[Bone stable isotope data of the Late Roman population \(4th–7th centuries CE\) from Mondragones](https://doi.org/10.1016/j.jasrep.2020.102566) [\(Granada\): A dietary reconstruction in a Roman villa context of south-eastern Spain](https://doi.org/10.1016/j.jasrep.2020.102566) © 2020 by Paula Fernandez-Martinez, Anne-France Maurer, Nicasio T. Jiménez-Morillo, Miguel C. Botella López, Belén López Martínez, Cristina Dias Barrocas is licensed under [CC BY-NC-ND 4.0](https://creativecommons.org/licenses/by-nc-nd/4.0/?ref=chooser-v1)

1 Bone stable isotope data of the Late Roman population (4th–7th centuries CE) from

2 **Mondragones (Granada): A dietary reconstruction in a Roman villa context of**

3 **south-eastern Spain**

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23 **Abstract**

24 The aim of this study is to examine the diet, using bone stable isotope analysis ($\delta^{13}C$ and δ^{15} N), of a Late Roman population (4th–7th centuries CE) from the Roman villa of Mondragones (Granada, Spain). This archaeological site presents an exceptionally high number (*n* = 121) of well-preserved skeletal remains (adults and non-adults), giving the 28 opportunity to study for the first time the nutritional and health conditions of a Late Roman population by the analysis of stable isotopes and pathologies in the context of the south-30 eastern Iberian Peninsula. Stable isotopes ratios of carbon ($\delta^{13}C$) and nitrogen ($\delta^{15}N$) were analysed in 46 individuals (21 adults and 25 non-adults) as well as in 7 faunal samples (2 *cows/ox,* 2 *goats/sheep,* and 3 *large mammals)*. Frequencies of cariogenic lesions, dental calculus, dental enamel hypoplasia, porotic hyperostosis, and cribra orbitalia were also explored. The anthropological study revealed a high presence of dental caries and calculus in adults, which are related to a diet rich in starch and carbohydrates, and non-specific stress markers in non-adults, probably pointing to the weaning process or childhood diseases. Collagen isotope ratios suggested that the 38 population of Mondragones had a diet rich in C_3 plants, with some meat intake from terrestrial herbivores. There were significant differences between non-adults and adults, 40 but no differences were detected by sex. The youngest non-adults (aged 1 year ± 4 41 months) showed the $\delta^{15}N$ mean value almost 4‰ above the adult female one, which could reflect the breastfeeding period.

Keywords: paleodiet, isotopes, collagen, breastfeeding, Late Antiquity, Spain.

1. Introduction

 The study of stable isotopes in skeletal remains has gained importance in recent years in the context of the Iberian Peninsula. While there are a lot of paleodietary data in Spain from prehistoric (Salazar García, 2011; Fontanals-Coll et al., 2016; Villalba-Mouco et al., 2018) and medieval sites (Inskip et al., 2019; Guede et al., 2017; Jiménez-Bobreil et al., 49 2020), there is a deep gap for the Roman and Late Roman period, except for some works such as López-Costas (2012). There is a lack of studies in Spain for this specific period in comparison with near geographical areas as Italy (Rutgers et al., 2009; Tafuri et al., 2018; Milella et al., 2019), maybe because it is usually very difficult to find many skeletal remains from the Late Roman period with good conservation conditions in the Iberian Peninsula as a result of cremation practices and taphonomic processes (Polo Cerdá and Garcia-Prosper, 2005; Heras Martínez et al., 2011; López-Costas, 2012; Diéguez Ramírez, 2015). This paleodiet study is focused on a sample of the Late Roman and 57 Late Antiquity population $(5th$ to $7th$ century CE [Common Era]) buried at the Roman villa of Mondragones, located in south-eastern Spain. Therefore, it represents a great opportunity to know more about the nutritional conditions of this period on the Iberian Peninsula.

 The study of stable isotopes in humans provides good quality data for the reconstruction of ancient populations' diets (Reitsema, 2013; Ma et al., 2016). Specifically, with this analytical technique it is possible to assess the type of vegetables that were consumed, as well as the sources of the dietary proteins (animal or vegetal). This knowledge provides direct information on aspects that otherwise could only be inferred by indirect evidence, such as food preparation utensils, storage vessels, or wall and vase paintings (Keenleyside et al., 2009). The stable isotope technique is based on the principle that the isotopic composition of tissues in both humans and animals is determined by the diet. Therefore, analysis of δ¹³C and δ¹⁵N can provide information about the diet of past populations (Budd et al., 2013; Müldner and Richards, 2007).

