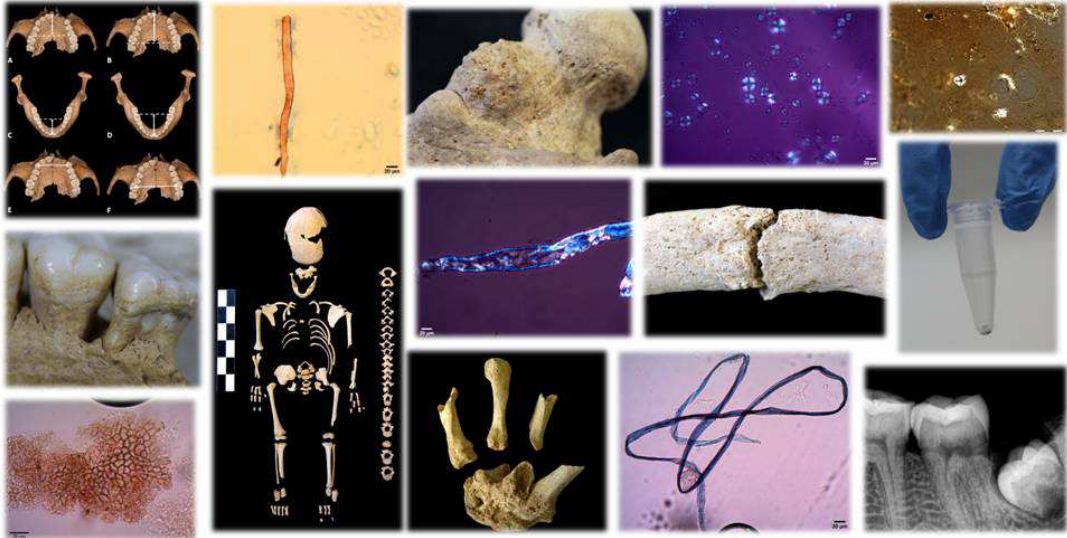


**THE NECROPOLIS OF CAN REINERS  
(7th c. AD, MALLORCA, SPAIN):  
DEMOGRAPHY, HEALTH, AND LIFESTYLE**



**PhD DISSERTATION**

**UAB**

**UNIVERSITAT AUTÒNOMA DE BARCELONA**

**UNITAT D'ANTROPOLOGIA BIOLÒGICA**

**DEPARTAMENT DE BIOLOGIA ANIMAL, DE BIOLOGIA VEGETAL I D'ECOLOGIA**

**DOCTORATE IN BIODIVERSITY, 2015**

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Elena Fiorin

PhD dissertation

2015



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SPAIN): DEMOGRAPHY, HEALTH, AND LIFESTYLE**

Dissertation presented by Elena Fiorin in fulfilment of the requirements for the Doctorate in Biodiversity of Departament de Biologia Animal, de Biologia Vegetal i d'Ecologia, Universitat Autònoma de Barcelona, directed by:

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## **ACKNOWLEDGEMENTS**

I would like to express my deep gratitude to Professor Assumpció Malgosa, my research supervisor, who has supported me throughout my thesis with her patience and helpful advices. One simply could not wish for a better or friendlier supervisor.

I would like to offer my special thanks to Albert Isidro who has offered many advices and insights regarding the palaeopathological analysis.

I am particularly grateful to Professor Rosa Maria Albert as she introduced me to the world of phytoliths and micro-remains analysis.

I am grateful for the assistance given by Joan Cadafalch, Dolors Ceperuelo, Maria Jose Adserias and Eduardo Chimenos regarding the dental occlusion study. They offered me the opportunity to learn new skills and to work in a multidisciplinary context. I would like to express my deep gratitude to Joan for his presence in every phase of the work and because he gave me helpful advices regarding all I wanted to know about teeth and occlusion features.

I would also like to extend my thanks to the laboratory technicians of the department, in particular to Nuria, for her help in offering me the resources in running the program and for helping me to handle the laboratory instruments. I wish to thanks various people for their contribution to this project; Mireia Ribó Amezcua, Meritxell Tur Bello, Maria Herrero and Anna Monzonís for helping me in the lab with the study of part of the skeletal remains. In particular, I'm grateful to Maria and Anna because they were amazing students: they were good workers and friendly, they helped me when I was involved in the final part of the practical work.

My best thanks are for the people who helped me when I was unhappy and stressed, the moments in which a person would have nearby good friends:

- Giacomo Capuzzo has been the best housemate that I ever had. We had very good moments, he is like a brother to me. When I had need of a friend he was always present. Thanks Giacomo!

- I am extremely thankful and indebted to Michael B. Toffolo for sharing your English knowledge with me. He is an extremely sincere and valuable friend. I'm very happy to have a friend like you!

- Pere Ibañez Gimeno was the best colleague and friend that I could be imagine to have. In my daily work I have been lucky to have nearby a friendly and cheerful friend. He provided me precious help regarding statistic analysis, English, Spanish, Catalan, goats jokes, etc. He helped me also providing a home in Cerdanyola when I needed to return to Barcelona for the thesis. Thank you very much!

- I am very grateful to my partner Paolo who supported me in....all! You are present in every moment of my life since eleven years. I could not wish a better person near me.

Finally, I thank my parents for supporting me throughout all my studies at University, and for providing a home in which to complete my writing up. They are the best supporters of my crazy passions and I'm so grateful to have parents like them.

## ABSTRACT

The present study focused on the reconstruction of the diet, lifestyle and health of the individuals that lived around 7th c. AD in the northern coast of the island of Mallorca (Spain). The skeletal remains came from the Necropolis of Can Reiners, located above the ancient forum of the Roman city of *Pollentia*. The excavation, conducted between 1980 and 1988, unearthed more than 200 burials of four different typologies. This thesis analyzed the osteological material of 156 of these burials, that contained a total of 226 individuals. Demographic analysis showed that the sample is composed of 63.9% of adults and 36.1% of non-adults. Regarding adults only 15.8% of the individuals were mature and no senile were present. Among the population, 37% were males, 25.5% females and 37.5% individuals of unknown sex. We observed that the mortality was not balanced between sexes, being the sex ratio (male/female=1.5) favourable to males. The crude mortality rate for Can Reiners individuals is 37.59% and the life expectancy at birth was approximately 26.6 years. Therefore, results of the analysis of our sample showed a high mortality and low life expectancy, which conforms to the expected values for ancient populations such as this one. Therefore, the low frequencies of pathologies such as osteoarthritis and traumas agree with the demographic data. We also analyzed other original bioanthropological aspects of the population. We elaborated a new method based on clinical dental practice suitable for fragmented skeletal remains in order to study the presence of malocclusion in ancient skeletal material. Our findings exhibited that the individuals of Can Reiners showed occlusal characteristics of ancient (such as high dental wear and molar relationship Class III) and modern (crowding, crossbite, among others) populations, although the normocclusion was present in 70% of the sample. Finally interesting data were discovered through the analysis of the microremains observed within the dental calculus. In this context the research followed two steps. Step one focused on

the characterization of several cereal seeds cultivated in the Mediterranean - since they are the plant organ with the highest starch content (about 70%) and one of the major sources of carbohydrates of the human diet- in order to eventually identify them in the microscopic analysis. Step two consisted in the microscopic analysis of different dental calculi sampled from skeletal remains recovered from the necropolis of Can Reiners. Within the sampled dental calculi the presence of different starch grains pointed to the use of various cereals crops in the island. Other botanical remains, such as spores and pollen grains, indicated the environment where they lived. The identification of these remains could be a powerful instrument for the reconstruction of the ancient diet, habits and the physical and social environment of the ancient populations.

## **PREFACE – STRUCTURE OF THE THESIS**

The thesis is structured in six sections.

First, the geographical and historical context of the necropolis of Can Reiners in *Pollentia* will be described. The former studies carried out on this necropolis will be presented schematically in order to better introduce the analysis undertaken by the present research, which, for the first time, has taken into account the entire sample of human bones from Can Reiners. Our research has addressed the study of Can Reiners proposing new methodologies and considering new evidence never applied and explored before.

Then, materials and methods will be discussed and therefore the final results will be presented. The results will be subdivided in three parts: (1) anthropological analysis, comprising palaeodemography, anthropometry, and palaeopathology of the entire population; (2) dental occlusion analysis of the skeletal remains (3) the study of the micro-remains observed in the dental calculus.

Since these three chapters address distinctive topics, each of them has paragraphs presenting an introduction, materials, methods, results, discussion and conclusions. Only in the chapter about the anthropological analysis of Can Reiners, because of the specificity of the addressed topic, results and discussions have been unified into a single paragraph.

The third chapter has been divided in two parts. The first part is an atlas of micro-remains of the Mediterranean cereals that were used since the Roman period. This analysis has been useful in order to describe and identify starch grains recovered in the sample of Can Reiners. The second part of the chapter presents the study of the micro-remains recovered in the dental calculus of some individuals of this necropolis.

Hereafter, the general discussion, conclusion and the bibliography used during the research will be presented.

Finally, in the Appendix, a paper published in the course of this research, has been attached. The article, published in the journal *Collegium Anthropologicum*, focuses on the description of a new methodology which addresses the recording of variables of the dental occlusion.

# TABLE OF CONTENTS

<b>1. Introduction .....</b>	<b>17</b>
1.1. Geographical context.....	19
1.2. Historical context .....	20
1.3. The forum of <i>Pollentia</i> .....	22
1.4. The necropolis of Can Reiners .....	25
1.5. Anthropological studies: the state of the art .....	29
<b>2. Objectives .....</b>	<b>33</b>
<b>3. Material and methods .....</b>	<b>37</b>
3.1. Material .....	39
3.2. Methods.....	40
<b>4. Results.....</b>	<b>43</b>
4.1. The bioanthropological profile of the individuals of Can Reiners .....	45
4.1.1. Introduction .....	45
4.1.2. Material and methods .....	45
4.1.3. Results and discussion.....	54
4.1.4. Conclusions .....	101
4.2. Dental occlusion variation in the population of Can Reiners.....	103
4.2.1. Introduction .....	103

4.2.2. Material and methods.....	104
4.2.3. Results.....	106
4.2.4. Discussion.....	121
4.2.5. Conclusions.....	124
4.3. Microscopic analysis of the dental calculus.....	125
4.3.1. Atlas of microremains of the Mediterranean cereals .....	125
4.3.1.1. Introduction .....	125
4.3.1.2. Material and methods .....	131
4.3.1.3. Results .....	132
4.3.1.4. Discussion.....	146
4.3.1.5 Conclusions .....	148
4.3.2. The significance of microremains recovered from dental calculus for anthropological analysis: the case study of Can Reiners .....	149
4.3.2.1 Introduction .....	149
4.3.2.2 Material and methods .....	150
4.3.2.3 Results .....	156
4.3.2.4 Discussion.....	176
4.3.2.5 Conclusions .....	180
<b>5. General discussion .....</b>	<b>181</b>
5.1 The population of Can Reiners: demography and life conditions on the Mallorca island in the 7th c. AD.....	183



5.2 The study of dental occlusion: a new approach based on dental clinical practice .....186

5.3 Dental calculus analysis: the creation of an atlas and the case study of Can Reiners .....189

**6. Conclusion.....197**

**7. Literature cited .....201**

**Appendix**

Document 1: Study of dental occlusion in ancient human remains: a methodological approach



## **1. INTRODUCTION**



There are many reasons laying behind the study the necropolis of Can Reiners. First of all, the human skeletons unearthed have never been studied entirely. The present study, for the first time, aimed to analyse all the anthropological remains from Can Reiners as a whole. Moreover, this study has explored the application of some methodologies never applied by the analyses previously undertaken, and therefore new, unknown results have been obtained through this innovative approach. Finally, the study of the necropolis of Can Reiners, dated back to the 7th century, allows to address one of the most challenging period of the past (Gnasso et al. 2015) embedded between the 6th century, the ‘last of the Roman centuries’ (Wickham 1998) and the 8th century, “which is indubitably a medieval one” (Macmaster 2015). Ultimately, the analysis of Can Reiners can contribute to the exploration of this significant period of transformations in a crucial area such the Balearic islands, gravitating in the middle of the Mediterranean, a sea which, precisely in this period, lost its political unity after centuries because of the Islamic conquest.

### **1.1 Geographical context**

The necropolis of Can Reiners was located above the forum of the Roman city of *Pollentia*. The archaeological remains of the Roman city are situated in the outskirts of Alcúdia, a municipality and township of the Balearic Islands (fig. 1.1), located in the north east of Mallorca (39°51'11"N; 3°07'16"E) at 19 m (62 ft) above the sea level.

The Roman city lies on a promontory on the isthmus which divides the two bays of the actual Alcúdia, in the south, and Pollença, in the north. These two towns later developed around the roman harbours of *Pollentia*: the *Portus Mayor* and the *Portus minor*. In the western area, *Pollentia* was flanked by a promontory named La Victoria, whereas along the other sides it was surrounded by marshlands and a hill named San Martí. Its position had several advantages

such as good visibility and the complete control of the sea surrounding the settlement (Orfila *et al.* 2008).



Figure 1.1: geographical context of the roman city of *Pollentia* within the western Mediterranean (A) and in Alcúdia bay, Mallorca (B).

## 1.2 Historical context

The *Gymnesian Islands*, the modern Mallorca and Minorca, were conquered by Q. Caecilius Metellus in 123 BC, appointed of the command of the campaign against the pirates by the Roman Senate. The *Pitiusas* (Ibiza and Formentera) were gradually included under the Roman rule after this date (Orfila *et al.* 2006), as the Romans were particularly interested in controlling these Islands because of their strategic position within the Mediterranean Basin (Orfila 2006).

Strabo, in the *Geographica*, and Pliny the Elder in the *Naturalis Historia* recorded several geographical and historical description about the *Gymnesian* and *Pytusae* Islands. Strabo, for example, mentioned the city of Pollentia in different excerpts of the *Geographica*: “...regarding the *Gymnesian*, the larger

*one has two cities, Palma and Pollentia, Pollentia at east and the other city in the west part...both are fertile and have good harbours ...”* (III, 5, 1). Recent studies (Cau and Chavez 2003, Chavez *et al.* 2010, Orfila *et al.* 1999, Orfila 2005) have placed the start of the construction of the city around 70-60 BC, probably after the Sertorian war. Since that time, *Pollentia* was a flourishing community and became the most important Roman city of the archipelago. The city suffered a great fire during the years 270-280 which caused the destruction of the forum and other areas such as *Sa Portella*, *Can Viver*, *Can Basser* and the house of *Polymnia* (Arribas and Tarradel 1987). However, the occupation of the urban area continued during the late Roman period, as clearly demonstrated, for instance, by archaeological evidence related with defensive structures in *Sa Portella* and in the Forum area (5th c. AD). Evidence of occupation of the forum during the 6th and the 7th centuries appears extremely shallow (fragments of pottery and one coin) and no data is so far available for the 8th century. Mallorca was conquered by the Muslims in the 10th century and the city of *Pollentia* was abandoned during the 12th century (Orfila 2000).

From the 16th century, local antiquaries started to be interested in finding the location of the abandoned city. In 1863 Joan Reiners in his book “Destruction of *Pollentia*” advanced a hypothesis about the ancient location of the city combining old archaeological finds and written sources. The recovery of the inscription (RE)SP POLL(entina) near Alcúdia suggested the hypothesis that the city of *Pollentia* was located in that area (Orfila 2000). The first excavations were carried out between 1923 and 1934 by Gabriel Labrés and Rafel Isasi under the supervision of the Superior Committee of Excavations and Antiquity of Madrid. These interventions did not produce any documentation and therefore the stratigraphical record of the forum was badly damaged, determining the loss of significant information (Arribas and Tarradell 1987). A second campaign started in 1957 with the economic support of the William L. Bryant Foundation, Vermont (USA) which funding will last in 1997. Since then, the supervision of the excavations was committed to Antoni Arribas,

Miquel Tarradell and Daniel Woods. From the '80 excavations were conducted into the rural property of Can Reiners, within of Camp de França. This is the area where the forum of the Roman city was found.

After 1996, the supervision of excavations was assigned to Antoni Arribas and Margarita Orfila with the collaboration of the municipality of Alcúdia.

Excavations unearthed several residential areas such as the district of La Portella, the house of *Polymnia*, and the district of Can Basser, some defensive structures such as the Finca de Can Ventayol and the fortification wall on the forum and the theatre. Several necropolises were discovered at Can Fanals, La Solana, Can Corrón and Can Reiners.

### 1.3 The forum of *Pollentia*

Many archaeological campaigns were carried out in the forum area of the Roman city of *Pollentia*, in which that was the area was the centre of the Roman public life. The most ancient level of the forum was represented by a leveling of the ground. During the Roman Republic (1th century BC) the *Capitolium*, some *Tabernae* and a *Porticūs* were built (fig. 1.2).

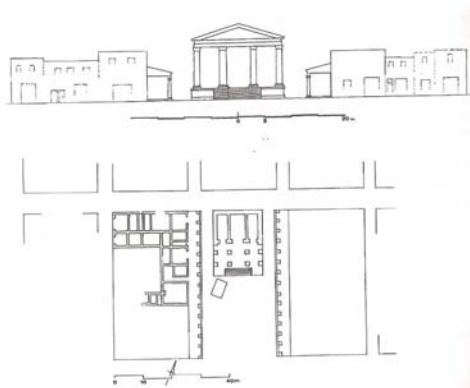


Figure 1.2: the forum during the Roman Republic (Mar and Roca 1998, p. 114)



Between the 1st and the 2nd centuries AD, the temple I (Templet I in figure 1.3) and the temple II (Templet II in figure 1.3) were built. The temples damaged the east side of the *Capitolium* and the east part of the square (fig. 1.3). Temple I was related to the imperial worship and interpreted as *Aedes Augusti* (Zucca 1998). Several elements, such as statues and inscriptions, adorned this area underlining the prestige of this period. To this phase, moreover, belongs the construction of the theatre, built outside the city.

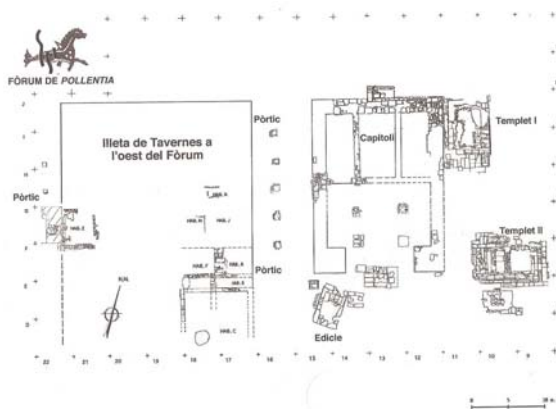


Figure 1.3: the forum between 1th and 2th centuries AD (Orfila 2000, p. 143)

The *tabernae* and the *porticūs* suffered several changes between the 2nd and the 3rd century AD (fig. 1.4). The compartments of the *tabernae*, for example, changed their size and become squared and irregular. As already mentioned, archaeological data suggest that in the 3rd century several fires occurred in the city and one of those destroyed the *tabernae* of the forum. Nevertheless, the fires did not determine the abandonment of the forum, which continued to be settled after these episodes. Remains of *opus signinum* paving the forum area were found above the layers of burning.

Between 365 and 369, the province of the Balearic Islands was created. In the *Notitia Dignitatum* (Dig. Oc. III, 5-139), it is reported that they were one of the

seven provinces of the *Hispania* dioceses under the rule of *praefectus praetorio Galliarum*.

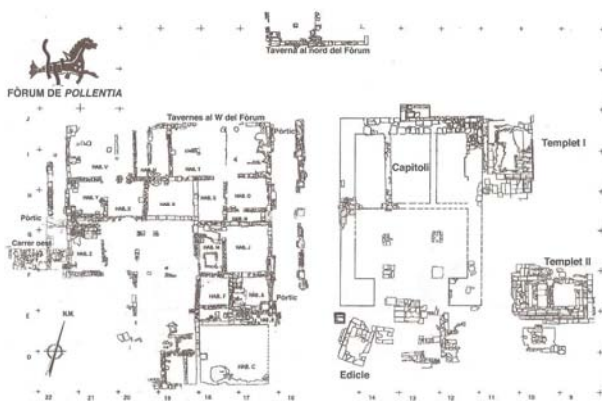


Figure 1.4: the forum in 3th century AD (Orfila 2000, p. 147)

In this historical phase, a fortify wall was built north of *tabernae* (fig. 1.5). The construction of a fortified wall strengthen the hypothesis of a reduction of the size of the city. The fortification was placed in the higher part of the city and the archaeological evidence arranged the chronology of this structure in the 5th century. Therefore, the urban area continued to be inhabited even after the Vandal occupation in 455 AD and after the Byzantine conquest in 534 AD.

From this phase, however, the area changed its function, as a huge necropolis occupied the fortification built in the public square. This is the necropolis of Can Reiners. One of the graves has cut off a part of a fortification tower, when it was in disuse. For this reason, it is supposed that the use of this area as a necropolis started around 600 century AD but, due the absence of grave goods, it is not clear how long the necropolis has been used (Orfila 2000).

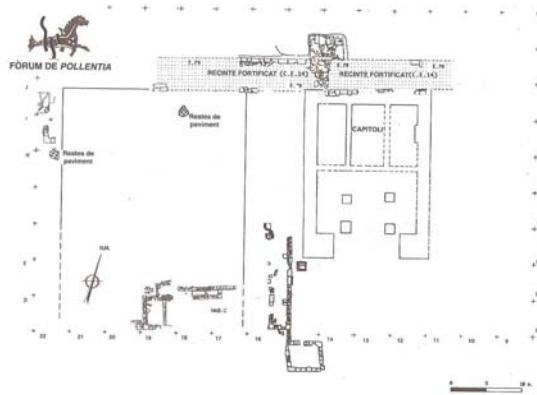


Figure 1.5: the forum in 4th century AD (Orfila 2000, p. 153)

From this phase the urban area suffered a radical transformation: the public square was turned into a cemeterial area. This process can be framed within the “disurbanisation” of many cities (Brogiolo, 1984) in the Western Europe which, between the 5th and the 7th century, were characterized by dramatic change in urban quarter destination, processes of abandonment and proliferation of areas devoted to arable land or pasture. Among these different processes, the installation of burials into the urban area appears as one of the most significant evidence of this transition.

#### 1.4 The necropolis of Can Reiners

The necropolis of Can Reiners was located in the central area of the forum and has been excavated since the 1980s. From 1980 to 2008, more than 250 graves were unearthed. Due to their characteristics, such as the different typologies of graves and the absence of grave goods, the exact chronology of the cemetery use is not clear so far. Whereas, through the stratigraphical evidence, it has been established that the appearance of burials in the forum started around 600 AD. The graves are located in the southern and eastern areas of the *Capitolium*. It is intriguing to note that neither the *Capitolium* nor the ruins of the *Tabernae* were

overlapped by the burials. In the 1980s, an archaeological team composed by Arribas, Tarradell and Wood excavated around 200 graves (fig. 1.6).

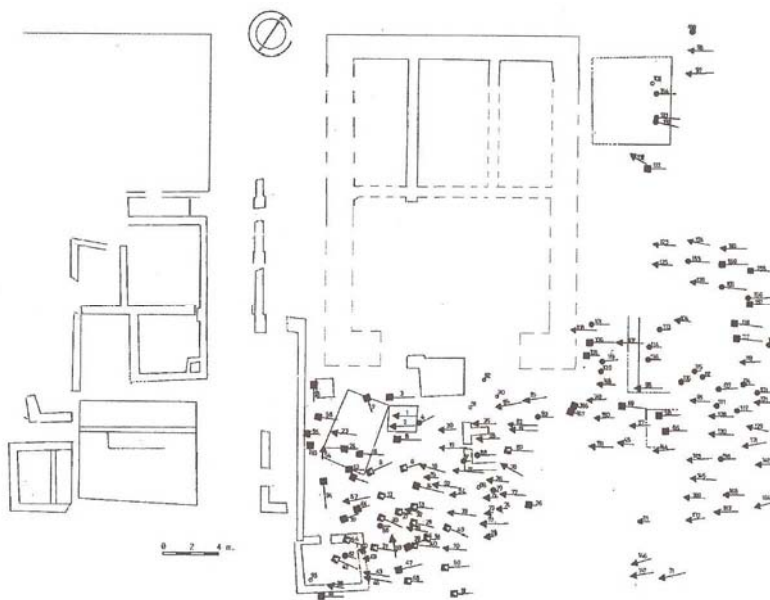


Figure 1.6: the necropolis of Can Reiners (Arribas and Tarradell 1987, p.134)

Thanks to their excavation, it was possible to understand the transformation of the forum during the centuries. For example, it was observed that several bases of honorary monuments were widely damaged by graves. Base n. 2 was carved to receive burials 1 (fig. 1.7) and 2, meanwhile the north side of base n. 3 was modified in order to contain grave n.7. The latter is a particular inhumation because its orientation is North-South whereas, the general orientation of individuals is West-East. Eventually, grave 23 recycles some stones of the same bases (n.3).



