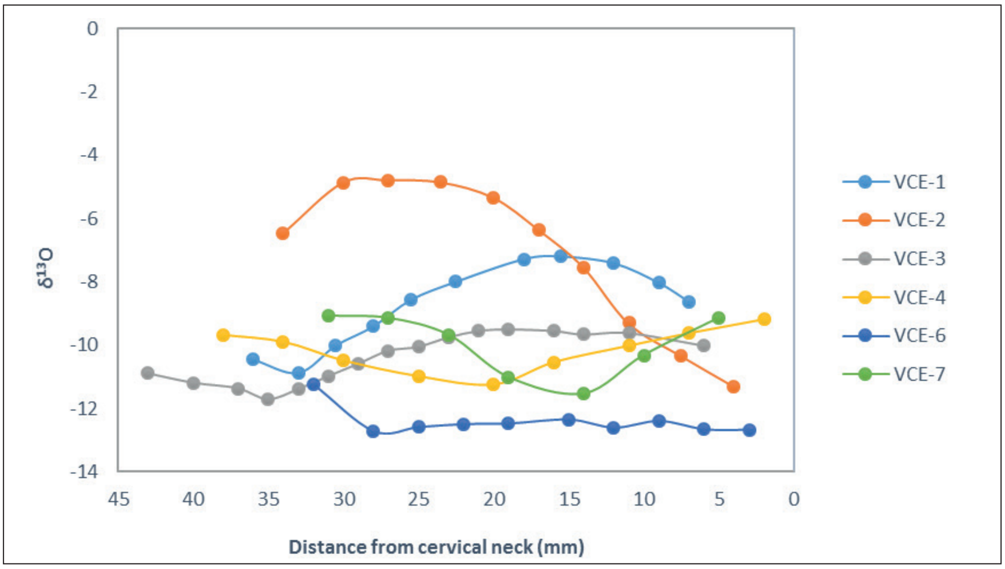

Fig. 6. Cattle tooth enamel δ13C results.

5. INTERPRETATION

The $\delta^{18}O_c$ and $\delta^{13}O_c$ values exhibited by the different tissues measured on humans (tooth enamel & bone hydroxyapatite) and animals (tooth enamel, cementum and dentine) reflect fairly accurately the environment surrounding the mega-site of Valencina. This is shown by the fact that, although bone and dentine – rather than enamel which is more resistant (Hillson, 1997) – , are normally subject to diagenesis, in this case dentine $\delta^{13}C$ and $\delta^{18}O$ values in cattle teeth are similar to those of enamel in their respective teeth, suggesting that bone still retains a biogenic signature.

Assuming that the climate was fairly stable during the Copper Age and no dramatic changes occurred, we can use $\delta^{18}O_c$ to investigate variability in water sources among individuals (human vs. cattle and intra-group), consequently inferring differences in mobility patterns. When comparing the results from $\delta^{18}O_c$ to the modern water precipitation recorded by the IAEA Water Resource Program through the closest GNIP stations to the site, which are located at 1) Morón (Sevilla) and 2) Beja (Portugal), results showed a $\delta^{18}O_c$ mean value of -4.38‰ ± 0.47‰ (1σ, n=11) and -5.06‰ ± 0.50‰ (1σ, n=2), respectively. Comparing the results from the Copper Age to modern precipitation, an increase of $\delta^{18}O_c$ 2‰ over the human enamel mean, albeit similar values for the faunal tooth enamel, can be seen. Therefore, the values are more or less consistent with the results offered by the modern annual precipitation.