 I_1 In C₃ plants, the δ¹³C value ranges between -20‰ and -35‰, while in C₄ plants the values vary between -7‰ and -17‰ (Pate, 2001). Consequently, carbon isotope values 73 can be used to distinguish between C_3 and C_4 plants. These differences in carbon isotope values are transferred along the food chain to animals and humans, making it possible 75 to determine the kind of plant ingested (Van der Merve and Vogel, 1978). Furthermore, 76 carbon isotope composition in the atmosphere reaches an average $δ^{13}C$ value of -7‰, while in the sea, dissolved carbonates display a value of 0‰ (Rullkötter, 2006).

 Therefore, carbon isotope values can differentiate the consumption of marine and terrestrial food sources (Craig et al., 2009; Reitsema et al., 2010).

 Nitrogen isotope in bone collagen provides information about trophic level (increase by 3–5‰ with increasing trophic level), distinguishing among herbivores, omnivores, and carnivores (Hedges and Reynard, 2007; Keenleyside et al., 2009). Moreover, an 83 increase in $\delta^{15}N$ bone collagen values is observed for individuals with significant 84 ingestion of marine products, either direct or indirect (i.e., sea spray) (Schoeninger and 85 DeNiro, 1984; Schoeninger, 1995). Indeed, atmospheric N₂ dissolved in water is 86 converted into ¹⁵N-enriched nitrates and ammonia, leading to generally more positive δ^{15} N values in the marine food web compared to terrestrial vertebrates (Pate, 1994). The nitrogen isotope ratios can also be used to estimate the duration of breastfeeding and 89 the timing of the weaning process (Fuller et al., 2006), which are very important because these processes have an impact on the health condition of a population and on its demography. The relation between nitrogen isotope ratios and the breastfeeding/weaning period has been analysed in many studies (Dupras et al., 2001; Turner et al., 2007; Prowse et al., 2008; Keenleyside et al., 2009; Bourbou et al., 2013), but it has limitations associated with the cross-sectional method applied. These studies are based on sampling non-survivors, which may not be representative for inferring population norms, and they do not consider the population and individual variation (Kendall, 2016), so such studies require caution.

 In addition, combining the stable isotope ratios with an osteological analysis of skeletal remains can provide information on health conditions and complement the dietary patterns (Toso et al., 2019). Pathological conditions caused by interruptions in growth are especially interesting in the paleodiet context, because they could suggest periods 102 of malnutrition or lack of specific essential nutrients (Katzenberg, 2012). Caries, dental calculus, and non-specific stress markers (such as cribra orbitalia, porotic hyperostosis, and dental enamel hypoplasia) are frequently considered in paleodiet studies (Buzon et al., 2012; Laffranchi et al., 2019), as well as in this research.

 Dental caries is considered one of the most important tools to reconstruct the diet of past populations, because its aetiology is related to fermentable carbohydrates from the diet (Hillson, 2001; Svyatko, 2014). It is an oral pathology characterized by demineralization and progressive destruction of calcified dental tissues by bacterial fermentation of carbohydrates (Hillson, 2019), although it is also affected by other factors such as 111 salivary glycoproteins, dental plaque, or deficient oral hygiene (Lopez et al., 2012). Dental calculus is produced from the accumulation of plaque that, if not removed, 113 becomes mineralized (Scott and Poulson, 2012). Plaque accumulates faster in an alkaline oral environment, which occurs when the diet is rich in proteins and/or

115 carbohydrates (Roberts and Manchester, 2010). Calculus is also influenced by salivary 116 flow, genetic factors, and dental care (Hardy et al., 2009).

 Cribra orbitalia and porotic hyperostosis are non-specific stress markers identified macroscopically as porous lesions of the orbital roof and cranial vault, respectively (Suby, 2014). While iron-deficiency anaemia is the most accepted aetiological factor for these pathologies (Oxenham and Cavill, 2010; Rivera and Mirazón, 2017), other studies suggest that cribra orbitalia and porotic hyperostosis could be linked to megaloblastic 122 anaemias, which are associated with deficiencies of vitamin B_{12} and vitamin B_9 (Walker et al., 2009). However, they are also related to multiple aetiologies like inflammatory, haemorrhagic, or tumoral processes (Ortner, 2003). Finally, dental enamel hypoplasia is another non-specific stress marker characterized by the formation of lines, pits, or grooves on the enamel surface (Roberts and Manchester, 2010). It can be related to 127 dietary deficiencies, childhood fevers, and infectious diseases (Hillson, 2019), and it can provide information about lifestyle and living conditions (Goodman and Rose, 1991; Laffranchi et al., 2019). These defects are formed only during enamel development, so 130 they can record the stress periods of childhood (Mays, 1998).

 The analysis of stable isotopes and the paleopathological variables from this particularly well-preserved Late Roman population from southern Spain provides the opportunity to assess the nutritional conditions of this little-known period and to compare it with chronologically contemporary populations.