Figure 1.7: grave 1

The graves excavated during the to excavation carried out between 1980 and 1988 are the subject of the present study. Generally, individuals were laid down in a supine position, with inferior limbs extended and upper limbs crossed above abdominal region (fig. 1.8). Skulls were oriented westward and the feet were oriented to east. In some cases superior limbs had a “pray” position that is when limbs over-flexed with the hands arranged near the cranium (fig. 1.9). In other cases, one superior limb was extended whereas the other one was flexed above the chest. Probably individuals were laid down directly in the ground with a shroud.



Figure 1.8: grave 13



Figure 1.9: grave 66

No elements of clothing such as buttons, pins and nails which testify the existence of a wood coffin were observed. By contrast, these elements are present into the burials of the necropolis of Can Fanals (2th– 3th centuries AD).

In the necropolis of Can Reiners several types of graves, including ossuaries, were excavated. Usually, burials contained a single skeleton although there are also graves containing more than one individual, such as the “cist” type. Burial typologies can be divided in:

- 1- Simple pit dug into the ground or within the bases of monuments or other structures. Sizes range from 1.70 to 2.00 meters. Simple pits represent 24.5% of the sample until the excavation carried out in 1984.
- 2- Pit covered by stone slab, usually recycled (37.7%).
- 3- Cist: stone lined grave covered by stone slabs (ca. 19.8% of the total). In two cases, a vertical stele was found above the grave.
- 4- A fourth typology has been defined “fosa con banqueta o túmulo”, i.e. a mound tomb or tomb with banquet, squared-shaped, filled with sediments. Within these tombs, no residual of food and offers were recovered, however. Arribas and Tarradell (1987) suggested that this typology should be the more recent one, as these tombs overlap the older burials and show similar characteristics with the palaeochristian inhumations.

Generally, no grave goods or other material were found in the burials, except for three of them in which three coins were recovered. In grave 77, in the south-east area of the necropolis, a bronze coin of *Probus* (276-282) was found. The coin was located near one of the hands of the individual. In the simple pit 38 and in grave 36 (mound tomb or tomb with banquet) two little bronze coins of Costantine were recovered, but they were unreadable. These findings, despite is

not clear if the coins were inserted into the grave or into the backfill, represents a *terminus post quem* for dating the first burials to around 300 AD.

### **1.5 Anthropological studies: the state of the art**

Individuals that belong to the Can Reiners necropolis excavated in the 1980s, have been analyzed by several researches. In 2003, Garcia and Subirá studied trace elements that allowed to reconstruct the diet of the ancient population. They extracted samples of compact tissue from 88 individuals of Can Reiners population. Among them, 35 were males and 29 females. The trace elements such as zinc, copper, magnesium, calcium and barium were analyzed along with the animal and human bones recovered in the same necropolis. Due to a possible contamination, strontium was excluded from the study. Results showed a diet basically composed by vegetables and the marine food contribution was poor in spite of their proximity to the coast. The female diet was a little bit different respect to the male, with a major supply of vegetables, cereals and dried fruit.

A second study undertaken by Ortega *et al.* (2003) studied the oral pathology. The analysis were carried out on 111 individuals, describing oral pathology according to Chimenos *et al.* (1999) criteria. The results were compared to other two populations of Mallorca: the pre-Talaiotic individuals of S'Aigua Dolça (Artá) and the Talaiotic individuals of S'Illot de Porros (Santa Margarita). Results showed an increasing of dental decay, *ante mortem* tooth loss, fistulas and enamel hypoplasia for the Can Reiners individuals. Nevertheless the results of Can Reiners oral pathologies are similar to those of other necropolis of the same historical period.

García, Subirá and Richards, in 2004, analyzed the stable isotopes of carbon, nitrogen and sulphur extracted from bone collagen of 39 individuals. Results showed a diet based on animal proteins, obtained from herbivorous such as

cows, goats and sheep and its milk, and on vegetal proteins obtained from plants such as wheat, rice, green bean and legumes. The percentages of marine proteins was significant only in 10 individuals whereas the others individuals have lower values. Once again, the values of female diet were rich in cereals and legumes respect to those of the males. In 2005, Subirá, García, and Berrocal analyzed the *cribra orbitalia* and the presence of calcium, barium, strontium, zinc, magnesium and copper in 83 individuals. The results showed that 64.5% of the individuals had *cribra orbitalia*, with no differences among sex. This pathology was present in subadults (57.14%) and in adults (66.67%).

In 2009, García Sívoli analyzed dental traits of this population. Results showed that there was a genetic continuity between the Catalan samples and the Majorcan samples. The author speculated that the individuals of Majorca descended from Catalan population and Iberian peninsula.

Finally, in 2010, Diaz de Villabona studied the mitochondrial DNA of three necropolis from Mallorca (Son Real, S'Illot de Porros and Can Reiners). Results showed that Can Reiners' samples have a high frequency of haplogroup H; the mitochondrial genetic diversity places it among other series of ancient and modern European context. In relation to the three series analyzed, Can Reiners exhibits the smaller genetic distance compared to other Catalan samples. It is also noticeable that a sequence of African lineage was observed (Cr 70.1).

Regarding the anthropological analysis of the graves excavated after 1988, some information has been recovered. In 2009, in the XVI Congress of the Spanish Society of Physical Anthropology (SEAF), a paleopathological study of the individual n. 1045 excavated in 1995 in *Pollentia* was presented. The individual is an adult male that present a traumatism in the cranial vault. The same individual exhibits several pathologies in the oral cavity, in the vertebral column and in the rest of the postcranial skeleton. Finally in 2009, Cardona



López presented a master thesis, directed by Cau and Orfila, focused on the anthropological study of the individuals excavated between 2004 and 2008.



## **2. OBJECTIVES**



The present study aims to characterize the bioanthropological structure of the population of Can Reiners in order to reconstruct demography, diet and lifestyle of the individuals that lived in the northern part of Mallorca around the 7th century AD. The following tasks have been established:

1. to reconstruct the bioanthropological dynamics of this population;
2. to study dental occlusion in order to establish if malocclusion represented an issue for the oral health of the individuals and if its pattern in this population provides information on the causes of malocclusion;
3. to investigate micro remains preserved into the dental calculus in order to obtain direct information about diet and habits of the individuals buried in this necropolis.



### **3. MATERIAL AND METHODS**





### 3.1 Material

The skeletal remains used for this work belong to the necropolis of Can Reiners (c. 7th century AD), located above the forum of the Roman city of *Pollentia* (Mallorca, Spain), the most important Roman city in the Balearic Islands. Although the exact chronology of the use of the cemetery is not clear yet, it dates back to around 600 AD, when the forum was abandoned (Orfila 2000). The excavations of the necropolis began in the 1980s and they continue nowadays.

The present study is based on the material recovered by the first archaeological campaigns carried out by professors Tarradell and Arribas from 1980 to 1988 (Arribas and Tarradell 1987). This anthropological sample consists in 226 individuals buried in 156 graves. The numbers of the graves range from 1 to 208. Some grave did not contain human skeletal remains but fragments of pottery or animal bones (Cr. 90.1, Cr. 91.1, Cr. 184.1, Cr. 192.1) whereas other tombs were found without skeletal remains (Cr.18, Cr. 21, Cr. 22, Cr. 28, Cr. 40, Cr. 44, Cr. 45, Cr. 48, Cr. 50, Cr. 51, Cr. 59, Cr. 60, Cr. 61, Cr.79, Cr. 80, Cr. 92, Cr. 110, Cr. 113, Cr. 114, Cr. 115, Cr. 134, Cr. 135, Cr. 136, Cr. 137, Cr. 138, Cr. 139, Cr. 140, Cr. 161, Cr. 162, Cr. 165, Cr. 167, Cr. 171, Cr. 173, Cr. 174, Cr. 175, Cr. 176, Cr. 177, Cr. 178, Cr. 179, Cr. 180, Cr. 181, Cr. 186, Cr. 188, Cr. 194, Cr. 195, Cr. 196, Cr. 197, Cr. 205). This justifies that the present work was partial but representative of the population buried in the necropolis during this period of transition between the Late Antiquity and the Middle Ages on Mallorca.

No field report is available from the excavations of these burials, but the records of some graves and photos done from 1980 to 1988 has been provided by the archaeologists (fig. 3.1). In these records were reported archaeological data and only occasionally anthropological information. For this reason, the description of skeletal material came uniquely from the anthropological laboratory study and it will be presented in the fourth chapter.

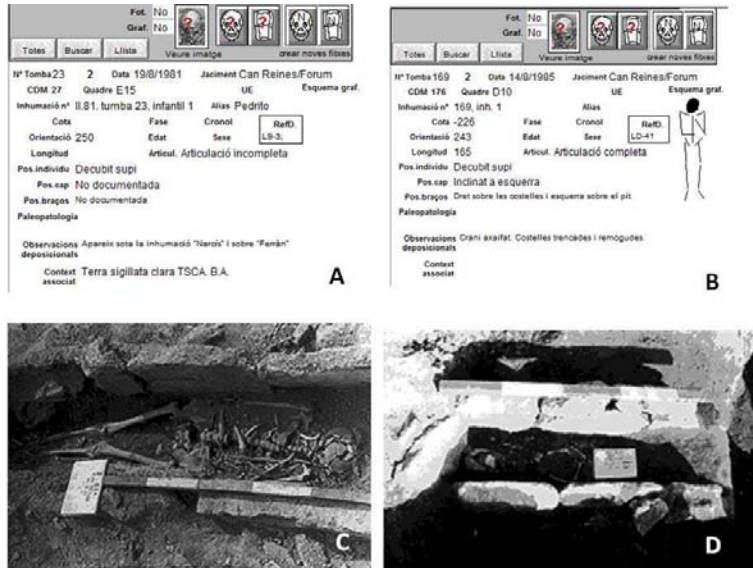


Figure 3.1.1: record of the burial 23.1 (A), record of the burial 169.1 (B), photo of the skeletal remains of the grave Cr.14 (C), photo of the skeletal remains of the grave Cr.53 (D)

### 3.2 Methods

The workflow followed in the study of the material of this thesis is presented in fig. 3.1.2. In the present study several topics have been debated and different methodologies have been employed, namely techniques for age and sex diagnosis, estimation of the stature, anthropological measurements and indices, paleodemography, paleopathology, analysis of dental occlusion, analysis of dental calculus microremains, and statistics. For sake of clarity it was necessary to describe the specific material and the methods employed for every analysis in the corresponding section of the chapter four.

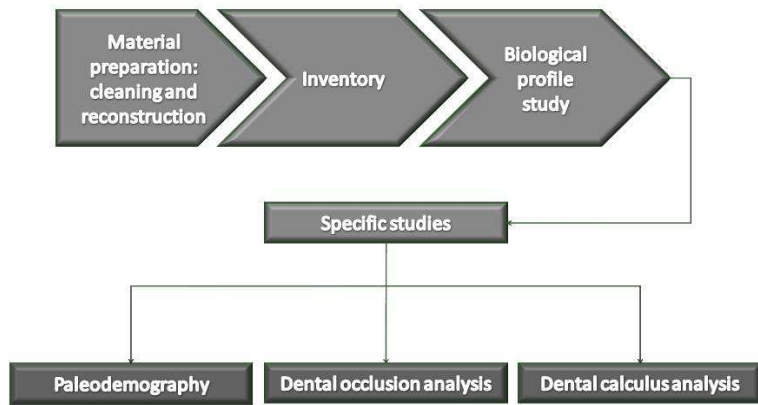


Figure 3.1.2: workflow followed in this thesis for anthropological study



## **4. RESULTS**



## **4.1 The bioanthropological profile of the individuals of Can Reiners**

### **4.1.1 Introduction**

Human remains have always represented an important source of information about many aspects of the past. In addition to the information regarding burial ceremonies, skeletal remains provide data relative to demography, diseases, dietary habits, daily activity, and behaviour.

Palaeodemographic studies offer, for example, an opportunity to link together variables such as sex and age at death which allow to reconstruct biological profiles and population size. This information is used to construct mortality curves, survivorship curves and life tables for any specific population represented. Palaeodemography data when compared with palaeopathological information provides a more complete interpretation of dynamics of the population in relation to the health status of the group of individuals. Therefore, the recovered information such as anthropometric measurements and preservation index acquire more value in the anthropological and social interpretation when compared with sex and age at death. For instance, questions such as diet, health, social position, or funerary care could be evaluated.

The aim of this section is to analyze the several anthropological variables recovered from the skeletal remains of the necropolis of Can Reiners in order to obtain a profile of the biological features, life style and diseases of the individuals of the sample.

### **4.1.2 Material and methods**

The analyzed sample came from 156 graves excavated in the Necropolis of Can Reiners (*Pollentia*, Mallorca, Spain) during the field campaigns conducted by

Tarradell and Arribas from 1980 to 1988. These burials contained 216 individuals whose skeletal remains were housed for study in the laboratory of Physical Anthropology of the Biological Anthropology Unit (BAVE) of the Universitat Autònoma de Barcelona.

The methods used for the anthropological study are described in the following sections:

### **Skeletal preservation**

The preservation of the skeletal material in this and other archaeological site is usually varied: some of the skeletons are fragmented, others incomplete, and some of them had the cortical bone damaged. These features are mostly due to the taphonomic processes that happens to the body between death and its recovering (Duday 2006). Causes of the damage are different: type of terrain, presence of roots and stones, water seepage, among others. Also human intervention, such as reopening of graves, could be responsible of damage and loss of information. To assess the level of preservation of bones, a preservation index (PI) determined by Walker *et al.* (1988) and modified by Armentano *et al.* (2012) was used. The PI was calculated according to the following equation:

$$PI_n = \Sigma (\text{elements preserved}) / \Sigma (\text{elements considered}) \times 100$$

where  $PI_n$  is the total percentage of every group of bones, the elements preserved are the number of the observed bones and the elements considered are the number of the expected bones. Three indices were calculated:

- $PI_1$  (N=12) where limbs elements (humerii, radii, ulnae, femurs, tibiae and fibulae) were considered
- $PI_2$  (N=19) where limbs ( $PI_1$ ) and the girdle bones (clavicles, scapulae, coxal bones and the sacrum) were considered.



- IP<sub>3</sub> (N=22) where PI<sub>2</sub> (limb and girdle bones) and three parts of the skull (neurocranium, splanchnocranium, and mandible) were considered.

These indices provided an insight about the presence of bony elements of every skeleton; this is important to understand the representation of skeleton or skeletons in one burial in order to evaluate the available information for several diagnoses or to assess intrusion cases. However, they do not denote integrity or kind of preservation of bones.

### **Sex determination**

For the determination of sex several methodologies were applied since not all features have the same discriminating value. Sometimes skeletal remains are fragmented and bones that are fundamental for the determination of sex are missing. In these cases the sex was determined only when the main criteria could be adopted. Individuals are classified as male or female. When the skeletons did not have the necessary elements useful for the diagnosis of sex they were classified as undetermined. Sex was determined from juvenile individuals onward; individuals from 0 to 12 years old, due to the incomplete sex maturation of the skeleton, have not yet developed necessary features to carry out a proper diagnosis of sex.

The classical parameters used for this diagnosis are described in Acsádi and Nemeskéri (1970), Buikstra and Ubelaker (1994), Ferembach *et al.* (1979), and Krogman and Iscan (1986). They include basically morphological analysis, but metric analyses are also useful. Also, due to the fragmentation of skeletal remains other methodologies and revised methods were used (Aleman *et al.* 1997, Brooks and Suchey 1990, Brusek 2002, Haun 2000, Olivier 1960, Safont *et al.* 2000).

## **Age determination**

In ancient skeletal remains estimation of age at death is not easy. Bones are frequently fragmented and those parts that are most useful could be missing or not clearly analysable. The manuals used for this work that include principal methodologies applied for estimation of age in ancient skeletal remains were Buikstra and Ubelaker (1994), Ferembach *et al.* (1979), Krogman and Iscan (1986) and Oliver (1960). Age at death categories have been defined according to Vallois (1960) and they are divided in six different groups: infant I (0-6 years old), infant II (7-12 years old), juvenile (13-20 years old), adult (21-40 years old), mature (41-59 years old), senile (more than 60 years old). Sometimes we divided the first group in two categories: perinatal (0-1 years old) and infant I (1-6 years old). The estimation of age in non-adults (until ca. 19 years old) was based on teeth eruption, growth phase of root and calcification of dental crown (Cretot 1978, Ubelaker 1978), diaphyseal length of bones (Alduc-Le Bagousse 1988, Fazeka and Kosa 1978, Kosa 1989, Scheuer and Black 2000, Stloukal and Hanakova 1978, Ubelaker 1989) and appearance and union of epiphysis and of centres of ossification (Fazeka and Kosa 1978, Scheuer and Black 2000). The different methods were generally calibrated on modern population due to the population variability. However, it is not possible to verify if modern bones growth phases are the same of those of the past.

There are several methods to estimate age at death in individuals that have reached skeletal maturity, but we have to take into account that each method is affected by individual variability, population and sex. Therefore, it is more advisable to use several criteria to estimate age at death. The methodologies employed in this study were the following:

- 1- The metamorphosis of pubic symphysis surface (Brooks and Suchey 1990, Gilbert and McKern 1973, McKern and Stewart 1957, Todd 1920 and 1921);
- 2- Changing of morphology on the auricular surface of ilium (Lovejoy *et al.* 1985, Buckberry and Chamberlain 2002);

- 3- Changing in sternal ends of the rib (Iscan *et al.* 1984, 1985, 1986, 1987);
- 4- Progressive closure and obliteration of sutures of the skull. (Meindl and Lovejoy 1985);
- 5- Dental wear (Brothwell 1981, Lovejoy 1985). It is based on the gradual attrition of occlusal surfaces of teeth, resulting from chewing, during the individual life.

### **Paleodemographic analysis**

Palaeodemography is a branch of the Physical Anthropology which deals with the reconstruction of the demography of ancient population, using the human skeletons recovered from archaeological contexts. Different obstacles constrict this reconstruction. On one hand, chronology of a necropolis is usually uncertain, and it is most unlikely to have information on social stratification, family size and population composition. On the other hand, there are some methodological difficulties to assure age and sex attribution. The estimation of age, indeed, depends on the nature of the material and is more open to error in adult age groups than in non-adult groups. By contrast, sex determination is more accurate in adult groups while for children is frequently impossible to determine, since they have not developed a proper sexual skeletal dimorphism (Brothwell 1971). Another important element that must be considered is that usually infant skeletal remains, particularly still-born and newborn infants, may be buried away from the usual cemetery area of adults. Therefore, and also because infant skeletal remains are more vulnerable to the taphonomic process, the palaeodemography work may be not completely correct.

Despite these restrictions, palaeodemography represents a rich field of investigation and offers an opportunity for skeletal material to reveal remarkable biological information of the population (Ubelaker 2008).

Usually, the demographical analysis employed to estimate mortality rates of ancient populations consist of calculation of life tables based on a model of stationary population (Acsádi and Nemskeéri 1970). The theoretical assumption

of a stable population is a methodological resource that allows standardizing demographic events of an ancient population, without affecting its parameters with additional mathematical methods. The assumption of a stable population means that the number of births and deaths of a population are similar and they stayed stable during the time. Therefore grow rate will be equal to zero ( $r=0$ ) (Márquez Morfín and Hernández 2001).

Starting from this assumption, curves of mortality, curves of survivorship and life tables were calculated using the methodologies proposed by Acsádi and Némkéri (1970), Márquez Morfín and Hernández (2001) and Weiss (1973). After the evaluation of our mortality data we compared our sample with other population which represent different mortality pattern.

### **Anthropometry and epigenetic traits**

The anthropometry is the discipline that deals with measurements of human body. The recovery of osteometric measurements allows us to reconstruct some characteristics of skull and body proportions of an individual or a population. Moreover, employing regression equations and skeletal indices (Martin and Saller 1957), it is possible to estimate stature, to diagnose sex and to evaluate robustness of individuals. In this study the morphometric data was recorded using the classical osteometrical variables described in Martin and Saller (1957) and Olivier (1963). The estimation of stature of Can Reiners population was calculated using the regression equations of Pearson (1899), Trotter and Gleser for Whites (1952, 1977), and tables of Manouvrier (1893) and Mendonça (2000).

The term “epigenetic” defined epigenetic variants as an expression of genes affecting development. They are also called: skeletal variants, non-metrical, discontinuous or discreta data. Some authors suggest that some traits (in particular the post-cranial traits) may could not have a genetic background but they could be modified by environment (Trinkaus 1978). The epigenetic identification method used in this study is based on morphological variation of

minor skeletal traits observed in skull and in post-cranial skeleton (Anderson 1968, Finnegan 1978, Hauser and De Stefano 1989). The epigenetic traits selected to be analyzed in the present work are:

For the cranium: frontal view: 1- *metopic suture*, 2- *metopic fissure*, 3- *supranasal suture*, 4- *medial supraorbital nutrient foramina*, 5- *lateral supraorbital foramen*, 6- *medial supraorbital foramen*, 7- *medial supraorbital notch*, 8- *supratrochlear notch*, 9- *supratrochlear foramen*, 10- *frontal groove*, 11- *trochlear spur*, 12- *nasal foramina*, 13- *ethmoidal foramina*, 14- *infraorbital suture*, 15- *infraorbital foramen*, 16- *zygomaxillary tubercle*, 17- *mental foramen*; lateral view: 18- *bipartite parietal bone*, 19- *inferior parietal foramen*, 20- *squamous ossicle*, 21- *superior squamous foramen*, 22- *inferior squamous foramen*, 23- *bipartite temporal squama*, 24- *sutura mendosa*, 25- *ossicle at asterion*, 26- *notch parietal bone*, 27- *occipitomastoid ossicle*, 28- *mastoid foramen*, 29- *squamomastoid suture*, 30- *suprameatal spine and depression*, 31- *auditory torus*, 32- *marginal tubercle*, 33- *zygomatofacial foramen*, 34- *bipartite zygomatic bone*; posterior view: 35- *ossicle at lambda*, 36- *incaic bone*, 37- *occipital foramen*, 38- *lambdoid ossicles*, 39- *parietal foramina*; basal view: 40- *palatine torus*, 41- *palatine bridging*, 42- *maxillary torus*, 43- *double condylar facet*, 44- *condylar foramen*, 45- *paracondylar process*, 46- *pharyngeal foveola*, 47- *pharyngeal tubercle*, 48- *transverse palatine suture*, 49- *premaxillary suture*, superior view: 50- *coronal ossicle*, 51- *sagittal ossicle*, 52- *symmetrical thinning of parietal bones*, 53- *bregma ossicle*.

For the postcranial: femur: 1- *Allen's fossa*, 2- *Poirier's facet*, 3- *plaque*, 4- *third trochanter*, 5- *hypotrochanteric fossa*, 6- *exostosis in trochanteric fossa*; humerus: 7- *supracondyloid process*, 8- *septal aperture*; scapula: 9- *acromial articular facet*, 10- *suprascapular foramen*, 11- *acromial bone*; tibia: 12- *lateral tibial squatting facet*, 13- *lateral widen tibial squatting facet*, 14- *medial tibial squatting facet*; calcaneus: 15a- *anterior calcaneal facet absent*; 15b- *anterior calcaneal double facet*, 16- *peroneal tubercle*; talus: 17a- *inferior talar articular surface absent*, 17b- *inferior talar articular double surface*, 18-

*medial talar facet and lateral talar extension, 19- os trigonum; atlas: 20- atlas facet form, 21- atlas posterior bridge; patella: 22- emarginated patella, 23- vastus notch, lumbar vertebra (L5): 24- spondylolysis.*

### **Palaeopathological analysis**

Palaeopathology is the study of ancient diseases that have left marks on ancient skeletal remains or mummified bodies. Palaeopathologists take advantage of directly study the remains of the diseases. By contrast, they have the disadvantage that their study is restricted largely to those diseases that affect the skeleton and some complementary analyses are not possible (anamnesis, soft tissues, etc). Usually skeletal diseases are uncommon, as most diseases affect the soft tissues.