The human $\delta^{18}O_c$ enamel data ranges from -8.3‰ to -4.7‰ and the faunal $\delta^{18}O_c$ data ranges from -6.94‰ to -3.47‰, excluding one individual which gives an outlying value of $\delta^{18}O_c$ -0.86‰. Intra-group, both humans and animals show a $\delta^{18}O_c$ variability of ~3.5‰. This difference might be related to the fractionation between diet and carbonate apatite in humans and cattle (Balasse, 2002; Cerling and Harris, 1999) as well as their different dietary habits, seasonal variations and ecological setting, with its subsequent use of the landscape resources. Both groups are quite homogeneous and no evidence of non-local individuals has been recorded according to the $\delta^{18}O_c$ results. In addition, no significant differences were found according to the sex and the age of the human individuals. This is rather interesting, since previous analysis of $^{87}Sr/^{86}Sr$ isotope analyses were performed on those same human individuals and some of them showed non-local values according to the strontium results (Díaz-Zorita Bonilla, 2017). Those outlying individuals (ALC-2, ALG-2, ALG-12 and ALG-13) are showing a difference of >2‰ between $\delta^{18}O_c$ enamel and $\delta^{18}O_c$ bone, which in one case (ALG-2) rises to >7‰. However, the $\delta^{18}O_c$ enamel data is consistent with those of the other individuals identified as local to the area based on their strontium signature (Díaz-Zorita Bonilla, 2017); this means that, while the environment and precipitations in the place where they come from might be similar to conditions in Valencina, the geological background was different.

According to the results of the cattle intra-tooth sampling, $\delta^{18}O_c$ enamel data shows that there is a large variability in the oxygen values for most of the samples, which might be in relation to mobility patterns and habitat distribution. While some individuals represent certain degree of mobility, including seasonal changes such as sample VCE-7, samples VCE-2, VCE-3 and VCE-4 show the most striking values.

The two cases of M1 show rather similar patterns (VCE-2 and VCE-6) with lower $\delta^{18}O_c$ values and more negative values towards the CEJ, reflecting the very first stage of an individual life, including the first bands which are reflecting the stage *in utero*. In contrast, assuming that for M2 and M3 mineralization reflects the post-birth phase, the initial bands close to the occlusal surface of the tooth are indeed reflecting the local values for Valencina, even considering that there is an offset due to the different seasons recorded, which explains why the VCE-4 values are more negative.

The VCE-7 individual, which is a M3, reflects changes through time and possibly suggesting a provenance from a different environment; the very early signature reflected in their occlusal surface does not correspond at all with the local environment of Valencina. However, later in life that individual migrated to a different ecological area (thus is reflecting the negative $\delta^{18}O_c$ values) with seasonal changes through time and the last stage, close to the CEJ, matches the local values; therefore, this individual was brought to Valencina from a different place, one which had different water sources and/or precipitation values.

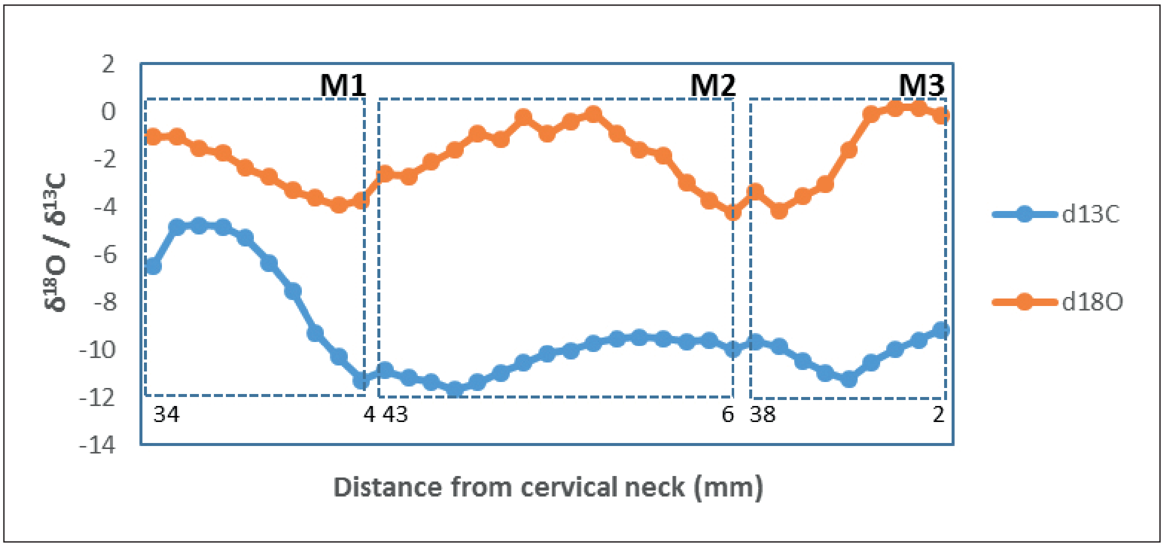

Fig. 7. Intra-individual cattle tooth $\delta^{13}C$ and $\delta^{18}O$ results.