2. Material and methods

2.1 Archaeological site

 The Roman villa of Mondragones is located in the city of Granada (Fig. 1), Andalusian region, Spain (37°11'26.46"N; 3°36'41.16"W), and it was first discovered and investigated in 2013 by Rodríguez Aguilera et al. (2014). This villa was built in the middle 140 of the 1st century CE, and the period of occupation was documented until the $7th$ century CE. It was a periurban villa, located 1.7 km from the urban nucleus of the *Municipium Florentinum Iliberritanum*, which is the Roman name of Granada. It showed the typical roman structures of agrarian villas, which have a productive function: the *pars rustica*, the *pars frumentaria*, and the *pars urbana* (Fornell-Muñoz, 1999). These villas were owned by the *dominus*, who lived in the *pars urbana* with his family and the service, while the labour force lived in the *pars rustica*, where the stables were also located (Joly, 2003)*.* The *pars frumentaria* was the centre of the agricultural production, and in Mondragones it was formed by an oil mill. It seems that Mondragones belonged to a group of villas dedicated to the agricultural exploitation of the fertile plain of Granada, mainly centred on agriculture, whose objective was to supply the area and probably export the excess 151 production (Sánchez López, 2013). From the 5th century CE, the villa was restructured

 with successive modifications, such as the reduction of space for oil production. In the 153 second half of the 6th century CE, a religious building and a cemetery were built. A total of 85 graves have been uncovered: 23 belong to the Imperial Roman time (1st century CE) and 62 belong to the Late Roman and Late Antiquity time. These 62 graves are 156 divided into 2 phases: [1] 23 graves dated from the $4th$ to $5th$ century CE, and [2] 39 157 graves dated from the 5th to 7th century CE (Fig. 2). Most individuals were inhumed in a *decubitus supinus* position with an East–West orientation (Rodríguez Aguilera et al., 2014).

- Fig. 1. Map showing the location of Mondragones (Google Earth V 7.3.2.5776 (64-bit) (May 17,
- 2018). Andalusian region, Granada, Spain. 37°11'26.46"N; 3°36'41.16"W, eye alt 247.03 km.
- Google 2018).

 Fig. 2. Plan of the Late Roman cemetery of Mondragones (adapted from Rodríguez Aguilera et al., 2014).

2.2 Archaeological samples

168 Although the site was used from the 1st to the $7th$ century CE, only Late Roman and Late 169 Antiquity ($4th$ to $7th$ century CE) individuals were selected to reduce cultural variability. The collagen turnover rate of sampled bones is an important key in the interpretation of 171 paleodiet (Hedges et al., 2007). Ribs and femurs are usually chosen for this kind of study owing to their faster turnover rates, which reflects diet from a recent period prior to death (Fahy et al., 2017). As many samples as possible were selected to make a representative analysis. Ribs from 46 individuals buried in Mondragones were sampled, including 21 adults (9 females, 3 males, 9 undetermined) and 25 non-adults (7 from 1 to 3 years old, 11 from 4 to 8 years old, and 6 from 10 to 18 years old). None of the selected ribs showed any signs of pathology or fractures. Of these 46 samples, 20 belong to phase 1 and 26 to phase 2.

- Seven samples of faunal remains recovered from the site were analysed for the baseline. Zooarchaeological characterization identified 2 different species: 2 samples of cow/ox (*Bos taurus*) and 2 samples of goat/sheep (*Capra hircus/Ovis aries*). The other remains (all ribs) were classified as large mammals (perhaps cattle).
- 2.3 Anthropological and paleopathological methods

 Human sex and age estimations were carried out using various standard methods based on cranium, mandible, and hip bones to increase the determination accuracy. Age 186 estimation methods in adults included the morphology of the pubic symphysis (Todd, 1920, in White et al., 2012) and the alterations of the auricular surface of the ilium (Schwartz, 1995). In non-adults, age estimation methods included dental development (Ubelaker, 1978, in Scheuer and Black, 2000) and long bone length (Maresh, 1970, in Scheuer and Black, 2000). Biological sex was determined only in adults using the morphology of the pelvis and different cranial and mandible features, including the nuchal crest, mastoid process, supraorbital margin, prominence of the glabella, and mental eminence (Buikstra and Ubelaker, 1994). Stature was estimated using the formulae developed by De Mendonça (2000) in a Portuguese adult population. This method measures the maximum and physiological length of the humeri and femora to determine stature in centimetres (cm).