Palaeopathological study starts with macroscopic observation and description of the lesion or anomaly. If the visual inspection it is not satisfactory, it can be supplemented by radiographies, micro-CT scanning, histological and DNA analysis. Furthermore, is important to decide if the lesion is monofocal or multifocal, this is if just involve one bone or also other parts of the skeleton, and if it is symmetrical or not. After the description of the lesion, it is fundamental know whether it is a pathognomonic sign of a specific disease or what diseases can present it. The prevalence of this/these disease/s at different sex, times or places is also essential. In this sense, it is useful make comparisons with modern day-data and with data published in bone reports from earlier periods. Finally, all the recovered information are assembled to proceed with the differential diagnostic.

The principal literature used in the present work was: Aufderhide and Rodriguez Martin (1998), Barnes (2012), Campillo (2001), Fornaciari and Giuffra (2009), Isidro and Malgosa (2003), Ortner (2003) and Waldron (2009).

## **Statistical analyses**

All data obtained from the analyses accomplished on skeletal materials of Can Reiners were recorded in a database. Statistical analyses were performed with SPSS 15.0 for Windows (SPSS Inc. 2006). First, it was tested the normality of variables distribution that helps us to identify the most appropriate statistical techniques. To assess normality of distributions, Kolmogorov-Smirnov test were applied with the significance correction of Lilliefors; when the sample was assembled by less than 50 variables, Shapiro-Wilk test was used. Depending on results, parametric or non-parametric tests were applied.

Other specific statistical tests were applied. Wilcoxon and Mann-Whitney test were used to assess differences between paired and unpaired variables, respectively. Friedman tests were used to assess differences among groups of paired variables. In order to determine between what pair of variables differences existed, we applied Wilcoxon test with Bonferroni correction. Kruskal-Wallis tests were used to assess differences among groups of unpaired variables. In order to determine between what pair of variables differences existed, we applied Mann-Whitney test with Bonferroni correction.

Regression analyses were employed to determine the relationship between groups of quantitative variables. Chi-square tests or Fisher's exact tests were used to assess the relationship between pairs of nominal variables and between nominal and ordinal variables.

Principal component analyses, separately for males and females, were used to assess the similitude among Can Reiners and other archaeological populations concerning several postcranial measurements.

### 4.1.3 Results and discussion

#### Skeletal preservation and funerary practices

In order to investigate relationship between skeletal preservation and variables such as age at death, sex and grave types, statistical analysis were applied. Table 4.1.1 shows descriptive statistics of preservation index of the entire sample. Skeletal remains have a global index (IP3) with a mean of 72.7% and a median of 77.2%.

Table 4.1.1: descriptive statistics of preservation index (PI) of Can Reiners skeletal remains

	<b>IP1</b>	<b>IP2</b>	<b>IP3</b>
<b>Mean</b>	77.10	73.42	72.69
<b>Median</b>	91.60	84.20	77.20
<b>Standard deviation</b>	27.88	26.77	25.61
<b>Minimum</b>	.00	.00	.00
<b>Maximum</b>	100.00	100.00	100.00

The variables had not a normal distribution, and therefore we used a non-parametric test. To evaluate if there were differences between variables IP1, IP2 and IP3, Friedman's test was used. Results showed differences ( $P < 0.05$ ) among the variables. To know exactly between which variables differences occur, Wilcoxon's test with Bonferroni correction was used (IP1- IP2:  $P < 0.016$ ; IP2- IP3:  $P < 0.016$ ; IP1- IP3:  $P < 0.016$ ). We observed significant differences between IP1-IP2 and IP1-IP3 whereas between IP2-IP3 no differences were found. Observing the IP1 median we noticed that it is higher compared to others. Therefore, we could conclude that superior and inferior limbs are better preserved compared to other skeletal remains (IP2 and IP3).

Thereafter, we evaluated if the index of preservation change according to the grave types (1. Simple pit; 2. Pit covered by stone slab; 3. Stone lined grave covered by stone slabs; 4. Mound tomb or tomb with banquet). Using a Kruskal-Wallis test, we observed statistically significant differences in the three preservation indices among the four grave types (IP1:  $P < 0.05$ ; IP2:  $P < 0.05$ , and



IP3:  $P < 0.05$ ). To know exactly which differences the preservation indices have among the grave types, Mann-Whitney test with Bonferroni correction was used. Between grave 1 and 2 type there were differences for all the preservation indices (IP1:  $P < 0.00833$ ; IP2:  $P < 0.00833$ ; IP3:  $P < 0.00833$ ) and between grave 2 and 4 type there were differences for the IP3 ( $P < 0.00833$ ). Therefore, all the skeletal remains are better preserved in the simple pit burials (grave 1) compared to those contained in pits covered by stone labs (grave 2) (table 4.1.2, IP1 mean: grave 1= 81.96, grave 2= 62.15; IP2 mean: grave 1= 77.55, grave 2= 56.73; IP3: grave 1= 76.40, grave 2= 58.61). In the grave type 4 (mound tomb or grave with banquet) crania are better preserved than those contained in the simple pits covered by stone labs (grave 2) (table 4.1.2, IP3 mean: grave 2= 58.61, grave 4= 85.64). This could be related with the preservative action of the soil in filled burials in contrast with body decomposition in the open space of covered burials.

Table 4.1.2: descriptive statistics of preservation index (PI) among the grave types of Can Reiners

	Grave type	N	Mean	Median	SD
<b>IP1</b>	<b>1</b>	74	81.96	100.00	27.35
	<b>2</b>	26	62.15	58.30	31.47
	<b>3</b>	43	76.33	83.30	26.73
	<b>4</b>	10	80.82	87.50	21.54
<b>IP2</b>	<b>1</b>	74	77.55	89.40	26.77
	<b>2</b>	26	56.63	60.45	28.54
	<b>3</b>	43	73.41	84.10	24.75
	<b>4</b>	10	81.70	86.85	19.13
<b>IP3</b>	<b>1</b>	74	76.40	86.30	25.89
	<b>2</b>	26	58.61	65.85	26.46
	<b>3</b>	43	71.90	77.20	24.50
	<b>4</b>	10	85.64	90.75	16.40

We also observed the relationship between preservation index and sex, using a Mann-Whitney test. Results showed differences between preservation index of males and females (IP1:  $P < 0.05$ ; IP2:  $P < 0.05$ ; IP3:  $P < 0.05$ ). In the following box plots (fig. 4.1.1) we can appreciate that the preservation indices are higher in males. Therefore, male's skeletal remains are better preserved than those of

females and this is probably due to the greater dimension and strength of male bones.

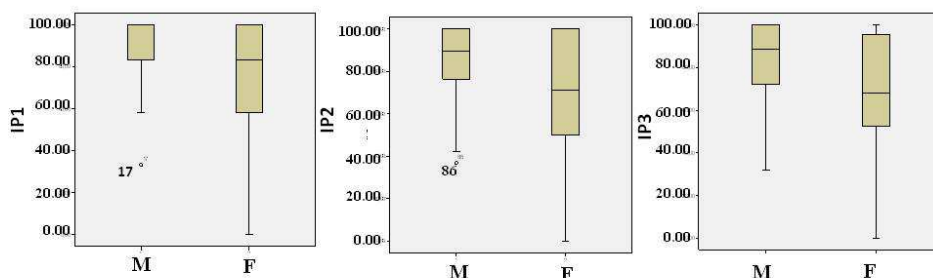


Figure 4.1.1: box plots of males and females preservation indices of Can Reiners

The regression analysis was applied to estimate the relationship between index of preservation and age at death of the individuals, where IP3 is the dependent variable and age is the independent variable. The results showed that P-values of the linear equation, square and cubic are  $<0.05$ . This means that there is a relationship between the variables, whereas it is important to be aware that the quadratic R has a small value ( $R^2=0.094-0.06$ ). Therefore, only around 10% of the IP3 variability depends on age being that non-adult individuals were less preserved than adults.

Finally, we analyzed the relationship between grave types and age at death and between the grave types and sex. Results exhibited a  $P>0.05$  (Kruskal-Wallis) in both tests, therefore, we could conclude that there are no relationship between the variables. This means that any type of burial is not exclusive to a certain groups of age and the type. Instead, it refers to other questions (familiar ties, chronology, fashions, etc)

Regarding the non-adult burials exclusively, the state of the preservation of skeletal remains is good (figure 4.1.2). The mean of the global index of preservation (IP3) is 69.76% and the median is 81.60%, more high than the mean (table 4.1.3).

Table 4.1.3: descriptive statistics of preservation index (PI) of Can Reiners non-adult skeletal remains

	<b>IP1</b>	<b>IP2</b>	<b>IP3</b>
<b>Mean</b>	68,63	68,28	69,76
<b>Median</b>	83,30	78,90	81,60
<b>Standard deviation</b>	31,28	28,62	26,90
<b>Minimum</b>	0,00	0,00	4,50
<b>Maximum</b>	100	100	100

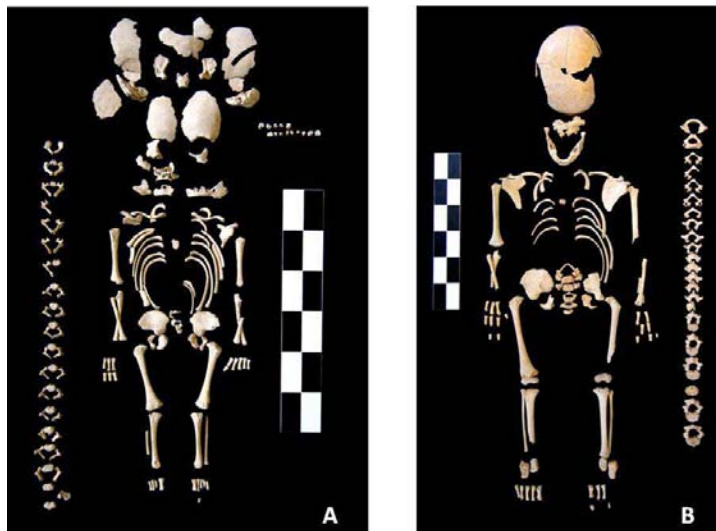


Figure 4.1.2: state of preservation of two non-adults from the necropolis of Can Reiners. An infant of 2-3 months (Cr. 53.1) (A), and a infant I of 2-3 years of age (Cr. 23.4) (B)

To evaluate if there are differences between variables IP1, IP2 and IP3, Friedman test was used. Results showed differences ( $P < 0.05$ ) among the variables. To know exactly between which variables differences occur, Wilcoxon test with Bonferroni correction was used (IP1- IP2:  $P > 0.016$ ; IP2- IP3:  $P < 0.016$ ; IP1- IP3:  $P > 0.016$ ). We observed significant differences between IP2 and IP3 being the IP3 median value higher than IP2 (table 4.1.4). Therefore, this indicates that crania are better preserved than the girdle bones (clavicles, scapulae, coxal bones and sacrum). Maybe it is the result of a differential recollection of non-adult remains because of minus knowledge that

archaeologist have of immature postcranial remains in contrast with skull bones.

Regarding the age class distribution among the grave types no statistically differences were observed (Kruskal-Wallis test  $P > 0,05$ ). This result corroborates the interpretation of adult burial types.

Table 4.1.4: non-adult distribution among the grave types

Age Class	N	%	Grave type				Total
			1	2	3	4	
<b>Infant I</b>	42	55,3%	10	12	14	4	40
<b>Infant II</b>	16	21,1%	8	1	6	1	16
<b>Juvenile</b>	18	23,7%	6	2	6	2	16
<b>Total</b>	76	100%	24	15	26	7	72

Concerning the funerary practices, we do not have the necessary elements to describe the position of most of the individuals, and to determined if the depositions were primary or secondary. However the few available photographs seem to indicate that burials were mostly primary. In any case, Arribas and Tarradell (1987) described the general types of the burials and positions of the individuals. They reported that individuals were generally laid down in a supine position, with inferior limbs extended and upper limbs crossed above abdominal region. Skulls were west oriented and the feet were oriented to east. In some cases superior limbs had a “pray” position or one superior limb was extended whereas the other was flexed above the chest. Probably individuals were laid down with a shroud directly in the ground. We observed that 72.7% of the burials were single and in the remaining 27.3% 2 or more individuals were buried. Table 4.1.5 shows the number of individuals contained within the burials among the four grave types.

We observed that within grave type 1 (simple pit) the majority of the burials contained 1 individual (86.1%) however, in some cases, until 6 individuals were buried. The grave type 1 is the only type where more than 4 individuals were buried. Within grave type 2 (pit covered by slab stones) 83% of the burials

contained 1 individual. In the grave type 3 (stone lined grave covered by stone slabs), the burials that contain 1 individual represent 59.4%. By contrast in grave type 4 (mound tomb or tomb with banquet) the 59.1% of the burials contained more than 2 individuals (just in one case we observed that there were 3 individuals). Regarding age at death and sex, we previously observed that no relationship between these variables and the types of graves were found.

Table 4.1.5: distribution of the individuals among the grave types

Grave type	N individuals within the grave						Total
	1	2	3	4	5	6	
1	56	3	2	2	1	1	65
2	20	1	2	1	0	0	24
3	19	6	4	3	0	0	32
4	9	12	1	0	0	0	22
<b>Total</b>	104	22	9	6	1	1	143

## Demographic structure

In total skeletal remains of 216 individuals were analyzed. Table 4.1.6 shows age and sex composition of the population from Can Reiners. The sample comprised 138 adults (63.9%) and 78 non-adults (36.1%). Regarding adult groups, excluding the individuals whose age at death was not determined (18.1%), the rest of population (84.2%) did not reach 40 years of age. Only 15.8% of the individuals were mature (41-59 years old) and no senile (more than 60 years old) were present.

Table 4.1.6: age and sex distribution of individuals of Can Reiners

Age at death categories	N	%	Male	Female	Unknown sex
<i>Infant I</i> (0-6 years old)	44	20.4%	-	-	44
<i>Infant II</i> (7-12 years old)	16	7.4%	-	-	16
<i>Juvenile</i> (13-20 years old)	18	8.3%	7	3	8
<i>Adult</i> (21-40 years old)	71	32.9%	46	22	3
<i>Mature</i> (41-59 years old)	28	13.0%	13	14	1
<i>Senile</i> (more than 60 years old)	0	0.0%	0	0	0
<i>Adult x</i> (undetermined age)	39	18.1%	14	16	9
<b>Total</b>	216	100.0%	80	55	81

Of the non-adult category, sex was determined only for the individuals that have reached skeletal maturity (ca.15 years old or older). Taking in account this distribution, the sample included 80 males (37%), 55 females (25.5%) and 81 individuals of unknown sex (37.5%). Therefore, the mortality is not balanced between sexes (80:55), being the sex ratio (male/female=1.5) favourable to males. Between 13 and 40 years, indeed, the probability of death of males is double compared to females. Meanwhile for mature individuals the ratio is balanced (13:14). These values are probably an artefact due to the reduced number of females in the sample either because there was a different place of burials for the females sample either because the community of Can Reiners had, for example, a military function. The area of the forum, indeed, was previously occupied by fortification structures.

An abridged life table of the individuals of Can Reiners, combined for both sexes, was created (table 4.1.7). Life tables are probabilistic models that describe the extinction by death of a *cohort* by ages. They also permit us to estimate conventional mortality parameters such as life expectancy at birth. Following current methodological practice, the life table of population from Can Reiners was built using 5 years age intervals, except for the first interval that represents the first 5 years of life that was divided in two interludes (0-1 and 2-4 years). For the adults individuals, who were not included in a particular group of age at death, we used a prorate technique to distribute this individuals in a determinate age class category (Márquez Morfín and Hernández 2001).

Observing the life table (table 4.1.7), the frequency of the non-adult mortality is 36.1% and from birth to first year is ca. 7%. Whereas, the expected values for the first year of life should have been between 30% and 50% (Angel 1969, Brothwell 1986-87). Therefore, there was a sub-representation of the immature individuals similar to that of the Roman population of Carthage (2th c. AD, N=87; 31.82%; Fauvelle 1890), and higher compared to the necropolis of Sta. Maria de Hito (6th- 12th c. AD, N=426, 19.25%; Galera 1989).

Table 4.1.7: life table reconstructed from Can Reiners age distribution

<b>x</b>	<b>d<sub>(x)</sub></b>	<b>D<sub>(x)</sub></b>	<b>l<sub>(x)</sub></b>	<b>d<sub>(x)'</sub></b>	<b>l<sub>(x)'</sub></b>	<b>q<sub>(x)</sub></b>	<b>L<sub>(x)</sub></b>	<b>T<sub>(x)</sub></b>	<b>e<sub>(x)</sub></b>
<b>0-1</b>	15	6.94%	216	70	1000	0.07	1930	26641.5	26.6
<b>2-4</b>	18	8.33%	201	83	930	0.089	3554	24711.5	26.6
<b>5-9</b>	16	7.40%	183	74	847	0.087	4050	21157.5	25
<b>10-14</b>	15	6.94%	167	69	773	0.089	3692.5	17107.5	22
<b>15-19</b>	15	6.94%	152	69	704	0.098	3347.5	13415	19
<b>20-24</b>	20	9.25%	137	93	635	0.146	2942.5	10067.5	15.8
<b>25-29</b>	19	8.79%	117	88	542	0.162	2490	7125	13
<b>30-34</b>	19	8.79%	98	88	454	0.194	2050	4635	10
<b>35-39</b>	32	14.81%	79	148	366	0.404	1460	2585	7
<b>40-44</b>	27	12.5%	47	125	218	0.573	777.5	1125	5
<b>45-49</b>	15	6.94%	20	70	93	0.753	290	347.5	3.7
<b>50-54</b>	5	2.31%	5	23	23	1.000	57.5	57.5	2.5

Legend: **x**= it is the age interval; **d<sub>(x)</sub>**= it is the number of deaths during interval **x**; **D<sub>(x)</sub>**= it is the percentages of the deaths during interval **x**; **l<sub>(x)</sub>**= it is the percentage of survivor during interval **x**; **d<sub>(x)'</sub>** and **l<sub>(x)'</sub>**= they are arithmetic resources used to adjust deaths; **q<sub>(x)</sub>**= it is the probability of death during interval **x**; **L<sub>(x)</sub>**= it is the number of years living during interval **x**; **T<sub>(x)</sub>**= it is the number of years lived in the interval **x** and all later years; **e<sub>(x)</sub>**= it is the life expectancy during interval **x**

Life expectancy at birth was calculated approximately to 26.6 years. This value is similar to the value obtained in the sample of Cathedral of Egara of 4th - 8th c. AD with a life expectancy at birth of 25.6 years (Jordana *et al.* 2010). Whereas, life expectancy of Can Reiners sample is higher respect the also contemporaneous population of Ruelles de Serris in France of the 7th - 10th c. AD, with a life expectancy at birth of 22.4 (Alesan *et al.* 1999). Life expectancy at birth of the Hungarian mediaeval series ( $e_{(x)} = 28.7$ ; Acsadi and Nemeskeri 1970) and skeletal remains of S'Illot des Porros of the 4th – 2th centuries BC ( $e_{(x)} = 28.3$ , Alesan *et al.* 1999) are higher respect those of Can Reiners population. The estimated values for the modern non-industrialized society show values that range between 20 and 30 years of age (Milner *et al.* 1989). Until the 17th century, the causes of the death were mainly various plagues, war or famine. These factors prevented the growth of the population and for this reason the life expectancy ranged between 20 and 40 years of age. In Europe this phase protracts until the 19th century (Schofield *et al.* 1991). This data confirmed that life condition of the antiquity population were hard.

The probability of death  $q(x)$ , during subadult phase of the individuals of Can Reiners was quite similar among the different ages. 64% of the population survives to the adult stage, and this group has a life expectancy of 15.8 years. Between 35 and 44 years, the probability of mortality range from 40% to 57%. From 50 years the number of survivors drastically decreases.

The crude mortality rate of a population is the average number of individuals that died per thousand per year. Assuming that the rate of deaths is constant, the crude mortality rate for the Can Reiners individuals is 37.59% ( $M=1000/ e_{(x)}$ ). This means that about 38 persons died out of each 1000 in the population each year. The crude rates of mortality calculated in Minorca Island between 17th and 18th century, for instance, were very high (45.81%). From the 20th century these values start to decrease until reach the current values of 5.5% (Muñoz-Tuduri and García-Moro 2007).

The mortality curve (fig. 4.1.3) shows a peak of mortality between two and four years of life, a decline through childhood to adolescence (until 19 years of age), and another peak of mortality between 20 and 24 years. The third peak, between 35 and 40 years, represents the maximum adult death frequency followed by a dramatic decline until the 54 years of age, where the curve is interrupted. Comparing our mortality curve to the curve of a roman population from Carthage (2nd c. AD, Tunisia) (Fauvelle 1890) we observed several differences (fig. 4.1.3). The first difference consists in the first peak of mortality of the Carthage sample that is placed between 5 and 9 years of age. The second difference is that the highest peak of mortality for individuals of Carthage is placed between 20 and 24 years of age whereas in Can Reiners population is between 35 and 40 years. The last difference is that the curve of mortality of the roman population has a steady decline after 24 years of age. The remaining differences between the two curves are largely products of greater longevity in population of Carthage (the individuals arrive until 102 years of age).



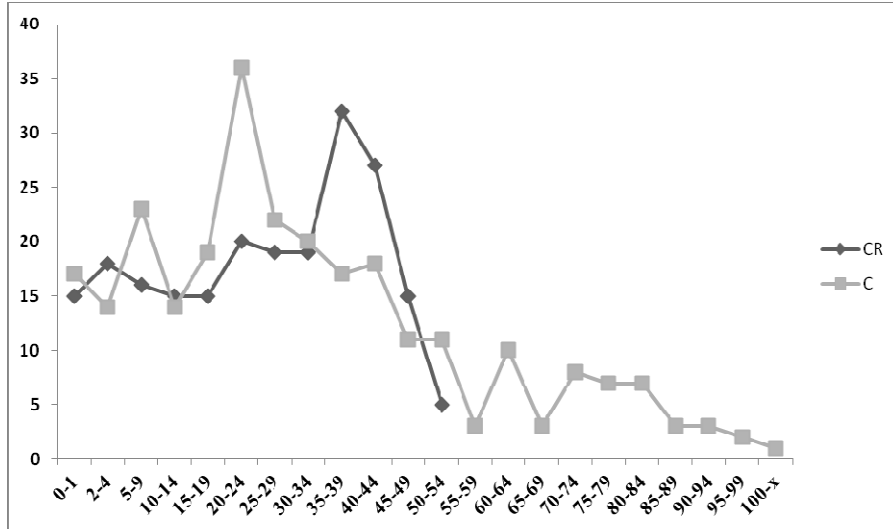


Figure 4.1.3: mortality curves reconstructed for the population of Can Reiners (CR) 7th century AD (Mallorca, Spain) and for the roman population of Cartage (C) 2nd century AD (Tunisia)

The sample of the population of Carthage is interesting because in each grave there was an epitaph in which the sex and the age of the individuals were written. Therefore, it has the same demographic information that a documented age and sex collection. Usually the peaks of mortality in the first years are related with secondary effects of the weaning. If our sample was sufficient representative of the population, it would mean that infants of Can Reiners were early weaned, but the probability of death at adult age was later than Carthaginian people.

Differences of mortality between Can Reiners and other ancient populations are clearly revealed through the survivorship curves (fig. 4.1.4). A curve of survivorship is the reverse of the mortality curve and plots the original population surviving after each five-year interval. To compare life tables of different populations we have adjusted the  $l_{(x)}=216$  (number of individuals of Can Reiners) establishing an original population of 1000 individuals ( $l_{(x)}=1000$ ). Therefore the three samples start with an original population of 1000. Considering this new data, in the sample of Can Reiners we observed that after the fourth year of life 83 non-adult individuals have died and 847 individuals

(84.7% of the sample) have survived at start of the fifth year. After the ninth years of life another 74 individuals have died whereas, 773 individuals have survived at the start of the tenth years. After the thirty-ninth year of life have died 782 individuals (78.2% of the sample) and only 218 individuals (21.8%) have survived at the start of the fortieth. Over 50, survivorship is insignificant, and there are no individuals over 55 years of age.