Samples VCE-2, 3 and 4 (which belongs to the same individual) display interesting values, since seasonal movements can also be detected (Fig. 7). While the M2 and M3 show a $\delta^{18}O_c$ difference of ~1‰ at the early stage which reflects the local values for Valencina, movement is detected during the first year of life possibly to areas with higher $\delta^{18}O_c$ values where there is less precipitation, higher temperature and more aridity; in the last stage, more negative $\delta^{18}O_c$ values are observed again which correspond to the Lower Guadalquivir valley.

In general, the $\delta^{13}C_c$ values are consistent for the human and for the animal group independently, but there is a difference of about 3‰ between both groups. This difference of 3‰ is due to the difference in fractionation between diet and carbonate apatite in humans and cattle (Passey *et al.*, 2005), even though they have a diet with a similar $\delta^{13}C_c$. The human enamel data show values of av. -12.72‰ ±0.5‰ (1σ) and the bone hydroxyapatite values of av. -11.39‰ ±0.7‰ (1σ). More positive values can be observed for the cattle, where the enamel values showed and average of -9.84‰ ±1.5‰ (1σ) (Fig. 6), the cementum av. -8.84‰ ±0.9‰ (1σ) and the dentine av. -9.80‰ ±0.7‰ (1σ). In any case, those $\delta^{13}C_c$ values are reflecting a diet based on C_3 plants, as is the case for Western Europe (Fig. 4). This difference in $\delta^{13}C_c$ possibly mirrors that of dietary habits of humans and cattle, but also the habitat of the species. Furthermore, it should be taken into account that physiological changes and metabolism patterns work differently for humans and for cattle. When analyzing the data in pairs (tooth enamel vs. bone apatite) it seems that, for those outlying individuals, based on the $^{87}Sr/^{86}Sr$ isotope analyses, there is a larger difference in $\delta^{13}C_c$, which in some cases is about ~2‰, the bone apatite samples being always more positive. Therefore, this might reflect the environment where individuals were spending their childhood, possibly a wetter landscape with more dense vegetation (tooth enamel signatures), a more open area and a drier environment (bone apatite), as is Valencina.

For the intra-tooth analysis of individual VCE-2, 3 and 4, although some changes are observed in the M1 (*in utero*-first phase of life of the individual) where C_4 plant consumption is reflected, the M2 and the M3 show similar values, therefore reflecting the same subsistence patterns from 2-3 months old through time reflecting a shifting to C_3 plants consumption (Fig. 7).

6. CONCLUSIONS

This study, which is the first to combine $\delta^{18}O_c$ and $\delta^{13}O_c$ human and animal isotopical data to investigate the mobility patterns ecological and dietary habits of the mega-site of Valencina, has proven the potential of biochemistry analysis. The $\delta^{18}O_c$ and $\delta^{13}O_c$ values exhibited by the different tissues measured on humans (tooth enamel and bone hydroxyapatite) and animals (tooth enamel, cementum and dentine) reflect fairly accurately the environment surrounding the mega-site of Valencina. Indeed all tissues (enamel, bone and dentine) were reflecting quite well the local signal meaning that bone were still biogenic. As a result, the multi-tissue analysis shows differences in relation to age, season and habits. Although the human and the faunal tooth enamel reflect the local $\delta^{18}O_c$ values for Valencina, some seasonal movement has been observed amongst the cattle. The potential of intra-tooth analysis showed also movements during the life of an individual and changes in the subsistence patterns. The $\delta^{13}O_c$ values are reflecting mostly a C_3 plants diet and differences in $\delta^{13}O_c$ among human and animals are related to subsistence strategies and habitat of the species.

This new application using carbon and oxygen isotopic analysis to investigate mobility and subsistence patterns among humans and animals opens new insights into the investigation of the subsistence patterns and social dynamics during the 3rd millennium BC in Iberia.

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