 To consider all variables that could affect the dietary interpretation, a paleopathological study of the individuals was carried out. Pathological conditions were analysed macroscopically using multiple descriptions. For this study, mainly pathologies related to diet were considered, such as dental (cariogenic lesions and dental calculus) and non- specific stress markers (cribra orbitalia*,* porotic hyperostosis, and dental enamel hypoplasia). Dental caries was analysed using the system of Moore and Corbett (1971), modified by Buikstra and Ubelaker (1994). Dental calculus was measured according to Brothwell's (1981) description. As for cribra orbitalia, porotic hyperostosis, and dental enamel hypoplasia, only the presence or absence of pathology was measured, with the limitation of not having radiographic data. Aufderheide and Rodríguez-Martín (1998), 207 Baxarias and Herrerin (2008), and Roberts and Manchester (2010) were followed in the description of these non-specific stress markers.

2.4 Bone collagen extraction

 Collagen was extracted using a modification of the method originally developed by Longin (1971) (Britton et al., 2008; Knipper et al., 2013; Salesse et al., 2014; Saragoça et al., 2016). In brief, around 0.5 g of human and faunal bone samples were collected and cleaned with a Dremel Rotary Tool. Bone samples were demineralized in 10 mL 0.5 214 M HCl at 4 \degree C for 14 days, with regular vortex and changing the acid after 1 week. To oxidize fulvic and humic acids, samples were rinsed to neutrality with Milli-Q water and soaked in 0.125 M NaOH for 20 h at room temperature. Samples were rinsed again to 217 neutrality with Milli-Q water and gelatinized in 0.01 M HCl at 70 °C for 48 h, with regular vortex. The liquid fraction containing solubilized collagen was filtered using Ezee-Filter separators (Elkay Laboratory Products), frozen with liquid nitrogen, lyophilized for 48 h,

and analysed. Collagen extraction was performed in the HERCULES Laboratory (Évora,

Portugal).

2.5 Stable isotope analysis

223 The carbon and nitrogen isotope composition $(^{13}C/^{12}C$ and $^{15}N/^{14}N$, respectively) of the collagen samples were determined by elemental analysis/isotope ratio mass spectrometry (EA/IRMS). The EA/IRMS system consisted of a Flash 2000 HT elemental analyser (Thermo Scientific, Bremen, Germany) with 2 reactors: i) combustion (C, N, and S) and ii) pyrolysis (H and O). The elemental analyser was coupled with a ConFlo IV (Thermo Scientific) continuous flow open split interface to a Delta V Advantage isotope ratio mass spectrometer (Thermo Scientific).

 Carbon and nitrogen isotope analysis used a helium carrier gas at a flow rate of 95 mL/min. Aliquots of collagen samples (between 0.5 and 0.6 mg) together with the calibration standards (approx. 0.6 mg) were weighed in tin cups (IVA Analysentechnik GmbH & Co. KG, Meerbusch, Germany). The cups were closed, folded, pressed to a small size, and loaded in a MAS 200R (Thermo Scientific).

 The stable isotope standard for carbon is Vienna Pee Dee Belemnite limestone (VPDB), 236 and it is Vienna air (V-Air) for nitrogen. The standards used (IAEA 600, IAEA CH $_6$, and 237 IAEA N₂) are recognized by the International Atomic Energy Agency (IAEA) (Valkiers et 238 al., 2007). The standard deviations of bulk δ^{13} C and δ^{15} N were ± 0.1‰ and 0.2‰, respectively. Carbon and nitrogen isotope composition was measured in duplicate for

each sample.

2.6 Statistical analysis

 Statistical analysis was performed using SPSS v.24.0 for Windows and Microsoft Excel for Windows. Stable isotope results were compared by age, sex, phase, population, and pathological condition using a non-parametric Mann–Whitney *U* test because the data do not follow a normal distribution.

- 3. Results and discussion
- 3.1 Anthropological and paleopathological results

 A total of 21 adults (9 females, 3 males, and 9 undetermined) aged between 18.5 years and 41.5 years were analysed (Table 1). Stature was estimated for 8 individuals whose sex determination was possible, using the physiological length of the femur. The male 251 average was 164.44 ± 6.90 cm $(n = 3)$, while the female average was 154.16 ± 5.92 cm (*n* = 5). Furthermore, stature was also estimated in females using the maximum length of the humerus, because this measure could be taken in more individuals of this sex (*n* 254 = 7), obtaining an average of 154.96 \pm 7.70 cm. These values are consistent with the 255 results of other populations of the same timeline that used the De Mendonça (2000) 256 method (López-Costas, 2012; Saragoça et al., 2016).

257 Nine adults (out of 21) showed cariogenic lesions, and 12 displayed dental calculus

258 (Table 1). Four individuals presented non-specific stress markers. Specifically, one

259 (MON-045A) shows cribra orbitalia, two (MON-059A and MON-059B) present dental

- 260 enamel hypoplasia, and one (MON-051A) shows both enamel hypoplasia and porotic
- 261 hyperostosis.