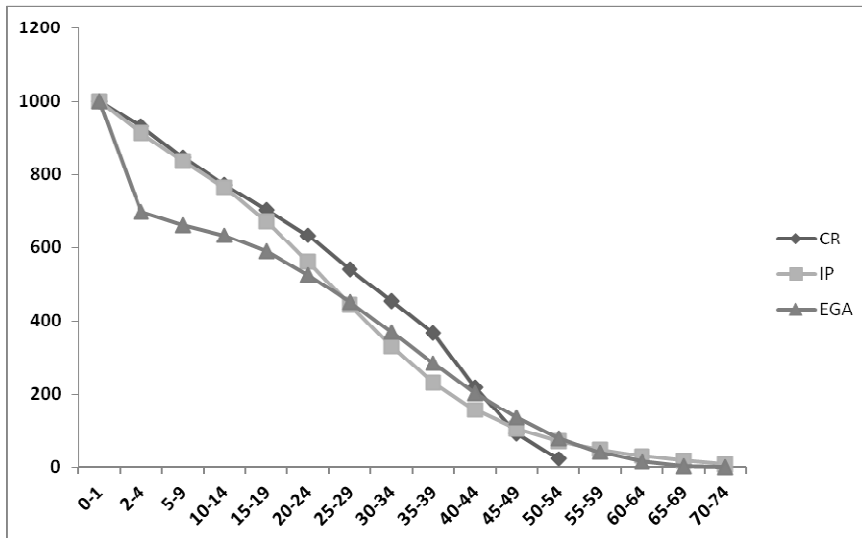


Figure 4.1.4: survivorship curves reconstructed for the population of Can Reiners (CR) 7th century AD (Mallorca, Spain), S'Illot des Porros (IP) 4th-2nd centuries BC (Mallorca, Spain) and Cathedral of Egara (EGA) 6th-8th centuries AD (Terrassa, Spain)

In order to better contextualise the mortality of Can Reiners population, we selected two ancient populations: one from the same geographic location and another from the same chronological frame. The sample of S'Illot the Porros (Talayotic period, Alesan *et al.* 1999) came from a place nearby the Necropolis of Can Reiners, in the same bay of Alcudia. The skeletal remains of the Cathedral of Egara came from Terrassa (Catalonia, Jordana *et al.* 2010) and they have a similar chronology to our sample. Comparing the curves, we observed that until 15 years of age the curves of Can Reiners and S'Illot des

Porros are very similar, whereas Egara curve shows a fewer number of infant of deaths. For adults, the curve of Can Reiners is interrupted after 54 years of age. For S'Illot de Porros and Egara the survivorship curve after this age is also insignificant, due to the few individuals represented in this category.

Results of the analysis of Can Reiners sample showed a high mortality and low life expectancy, which conforms to the expected values for ancient populations such as this one (Acsádi and Nemeskéri 1970, Weiss 1973). Our findings were similar to those of the series of S'Illot de Porros and this is an interesting result because they are two populations that lived in the same place (in the same bay of Alcúdia in the north of Mallorca) in different chronological periods. Therefore, it seems that there were no differences in life conditions between the 4th c. BC and the 7th c. AD in the Island of Mallorca. The deficit of old age individuals, indeed, could reflect a population with low probability of reaching advanced ages and not only a methodological problem related to the difficulty in determining the age at death in this group.

By contrast, sex-ratio was different among the two populations. In S'Illot de Porros both sexes were equally represented, whereas in Can Reiners' sample the sex ratio was favorable to males, possibly due to a different use of the cemetery.

### **Anthropometry**

In this section we analysed the metric variations of the individuals of the necropolis of Can Reiners. Results, such as the estimation of stature and postcranial measurements, were compared to values of other populations of Mallorca, Spain, and other geographical areas.

For the cranial measurements we observed 44 cranial variables and 8 cranial indices. The descriptive statistics of the variables analyzed in this study are described in tables 4.1.8 and 4.1.9.

Table 4.1.8: descriptive statistics of cranial measurements of the individuals of Can Reiners

Variable	Males			Females		
	N	Mean	SD	N	Mean	SD
Maximum length	16	188.43	5.24	11	181.27	2.53
Basal length	7	106.14	6.36	4	93.75	4.85
Maximum breadth	20	140.85	4.86	12	136.58	4.96
Minimum frontal breadth	15	97.86	4.80	10	91.80	4.49
Maximum frontal breadth	14	102.92	7.53	8	106.87	13.05
Biasteric breadth	21	111.57	5.79	10	107.50	3.37
Basion-bregma height	9	139.22	8.18	4	131.00	9.05
Porion-asterion length r.	15	46.50	4.37	12	41.16	3.45
Porion-asterion length l.	15	45.80	5.31	11	41.45	2.94
Auricular height r.	17	129.23	5.14	9	117.44	21.06
Auricular height l.	20	128.65	5.03	9	117.22	19.83
Head circumference	11	497.90	123.54	5	517.60	26.5
Foramen magnum length	9	35.77	2.48	5	33.00	4.35
Foramen magnum breadth	9	29.77	2.68	5	27.60	3.04
Transversal arc	13	268.76	83.23	10	291.20	63.2
Sagittal transversal arc	12	359.83	70.8	9	372.22	20.32
Frontal sagittal arc	16	124.56	6.51	8	125.50	9.62
Parietal sagittal arc	29	126.89	9.12	11	124.63	9.97
Frontal sagittal cord	17	115.82	6.68	8	110.25	6.47
Parietal sagittal cord	29	117.51	6.41	11	103.45	31.33
Occipital sagittal arc	25	97.72	15.87	10	96.00	5.92
Transversal cord	16	130.25	54.28	10	114.20	7.22
Mastoid height r.	23	30.30	7.51	13	26.53	5.31
Mastoid height l.	19	28.15	5.15	10	29.40	5.46
Facial breadth	2	96.00	9.89	0	-	-
Facial height	1	132.00	-	1	118.00	-
Facial total height	2	115.50	3.53	0	-	-
Facial superior height	3	71.00	5.29	0	-	-
Orbital breadth right	5	36.80	3.34	3	38.00	2.00
Orbital breadth l.	4	39.25	1.5	1	38.00	-
Orbital height r.	5	32.40	0.89	1	36.00	-
Orbital height l.	4	32.75	1.25	1	31.00	-
Interorbital breadth	5	26.66	1.15	2	24.00	4.24
Nasal height	3	55.66	2.88	1	47.00	-
Nasal breadth	8	25.00	1.51	2	22.50	2.12
Bicondylar breadth	8	123.50	4.53	5	117.40	4.72
Bigonial breadth	18	103.61	7.36	5	99.80	3.56
Mandibular ramus height r.	10	60.60	7.47	7	56.14	8.27
Mandibular ramus height l.	12	61.66	7.79	6	53.66	7.47
Mandibular ramus breadth r.	22	33.18	2.63	9	31.66	3.04
Mandibular ramus breadth l.	26	33.61	2.4	9	32.55	2.96
Mandibular length	18	81.61	11.62	9	83.66	15.19
Mandibular angle	17	109.94	38.59	9	114.22	30.72
Mentonian symphysis height	21	31.19	2.63	7	28.42	3.50

Legend: N= number of individuals, SD= standard deviation, r.= right, l.= left

Table 4.1.9: descriptive statistics of cranial indices of the individuals of Can Reiners

Indices	Males					Females				
	N	Min	Max	Mean	SD	N	Min	Max	Mean	SD
<b>Cephalic</b>	15	71.19	81.14	74.37	2.86	11	69.23	78.45	75.63	2.61
<b>Cranial b-h</b>	8	83.56	106.47	97.63	7.11	6	71.42	100.70	92.23	11.1
<b>Cranial l-h</b>	7	70.00	80.00	74.62	3.71	5	68.75	78.57	73.82	4.21
<b>Fronto-t</b>	12	76.42	98.87	92.45	6.58	7	80.17	107.79	90.19	10.28
<b>Fronto-p</b>	12	64.13	75.18	69.19	3.76	8	64.58	71.96	68.74	2.41
<b>Orbital</b>	4	84.71	103.22	90.98	8.35	2	79.48	90.00	84.74	7.43
<b>Nasal</b>	2	40.67	40.74	40.70	0.04	2	48.14	51.06	49.60	2.06
<b>Mandibular</b>	15	66.34	100.97	78.24	11.65	5	63.63	108.00	80.74	18.14

Legend: N= number of individuals, SD= standard deviation, b-h= breadth-height, l-h= length-height, Fronto-t= transversal, Fronto-p= parietal

Regarding the cranial measurements, the variables do not follow a normal distribution (Kolmogorov-Smirnov) showing statistical differences between sexes (Mann-Whitney's test,  $P < 0.05$ ) in: maximum length and breadth, basal length, minimum frontal breadth, porion-asterion length, auricular height, parietal sagittal cord ,transversal cord, bicondylar breadth of the mandible. As expected, results showed that some measurements exhibited sexual dimorphism being the values higher in males than in females (table 4.1.8). Determination of sex is important in forensic and anthropology fields and some measurements, such as the bicondylar breadth, are good indicators for sexual dimorphism (Vinay *et al.* 2013).

Due to the state of preservation of the craniums of Can Reiners, we could calculate only 8 cranial indices. The frequencies of the indices among males and females are represented in figure 4.1.5. Regarding the cephalic index we observed that brachycranial morphology is more prevalent in females while in males more of 50% of the crania were dolichocranic. Generally, crania of both sexes have a low skull and flattened in front with a cranial breadth-height index of less than 92 (tapeinocranic). In the cranial breadth-height index we observed that the female skulls are 50% chamaecranic (low skull) and 50% hypsicranic (high skull); the males exhibit more than 60% of hypsicranic skulls. The frontal index exhibits average values in both sexes. In the transversal frontal-parietal index a particular morphology does not prevail. Regarding the mandibular

index, the mandibles of males are wider (80% of wide mandible) than those of females (50% of wide mandible). However, these cranial indices do not present statistical differences (Mann-Whitney test  $P > 0.05$ ) between sexes

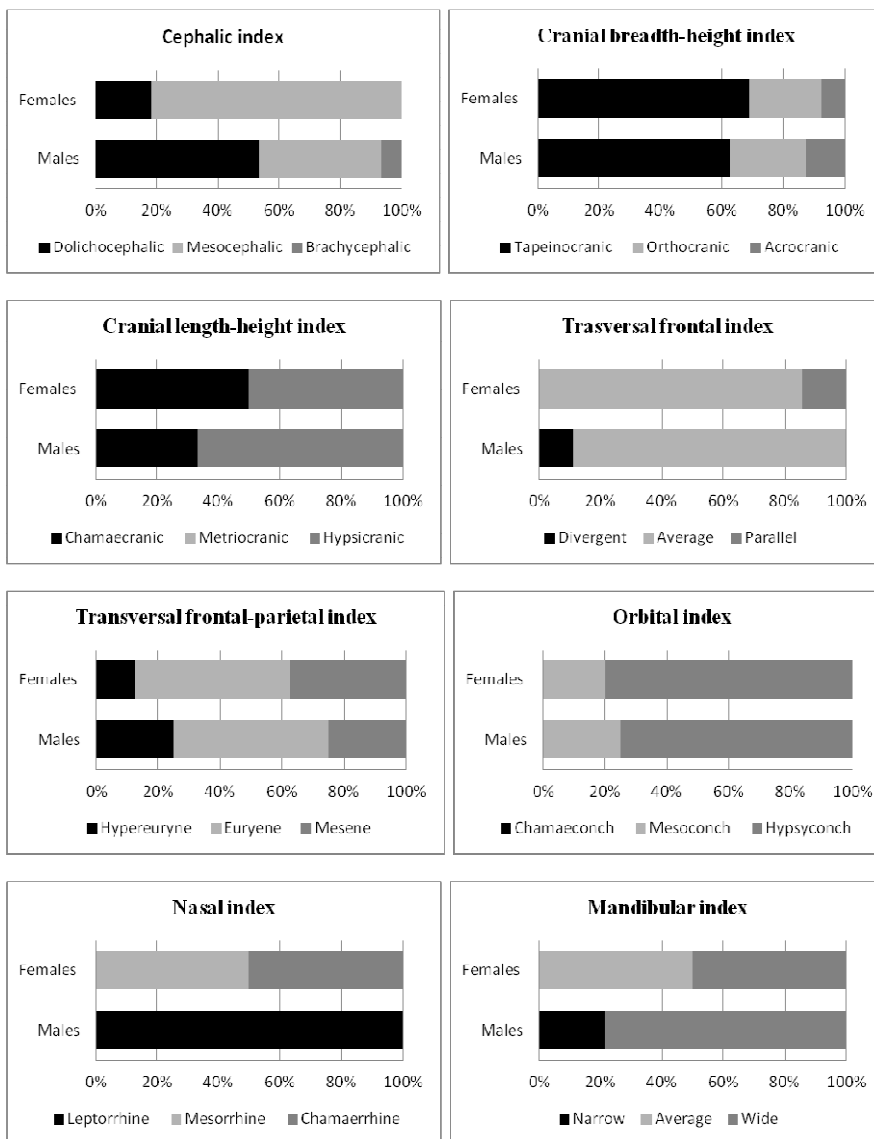


Figure 4.1.5: relative frequencies of the cranial indices among the adult individuals of the necropolis of Can Reiners

The postcranial metrical variation of the adult individuals of Can Reiners was analyzed observing 119 measurements and 9 indices representing the morphometric variation of a population and the skeletal proportion of the bones. The descriptive statistics, divided for males and females were described in the tables 4.1.10 and 4.1.11.

The postcranial variables do not follow a normal distribution (Kolmogorov-Smirnov). We decided to exclude sternum, coxal bones, sacrum and scapula measurements due to the few number of elements that could be measured.

All the measurements, except the left minimum diameter at mid-shaft of the humerus and the left maximum length of the ulna present statistical significant differences (Mann-Whitney's test,  $P < 0.05$ ) between sexes; The results showed that generally measurements of bones exhibited a statistically significant sexual dimorphism and that values of males were higher than those of females.

Statistical differences between sides were tested for all the variables (Wilcoxon's test), in order to determine whether both sides could be pooled together in the analyses. Results showed statistical significant differences ( $P < 0.05$ ) between sides in the maximum length of the femur, the width of the talus, the height of the patella, the median height of the calcaneus, the minimum and the midshaft circumferences of the ulna, the minimum and the midshaft circumferences the sagittal and the transversal diameters at midshaft of the radius, and for the almost all measurements of the humerus: the maximum length, the minimum and the midshaft circumferences, the maximum diameter at the midsahft and the vertical diameter of the head. Therefore, laterality is notable in the arms.

Table 4.1.10: descriptive statistics of the postcranial measurements of males and females individuals of the necropolis of Can Reiners

Variable	Males			Females		
	N	Mean	SD	N	Mean	SD
<b>Clavicle</b>						
Maximum length, right	22	148.63	13.32	12	135.41	9.91
Maximum length, left	18	146.00	7.72	11	133.18	6.83
Midshaft circumference, right	30	39.70	3.17	16	34.62	4.20
Midshaft circumference, left	30	39.83	3.68	16	35.62	2.94
<b>Humerus</b>						
Maximum length, right	29	326.58	14.14	14	303.14	17.60
Maximum length, left	26	319.46	13.08	9	308.88	29.40
Physiological length, right	29	321.20	13.73	14	297.57	18.29
Physiological length, left	26	313.80	13.77	9	295.55	12.55
Minimum circumference, right	57	67.35	4.91	30	57.66	3.60
Minimum circumference, left	53	65.24	4.86	25	58.24	4.46
Midshaft circumference, right	50	71.18	6.11	29	61.48	4.38
Midshaft circumference, left	48	69.02	6.68	24	62.04	5.93
Minimum diameter, right	52	19.30	2.26	30	16.26	1.91
Minimum diameter, left	49	18.89	2.40	25	16.08	1.86
Maximum diameter, right	52	23.63	2.23	30	20.96	1.99
Maximum diameter, left	49	22.71	2.25	25	21.32	1.97
Inferior epiphysis breadth, right	47	63.55	3.60	20	56.30	2.15
Inferior epiphysis breadth, left	40	63.52	3.24	20	54.50	2.64
Transversal head diameter, right	18	42.89	2.49	13	39.92	6.38
Transversal head diameter, left	18	42.44	2.20	7	41.42	7.41
Vertical head diameter, right	29	46.82	2.76	16	40.12	1.82
Vertical head diameter, left	28	45.64	2.43	9	40.55	1.74
<b>Ulna</b>						
Maximum length, right	22	273.00	9.72	2	254.00	2.82
Maximum length, left	19	264.10	11.41	2	253.33	5.50
Physiological length, right	24	239.41	10.48	6	223.83	14.90
Physiological length, left	23	236.52	14.07	4	210.00	15.42
Minimum circumference, right	39	39.64	3.19	12	34.66	2.14
Minimum circumference, left	39	39.69	3.37	12	34.33	2.57
Midshaft circumference, right	35	52.28	6.56	12	44.33	3.20
Midshaft circumference, left	33	49.57	5.29	11	44.18	3.97
Minimum diameter, right	35	17.65	1.73	12	16.00	1.75
Minimum diameter, left	34	17.38	2.00	11	15.72	1.84
Maximum diameter, right	35	13.80	1.85	12	11.66	1.23
Maximum diameter, left	33	13.48	1.85	11	11.81	1.53
Inferior epiphysis breadth, right	28	20.21	2.36	5	17.40	0.89
Inferior epiphysis breadth, left	32	24.65	28.12	8	17.25	1.90

Legend: N= number of individuals, SD= standard deviation, AP=anterior-posterior, ML= medio-lateral, NF= nutrient foramen



Variable	Males			Females		
	N	Mean	SD	N	Mean	SD
<b>Radius</b>						
Maximum length, right	22	246.13	9.59	5	232.60	5.98
Maximum length, left	22	244.50	10.73	10	223.40	10.37
Physiological length, right	24	237.75	8.54	6	223.66	8.33
Physiological length, left	24	235.62	10.60	10	215.30	9.97
Minimum diameter, left	34	12.08	1.53	19	10.47	1.07
Maximum diameter, right	41	16.31	1.94	14	15.00	1.96
Maximum diameter, left	34	16.20	1.98	19	14.42	1.38
Inferior epiphysis breadth, right	33	33.00	3.53	12	29.25	1.48
Inferior epiphysis breadth, left	30	32.96	2.17	16	29.87	4.42
<b>Femur</b>						
Maximum length, right	26	448.11	18.59	12	415.08	23.00
Maximum length, left	26	451.38	22.09	11	412.81	19.86
Physiological length, right	24	442.29	27.39	12	409.83	22.49
Physiological length, left	25	442.80	29.62	11	411.18	24.82
Midshaft circumference, right	43	93.86	5.95	23	83.17	5.23
Midshaft circumference, left	42	94.69	5.96	25	81.96	3.88
Subtrochanteric circumference, right right	49	99.77	5.36	28	91.85	5.16
Subtrochanteric circumference, left	45	100.26	5.10	29	90.51	4.54
Vertical head diameter, right	30	46.83	1.91	12	42.91	1.62
Vertical head diameter, left	32	47.21	2.33	15	41.33	2.31
Transversal head diameter, right	24	51.03	21.57	10	42.20	1.22
Transversal head diameter, left	25	47.20	1.91	12	41.33	2.18
Subtrochanteric AP diameter, right	53	29.00	5.00	30	25.53	1.81
Subtrochanteric AP diameter, left	49	28.85	4.29	33	25.24	1.52
Subtrochanteric ML diameter, right	53	34.07	2.82	30	31.56	2.26
Subtrochanteric ML diameter, left	40	34.59	3.12	32	31.09	2.63
AP diameter, right	43	31.53	4.82	22	26.45	2.50
AP diameter, left	44	31.70	4.17	25	26.16	2.19
ML diameter, left	43	30.44	8.27	25	26.08	2.05
Inferior epiphysis breadth, right	26	82.07	4.35	13	75.92	3.56
Inferior epiphysis breadth, left	23	82.78	3.72	17	74.23	3.50
<b>Patella</b>						
Maximum breadth, right	17	44.64	3.88	12	40.00	3.97
Maximum breadth, left	23	44.08	2.81	9	39.33	3.67
Maximum height, right	20	42.30	3.48	10	38.60	3.80
Maximum height, left	22	42.59	2.98	11	37.54	2.33
Depth, right	21	20.04	1.85	15	18.33	2.05
Depth, left	18	20.28	1.43	13	18.84	1.72

Legend: N= number of individuals, SD= standard deviation, AP= anterior-posterior, ML= medio-lateral, NF= nutrient foramen

Variable	Males			Females		
	N	Mean	SD	N	Mean	SD
<b>Tibia</b>						
Maximum length, right	29	376.55	15.51	16	344.31	13.36
Maximum length, left	27	376.96	17.46	14	345.21	17.75
Physiological length, right	29	370.58	15.19	15	339.60	13.07
Physiological length, left	26	370.26	15.91	14	339.00	16.70
Minimum circumference, right	51	80.05	6.23	29	70.00	5.16
Minimum circumference, left	49	79.48	6.00	26	69.30	5.11
Midshaft circumference, right	35	86.65	6.52	18	72.00	10.72
Midshaft circumference, left	42	86.97	7.11	20	74.45	4.00
AP diameter at NF, right	48	36.54	4.04	27	30.85	3.07
AP diameter at NF, left	51	35.72	3.20	26	30.80	3.16
ML diameter at NF, right	47	25.02	2.34	27	21.88	2.80
ML diameter at NF, left	51	24.62	2.41	26	21.84	2.42
AP diameter, right	43	31.46	3.57	20	26.75	1.88
AP diameter, left	48	31.64	3.81	21	27.19	1.72
ML diameter, right	43	23.06	2.49	19	20.05	1.31
ML diameter, left	48	22.79	2.55	21	19.61	1.46
Circumference at NF, right	46	97.52	6.65	26	84.69	7.16
Circumference at NF, left	49	97.08	6.18	26	84.88	6.85
Superior epiphysis breadth, right	21	76.95	3.29	15	70.86	5.12
Superior epiphysis breadth, left	19	76.52	3.80	15	69.86	4.82
Inferior epiphysis breadth, right	30	51.33	3.48	13	46.07	2.9
Inferior epiphysis breadth, left	32	50.87	3.11	15	44.86	2.77
<b>Fibula</b>						
Maximum length, right	12	366.58	14.27	9	335.77	14.83
Maximum length, left	13	364.82	16.49	6	329.83	22.03
Minimum circumference, right	30	38.60	3.70	13	35.84	2.15
Minimum circumference, left	33	38.33	4.26	15	35.46	2.69
Inferior epiphysis breadth, right	32	26.96	1.82	15	23.46	1.35
Inferior epiphysis breadth, left	31	26.09	2.11	13	23.23	1.58
<b>Calcaneus</b>						
Maximum length, right	28	81.64	4.85	19	74.15	4.07
Maximum length, left	34	82.35	4.65	15	76.00	4.39
Minimum height, right	33	40.48	9.05	19	33.05	4.91
Posterior breadth, right	29	34.27	3.65	17	31.17	3.32
Posterior breadth, left	27	33.59	2.73	12	31.16	4.19
<b>Talus</b>						
Breadth, right	29	46.03	3.39	20	41.30	3.59
Breadth, left	32	44.43	3.43	18	40.61	3.01
Length, right	31	60.00	3.25	21	53.61	3.72
Length, left	34	58.97	3.14	18	54.11	3.61
Height, right	34	32.94	4.63	19	29.52	1.92
Height, left	33	33.57	5.14	20	29.75	1.91

Legend: N= number of individuals, SD= standard deviation, AP=anterior-posterior, ML= medio-lateral, NF= nutrient foramen

Table 4.1.11: descriptive statistics of postcranial indices of the individuals of Can Reiners

Indices	Males					Females				
	N	Min	Max	Mean	SD	N	Min	Max	Mean	SD
<b>Clavicle</b>										
Robusticity	2	21.3	32.35	26.96	2.58	1	17.5	33.33	25.97	4.40
Robusticity	1	21.5	31.46	27.15	2.66	1	19.8	30.00	26.39	2.82
Claviculohumer	1	44.4	51.37	47.18	2.45	4	41.7	48.00	44.94	2.89
Claviculohumer	1	44.5	49.35	47.50	1.81	4	41.9	46.61	44.27	2.02
<b>Humerus</b>										
Diaphyseal	5	64.0	131.2	81.94	11.3	2	63.6	95.00	77.30	6.61
Diaphyseal	4	64.0	131.2	83.57	12.5	2	63.6	86.36	74.77	6.11
<b>Radius</b>										
Diaphyseal	4	61.1	130.0	76.08	10.6	1	64.0	84.61	73.78	7.12
Diaphyseal	3	58.8	94.11	75.93	7.23	1	64.0	84.61	71.78	5.71
Brachial index	1	71.3	80.61	76.09	2.47	3	75.9	77.13	76.73	0.65
Brachial index	1	70.9	82.06	76.03	2.99	6	73.9	78.52	75.84	1.56
<b>Ulna</b>										
Platolenia index	3	61.9	121.4	78.33	11.6	1	64.7	78.94	73.33	4.86
Platolenia index	3	61.9	121.4	79.08	11.0	1	64.7	86.66	75.82	6.88
<b>Femur</b>										
Pilastric index	4	72.9	139.2	107.8	11.7	2	74.1	131.8	102.9	12.5
Pilastric index	4	86.2	125.0	106.8	9.00	2	74.1	121.7	100.7	12.1
Platymeric	5	66.6	152.0	85.53	15.1	2	71.4	100.0	81.57	6.43
Platymeric	4	57.1	146.1	83.12	14.4	2	69.4	100.0	82.32	7.40
Robusticity	2	12.0	17.99	13.61	1.25	1	11.6	14.05	12.65	0.65
Robusticity	2	12.2	17.99	13.52	1.12	9	11.1	14.46	12.86	1.05
<b>Tibia</b>										
Cnemic index	4	50.0	80.64	68.54	6.91	2	60.6	88.88	71.65	7.89
Cnemic index	5	53.4	86.68	69.10	6.80	2	60.6	92.30	71.70	8.69

Legend: N= number of individuals, SD= standard deviation

Regarding postcranial index (fig. 4.1.6) the superior limb was generally characterized by a robust clavicle. Usually in Can Reiners the clavicle was short (claviculohumeral index) except for the left side of the males, which was long in relation to high differences between both arms. The diaphyseal humerus index showed intriguing values: females were characterized by high percentages of platibrachia which is a medio-lateral flattening of the shaft of the humerus caused by a repetitive use of the biceps and the deltoid. By contrast,

the percentages of the males showed high values of euribrachia which represent normal values of the shaft of the humerus. The differences in the values of the indices among sexes was statistically significant (Mann-Whitney  $P < 0.05$ ). Regarding the brachial index of the ulna we observed high percentages of platolenia in both sexes. Platolenia is present when the superior extremity of the ulna is flattened. These results show robust arms in both sexes but probably related with different task.

For the inferior limbs, the pilastric index of the femour, was generally strong or average in both sexes. High values of this index indicate a strong developing of the *linea aspera*, related to an intense work of the thigh muscles. Also, the robusticity index showed robust values in both sexes. In males these values were more frequent but no significant statistical differences were observed (Mann-Whitney  $P < 0.05$ ). Platymeria (flattening of the superior end of the diaphysis of the femur) was the most represented form in Can Reiners's femurs. Finally, that the platycnemic index of the tibia showed low percentages and mesocnemic and eurycnemic values were more frequent.

The postcranial indices of the skeletal remains of this necropolis showed that in most cases bones were robust, with diaphysis flattened which indicates an intense work of the musculature. The differences between sexes were not significant except for the humeral diaphyseal index where the platybrachia is more intense in females than in males.

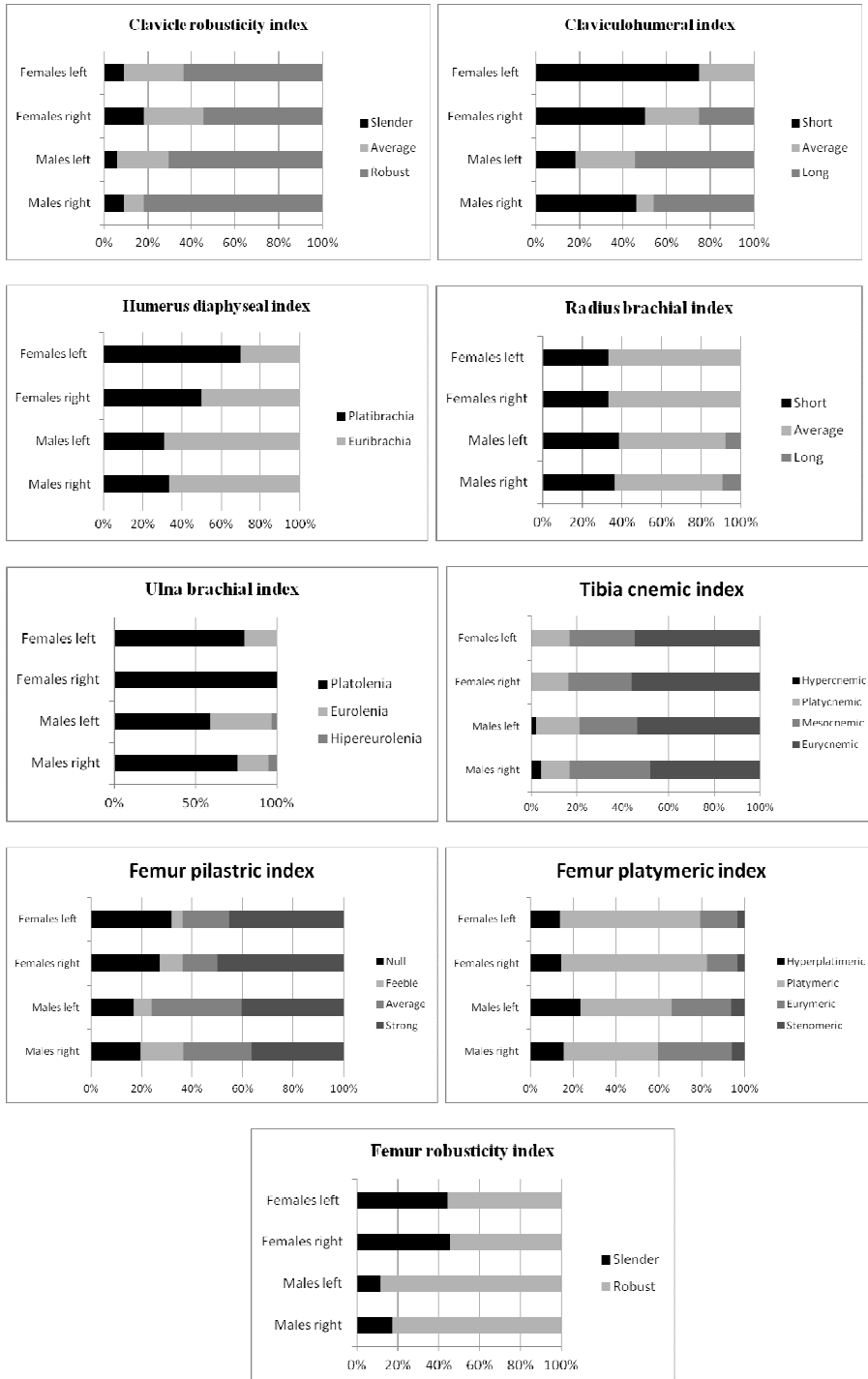


Figure 4.1.6: relative frequencies of the postcranial indices among the adult individuals of the necropolis of Can Reiners

## Biometric comparison among populations

Can Reiners was compared to some populations of Mallorca, Catalonia and Castile and León using selected postcranial measurements. We decided to use right and left lengths and minimum circumferences of long bones, due to the significant differences that we observed between sides in several measurements of long bones of Can Reiners individuals. Principal component analysis (PCA) was applied using mean values of the measurements of all the populations. Table 4.1.12 shows the populations employed for this analysis and table 4.1.10 shows the measurements of Can Reiners used in the PCA.

Table 4.1.12: skeletal samples employed for the principal component analysis (PCA)

Code	Sample	Location	Chronology	Reference
SMM	<b>Santa Maria de Matadars</b>	Barcelona, Catalonia	9th-10th	(Armentano <i>et al.</i> 2005)
SE	<b>Sant Esteve</b>	Barcelona, Catalonia	9th-12th	(Fadrique <i>et al.</i> 2005)
MSL	<b>Monestir de San Llorenç</b>	Barcelona, Catalonia	7th-8th	(Armentano <i>et al.</i> 2004)
IP	<b>S'Illot de Porros</b>	Mallorca, Balearic Islands	6th-2th BC	(Malgosa 1992)
SR	<b>Son Real</b>	Mallorca, Balearic Islands	6th-2th BC	(Font 1977)
EGA	<b>Cathedral of Egara</b>	Terrassa, Catalonia	4th-8th	(Jordana 2007)
TER	<b>Parish churches of Terrassa</b>	Terrassa, Catalonia	9th-13th	(Jordana 2007)
OLM	<b>La Olmeda</b>	Palencia, Castile and León	7th-13th	(Hernández and Turbón 1991)

The first two components of the PCA for males explain 72.85% of the variance (component 1= 58.47%, component 2= 14.38%). Concerning the first component (fig. 4.1.7) we observed that the population of Can Reiners exhibits similar values of the population of the Monestir de San Llorenç, located in Catalonia. The chronologies of the two necropolis are similar. Regarding the second component, our population seems to remain isolated from the other skeletal samples. The necropolis that shows the most different values is the necropolis of Sant Esteve (9th-12th c. AD). This difference is explained by the

values of the minimum circumferences of the ulnae which are the largest in this population. Nevertheless, it is intriguing to observe that the population of S'Illot de Porros and Son Real exhibit different values than Can Reiners. These skeletal samples share the same location, although the populations lived in different period of time.

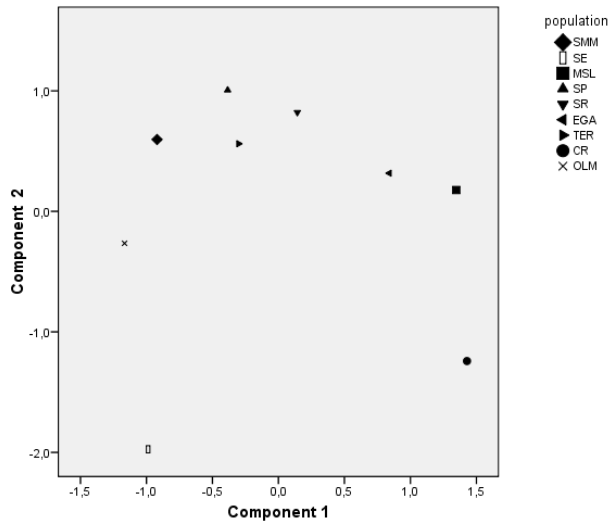


Figure 4.1.7: principal component analysis of males using postcranial measurements: component score of the populations. For the population code see table 4.1.12

For females, the first two components of the PCA explain the 61.75% of the variance (component 1= 41.20%, component 2= 20.54%). For the first component (fig. 4.1.8) we observed that the population of Can Reiners shows similar values to Son Real and Santa Maria de Matadars. This result is different from the male sample, where we observed that Can Reiners and the talayotic populations that lived in Mallorca nine centuries before were not similar. For the female sample we observed that Son Real and Can Reiners are similar. Regarding the second component we noticed a similarity between Can Reiners and the population of Son Real and the Monestir de San Llorenç.

Therefore, we can conclude that for the male sample we observed a similarity with the sample of Monestir de San Llorenç and dissimilitude with the talayotic

populations of Mallorca. By contrast, for the female sample, we observed strong resemblance between our population and Son Real female skeletal remains.

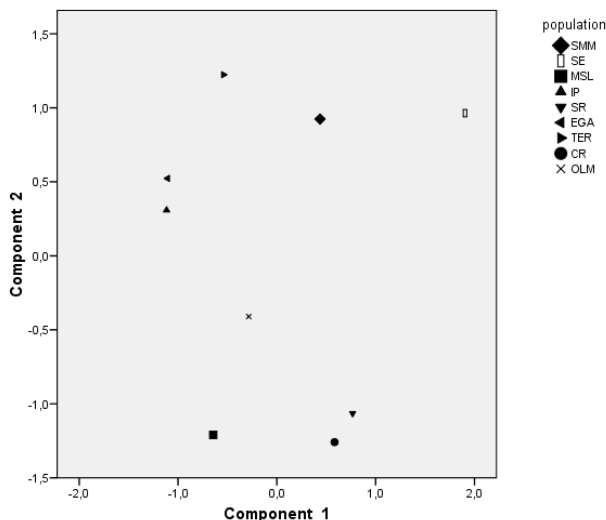


Figure 4.1.8: principal component analysis of females using postcranial measurements: component score of the populations. For the population code see table 4.1.12

### Estimation of the height

Stature has a genetic base however it is also influenced by socioeconomic and environmental factors. The diet plays also an important role: a poor nutrition might be the cause of the onset of a metabolic disease which could influence the stature of the individual. The study of stature and its temporal changes can provide an important contribution to the reconstruction of life and health condition in the archaeological sample. For this reason it is important to study the height in ancient skeletal materials.

The estimation of stature of the individuals of Can Reiners was calculated using four different methodologies in order to analyze if there were differences among them. The methods used were: the regression equations of Pearson



(1899), the regression equations of Trotter and Gleser for Whites (1952, 1977), the tables of Manouvrier (1893) and Mendonça (2000).

Manouvrier (1893) determined the average stature of individuals who presented the same length for a given long bone. The method received some disapproval such as the calculation of the height was approximate and because a statistical analysis was not employed (Olivier, 1963). Pearson, six years later, published his regression theory which used the correlation between long bone length and stature to obtain a linear equation that would predict stature from long bone length. The method was tested in a Chinese population and it did not perform well. Pearson, indeed, obtained his regressions equations using European materials. Trotter and Gleser (1952, 1977) created their regression equations using a sample of American white and Afroamerican cadavers. The problems of this method are related to its application to the ancient European population: it is widely employed in forensic contexts in the United States, however may not performed well with ancient European skeletal material. Nevertheless, in Europe, it is largely employed by anthropologists. Finally, the recent method of Mendonça (2000), based on the study of modern Portuguese cadavers was used for Can Reiners material due to its geographical context: the Iberian Peninsula.

The measurements employed for the estimation of stature were: maximum length and physiological length of the femur, total length of the tibia, maximum length of the fibula, humerus, ulna and radius and physiological length of the humerus (table 4.1.10).

In order to investigate relationship between the methodologies of Manouvrier, Trotter and Gleser, Pearson, Mendonça and MendonçaF (F= physiological), statistical analyses were applied. The estimation of stature of each method did not exhibit a normal distribution (Kolmogorov-Smirnov) and Friedman test was employed to evaluate if there were differences between methods. Results showed differences ( $P < 0.05$ ) when all the individuals were analysed, for males

and females separately, for estimations of right and left femora of males separately, and between estimations from femora and humeri.

To know exactly between which methods differences occur, Wilcoxon test with Bonferroni correction was used. Using all the individuals, we observed significant differences among all pairs of methods ( $P < 0.005$ ), except for the Mendonça-Manouvrier ( $P > 0.005$ ), where no differences were found.

For both males and females, the results showed significant differences between all the methods ( $P < 0.005$ ), except for the pairs Mendonça-Manouvrier ( $P > 0.005$ ) and Mendonça- MendonçaF ( $P > 0.005$ ).

Stature estimations obtained from the right femur of males showed no statistical differences between methods for: Mendonça-Manouvrier ( $P > 0.005$ ), MendonçaF- Mendonça ( $P > 0.005$ ), and MendonçaF-Pearson ( $P > 0.005$ ). For the left femur of males we observed no statistical differences for: Mendonça-Manouvrier ( $P > 0.005$ ), MendonçaF-Mendonça ( $P > 0.005$ ), MendonçaF- Pearson ( $P > 0.005$ ), and Mendonça- Pearson ( $P > 0.005$ ).

No statistical differences between methods obtained from the femurs and the humeri were observed for: Mendonça-Manouvrier ( $P > 0.005$ ) and MendonçaF-Manouvrier ( $P > 0.005$ ), and MendonçaF- Pearson ( $P > 0.005$ ) Mendonça-Pearson ( $P > 0.005$ ), and MendonçaF- Mendonça ( $P > 0.005$ ).

In conclusion, in all the cases we observed statistically differences between Trotter and Gleser and all the other methods ( $P < 0.005$ ), and between Pearson-Manouvrier ( $P < 0.005$ ). There are no statistically differences between Mendonça-Manouvrier and Mendonça-MendonçaF

These findings showed significant differences between the four methodologies and the estimation of the stature changed among these methods. Using ancient skeletal remains we did not know which of these methodologies is correct and which is not. In the last decades, the method of Trotter and Gleser was largely employed for the American and European sample. By contrast the table of Manouvrier were used in the past and not in recent studies.

Therefore when comparing data from different sites we faced several problems such as the selection of the bone and the choice of the method to calculate height. Ortner (2003) suggests comparing long bones lengths directly rather than converting long bone lengths to stature estimates. Nevertheless, estimation of stature provides a specific trait of the individuals and populations, and physical anthropologists continue to use them.

Tables 4.1.13 and 4.1.14 display the estimation of stature of Can Reiners individuals divided for sex, bone side and method employed. We noticed that in all the cases the estimate stature calculated with the regression equations of Trotter and Gleser were higher compared to the others. By contrast the method of Pearson always displayed the lowest values.

Table 4.1.13: male estimation of stature

<b>Bone</b>	<b>Variables</b>	<b>N</b>	<b>Min</b>	<b>Max</b>	<b>Mean</b>	<b>SD</b>
Femur-right	Manouvrier	24	160.5	173.0	<b>167.4</b>	3.15
	Pearson	24	159.1	170.8	<b>165.2</b>	3.08
	Mendonça	24	158.0	175.0	<b>166.5</b>	4.68
	MendonçaF	24	158.0	174.0	<b>166.1</b>	4.34
	Trotter&Gleser	24	160.4	175.9	<b>168.5</b>	4.16
Femur-left	Manouvrier	25	160.5	178.5	<b>167.4</b>	4.11
	Pearson	25	158.9	172.7	<b>165.3</b>	3.80
	Mendonça	25	155.0	180.0	<b>166.5</b>	6.13
	MendonçaF	25	157.0	177.0	<b>166.2</b>	5.52
	Trotter&Gleser	25	159.7	180.2	<b>168.7</b>	5.22
Humerus-right	Manouvrier	30	153.0	176.7	<b>165.7</b>	5.39
	Pearson	30	155.7	173.7	<b>164.8</b>	4.35
	Mendonça	30	156.0	176.0	<b>165.7</b>	4.87
	Trotter&Gleser	30	161.1	180.1	<b>170.7</b>	4.63
Humerus-left	Manouvrier	25	153.0	168.6	<b>164.1</b>	4.30
	Pearson	25	154.6	168.2	<b>163.4</b>	3.57
	Mendonça	25	154.4	170.0	<b>164.2</b>	4.19
	Trotter&Gleser	25	161.1	174.2	<b>169.1</b>	3.79

Table 4.1.14: female estimation of stature

<b>Variables</b>	<b>Bone</b>	<b>N</b>	<b>Min</b>	<b>Max</b>	<b>Mean</b>	<b>SD</b>
Femur-right	Manouvrier	12	148.8	165.0	<b>155.1</b>	5.37
	Pearson	12	146.4	160.4	<b>152.5</b>	4.37
	Mendonça	12	149.0	166.0	<b>158.8</b>	5.52
	MendonçaF	12	148.0	165.0	<b>155.2</b>	5.42
	Trotter&Gleser	12	149.7	166.7	<b>156.6</b>	5.68
Femur-left	Manouvrier	11	148.8	165.0	<b>154.8</b>	4.64
	Pearson	11	145.4	160.9	<b>152.8</b>	4.82
	Mendonça	11	150.0	167.0	<b>155.5</b>	4.84
	MendonçaF	11	146.0	166.0	<b>155.5</b>	6.18
	Trotter&Gleser	11	150.2	167.2	<b>156.1</b>	4.90
Humerus-right	Manouvrier	14	149.7	171.5	<b>156.5</b>	6.07
	Pearson	14	149.1	166.2	<b>155.5</b>	4.66
	Mendonça	14	151.0	170.0	<b>157.4</b>	5.37
	Trotter&Gleser	14	152.7	173.5	<b>160.4</b>	5.67

Due to our results and also due to the theory in which inferior long bones give a major reliability respect to the superior limbs (Krogman and Iscan 1986), we decided to employ the values of the right femur, for both sexes, to compare the estimation of stature with other populations. The difference between the estimation of height between sexes ranges from 7.70 cm (Mendonça) to 12.7 cm (Pearson). These values are considered average values by Manouvrier (1893). The mean of the stature of males among the methodologies ranges from 165.2 to 168.5 whereas, for females ranges from 152.5 to 156.6.

In table 4.1.15 the estimation of heights from contexts which have in common the geographic area or the chronology of the population of Can Reiners are displayed. These tables show that Pearson (1899) is the most employed method in Spain. By contrast, in Italy the methodology of Trotter and Gleser is mostly used. In the French series the methodologies employed were not reported.

Table 4.1.15: comparison of the estimation of the stature of both sexes, among different series of populations

Population	Chronology	Pearson (1899)		Other Methodologies	
		Mean males	Mean females	Mean males	Mean females
S'Illot de Porros (Malgosa 1992)	Talayotic (6th -2th BC)	166.69	153.53		
Son Real (Font 1977)	Talayotic (6th -2th BC)	163.61	153.77		
Tarragona (Pons 1949)	Roman	163.18	151.82		
Ampurias (Pons 1949)	Roman	165.87	147.32		
El Cerro del Castellón (Luna and Bertranpetit 1983)	Late Roman	164.2	155.7		
La Solana (Safont <i>et al.</i> 1997)	6th-8th AD	166.1	154.1		
Cathedral of Egara (Jordana 2007)	4th-8th AD	167.16	153.82		
Parish church of Terrassa (Jordana 2007)	9th-13th AD	166.23	155.15		
La Olmeda (Hernández and Turbón 1991)	7th-13th AD	163.3	155.4	166.82*	160.67*
Santa Maria de Hito (Galera and Garralda 1992)	6th-13th AD	165.4	155.3		
Le Palazette (Facchini and Brasili 1977-79)	1th-3th AD	163.18	151.82		
Osteria del Curato II (Catalano and Minozzi 2003)	Roman Imperial			170.0*	157.0*
San Martino in Collinaia (Amadei 1993)	Late Roman			168.0*	159.7*
San Pietro in Mavinas (Fiorin and Canci 2011)	6th-9th AD			176.5*	166.0*
Collegno (Bedini and Bertoldi 2004)	6th-7th AD			173.6*	163.3*
Cadarache (Pouyé <i>et al.</i> 1994)	Late Roman			167.0**	160.7**
Giberville (Pilet <i>et al.</i> 1990)	6th AD			172.2**	159.2**
Frénouville (Buchet 1978)	5th-7th AD			167.0**	155.0**
<b>Can Reiners</b>	<b>7th-</b>	<b>165.2</b>	<b>152.5</b>	<b>168.5*</b>	<b>156.6*</b>

Legend: \*= Trotter and Gleser method (1952,1977), \*\*= unspecified method

Starting from the estimation of stature of female individuals, we can advance some inferences. Regarding Pearson method (1899), the mean value of Can Reiners height was one of the lowest. All the early medieval and medieval samples show higher values, and also the samples of the Talayotic period of Son Real (Font 1977) and S'Illot de Porros (Malgosa 1992) exhibit higher

values than our sample. However, the values of the Roman period of the necropolis of Ampurias and Tarragona in Spain (Pons 1949) and the necropolis of Le Palazette in Italy (Facchini and Brasili 1977-79) display lower values than Can Reiners. From a chronological point of view we observed that the female stature diminish from Talayotic to Roman period, and increase again at the Medieval time. The height of the females of Can Reiners is more similar to those of the Roman period and different from those of the medieval time.

Observing the estimation of stature calculated with Trotter and Gleser (1952, 1977), the value of the height of females increase considerably from 152.5 to 156.6. Clearly, we cannot compare the values obtained with Trotter and Gleser with the value calculated with Pearson. Compared to the other values of the Italian samples with same methodologies we noticed that our value is also the lowest value.

The necropoles of Cadarache and Giberville from France show again, higher values of height than Can Reiners if compared with Pearson (1899) and Trotter and Gleser (1952, 1977) methods.

Regarding males, and more in particular the values of Pearson (1899), we observed that the estimation of stature of Can Reiners individuals is similar to the males of the Roman period of Ampurias (Pons 1949) and Santa Maria de Hito (6th-13th century AD) (Galera and Garralda 1992). Considering Trotter and Gleser (1952, 1977), we noticed that the mean of height of Can Reiners is similar to the Italian late Roman necropolis of San Martino in Collinaia (Amadei 1993). The others values are higher than ours. Finally, observing the values from the French cemeteries, they are higher (Pearson method) than Can Reiners or not (Trotter and Gleser) depending of the method of comparison.

Considering the methods of Pearson and Trotter and Gleser separately, we can conclude that the values of males and females showed different trends. The mean of male stature appears coherent with the mean values of the time (7th c. AD) whereas the mean value of female showed a different trend. Female stature

of Can Reiners exhibits lower values than those of the Talayotic period in the Balearic Islands, the late Roman and early medieval period of the analyzed cemeteries, being more similar to the mean of the Roman period.

### **Epigenetic analysis**

The morphological skeletal variants analysis was accomplished using 77 epigenetic traits, belonging to cranial and postcranial skeleton. We observed frequencies in the population and differences between sexes.