262 Among the 25 non-adults analysed, 5 presented cariogenic lesions and 3 showed dental

- 263 calculus. Seven non-adults displayed cribra orbitalia*,* and one non-adult showed porotic 264 hyperostosis. Furthermore, 2 cases of dental enamel hypoplasia were detected.
- 265 All these pathologies have been observed in similar contemporary populations from
- 266 Iberian and Italian Peninsulas (i.e., Ortega Pérez and De Miguel Ibáñez, 1997; Facchini

267 et al., 2004; Belcastro et al., 2007; Cardona López, 2009).

- 268 Table 1. Anthropological (age-at-death, sex, and stature) and paleopathological data of the
- 269 individuals from Mondragones. n.d.: not determined; n.a.: not assessable; -: feature absent; LR:
- 270 Late Roman; LA: Late Antiquity.

271 3.2 Collagen quality

 According to well established criteria, collagen extraction was successful for human and faunal bone samples: collagen yields >1%, (van Klinken, 1999); carbon content between 15.3% and 47.0% (Ambrose, 1990); nitrogen content between 5.5% and 17.3% (Ambrose, 1990); and C/N ratios between 2.9 and 3.6 (DeNiro, 1985). All samples displayed collagen content ranging from 5% to 15%. The carbon and nitrogen content in the bone collagen showed ranges between 37.1% and 43.8%, and between 13.6% and 16.1%, respectively. The atomic C/N ratios of bone collagen ranged between 3.0 and 3.3. Consequently, all the samples analysed in this study were considered well preserved (Table 2).

- 281 Table 2. Carbon and nitrogen stable isotope results and collagen quality indicators for human
- 282 and faunal samples.

283 3.3 Dietary patterns

284 The isotopic composition (δ¹³C and δ¹⁵N) of human and faunal samples are listed in 285 Table 2 and represented in Fig. 3.

286 δ^{13} C data from animal samples ranged from -21.0‰ to -19.1‰ and thus related to a diet 287 based predominantly on C_3 plants. Most of the faunal samples displayed more depleted 288 δ^{13} C values ($\delta^{13}C_{\text{average}} = -20.1 \pm 0.7\%$) than the human ones ($\delta^{13}C_{\text{average}} = -18.6 \pm 0.5\%$), 289 reflecting a δ^{13} C increase for one trophic level. However, there was one outlier (UE-750, 290 a large mammal) with a δ^{13} C value of -19.0‰ (Fig. 3). The δ^{13} C value of this sample may 291 be due either to the consumption of C_4 plants, such as millet, or to the type of plant tissue 292 consumed, because there are organs more enriched with δ^{13} C than others (Dungait et 293 al., 2011). Another possibility may be that this sample was incorrectly classified as a 294 large mammal when it could belong to a species like domestic pig, which is normally 295 clustered with human data (Ren et al., 2017). According to evidence from the literature, 296 these animals had a conspicuous importance in the Roman economy and diet (Prowse 297 et al., 2004). Although the identification of this rib was not possible, the presence of pigs 298 in the Iberian Peninsula has been recorded for the Roman period (Grau-Sologestoa, 299 2015), so this may support the idea that in Mondragones there were domesticated pigs. 300 Concerning the ¹⁵N composition, the values recorded in the collagen of the faunal 301 samples ranged from 6.4‰ to 8.6‰ ($\delta^{15}N_{\text{average}} = 7.4 \pm 0.8$ %), where the highest $\delta^{15}N$ 302 values belonged to large mammals (7.8–8.6‰). If it is assumed that these faunae were 303 cattle, these results may be linked to the manuring practices. The application of animal 304 dung to fields where domesticated animals were kept would generate a $\delta^{15}N$ enrichment 305 in the soil and plants (Bogaard et al., 2007) as well as in the bone of animals and human 306 consumers (van Klinken et al., 2002; Bogaard et al., 2007). However, the highest 307 nitrogen value of these large mammals belonged to the outlier UE-750 (8.6‰), so these

308 results must be interpreted cautiously, as species determination for these samples was 309 not possible.

 A diachronically isotopic study was realized to establish whether there were differences among individuals from the 2 chronological phases. The *U* test showed that there were 312 no significant differences between Late Roman and Late Antiquity individuals ($P_{\text{carbon}} =$ 0.367, *P*nitrogen = 0.929), so both phases were analysed together. The lack of significant differences between phases could be related to continuities of dietary and/or farming practices since the population of Mondragones lived mainly from their agriculture production.