Regarding frequencies of cranial traits, we observed that in 10 cases (supranasal suture, medial supraorbital notch, infraorbital foramen, zygomaxillary tubercle, mental foramen, suprameatal spine and depression, zygomaticofacial foramen, condylar foramen, transverse palatine suture, premaxillary suture) epigenetic traits were present with a relative frequency of more than 50% (table 4.1.16) because represent the normal trait. By contrast, in the postcranial skeleton discrete traits were less frequent. Only in two cases (acromial articular facet and lateral tibial squatting facet) they had a relative frequency higher than 40% (table 4.1.17). Nevertheless, the relative high frequency of these traits could be related to activity skeletal markers (Trinkaus 1978, Capasso *et al.* 1999). Both facets (acromial articular and lateral tibial squatting) were equally present in females and males individuals in right and left sides. The acromial facet is located in the inferior surface of the acromion process of the scapula and it is created by the impingement of the humeral head in extension of the arm overhead. The squatting facet is due to impingement of the tibia on the talar head in hyper-dorsiflexion of the ankle, typical of squatting or kneeling position posture. The medial and/or lateral *fossa* is positioned in the anterior margin of the distal tibia articular surface. We also observed a relative frequency of 35.1% of variations in the articular surface of the talus, related to this posture. Lateral squatting facets are common in several ancient and modern skeletal series. For example, in the skeletal remains of the prehistoric village site in northeast of

Thailand, tibia lateral squatting facets were present in 92.6% of the individuals (Pietrusewsky and Douglas 2002).

Table 4.1.16: presence of cranial epigenetic traits in the individuals of Can Reiners

<b>Cranial</b>	<b>N</b>	<b>+</b>	<b>%</b>	<b>Cranial</b>	<b>N</b>	<b>+</b>	<b>%</b>	<b>Cranial</b>	<b>N</b>	<b>+</b>	<b>%</b>
<b>1</b>	45	4	8.9	<b>19</b>	56	1	1.8	<b>37</b>	59	14	23.7
<b>2</b>	41	1	2.4	<b>20</b>	50	1	2.0	<b>38</b>	58	20	34.5
<b>3</b>	37	24	64.9	<b>21</b>	47	1	2.1	<b>39*</b>	61	19	31.1
<b>4</b>	47	16	34.0	<b>22</b>	47	2	4.3	<b>40</b>	30	1	3.3
<b>5</b>	47	6	12.8	<b>23</b>	52	3	5.8	<b>41</b>	33	15	45.5
<b>6</b>	47	16	34.0	<b>24</b>	54	5	9.3	<b>42</b>	30	0	0.0
<b>7*</b>	47	26	55.3	<b>25</b>	54	4	7.4	<b>43</b>	34	2	5.9
<b>8</b>	31	3	9.7	<b>26</b>	52	3	6.8	<b>44</b>	18	10	55.6
<b>9</b>	39	3	7.7	<b>27</b>	50	1	2.0	<b>45</b>	17	5	29.4
<b>10</b>	45	4	8.9	<b>28</b>	72	29	40.3	<b>46*</b>	21	2	9.5
<b>11</b>	9	0	0.0	<b>29</b>	72	27	37.5	<b>47</b>	22	3	13.6
<b>12</b>	10	3	30.0	<b>30</b>	71	49	69.0	<b>48</b>	19	12	63.2
<b>13</b>	7	2	28.6	<b>31</b>	69	6	8.7	<b>49</b>	24	12	50.0
<b>14</b>	31	9	29.0	<b>32</b>	32	11	34.4	<b>50</b>	50	0	0.0
<b>15</b>	37	34	91.9	<b>33*</b>	44	35	79.5	<b>51</b>	50	0	0.0
<b>16</b>	41	22	53.7	<b>34</b>	41	1	2.4	<b>52</b>	60	0	0.0
<b>17</b>	55	51	92.7	<b>35*</b>	67	7	10.4	<b>53</b>	47	0	0.0
<b>18</b>	58	0	0.0	<b>36</b>	66	3	4.5				

Legend: N= individuals of the sample, += absolute frequency of presence of the epigenetic trait, %= relative frequency of presence of the epigenetic trait, \*= statistically significant differences between sexes (Fisher's exact test:  $p < 0.05$ ). For legend of traits see material and methods section.

Table 4.1.17: presence of postcranial epigenetic traits in the individuals of Can Reiners

<b>Postcranial</b>	<b>N</b>	<b>+</b>	<b>%</b>	<b>Postcranial</b>	<b>N</b>	<b>+</b>	<b>%</b>
<b>1</b>	76	7	9.2	<b>14</b>	69	13	18.8
<b>2</b>	62	4	6.5	<b>15a</b>	70	33	47.1
<b>3</b>	65	19	29.2	<b>15b</b>	70	37	52.9
<b>4</b>	85	20	23.5	<b>16</b>	64	25	39.1
<b>5</b>	86	8	9.3	<b>17a</b>	58	12	20.7
<b>6</b>	78	14	17.9	<b>17b</b>	58	46	79.3
<b>7</b>	91	0	0.0	<b>18</b>	57	20	35.1
<b>8*</b>	92	16	17.4	<b>19</b>	73	12	16.4
<b>9</b>	44	20	45.5	<b>20</b>	57	20	35.1
<b>10</b>	25	9	36	<b>21</b>	41	1	2.4
<b>11</b>	35	2	5.7	<b>22</b>	52	1	1.9
<b>12</b>	70	30	42.9	<b>23</b>	52	6	11.5
<b>13</b>	68	4	5.9	<b>24</b>	39	2	5.0

Legend: N= individual of the sample, += absolute frequency of presence of the epigenetic trait, %= relative frequency of presence of the epigenetic trait, \*= statistically significant differences between sexes (Fisher's exact test:  $p < 0.05$ ). For legend of traits see material and methods section.



The epigenetic traits which showed statistically significant differences (Fisher's exact test:  $p < 0.05$ ) between males and females are described in table 4.1.18.

Table 4.1.18: epigenetic traits with statistically significant differences among sex (Fisher's exact test:  $p < 0.05$ )

	<b>Epigenetic traits</b>	<b>Sex</b>	<b>Absent</b>	<b>Present</b>	<b>Total</b>
<b>Cranial</b>	7-medial supraorbital notch	male	17	13	30
		female	4	13	17
		Total	21	26	47
	33-zygomaticofacial foramen	male	9	22	31
		female	0	13	13
		Total	9	25	44
	35-ossicle at lambda	male	42	1	43
		female	18	6	24
		Total	60	7	67
	39- parietal foramina	male	24	16	40
		female	18	3	21
		Total	42	19	61
<b>Postcranial</b>	8- humeral septal aperture	male	55	7	62
		female	21	9	30
		Total	76	16	92

Ossicle at lambda, for example, seems to be more prevalent on female sample. The ossicles at lambda and lambdoid ossicles represent fontanelle ossicle characterized by a later onset developmentally and the premature fusion of secondary additional centre of ossification along the lambdoid suture. Both may occur in the postnatal period and may be subjected to mechanical influences (Mirazón Lahr 1996). A study of sexual dimorphism among the wormian bones in adult skulls (Patil and Sheelavant 2012) revealed that the overall incidence of the wormian bones was higher in female (64.80%). Regarding the ossicle at lambda, they observed a higher incidence in the female skulls. A study of the frequencies variations of the cranial epigenetic in human population (Hanihara and Ishida 2001) showed that a good number of samples from the western hemisphere of the Old World (from subsaharian Africa to Europe), western Oceania, and the New World have generally high frequencies of the ossicle at lambda (10%-20%).

In relation to medial supraorbital notch, foramen zygomaticofacial and parietal foramina, different results were obtained depending of the author (Hauser and De Stefano, 1989). In general, most works reported no significant differences between sexes or sides, whereas others suggested slight differences between sexes.

Regarding the postcranial non-metrical traits, we observed that septal aperture of the humerus is more frequent among females. The septal aperture is a deficit of the bony septum of the humerus that separates the coronoid fossa from the olecranon fossa. Its incidence in adults ranges from 6.9% in American Whites, to almost 60% in some Northern African and West African groups (Paraskevas *et al.* 2012). It is more frequent in females of modern (Paraskevas *et al.* 2010) and ancient (Jordana 2007, Mays 2008) populations.

The lack of systematic works on this subject prevents more accurate comparisons with other archaeological series of the context of Can Reiners.

### **Palaeopathology**

The term palaeopathology derived from the Greek and its literary means “study of the ancient pathologies”. This discipline employed the ancient human skeletal and mummified remains to reconstruct the life-style and health of the past population (Fornaciari and Giuffr  2009).

Ortner (2003) reported that a relative small percentage (ca. 10-15%) of skeletal remains exhibits significant evidence of disease. One of the most common type of diseases, infections, can be barely traced in bones. The prevalence of skeletal infectious disease, therefore, could be a poor indicator of the overall health of an assemblage of skeletal remains. Nevertheless, paleopathologists believe that infection diseases were common in the past and many of the deaths of the infants, seen in the archaeological skeletal material, would have resulted from gastro-intestinal infections as in the case of the poor countries nowadays (Black *et al.* 2003, Waldron 2009).

As described above, we observed a small prevalence of skeletal disease on the skeletal remains of the cemetery of Can Reiners. The pathologies of our sample are the same that frequently affect ancient population. The different types of disease will be described separately.

### **Osteoarthritis and eburneation**

Osteoarthritis is one of the most common conditions seen in skeletons. It is a low grade of inflammatory disease, chronic and degenerative, that affects synovial joints. Factors that contribute to the formation of osteoarthritis are age, genetics, sex, obesity, associated diseases, trauma and most importantly, overload. Generally, the changes that take place in the articulating bones include: a) formation of new bone around the margins of the bone (osteophytes) and the joint surface; b) pitting on the joint surface; c) changes in the normal contour of the joint and production of eburneation. An eburneation is a highly polished area which develops in areas of the joint where the articular cartilage has been completely lost (Waldron 2009). In Can Reiners we observed the presence of eburneation in only 4 individuals (1.85%). The individuals were two adult males and two mature females. Eburneations were located on the condyle of the femurs (2 cases), on a right scaphoid and on a left glenoid fossa of scapula (Cr. 8.1, Cr.169.1, Cr.126.1, Cr. 201.1).

Osteoarthritis was uncommon in the individuals of Can Reiners except in the vertebral spine. We analyzed signs of the degeneration of the intervertebral disc and the articular facets of vertebrae, and the formation of the nodes of Schmörl.

Degeneration of the disc is particularly associated with aging and occurs equally in both sexes. Usually, there are formation of marginal osteophytes and pitting on the superior and inferior surfaces of the vertebrae most commonly seen in the lower cervical and lumbar regions (Waldron 2009). Schmörl's nodes are protrusions of the intervertebral disc through the vertebral body endplate and into the adjacent vertebra. They are more common in individuals who impose great stresses on their lower spine.

The presence of osteoarthritis on the spine was analyzed on 1258 vertebrae, 827 from male individuals and 425 to females (table 4.1.19). 111 male and 85 female vertebrae were affected by some degree of degenerative diseases.

Table 4.1.19: frequencies of the osteoarthritis in the vertebral column

	Total vertebrae			Affected vertebrae			% affected vertebrae		
	Males	Females	x	Males	Females	x	Males	Females	x
<b>Cervical</b>	264	137	18	25	28	0	9.46%	20.43%	0.00%
<b>Thoracic</b>	391	205	41	47	31	0	12.02%	15.12%	0.00%
<b>Lumbar</b>	172	83	14	39	26	0	22.67%	31.32%	0.00%

Legend: x= undetermined individuals

The presence of osteoarthritis on the cervical vertebrae was individuated and analysed on 71 individuals. Results showed that 24.54% of the individuals suffered of cervical osteoarthritis. Of the 16 individuals with osteoarthritis 9 were males and 7 were females (fig. 4.1.9). 6 females and 5 male were mature and 1 female and 4 males were adult. No juvenile or children were affected.



Figure 4.1.9: cervical osteoarthritis on the articular surface in a mature female (Cr. 201.1)

Regarding frequencies of the number of cervical vertebrae affected by osteoarthritis, 20.43% belonged to females, whereas only 9.46% belonged to

males (table 4.1.19). In both cases we observed that osteoarthritis was related to the age at death.

Of a sample of 70 individuals, 24 of them (34.28%) exhibited osteoarthritis in the thoracic vertebrae. 9 of them were mature (5 males and 4 females) and 15 of them were adults between 27 and 40 years of age (10 males and 5 females). Despite the high number of males, the percentages of the affected vertebrae were similar in both sexes (table 4.1.19). This is due to the number of vertebrae affected in the same individual with the increase of age at death.

In relation to the lumbar vertebrae affected by osteoarthritis, 21 over 61 individuals, (34.42%) showed affectation. In this segment of the vertebral column females showed also a higher percentage of osteoarthritis (table 4.1.19).

Nevertheless, no differences between the presence and absence of osteoarthritis in the vertebrae (cervical, thoracic and lumbar) among sexes were observed (Chi-squared test  $P > 0.05$ ). However, correlations between the osteoarthritis of vertebrae among the class of age at death has been found, Fisher's exact test shows an association ( $P < 0.05$ ) between cervical (table 4.1.20) and lumbar (table 4.1.21) osteoarthritis and age at death. No association (Fisher's exact test  $P > 0.05$ ) was observed between thoracic osteoarthritis and age at death (table 4.1.22).

Table 4.1.20: Analysis of association between cervical osteoarthritis and class of age at death (Fisher exact test=  $P < 0.05$ ). N= number of vertebrae

		Cervical osteoarthritis		Total	
		Absence	Presence		
<b>Age class</b>	Juvenile	N	6	0	6
		% age class	100.0%	.0%	
	Adult	N	28	5	33
		% age class	84.8%	15.2%	
	Mature	N	8	11	19
		% age class	42.1%	57.9 %	
<b>Total</b>		N	42	16	58
		% age class	72.4%	27.6%	

Table 4.1.21: Analysis of association between thoracic osteoarthritis and class of age at death. (Fisher exact test= P>0.05). N= number of vertebrae

		<b>Lumbar osteoarthritis</b>		<b>Total</b>	
		Absence	Presence		
<b>Age class</b>	Juvenile	N	5	0	5
		% age class	100.0%	.0%	
	Adult	N	24	8	32
		% age class	75.0%	25.0%	
	Mature	N	6	11	17
		% age class	35.3%	64.7%	
<b>Total</b>		N	35	19	54
		% age class	64.8%	35.2%	

Table 4.1.22: Analysis of association between lumbar osteoarthritis and class of age at death. Fisher exact test= P<0.05. N= number of vertebrae

		<b>Thoracic osteoarthritis</b>		<b>Total</b>	
		Absence	Presence		
<b>Age class</b>	Juvenile	N	5	0	5
		% age class	100.0%	.0%	100.0%
	Adult	N	26	9	35
		% age class	74.3%	25.7%	100.0%
	Mature	N	9	9	18
		% age class	50.0%	50.0%	100.0%
<b>Total</b>		N	40	18	58
		% age class	69.1%	31.0%	100.0%

The presence of Schmörl's nodes was analyzed on 1341 vertebrae, 841 from male individuals and 500 to females (table 4.1.23). 111 male and 38 female vertebrae were affected.

Table 4.1.23: frequencies of Schmörl's nodes in the vertebral column

	<b>Total vertebrae</b>			<b>Affected vertebrae</b>			<b>% of affected vertebrae</b>		
	Males	Females	x	Males	Females	x	Males	Females	x
<b>Cervical</b>	259	137	20	3	2	0	1.15%	1.45%	0.00%
<b>Thoracic</b>	410	191	35	79	16	0	19.26%	8.37%	0.00%
<b>Lumbar</b>	172	172	14	29	20	0	16.86%	26.31%	0.00%

Legend: x= undetermined individuals

The analysis of the frequencies of Schmörl's nodes on the cervical vertebrae was carried out in 72 individuals. Results showed that only 5 individuals (6.94%) suffered this lesion and the vertebrae affected were few (table 4.1.23). In the thoracic vertebrae Schmörl's nodes were more frequent (14.3%, N=31) in males (N=24) than in females (N=7). Also the number of male affected vertebrae was higher (19.26%) than females ones. In the lumbar vertebrae nodes were more frequent in males (N=14) than in females (N=9). Whereas, the number of affected vertebrae were higher in females (26.31%) (table 4.1.23). Nevertheless, no differences between the presence and absence of Schmörl's nodes in the vertebrae (cervical, thoracic and lumbar) among sexes and among age at death, were observed (Chi-squared test  $P>0.05$ ).

## **Fractures**

Fractures are the most common form of trauma found in human skeletal remains. A fracture is a break in the continuity of bone and it is almost always associated with damage to overlying or adjacent soft tissues (Waldron 2009). We observed fractures on 6.48% of the population (14 individuals) corresponding to 10 adult males and 4 females. Fractures affected clavicles (2 individuals), radius (1 individual), ulnae (2 individuals), ribs (2 individuals), metacarpal bones (5 individuals) and metatarsal bones (2 individuals). Some individual were affected for more than one fracture. Fractures were well-reduced except for one of the two individuals with the ribs fracture: the several fractures located on the right side were still fractured, developing periostitis near the border of the fracture.

*Osteochondritis dissecans* (OCD) is a joint condition in which bone underneath the cartilage dies due to lack of blood flow. This bone and cartilage can then break loose, causing pain and possibly hinder joint motion. It may be caused by direct trauma or repetitive microtrauma but the exact etiology is unclear (Waldron 2009). Usually the onset of disease occurs in adolescents and young adults and it is much more common in males than in females (Ortner 2003). The

most common sites for these anomalies are: the knee, the glenoid fossa, and the proximal surface of the first phalanx of the great toe. The *osteochondritis dissecans* of the wrist is rare and most confined in the radius-scaphoid joint. We observed the presence of osteochondritis in 16 individuals (7.40% of the sample). The majority of the individuals affected were adult male (12 males, 75%). The bones affected by osteochondritis were: 9 the first phalanx of the great toe, 2 third cuneiform, 1 third metatarsal, 1 condyle of the femour, 2 glenoid fossae, 1 lunate bone.

### **Infections**

Periostitis is a non-specific bone reaction that could be due to an infection. When the periosteum is inflamed, new bone is commonly formed, sometimes as a concomitant of well-recognized disease. Periostitis was observed in 8 individuals (83.7% of the sample) and was noticed only in adult males between 30 and 40 years of age. It affected mainly tibiae and fibulae. In one adult male (Cr 23.1) periostitis was observed on the superior and inferior limbs and on the ribs. Several right ribs were fractured (fig 4.1.10).

In Can Reiners sample only one case of osteomyelitis was observed (0.5% of the individuals). The individual was an adult male of 40 years of age (Cr 8.1) and the disease affected the tibia. Osteomyelitis is a term used for any form of infection of bone and marrow bone which results in its inflammatory destruction. Generally, a drainage channel called *cloaca* is formed to drain the pus out of the bone. Osteomyelitis could be consequence of a fracture and is often the result of pyogenic bacteria into the bone either from outside or hematogenous origin. However other infections agents such as viruses, fungi and multicelled parasites can also infect marrow bone (Waldron 2009).



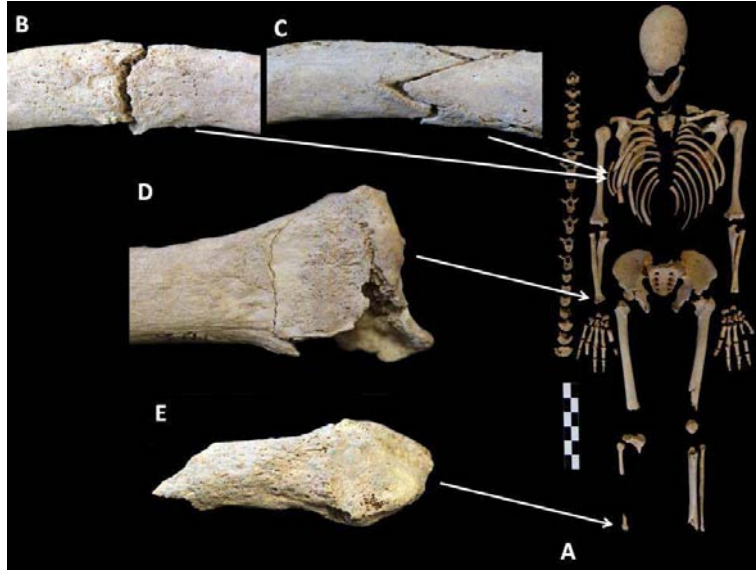


Figure 4.1.10: the adult male Cr. 23.1: skeletal preservation (A), ribs fractures and traces of periostitis (B and C), periostitis on the distal epiphysis of the right radius (D), periostitis on the distal epiphysis of the right fibula (E)

## Tumours

Benign tumours of bones may arise from bone or cartilage. Osteomas are overgrowths of normal bone that are formed in the periosteum. They are common, usually small and easy to recognize, and generally they have no clinical significance. Osteoma occurs almost exclusively in the skull and the most frequent locations are the frontal and the parietal bones (Waldron 2009). In Can Reiners sample only one osteoma (0.5% of the population) located in the frontal bone was observed (Cr. 72.1). It consists in a smooth lump of compact bone and it is present in an adult male about 25-30 years old.

Within the tumours that arise from cartilage we observed only one osteochondroma (Cr. 72.1; 0.5% of the sample). Osteochondromas are bony excrescences covered with cartilage that arise from the surface of bone at metaphysary level. They are the most common benign bone tumours and are found most frequently on the femur, humerus and tibia, especially the areas

with the most rapid growth such as the distal femur, and the proximal humerus and tibia. Its initiation is limited to the growing period of the skeleton, beginning most exclusively in childhood. There is no sex predilection (Ortner 2003; Waldron 2009).

In the adult male Cr 72.1, indeed, an osteochondroma was observed in the superior metaphysis of the femur (figure 4.1.11 A, B and C). In the same individuals we previously described the presence of an osteoma.



Figure 4.1.11: Intrarticular osteochondroma of the right femur in an adult male (Cr. 78.1). Posterior view of the femur (A), particular of the tumour, medial view (B), particular of the osteochondroma, posterior view (C)

### **Congenital anomalies**

Variation in skeletal structure reflects disturbances during morphogenesis, usually resulting from genetic mutations. Sometimes, also other factors such as maternal infection or nutritional disorders can behave as epigenic factor acting like mutant genes during embryonic development (Barnes 2012). The analysis

of skeletal variation in the sample of Can Reiners showed that 10 individuals (4.6%) exhibited congenital anomalies mainly in males (N=7). The anomalies identified were: one case of caudal shift of the L-S border (fig. 4.1.12 A), 4 cases of cranial shifts of the lumbar-sacral (L-S) border (or L5 sacralisation), 2 cases of single block vertebra (fig. 4.1.12 B and C), 1 probable case of Klippel-Feil multiple block vertebra, 2 sternum anomalies, and one carpal coalition vs fusion.

Caudal shift of the L-S border (figure 4.1.12 A) causes the first sacral segment takes on lumbar features. In our case we observed a partial separation of S1 from the sacrum. When adjacent vertebral segments fail to separate, the fissure between precursor developing units of resegmented does not appear, and a unification of the vertebra occurs. This is the case with two completely blocked vertebrae. The caudal shift of the L-S border and a case of two block vertebrae were observed in the same individual (Cr 6.1).

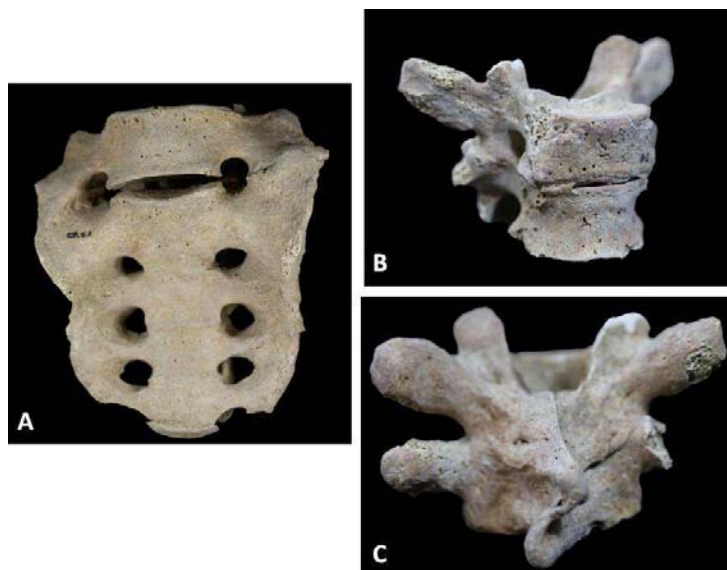


Figure 4.1.12: caudal shift of the L-S border (Cr.6.1) (A), a frontal (B) and a posterior (C) view of two block vertebrae (Cr 6.1)

We also observed a probably case of Klippel-Feil, but the state of preservation of the bones did not allow a properly description of the anomaly. Regarding sternum anomalies, we observed a case of sternal aperture and a curious case of a congenital non-fusion of the caudal portion of the sternum (Cr 16.1) (fig. 4.1.13). This type of congenital defect is unusual and there are few cases reported in the palaeopathological literature (Barnes 2012, Capasso 2001). This represents a rare malformation, but no other skeletal abnormalities was found and no associated pathological conditions affecting other structures was detected. The reason for this incomplete fusion in the midline of the mesosternum is most certainly idiopathic.