317 The δ^{13} C values of non-adults ranged from -19.3‰ to -17.4‰ (δ^{13} C_{average} = -18.5 ± 0.6‰), 318 while their $\delta^{15}N$ values ranged from 8.8‰ to 14.6‰ ($\delta^{15}N_{\text{average}} = 10.7 \pm 1.7\%$). The 319 averages of carbon and nitrogen isotope composition, respectively, in non-adults by age 320 groups were: $[1] -17.9 \pm 0.5\%$ and $12.8 \pm 1.8\%$ for non-adults from 0 to 3 years $(n = 7)$, 321 [2] -18.6 ± 0.4‰ and 10.1 ± 0.5‰ for non-adults from 4 to 8 years (*n* = 11), and [3] -18.9 $\pm 0.3\%$ and $9.4 \pm 0.5\%$ for non-adults from 9 to 18 years ($n = 6$). The $\delta^{13}C$ values for 323 adults (males and females) ranged from -19.4‰ to -18.0‰ ($\delta^{13}C_{average}$ = -18.7 ± 0.3‰), 324 and their $\delta^{15}N$ values ranged from 8.6‰ to 11.2‰ ($\delta^{15}N_{\text{average}} = 9.9 \pm 0.6$ ‰) (Table 3).

325 The δ^{13} C and δ^{15} N values were significantly different between adults and non-adults 326 between 0 and 3 years ($P_{\text{carbon}} = 0.000$; $P_{\text{nitrogen}} = 0.000$), but there were no differences 327 between adults and the other groups of non-adults (*P* > 0.05). These results show that 328 as the age of the non-adults increases, the results become more similar to those 329 obtained in adults. Furthermore, the δ^{13} C and δ^{15} N values of human samples showed no 330 significant differences in the diets of different sex ($P_{\text{carbon}} = 0.482$, $P_{\text{nitrogen}} = 0.864$).

332 Human samples displayed relatively higher δ^{13} C and δ^{15} N values in comparison to the 333 fauna ($P_{carbon} = 0.000$; $P_{nitroden} = 0.000$), approximately 1.4‰ and 2.5‰ for C and N, respectively (Fig. 3). This is indicative of an increase of one trophic level between fauna and humans (Katzenberg, 2008), which suggests that animal meat may be a prominent protein resource in the diet of the population of Mondragones. In Roman culture, domesticated animals were kept not only for food provision but also as important beasts

338 of burden, for example in the case of cattle (Cool, 2006). The meat component of the diet in this period came primarily from pigs, with a minor component of sheep and goats (approximately 25% to 35%), whose main function was to produce wool, milk, and cheese (Prowse et al., 2004). However, in the Iberian Peninsula, there is a good degree of variability that suggests that a local pattern persisted (King, 1999). This pattern, in most of the studied sites, consisted in a relatively low pig percentage (20% or less) and a higher percentage of sheep/goat and ox (King, 1999). In any case, the meat influence on the Roman diet is well documented (Alcock, 2006; Cool, 2006; Faas, 2013) and illustrated by the stable isotopic composition of the population from Mondragones.

347 The collagen average δ^{13} C values of individuals with a diet based on C₃ plants are 348 around -20‰, while for individuals who consume a diet rich in C_4 plants this average is 349 around -10‰ (Keenleyside et al., 2009). Taking this into account, the δ^{13} C values in adult 350 humans ($\delta^{13}C_{average}$ = -18.7 ± 0.3‰) could be indicative of C₃ consumption with some 351 contribution of C₄ plants, possibly millet. Although there is no archaeobotanical evidence 352 of millet cultivation in Mondragones, there is evidence of its cultivation in the south-353 eastern Iberian Peninsula during the Bronze Age in locations close to Mondragones 354 (Moreno-Larrazabal et al., 2015). Moreover, its presence is widely documented in most 355 of the archaeological sites from the Bronze Age in Mediterranean geography (Peña-356 Chocarro, 1999; Rovira Buendía, 2007; Buxó and Piqué, 2008). Nevertheless, in 357 Mondragones an increase in $\delta^{13}C$ values was observed along with the $\delta^{15}N$ values (see 358 Fig. 3). C_4 consumption increases the collagen δ¹³C values, while δ¹⁵N values should not 359 be affected, so this allows distinguishing between the consumption of C_4 plants and 360 aquatic resources, which are usually also enriched with ¹⁵N (Schoeninger and DeNiro, 361 1984; López-Costas et al., 2015). The pattern observed in Mondragones, of concomitant and significant increase in human bone δ¹³C and δ¹⁵N values (R^2 = 0.2801, *P* = 0.0005), 363 can be due to fish consumption, which is consistent with its proximity to the 364 Mediterranean Sea. The inclusion of other isotopes (e.g. sulphur, $\delta^{34}S$) could clarify the 365 effective presence of a marine contribution in the diet of this population (Nehlich et al., 366 2010, Curto et al., 2019).