Figure 4.1.13: congenital sternal anomaly in an male between 17 and 21 years of age (Cr. 16.1)

Finally we reported a case of carpal fusion. An adult male individual (Cr 78.1) exhibited a massive carpal coalition on the left hand (figure 4.1.14) while the right hand had a normal development. Figure 4.1.14 B displays the left hand in dorsal view: all carpals except the pisiform bone, are united with the second and third metacarpals. This anomaly could be confused with an ankylosis of the wrist due to rheumatic diseases.



Figure 4.1.14: unilateral massive carpal coalition in an adult male (Cr 78.1). Left and right hand of the individual (A), detail of the carpal coalition of the left hand (B)

Our study of degenerative vertebra changes in the necropolis of Can Reiners showed age correlation between osteoarthritis and cervical and lumbar vertebrae. No statistically significant correlation was observed between osteoarthritis and sex or thoracic vertebrae. Neither statistical correlation was observed between nodes of Schmörl and sex and age at death. These results allow us to hypothesise that the onset of osteoarthritis on the thoracic vertebrae and the formation of the Schmörl's nodes in all the vertebrae are related with the mechanical stress that the individuals were exposed. In relation with other populations of the same geographical frame, there are so few paleopathological studies. In this sense, the study of the population of S'Illot des Porros (Malgosa and Campillo, 1991) and Son Peretó (Alapont *et al.* 2012) can be used for comparison. In S'Illot des Porros 36.9% of individuals exhibit some form of osteoarthritis, mainly in spine of male individuals. In this prehistoric necropolis, some cases of Schmörl'nodes, kyphoscoliosis and scoliosis were found (Castellana *et al.* 1999). Osteoarthrosis is the most predominant disease in Son Peretó. As expected, if the affectation is not exactly the same, the three necropoles have a same pattern of adaptation to a hard style of life.

The frequency of traumatismos observed in the individuals of Can Reiners (14 affected individuals) is not so high, and definitively lower than those found in

S'Illot des Porros (27 affected individuals; Pérez *et al.* 1991) that have a similar sample size (219 individuals taken into consideration in this work). However it coincides in the higher affectation of males than females (1.9 M/F); this evidence was observed also in other studies (Bennike 1985). Also Can Reiners coincides with S'Illot des Porros in the lack of evidences for lesions suspected to be secondary to violent/war actions, in contrast with those found in Son Real, also in the Alcudia bay (Campillo 1985). On the other hand, it is interesting to note the presence of trepanations, a ritual traumatism, in Son Real necropolis where several spectacular cases were found (Campillo 1985). By contrast, in only two cases were found in S'Illot Porros (Subirà *et al.* 1991). This type of ritual traumatism is completely missing within the sample of Can Reiners.

Regarding other pathologies, we observed sporadic cases of infections (0.5%), two cases of benign tumours located in the same individual, rare examples of eburnations (1.85%), some cases of congenital anomalies (4.6%). Aside from a case of bizarre sternal congenital anomaly, in S'Illot des Porros two possible cases of congenital anomalies were found: one only case of *spina bifida* (Malgosa and Campillo 1991) and a case of dysplasia maybe related with mucopolysaccharidosis (Campillo and Malgosa, 1991). It is interesting to mention a possible case of dwarfism from the sepulchral cave of Bronze Age of Can Martorellet (Pollença, Mallorca; Gómez, 2002).

Aside of some few rare anomalies, in general we observed pathologies that commonly affected an ancient community as osteoarthritis and oral pathologies (Ortega *et al.* 2003). However, we did not found cases of degenerative diseases compatible with an advanced age at death.

In addition, it is important to note the scarcity of systematic studies of pathologies in the ancient populations of the Balearic Islands. In this sense, the study of Can Reiners sample is the only contribution to the knowledge of health of population of Mallorca between the Late Antiquity and the medieval period from a holistic point of view.

#### **4.1.4 Conclusions**

The global index of preservation of the skeletal remains of the necropolis of Can Reiners is 72.7% and superior and inferior limbs are better preserved than other part of the skeleton. The skeletal remains of males are better preserved than those of females. Regarding the age class and sex distribution among the grave types no statistically differences were observed. Results of the demographic analysis of Can Reiners showed a high mortality and low life expectancy, which conforms to the expected values for ancient populations such as this one. In addition, the pathologies, that could be observed in the skeletal remains, were not frequent as expected from a population with a high crude mortality rate. Therefore, we may speculate regarding the presence of pathologies that affected other parts of the body, such as the gastro-intestinal diseases, or respiratory diseases and others that affected soft tissues. Regarding the estimation of height we observed significant differences between the four methodologies employed in this study and the values of stature changed among these methods. We calculated that the mean of the stature of males range from 165.2 to 168.5, and for females range from 152.5 to 156.6.





## **4.2 Dental occlusion variation in the population of Can Reiners**

### **4.2.1 Introduction**

Dental occlusion is the relationship between both dental arches where the maximum number of contacts between upper and lower teeth is achieved. A correct dental occlusion “refers to how well the teeth are arranged individually and one-to-another within and between the dental arches” (Harris and Corruccini 2008). Conversely, malocclusion is considered a discrepancy between teeth that can modify the maxillary shape, sometimes affecting the mastication. A good occlusion is fundamental for a correct mastication, diet and health. It is relevant to the acquisition of food and has changed during evolution, in relation to the adjustment to the availability of food.

Although the importance of the study of dental occlusion, it is not frequently included in anthropological maxillary analyses due to often fragmented and incomplete skeletal remains, and in particular, due to the lack of a suitable methodology for the study of such ancient remains. The study of occlusion is generally conducted only when jaws are occluded with condyles placed in their fossae and taking patterns of attrition as a reference of dental position. However, complete skulls from ancient context are unusual; in the most habitual scenario, samples suitable for dental occlusion analysis are just a small percentage of the total number of skeletons available. This relates to the vulnerability of bones and the importance of a proper recovering and management of samples, from excavation to laboratory. In several cases, taphonomic process, which is what happens to the body between death and its recovery (Duday 2006), is liable for fragmentation and lack of archaeological skeletal material.

Therefore, a methodology based on clinical dental practice suitable for fragmented skeletal remains was elaborated (Fiorin *et al.* 2014, appendix:

document 1). The present work is based on the analysis of the “static” occlusion, which means studying jaws with maximum dental contact and maximum intercuspitation. A lab sheet has been developed including dental and maxillary parameters that would be recommended to evaluate occlusion and its anthropological interest. Different biological traits, such as age, sex and preservation status have also been considered.

The purpose of this study is to define the occlusal characteristics of the population of Can Reiners in order to study the factors that could lead to malocclusion as well as other diagnostic features for the interpretation of health and lifestyle in an anthropological context. The analysis will be conducted by applying the specific methodology on ancient fragmentary material and elaborated in the frame of this project (Fiorin *et al.* 2014).

#### **4.2.2 Material and methods**

The material analyzed in this study belongs to the necropolis of Can Reiners (ca. 600 AD, Mallorca, Spain). The skulls were frequently incomplete and fragmented, and thus the material employed for occlusion analysis was not numerous (33 of 226 individuals, 14.6%). The samples were chosen according to the state of preservation of jaws (fig. 4.2.1 A). In some cases, jaws and mandibles were well preserved whereas in other cases only one fragment or side of the dental arch was present. Mandibular condyles, for example, were preserved in only 13 individuals. The sample included the following individuals: Cr 11.1, Cr 13.1, Cr 15.1, Cr 15.2, Cr 17.1, Cr 23.3, Cr 30.1, Cr 32.1, Cr 39.1, Cr 41.37, Cr 43.1, Cr 46.1, Cr 47.57, Cr 49.1, Cr 67.1, Cr 70.1, Cr 77.1, Cr 89.1, Cr 96.1, Cr 107.1, Cr 119.1, Cr 123.1, Cr 124.1, Cr 129.1, Cr 132.1, Cr 141.1, Cr 144.1, Cr 145.1, Cr 147.1, Cr 160.1, Cr 169.1, Cr 191.1, Cr 203.1.

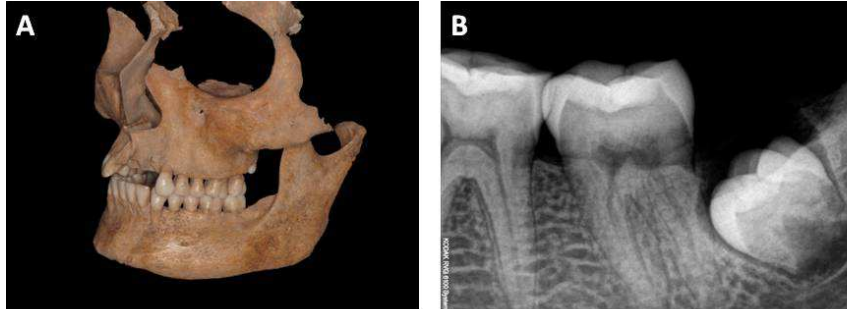


Figure 4.2.1: a well-preserved pair of jaws (Cr 191.1) in “static” occlusion (A), a x-ray which shows formation and eruption of the third molar (tooth 38, Cr 191.1) (B)

Regarding data collection, it is possible to gather information from five main sections: anthropological information, dental features such as dental pathology, tooth wear, diastema or dental crowding, dental arches features such as morphology, symmetry and measurements, occlusal features such as overbite, openbite, overjet, molar and canine relationship, and articulation characteristics such as temporo-mandibular joint pathology (Fiorin *et al.* 2014). Not all characteristics could be recorded for all individuals, and therefore the total number of individuals analyzed was different in each analysis.

Sex determination was conducted using the skeletal parts that express the most pronounced sexual dimorphism (Acsádi and Nemeskéri 1970, Buikstra and Ubelaker 1994, Ferembach *et al.* 1979, Krogman and Iscan 1986). The individuals were classified as male, female and undetermined. The undetermined individuals are those that do not conserve the necessary elements for sex diagnosis. Age at death categories were defined according to Vallois (1960) and they were divided into six different groups: infant I (0-6 years old), infant II (7-12 years old), juvenile (13-20 years old), adult (21-40 years old), mature (41-59 years old), senile (more than 60 years old). To describe maxillary and dental characteristics, morphological analysis was conducted. Nevertheless, to assess some dental features such as the agenesis, radiographic examinations of the jaws were done (fig. 4.2.1 B).

To achieve a consensus in this analysis, intra- and inter-observer differences were previously calibrated. Four examiners analyzed the sample and the measurements were repeated in all the jaws two times. All statistical analyses were performed with SPSS 15.0 for Windows (SPSS Inc. 2006). For metrical and ordinal parameters, differences among groups were evaluated with Mann-Whitney's test and Kruskal-Wallis's test, as some of them were not normally distributed (Shapiro-Wilk:  $P < 0.05$ ). Bivariate correlations for metric and ordinal variables were assessed using the Spearman's rank correlation coefficient. Association among nominal variables and between nominal and ordinal variables was assessed using Fisher's exact test. As female sample size is low, differences between sexes could not be properly assessed. Therefore, both genders are pooled together in the analysis of data.

In order to compare maxillary occlusion of Can Reiners, specific literature was searched. Papers containing the variables analyzed in ancient remains are rare because of the methodological difficulties previously explained. When data were available, some frequencies and metric data were compared with modern and ancient populations.

### **4.2.3 Results**

#### **Anthropological information**

Regarding sex of the 33 individuals examined, 66.6% were male, 21.2% were female and the remaining 12.2 % were considered undetermined. The sample is composed by 6.1% of infant II (7-12 years old), 21.2% of juvenile (13-20 years old), 60.6% of adult (21-40 years old) and the remaining 12.1% of mature (41-59 years old) (table 4.2.1).

Table 4.2.1: sex and age at death of the individuals of Can Reiners site employed for dental occlusion analysis

<b>Sex</b>	<b>Infant II</b>	<b>Juvenile</b>	<b>Adult</b>	<b>Mature</b>	<b>Total</b>
<b>Male</b>	0	3	15	4	22
<b>Female</b>	0	2	5	0	7
<b>Undetermined</b>	2	2	0	0	4
<b>Total</b>	2	7	20	4	33

### **Dental features**

Among the jaws of the 33 individuals, 93.5% of alveolar cavities and 75% of teeth were preserved. *Ante mortem* tooth loss was found in 2.3% of the alveolar cavities and they were present in adult and mature individuals of both sex. Teeth more involved were the maxillary molars (31.8%) and premolars (40.9%). Dental decay was present in 5.2% of teeth and involved almost exclusively molars and premolars (97.5%). Agenesis affected seven individuals (21.2%) and rotation and dental movements involved 1.3% of the total teeth. Usually medial movements of tooth have a relationship with an *ante mortem* tooth loss.

**Spacing characteristics:** conditions of space were evaluated by the presence of a diastema or dental crowding. Central diastema, which is the inter-incisal space in the upper arch midline, was present in six individuals (27.3%). The values ranged from a minimum of 3 mm to a maximum of 6 mm. Dental crowding was present in only four individuals (12.1%) and involved the inferior central teeth. Low values of 2-3 mm were not considered for this study.

**Tooth wear:** it was evaluated according to Brothwell (1981) and Lovejoy (1985) taking into account the position of teeth in occlusion. Dental arch was divided in sextants in order to perform an accurate analysis of central and lateral dental wear. To assess if there were differences in gradual and metrical variables among groups of age at death, Kruskal-Wallis test was used. Results of the test showed that there were differences ( $P < 0.05$ ) among groups for tooth wear of upper and lower jaws. To know exactly between which groups

differences occur, Mann-Whitney test with Bonferroni correction was used. We found that there were differences between juvenile and adult individuals for tooth wear of the superior right dental arch and tooth wear of the inferior right dental arch ( $P < 0.017$ ). For all of them, the adult individuals presented higher values (table 4.2.2). Between juvenile and mature groups there were differences for tooth wear of the upper molars and premolars and for tooth wear of lower right molars and premolars ( $P < 0.017$ ). All of the values were higher in the mature group as expected (table 4.2.2). Between adults and matures there were no significant differences. The association between age at death and others variables was not significant (Fisher's exact test:  $P > 0.05$ ). In Can Reiners sample, tooth wear under 20 years old was absent or had only some points of enamel wear. Tooth wear increases with age at death, but this was not constant in Can Reiners individuals because there were also mature individuals with a low tooth wear.

Table 4.2.2: mean, standard deviation and median of the tooth wear divided for sextants and age group

<b>Sextant</b>	<b>Age group</b>	<b>Mean</b>	<b>SD</b>	<b>Median</b>
<b>Superior 1</b>	Juvenile	0.29	0.76	0.00
	Adult	1.70	1.08	1.50
	Mature	2.50	1.00	2.00
<b>Superior 2</b>	Juvenile	0.43	0.79	0.00
	Adult	2.15	1.50	2.00
	Mature	2.75	1.50	3.00
<b>Superior 3</b>	Juvenile	0.43	0.53	0.00
	Adult	1.45	1.15	1.50
	Mature	2.50	1.00	2.00
<b>Inferior 1</b>	Juvenile	0.57	0.78	0.00
	Adult	1.90	0.78	1.50
	Mature	2.50	1.00	2.00
<b>Inferior 2</b>	Juvenile	0.43	0.78	0.00
	Adult	1.90	1.02	2.00
	Mature	2.50	1.30	2.50
<b>Inferior 3</b>	Juvenile	0.71	0.95	0.00
	Adult	1.80	0.95	2.00
	Mature	2.50	1.00	2.00

Legend: sextant superior 1= superior right molars and premolars, superior 2= superior canines and incisors, superior 3= superior left molars and premolars, inferior 1= inferior right molars and premolars, inferior 2= inferior canines and incisors, inferior 3= inferior left molars and premolars

## Dental arches features

Shape of dental arch was evaluated in 23 maxillae and in 33 mandibles. The values for the mandibles were: 72.7% parabolic in shape, 12.1% oval-shaped and 15.2% square-shaped. In the maxillae 69.6% were parabolic in shape, 17.4% oval-shaped and 13% square-shaped. Generally jaws were more or less symmetrical, but there were three individuals with clear asymmetry: the adult male Cr 49.1 showed asymmetry in the left superior dental arch, the adult male Cr 160.1 had a left inferior dental asymmetrical arch, and the adult male Cr 32.1 exhibited an asymmetry in the right superior dental arch. The maxilla asymmetry of individual Cr 49.1 could be caused by an ancient trauma that also affected dental occlusion.

Tables 4.2.3 and 4.2.4 show minimum, maximum and average values of the sagittal (fig. 4.2.2 A, B, C and D) and transversal measurements (fig. 4.2.2 E and F).

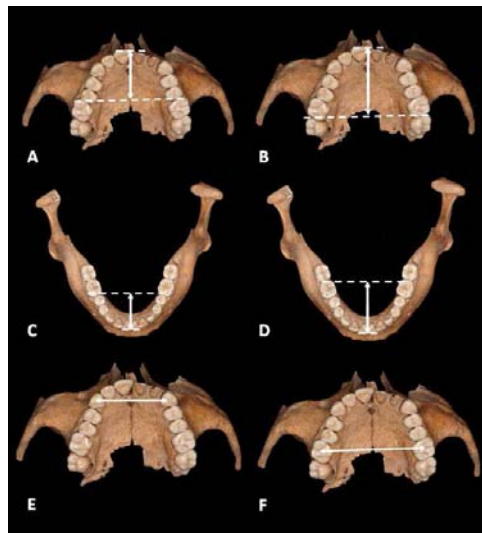


Figure 4.2.2: sagittal measurements: maxillary depth (A), and length (B); mandible depth (C) and length (D); transversal measurements: intercanine (E) and intermolar (F) distance

The lowest value of upper sagittal depth measurement corresponded to a juvenile female (Cr 191.1) and the highest value belonged to an undetermined infant II (Cr 141.1). While the lowest value of lower sagittal depth measurement corresponded to an adult male (Cr 77.1), the highest values belonged to an adult female (Cr 67.1) and a juvenile male (Cr 23.3). The lowest value of upper sagittal length measurement corresponded to an adult male (Cr 145.1) and the highest values belonged to an adult female (Cr 67.1) and an adult male (Cr 19.1). The lowest value of lower sagittal length measurement corresponded to an undetermined infant II (Cr 141.1) and the highest value belonged to an adult female (Cr 67.1).

Table 4.2.3: sagittal measurements

	<b>N</b>	<b>Mean</b>	<b>Median</b>	<b>SD</b>	<b>Min</b>	<b>Max</b>
<b>Upper depth</b>	19	25.4	25	2.43	22	31
<b>Lower depth</b>	33	19.6	20	2.88	13	24
<b>Upper length</b>	18	35.7	36.5	2.84	32	41
<b>Lower length</b>	33	29.2	30	3.46	22	34

The lowest value of upper transversal intercanine measurement corresponded to an adult female (Cr 67.1) and the highest value to an undetermined infant II (Cr 141.1). While lowest values of lower transversal intercanine measurement belonged to an adult male (Cr 77.1) and an adult female (Cr 67.1) and highest values corresponded to an adult male (Cr 132.1), an adult female (Cr 30-1) and an undetermined infant II (Cr 141.1). The lowest value of upper transversal intermolar measurement belonged to an undetermined juvenile (Cr 89.1) and the highest value corresponded to an adult male (Cr 43.1). The lowest values of lower transversal intermolar measurement corresponded to an undetermined infant II (Cr 11.1) and an adult female (Cr 30.1), while the highest value belonged to two adult male (Cr 147.1 and Cr 49.1).



Table 4.2.4: transversal measurements

	<b>N</b>	<b>Mean</b>	<b>Median</b>	<b>SD</b>	<b>Min</b>	<b>Max</b>
<b>Upper intercanine</b>	19	32	31	3.12	27	38
<b>Lower intercanine</b>	32	25.3	25	2.51	21	31
<b>Upper intermolar</b>	17	47.3	48	3.88	39	54
<b>Lower intermolar</b>	33	45.9	46	2.62	38	50

Table 4.2.5 shows a comparison of the sagittal and transversal measurements among the present study and other ancient and modern populations. The series of adult individuals for comparison were: a sample of individuals (N= 43) from the archaeological site of Roaix, located in the south of France, from the Copper Age (Mockers *et al.* 2004); a modern population (N= 27) of Northern European origin (Harper 1994); a modern population (312 individuals) and soldiers (N= 35) who died during a epidemic of typhus in 1810 from west Sweden (Ingervall *et al.*1972) and a modern population (N= 100) from Pakistan (Amin *et al.* 2012). The results showed that Can Reiners series have a smaller intercanine width than modern Pakistani population (Amin *et al.* 2012). The lower intercanine width of our sample is slightly greater than the prehistoric population of Roaix (Mockers *et al.* 2004), although this differences is not statistically significant, and similar to that of the modern European sample (Harper 1994). In relation to intermolar width, Can Reiners sample have smaller average than the two Swedish samples (Ingervall *et al.*1972), although differences are only significant for lower intermolar width, and higher than the Pakistani population. Regarding upper and lower length, our sample has higher values than the other samples, except for lower length of the Swedish modern sample, which is similar. This last data are interesting because generally the decrease of the jaws size is related to insurgence of malocclusion and the values of the length of Can Reiners samples are higher than the others.

Table 4.2.5: mean and standard deviation (SD) for transversal and sagittal measurements of Can Reiners compared with ancient and modern population

	<b>Ancient population (before XX century)</b>				<b>Modern population (After XX century)</b>							
	Can Reiners		(Mockers <i>et al.</i> 2004)		(Ingervall <i>et al.</i> 1972)		(Ingervall <i>et al.</i> 1972)		(Amin <i>et al.</i> 2012)		(Harper 1994)	
	Mean	SD	Mean	SD	Mean	SD	Mean	SD	Mean	SD	Mean	SD
<b>UIC</b>	32	3.1							34.6*	2.6		
<b>LIC</b>	25.3	2.5	24.6	1.9					26.7*	2.5	25.4	1.9
<b>UIM</b>	47.3	3.9			48.9	3.0	47.4	3.4	44.6*	2.8		
<b>LIM</b>	45.9	2.6			50.8*	3.1	48.8*	2.5	41.2*	3.8		
<b>UL</b>	35.7	2.8			29.1*	2.8	32.4*	2.8	27.8*	3.9		
<b>LL</b>	29.2	3.5			26*	1.8	29.9	2.8	23.1*	2.5		

Legend: UIC= upper intercanine, LIC= lower intercanine, UIM= upper intermolar, LIM= lower intermolar, UL= upper length, LL= lower length. Asterisks indicate statistically significant differences with Can Reiners (Student's t-test: p<0.05)

## Occlusal features

Tables 4.2.6 and 4.2.7 show the main characteristics of the occlusion of individuals of Can Reiners. For sake of clarity each one was analyzed separately.

Table 4.2.6: Main dental occlusal characteristics of Can Reiners ancient population. The number of observed cases (N) varies according the state of preservation of the maxillary remains

	Overbite	Open bite	Overjet	Anterior cross bite	Midline displacement	Posterior open bite		Lingual posterior cross bite		Lingual posterior cross bite	
						R	L	R	L	R	L
<b>N</b>	20	20	18	22	25	27	30	27	31	28	30
<b>Present</b>	1	3	4	2	0	0	0	2	2	0	1
<b>Absent</b>	19	17	14	20	25	27	30	25	29	28	29

Legend: R= right, L= left

**Overbite:** it is the vertical covering of lower incisors by upper incisors, which is considered normal when covering is about 1/3. In this sample, 39.4% was not

recordable and 95 % was considered normal. Only in one case, an undetermined juvenile (Cr 89.1), had more than 2/3 of dental crown of lower teeth covered (9 mm).

**Open bite:** it is the occlusion characterized by the presence of a space between upper and lower anterior teeth incisal edges when jaws are closed. The analysis showed that open bite was not recordable for 39.4%, absent in 85% and present in 15% of the individuals. Open bite was present in a juvenile female (Cr 15.2), in an adult male (Cr 145.1) and in a juvenile male (Cr 16.1). The space evaluated was about 3 mm in all of them.