367

368 Fig. 3. Carbon and nitrogen isotope data from humans and faunal remains recovered at 369 Mondragones.

370 Most of the human collagen δ^{13} C and δ^{15} N values were closely clustered, with the exception of 5 outliers (MO-041A, MO-043B, MO-056B, MO-086B, and MO-093A) 372 associated with an enriched $\delta^{15}N$ value (Fig. 3). All of them were non-adults (1 year ± 4 373 months), whose average $\delta^{15}N$ value of 13.7 \pm 0.8‰ was almost 4‰ above the average δ^{15} N value for the adult female population of Mondragones (9.9 \pm 0.6‰) ($P = 0.000$). 375 This high average $\delta^{15}N$ is probably indicating the period of breastfeeding. Values for $\delta^{15}N$ have been used to study breastfeeding and weaning patterns, because when infants are at breastfeeding age, their trophic level is one unit above their mothers. This is due to 378 they are basically consuming their mother's tissue by the breast milk (Fuller et al., 2006). But the association between isotope data and the breastfeeding/weaning process involves the assumption that the bone collagen isotopes are representative of diet at approximately the time of death and that non-adults who died are representative of their age group (Beaumont et al., 2015). This is not considering the "Osteological Paradox" (Wood et al., 1992), which suggests that non-adults who have died may not be representative of the health conditions of the whole population. Increased nitrogen ratio could also reflect other conditions than the period of breastfeeding, such as a metabolic disorders or maternal stress episodes during pregnancy (Siebke et al., 2019). Moreover, 387 a negative nitrogen balance could also produce an increase in $\delta^{15}N$ values (Laffranchi et al., 2018). It is caused by an imbalance between nitrogen intake and excretion (more catabolic than anabolic processes), which could be related to starvation, protein malnutrition or disease (Long et al., 1979; Fuller et al., 2005). An individual with some of these stress conditions loses tissue due to an excessive catabolic activity to maintain 392 protein synthesis in other parts of the body, which could lead to increased $\delta^{15}N$ ratios

 (D'Ortenzio et al., 2015). Henceforth, although an increase in nitrogen levels is generally observed in non-adults between 0 and 1 years of age, and it subsequently drops to the adult average, the observed increase does not allow us to conclude definitively that it is due to the breastfeeding period. Other techniques are recommended to complement the information obtained by analysing stable isotope ratios from bone, such as the application of incremental dentine micro-samples from teeth (Burt, 2015) and the investigation of other stable isotopes ratios e.g. oxygen (Reynard and Tuross, 2015; Britton et al. 2015).

401 On the other hand, there is another outlier (MO-050A) that displays a similar $\delta^{13}C$ value 402 to non-adults and a similar $\delta^{15}N$ value to adults (-17.6‰ and 10.4‰, respectively) (Fig. 403 3). This individual was 4 years \pm 12 months in age, and its δ^{13} C value could be due to the weaning process, assuming the limitations explained above. Its nitrogen and carbon isotope values were intermediate between non-adults and adults. This could be related to, in some cases, the transition to an adult diet, which goes through the introduction of 407 supplementary foods enriched in ${}^{13}C$ (Dupras et al., 2001). This is consonant with the description of weaning practices of the Roman Era realized by Soranus and Galen (Greek and Roman physicians, respectively). They described it as a gradual process based on the introduction of supplementary foods (such as boiled honey or a mixture of honey and goat's milk) from 6 months of age to 3 years of age, when the weaning was completed (Dupras et al., 2001; Fuller et al., 2006; Saragoça et al., 2016).

 Furthermore, a comparison of Mondragones dietary patterns with other Roman and Late Roman sites from the Iberian and Italian Peninsulas was also realized (Table 4). The 415 adult results have been compared with the sites of Joan Planells (Ibiza, Spain) (Alaica et al., 2019), Tossal de les Basses (Valencia, Spain) (Salazar-García et al., 2016), Monte da Cegonha (Alentejo region, in southern Portugal) (Saragoça et al., 2016), and Port of Velia (Velia, Italy) (Craig et al., 2009). For the comparison of non-adult results, the sites of Isola Sacra (on the coast near Rome) (Prowse et al., 2008) and Monte da Cegonha (Saragoça et al., 2016) were selected. Dietary patterns from these populations are based 421 predominantly on C_3 plants, with variable meat or dairy consumption. There are also 422 differences in the consumption of C_4 plants and marine resources. In addition, the population from Port of Velia can be divided into 2 groups: [I] with a diet rich in cereals and relatively poor in meat and marine resources and [II] with much more meat and fish 425 intake, and a consequent $\delta^{15}N$ increase. These populations have similar faunal isotope values to those obtained in this study, which enables a comparison of the human values. 427 Even though there are significant differences between the δ¹⁵N and δ¹³C values in most adult populations (*P* < 0.05), the variation is more pronounced in nitrogen values (Fig. 4), especially in coastal populations. This variation appears to be associated with the availability of marine resources, because the only population that does not show significant differences is Monte da Cegonha, which is also located inland. Related to this, there are significant differences between nitrogen values of non-adults from Mondragones and non-adults from Isola Sacra (*P* = 0.000) (Table 4), who present higher 434 values of $\delta^{15}N$. However, there are no differences between carbon values, so the nitrogen increase is probably due to seafood intake detected in their mothers (Prowse et al., 2008).

437 Table 4. Comparison of stable isotope results in different European sites.

Fig. 4. Plot of δ¹⁵N versus δ¹³C values of the adult human (and faunal) samples from

440 Mondragones compared with the mean values from other published Roman populations.

441 3.4 Pathological conditions and diet

 An attempt to establish a relationship between the different pathologies detected and dietary patterns at the population level was realized. It was possible to infer the influence of diet in some pathologies attending to the detected cases (Table 5). However, a limitation of these analysis is that not all individuals had a complete skeleton, so only individuals with good skeletal representation (mainly cranium and teeth well-preserved)

 were considered. Therefore, these results are only an approximation of the relation between diet and pathological conditions.

 Cariogenic lesions, dental calculus, dental enamel hypoplasia, and cribra orbitalia were analysed. Porotic hyperostosis could not be included in the statistical study because it was observed macroscopically in only 2 individuals and this result was not be confirmed by a further radiographical analysis. Caries was observed in at least 14 individuals (5 non-adults, 5 females, 3 males, and 1 undetermined), as well as dental calculus, which was observed in 12 individuals (3 non-adults, 7 females, 3 males, and 2 undetermined) (Table 5). Significant differences in carbon ratios between individuals with and without 456 caries were observed $(P = 0.018)$. Even though with this result and the dental calculus (*P* > 0.05) it was not possible to assume a relation between diet and this kind of dental lesions, the presence of caries and dental calculus in several individuals was indicative of starchy food and/or carbohydrate consumption (Lieverse, 1999; Featherstone, 2000). Regarding non-specific stress markers, these conditions were found more frequently in non-adults (7 non-adults and 1 undetermined) (Table 5). Significant differences in terms of cribra orbitalia were observed (*Pcarbon* = 0.001; *Pnitrogen* = 0.006). In this case, individuals affected show higher nitrogen and less negative carbon averages than individuals without cribra, which may be related to the fact that most individuals affected by this 465 condition are non-adults (all aged: 1 year \pm 4 months). Cribra orbitalia and porotic hyperostosis have been traditionally related to anaemic conditions (Ortner, 2003), among others also, and the high prevalence of these pathologies in non-adults could be related to the Infant weaning transition described by Roman authors as Galen and Soranus, who suggest the introduction in the diet of infants of a mixture of goat's milk 470 and honey (Fairgrieve and Molto, 2000). Goat's milk has a lower content of folate than 471 human milk (Chanarin, 1990), which has a direct impact on iron absorption and may lead to megaloblastic anaemia (Dupras et al., 2001). Dental enamel hypoplasia was observed in only 5 individuals (2 non-adults, 2 females, and 1 male). However, the relation between diet and presence/absence of dental enamel hypoplasia did not show significant differences (*P* > 0.05). None of these 5 individuals showed signs of cribra orbitalia or porotic hyperostosis, so this condition may be related to other dietary deficiencies or childhood diseases (Roberts and Manchester, 2010). In fact, enamel hypoplasia has also been linked to trauma to the developing tooth, genetic conditions, and specific environmental factors (Towle and Irish, 2020), so all these issues should be considered. 480 Table 5. Association between δ^{13} C and δ^{15} N values and pathologies.

481 4. Conclusions
482 This study prov This study provided the first paleodietary information on a Late Roman population from 483 the south-eastern of the Iberian Peninsula. The results indicated a diet rich in C_3 plants 484 supplemented with meat from terrestrial herbivores, although it is possible that this 485 population had minimum C_4 plant or fish intake.

486 Dietary differences were not observed according to sex, but these differences were 487 observed in age where the youngest non-adults in the breastfeeding period formed a 488 well-defined group and showed significant differences with adults.

489 The palaeopathological study revealed the presence of diet-related diseases and non-490 specific stress markers. The high presence of caries and dental calculus are indicative

491 of a diet rich in starch and carbohydrates, while cribra orbitalia, porotic hyperostosis, and

492 dental enamel hypoplasia seem to be more related to dietary deficiencies (e.g. effect of

493 supplementary food during the weaning period, malnutrition, diarrhea…).

494 Finally, considering the dietary variation and the geographical location of different Roman

495 populations, although it is possible to establish similarities among all of them (for 496 example, C_3 plants are common to all), it seems that the diet depended more on the

497 environment and the local availability of food than on cultural habits.

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