**Overjet:** it quantifies the distance between lower incisor buccal surface and upper incisor incisal edge evaluated parallel to the occlusal plane considering the starting point of the measurement at the superior incisor. Up to 4 mm is considered as a normal value, while higher values are considered malocclusion. In Can Reiners sample, 45.5% of individuals were not recordable, 77.8% of values were up to 4 mm, and 22.2% of values were between 4 and 8 mm. The juvenile male Cr 16.1 and the adult male Cr 19.1 had an overjet of 5 mm, the adult male Cr 77.1 had an overjet of 6 mm and the undetermined juvenile Cr 89.1 of 7 mm.

**Anterior cross bite:** it is a malocclusion caused by a mandible or maxillary bone sagittal displacement. Anterior cross bite was not recordable in 33.3%, was absent in 90.1% and was present (it affects just one tooth) in 9.1% of the individuals (two adult males: Cr 43.1 and Cr 17.1).

**Midline displacement:** it can be observed by the asymmetry between the dental arches in the incisor area. In these samples, the midline displacement was not recordable in the 24.2% of the individuals, and it was absent in 100%.

**Posterior open bite:** it is the presence of a space between upper and lower posterior teeth when jaws are closed. It was not recordable in 18.2% of

posterior right side of the arch and in 9.1% of posterior left side. It was absent in the remaining samples.

**Lingual posterior cross bite:** it is caused by a transversal malocclusion, where upper molar and premolar buccal cuspids occlude in lower molar and premolar pits. Lingual posterior cross bite was not recordable in 18.1% of the right and in 9.1% of the left posterior dental arch. On the right side it was absent in 92.6% of samples and present in 7.4% (Cr 17.1 and Cr 145.1, which were an adult mature male and an adult male respectively). On left side, lingual posterior cross bite was absent in 93.5% and it was present in 6.5% (Cr 49.1 and Cr 145.1, which were an adult mature male and an adult male respectively).

**Buccal posterior cross bite:** it occurs when upper molar and premolar palatine cuspids contact with lower molar and premolar buccal cuspids. It was present only in one case, in the left posterior dental arch of an adult female (Cr 47.57). In the right dental arch it was not observable in 15.1% and it was absent in 100%. In the left dental arch it was absent in 96.7% and present in one case (3.3%).

**Canine relationship:** it is the relation between upper canine cuspid and lower canine and first premolar cuspids. Angle's classification (1899) established that Class I is considered normocclusion (the upper canine cuspid is located between the lower canine and the lower first premolar, when dental arches are occluding), while the other two (Class II and III) are considered as anterior or posterior movement of the mandible. 24.4% of samples were not recordable for both dental arch sides, 42.4% were not recordable for right side and 27.3% were not recordable for left side. Regarding the right dental arch, 73.7% were considerate Class I, 10.5% were considered Class II and 15.8% were considered Class III. In the left dental arch, 66.6% were considerate Class I, 20.8% were considered Class II and 12.5% were considered Class III.

Table 4.2.7: distribution of Angle Class occlusal characteristics of Can Reiners ancient population. The number of observed cases (N) varies according to the state of preservation of the maxillary remains

	Canine relationship		Molar relationship	
	Right	Left	Right	Left
<b>N</b>	19	24	25	29
<b>Class I</b>	14	16	19	20
<b>Class II</b>	2	5	1	6
<b>Class III</b>	3	3	5	3

**Molar relationship:** like canine relationship, molar relationship is classified by Angle in Class I, Class II and Class III. Class I, or normocclusion, is present when the upper first molar mesiovestibular cuspid occludes in the lower first molar buccal sulcus. Class II is observed when maxillary arch is displaced forward in relation to the mandible, while Class III is present when maxillary bone is displaced backwards. The 6.1% of the sample was not recordable for both dental arch sides, the 24.2% were not recordable for right side and the 12.1% were not recordable for left side. The right dental arch showed Class I molar in 76% of the arches, Class II in 4% and Class III in 20%. On the left side, Class I was present in 69% of the arches, Class II in 20.7% and Class III in 10.3%.

Table 4.2.8 shows a comparison among molar relationship of Can Reiners and other populations. The samples used were: individuals (N= 58) that came from 5 archaeological sites (from 8th to 17th c. AD) and a present-day group living population (N= 82) from the Southeast France (Guichard *et al.* 2001), 52 Gallic and Gallo-Roman skulls from north France (Benauwt 1974) and a modern group of 137 Swedes (Seipel 1946). Comparing these groups, it is clearly visible that the “normal” trend is Class I occlusion. The highest values are those of Can Reiners and of the modern Swedes. Class III has higher percentages in ancient sample than in the modern Sweden group, whereas it shows similar values with the present-day sample. Comparing Class II, the value of Can Reiners (left dental arch) is higher than the medieval and Gallo Roman samples and similar to the modern Sweden group. The interesting result is that usually in

modern populations the values of Class II are higher than those of Class III, whereas in ancient samples the opposite occurs. In Can Reiners, these percentages are different. If we observe the right dental arch for Class III (N= 5) and the left dental arch for Class II (N= 6), the two values are rather similar.

Table 4.2.8: distribution of molar relationship (Angle classification) in Can Reiners and other populations

Population	Class I		Class II		Class III		Total	
	N	%	N	%	N	%	N	%
Can Reiners right dental arch	19	76	1	4	5	20	25	100
Can Reiners left dental arch	20	69	6	20.7	3	10.3	29	100
Galic/Gallo Roman (Benauwt 1974)	30	57	7	13.4	15	28.8	52	100
Medieval (Guichard <i>et al.</i> 2001)	25	43.1	11	18.9	22	38	58	100
Present-day (Guichard <i>et al.</i> 2001)	37	45	28	34	17	21	82	100
Modern Swedes (Seipel 1946)	102	74.15	27	20.10	8	5.75	137	100

## Occlusal plane

**Spee curve:** it is a line defined by occlusal surfaces of the teeth of the mandibular hemiarch, joining anterior teeth incisal edges with posterior teeth buccal cuspids (fig. 4.2.3 A). Spee curve was normal in 88%, flat in 3%, and inverted in 9% of the mandibles.

**Wilson curve:** it is the transversal curvature of the occlusal plane in frontal view (fig. 4.2.3 B). In Can Reiners samples, it was normal in 9.4% of the mandibles, flat in 40.6%, and inverted in 50%.

## Articulation

In this section, pathologies affecting the temporo-mandibular joint (TMJ) were analyzed. The mandibular condyles were absent in many individuals (42.4% on right side, 33.3% on left side). Only two individuals exhibited signs of pathologies. The left condyle of an adult male (Cr 147.1) and the right of another adult male (Cr 123.1) showed osteoarthritis on the surface.

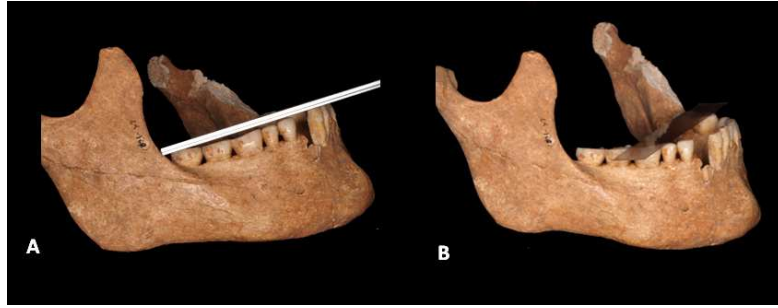


Figure 4.2.3: Spee curve (A) and Wilson curve (B)

### Statistical correlations

Correlation between variables (metrical and gradual) was determined using Spearman's correlation coefficient. A strong correlation was observed among the tooth wear of all teeth (N=33;  $R^2$  ranging between 0.5 and 0.87;  $P<0.05$ ). Another strong correlation was observed between superior intercanine width and inferior intercanine width (N=19;  $R^2=0.75$ ;  $P<0.05$ ) and inferior intermolar width (N=17;  $R^2=0.61$ ;  $P<0.05$ ).

Association between nominal and other variables using Fisher's exact test ( $P<0.05$ ) was observed between the Spee and Wilson curves and dental wear, between molar relationship and dental wear and between molar and canine relationship. A general trend is recognized observing the frequency distribution in the contingency table for the variables Spee curve and dental wear: the increase of dental wear is linked to the inversion of the Spee curve. When dental wear is not heavy (from grade 0 to 3), Spee curve tends to be normal. This interrelation is observed between Spee curve and dental wear of the inferior canines and incisors (table 4.2.9) and inferior left molars and premolars (table 4.2.10), and between Spee curve and dental wear of superior right molars and premolars (table 4.2.11) and superior left molars and premolars (table 4.2.12) sextant. Fisher's exact test also showed an association ( $P<0.05$ ) between Wilson curve and dental wear of the inferior left molars and premolars wear.

**Table 4.2.9:** Association between Spee curve and dental wear of the inferior canines and incisors (Isext2)

			Curve of Spee			Total
			Normal	Flat	Inverted	
<b>Isext2</b>	0	N	7	1	0	8
		% Isext2	87.5%	12.5%	.0%	100.0%
	1	N	7	0	1	8
		% Isext2	87.5%	.0%	12.5%	100.0%
	2	N	11	0	0	11
		% Isext2	100.0%	.0%	.0%	100.0%
	3	N	3	0	0	3
		% Isext2	100.0%	.0%	.0%	100.0%
	4	N	1	0	2	2
		% Isext2	33.3%	.0%	66.7%	100.0%
<b>Total</b>		N	29	1	3	33
		% Isext2	87.9%	3.0%	9.1%	100.0%

Legend: Isext2, dental wear: 0= absence, 1= point of enamel wear, 2= islands of visible dentine, 3= confluence of islands, 4= absence of enamel

Table 4.2.10: Association between Spee curve and dental wear of the inferior left molars and premolars (Isext3)

			Curve of Spee			Total
			Normal	Flat	Inverted	
<b>Isext3</b>	0	N	7	1	0	8
		% Isext3	87.5%	.0%	.0%	100.0%
	1	N	5	0	0	5
		% Isext3	100.0%	.0%	.0%	100.0%
	2	N	15	0	1	16
		% Isext3	93.8%	.0%	6.3%	100.0%
	3	N	2	0	0	2
		% Isext3	100.0%	.0%	.0%	100.0%
	4	N	0	0	2	2
		% Isext3	.0%	.0%	100.0%	100.0%
<b>Total</b>		N	29	1	3	33
		% Isext3	87.9%	3.0%	9.1%	100.0%

Legend: Isext3, dental wear: 0= absence, 1= point of enamel wear, 2= islands of visible dentine, 3= confluence of islands, 4= absence of enamel



Table 4.2.11: Association between Spee curve and dental wear of the superior right molars and premolars

			Curve of Spee			Total
			Normal	Flat	Inverted	
<b>Ssxt1</b>	0	N	9	1	0	10
		% Ssxt1	90.0%	10.0%	.0%	100.0%
	1	N	7	0	1	8
		% Ssxt1	87.5%	.0%	12.5%	100.0%
	2	N	9	0	0	9
		% Ssxt1	100.0%	.0%	.0%	100.0%
	3	N	4	0	0	4
		% Ssxt1	100.0%	.0%	.0%	100.0%
	4	N	0	0	2	2
		% Ssxt1	.0%	.0%	100.0%	100.0%
<b>Total</b>		N	29	1	3	33
		% Ssxt1	87.9%	3.0%	9.1%	100.0%

Legend: Ssxt1, dental wear: 0= absence, 1= point of enamel wear, 2= islands of visible dentine, 3= confluence of islands, 4= absence of enamel

Table 4.2.12: Association between Spee curve and dental wear of the superior left molars and premolars (Ssxt3)

			Curve of Spee			Total
			Normal	Flat	Inverted	
<b>Ssxt3</b>	0	N	10	1	0	11
		% Ssxt3	90.9%	9.1%	.0%	100.0%
	1	N	8	0	0	8
		% Ssxt3	100.0%	.0%	.0%	100.0%
	2	N	9	0	1	10
		% Ssxt3	90.0%	.0%	10.0%	100.0%
	3	N	2	0	0	2
		% Ssxt3	100.0%	.0%	.0%	100.0%
	4	N	0	0	2	2
		% Ssxt3	.0%	.0%	100.0%	100.0%
<b>Total</b>		N	29	1	3	33
		% Ssxt3	87.9%	3.0%	9.1%	100.0%

Legend: Ssxt3, dental wear: 0= absence, 1= point of enamel wear, 2= islands of visible dentine, 3= confluence of islands, 4= absence of enamel

As well as the Spee curve, the right molar relationship has a statistically significant association (Fisher's exact test:  $P < 0.05$ ) with inferior left molars and premolars dental wear. Contingency table shows that Class I or normocclusion (RAMD = 0) is present until dental wear grade 2. When dental wear has a value of 4, a Class III (RAMD = 3) is present (table 4.2.13). The last two Fisher's

exact tests with statistical significance ( $P < 0.05$ ) show an association between right molar and right canine relationship and between left molar and left canine relationship (tables 4.2.14 and 4.2.15).

Table 4.2.13: Association between right molar relationship and inferior left molars and premolars dental wear (Isext3)

		Right molar relationship				Total
		0	1	3	4	
<b>Isext3</b> 0	N	4	1	1	1	7
	% Isext3	57.1%	14.3 %	14.3 %	14.3%	100.0%
1	N	3	1	1	0	5
	% Isext3	60.0%	20.0%	20.0%	.0%	100.0%
2	N	11	0	0	0	11
	% Isext3	100.0%	.0%	.0%	.0%	100.0%
4	N	0	0	0	2	2
	% Isext3	.0%	.0%	.0%	100.0%	100.0%
<b>Total</b>	N	18	2	2	3	25
	% Isext3	72.0%	8.0%	8.0%	12.0%	100.0%

Legend: Right molar relationship: 0= Class I, 1= incomplete Class II, 2= complete Class II, 3= incomplete Class III, 4= complete Class III. Isext3: tooth wear: 0= absence, 1= point of enamel wear, 2= islands of visible dentine, 3= confluence of islands, 4= absence of enamel

Table 4.2.14: Association between right canine relationship and right molar relationship

		Right canine relationship			Total
		0	1	2	
<b>Right molar relationship</b> 0	N	12	0	1	13
	% RMR	92.3%	.0%	7.7%	100.0%
3	N	0	1	0	1
	% RMR	.0%	100.0%	.0%	100.0%
4	N	0	0	2	2
	% RMR	.0%	.0%	100.0%	100.0%
<b>Total</b>	N	12	1	3	16
	% RMR	75.0%	6.3%	9.1%	100.0%

Legend: Right canine relationship: 0= Class I, 1= Class II, 3= Class III. Right molar relationship (RMR): 0= Class I, 1= incomplete Class II, 2= complete Class II, 3= incomplete Class III, 4= complete Class III

Table 4.2.15: Association between left canine relationship and left molar relationship

		Left canine relationship			Total	
		0	1	2		
<b>Left molar relationship</b>	0	N	13	1	1	15
		% LMR	86.7%	6.7%	6.7%	100.0%
	1	N	1	2	0	3
		% LMR	33.3%	66.7%	.0%	100.0%
	2	N	1	2	0	3
		% LMR	33.3%	66.7%	.0%	100.0%
<b>Total</b>	4	N	0	0	1	1
		% LMR	.0%	.0%	100.0%	100.0%
		N	15	5	2	22
		% LMR	68.2%	22.7%	9.1%	100.0%

Legend: Left canine relationship: 0= Class I, 1= Class II, 3= Class III. Left molar relationship (LMR): 0= Class I, 1= incomplete Class II, 2= complete Class II, 3= incomplete Class III, 4= complete Class III

#### 4.2.4 Discussion

The analysis of Can Reiners population was performed in 33 individuals with permanent dentition. The 66.6% of the sample was composed by males, 21.2 % by females and 12.2% by undetermined skeletons. Statistical comparisons between sexes were not conducted due to the low number of female skeletons. The groups of age used for this study were infant II, juvenile, adult and mature; the age at death ranges from 12 to 51 years old. Pathologies such as dental decay and tooth loss were not frequent in this sample and affected only adults and mature individuals, the involved teeth were usually premolars and molars. The analysis of oral pathologies of the population of Can Reiners (Ortega *et al.* 2003) showed relatively high values for the frequency of dental decay and tooth loss. This fact is related to the subsample used in this analysis composed by maxillaries with more or less complete dental arch suitable for occlusion examination.

Statistical analysis showed that dental wear increases with age at death, but no statistical differences between dental wear among adults and matures groups were observed. Differences among age groups in spacing characteristics, dental arches features and TMJ pathologies variables were not statistically significant.

Nevertheless, the results may be influenced by the low number of mature individuals.

Sagittal and transversal measurements are important for dentition development and diagnosis and treatment planning, which are closely related factors. For instance, the intermaxillary intercanine relation is an influential factor in the development of a posterior crossbite (Lindsen *et al.* 2002). In a recent study carried out in Pakistani patients without orthodontic treatment (Amin *et al.* 2012), data showed that increase of intercanine width was associated to an increase of intermolar width (between upper intercanine and intermolar:  $R^2=0.35$ ; and between lower intercanine and intermolar:  $R^2=0.33$ ). A weak correlation was observed between upper intercanine and upper arch length, as well as lower arch length ( $R^2=0.01$  and  $R^2=0.12$ ), and a high correlation was observed between upper arch length and lower arch length ( $R^2=0.61$ ).

In Can Reiners sample, the superior intercanine width was correlated with the inferior intercanine width and the inferior intermolar width. These correlations are stronger than those found by Amin (2012). Similarly to what Amin (2012) observed, the increase of superior length values is linked to an increase of inferior length values. Furthermore, the data of Can Reiners showed that an increase of inferior and superior depth was related to an increase of inferior and superior length respectively. Finally the decrease of the superior length is associated to an increase of dental wear of upper canines, incisors and left molars and premolars and lower canines and incisors, and the decrease of the inferior intermolar width is related to a decrease of dental wear of the superior canines and incisors and lower molars and premolars.

Usually transversal and sagittal measurements are dependent on each other (Lindsten *et al.* 2012), whereas in Can Reiners no correlation was observed between the arch width and the arch depth measurements. Comparing our measurements with other samples, sagittal length were generally higher than other samples.

The curve of Spee has a biomechanical function during food processing, because it increases the crush-shear ratio between the posterior teeth and the efficiency of occlusal forces during mastication. Even though leveling of the Spee curve is an everyday occurrence in orthodontic practice, few research has been done to assess the relationship of the Spee curve and multiple factors that could influence its depth. Batham *et al.* (2013) observed that the depth of Curve of Spee was greatly influenced by sagittal maxillomandibular discrepancies, overbite, overjet and the inclination of mandibular first molar. Baydas *et al.* (2004) observed statistically significant correlation between the depth of curve of Spee and overjet and overbite. By contrast, the sample of Can Reiners does not show correlation among these variables maybe due to sample size. Few studies are devoted to the Spee Curve analysis in ancient remains. Sengupta *et al.* (1999) examined the effect of dental wear on the curve of Spee in an archaeological population with a high rate of dental wear. Their data suggested that there is no straightforward relation between the Spee curve and occlusal wear. Conversely, the Can Reiners findings show a significant association between Spee curve and dental wear, involving in particular the upper premolars and molars, and also the teeth of the right mandibular dental arch. This association is not present in the modern sample due to the absence, even in the mature individuals, of a heavy dental wear.

Statistically significant correlation was observed between canine and molar relationship (Angle's classification) and between molar relationship and inferior molars and premolars dental wear. Dental wear could be the influencing factor of the onset of molar malocclusion (Class III) into Can Reiners sample.

The general trend indicates that prevalence of malocclusion in modern population is higher than in the samples from antiquity (Evensen and Øgaard 2007, Weiland *et al.* 1997). Actually, our sample reflects this tendency especially in variables such as spacing characteristics and some occlusal features such as overjet, overbite and openbite. Concerning molar relationship (Angle classification), this tendency is not so simplistic. Generally,

normocclusion is the normal trend (Benauwt 1974, Guichard *et al.* 2001, Seipel 1946) for all historical periods, and in modern populations the values of Class II increase at the expense of Class III. By contrast, in our population these percentages are rather similar (right dental arch for Class III and left dental arch for Class II) and the values of Class III are not as high as in the other ancient samples.

#### **4.2.5 Conclusions**

The findings of the analysis of Can Reiners (ca. 600 AD) showed that normocclusion is present in ca. 70% of the samples, whereas some values, such as molar relationship, are not so different to the modern ones. Therefore, the general trends that indicate that malocclusion is higher in modern than in ancient populations is too simplistic. Dental wear influences some variables such as the curve of Spee, the superior length, the inferior intermolar width and the right molar relationship. We observed that there are correlations between intercanine and intermolar width and between length and depth, whereas no correlation was observed between transversal and sagittal measurements. Although dental wear was not high in Can Reiners individuals, it seems to be the most decisive determinant of the buccal characteristics. For instance, it may be the most influencing factor for the change of molar relationship. Other analyses, performed in archaeological materials, would be helpful for the future studies aiming to clarify the relationship between occlusal features and the factors that influence them.

### **4.3 Microscopic analysis of the dental calculus**

#### **4.3.1 Atlas of microremains of the Mediterranean cereals**

##### **4.3.1.1 Introduction**

Dental calculus formation is due to the mineralization of the dental plaque. The driving force for plaque mineralization is the super saturation of saliva, particularly plaque fluid, with respect to calcium phosphates. Microorganisms are involved in the formation of calculus but their role is not clear, for instance, some oral bacteria seem to be associated with the absence of calculus. The development of calculus, its composition and quantity vary among people and its amount is influenced by diverse factors such as oral hygiene, diet, age, gender, genetics, bacterial composition and many others (Jepsen *et al.* 2011).

Dental calculus is composed of organic and inorganic components. The inorganic part consists primarily of calcium phosphate salts organized into four crystals structures: octacalcium phosphate ( $\text{Ca}_8(\text{PO}_4)_4(\text{HPO})_25\text{HO}$ ), hydroxyapatite ( $\text{Ca}_{10}(\text{PO}_4)_6(\text{OH})_2$ ),  $\beta$ -tricalcium phosphate or whitlockite ( $\text{Ca}_{10}(\text{HPO}_4)(\text{PO}_4)_6$ ), and brushite ( $\text{CaHPO}_4 \cdot 2\text{H}_2\text{O}$ ), the latter being present in the early stage of formation of the calculus (Jin and Yip 2002). The organic matrix is composed of lipids, carbohydrates and proteins (bacteria and salivary proteins). Usually, the supra gingival calculus is stockpiled on the lingual surfaces of mandibular inferior teeth (in particular in the incisors) and decreases towards the third molars, whereas in the maxilla it is most present in the buccal surfaces of the first molars. Saliva is associated with the distribution pattern of supra gingival calculus. The sub gingival calculus is higher on the lingual than on the buccal surfaces (Jin and Yip 2002).

Trace amounts of food and environmental micro-debris such as starch grains, phytoliths, pollen, spores and fibres are preserved within dental calculus, together with organic components such as lipids and proteins. The recovery and identification of these elements appear fundamental for the reconstruction of the palaeodiet, lifestyle and paleoenvironment of ancient populations. In the last decade many studies have been carried out in order to increase the knowledge regarding this new and innovative field of study (Blatt *et al.* 2011, Eerkens *et al.* 2014, Hardy *et al.* 2009, Hardy *et al.* 2012, Henry and Piperno 2008, Mickleburgh and Pagán-Jiménez 2012, Power *et al.* 2014, Warinner *et al.* 2014, Wesolowski *et al.* 2010). The dominant inclusions within calculus are fragments of vegetal tissue, phytoliths, starch grains and fibres.

Phytoliths are mineral components produced by several higher plants, especially grasses. The term “phytolith” includes different mineral forms precipitated by plants, i.e. calcium oxalate monohydrate ( $\text{CaC}_2\text{O}_4 \cdot \text{H}_2\text{O}$ ) and dihydrate ( $\text{CaC}_2\text{O}_4 \cdot 2\text{H}_2\text{O}$ ), calcium carbonate ( $\text{CaCO}_3$ ) and opaline silica ( $\text{SiO}_2 \cdot n\text{H}_2\text{O}$ ) (Piperno 2006a). Once buried, after plant death and decay of organic matter, oxalates and carbonates are prone to chemical dissolution and therefore are poorly preserved in archaeological settings, except under special circumstances (Reinhard and Danielson 2005). Opaline phytoliths can form within the cell lumen, cell wall, or in between cells through precipitation of monosilicic acid ( $\text{H}_4\text{SiO}_4$ ) absorbed by the root system. Silica is much more resistant to weathering, and for this reason opaline phytoliths are usually preserved in anthropogenic deposits. Furthermore, these components exhibit specific morphologies according to the part of the plant that is silicified and to the taxon the plant belongs to. For instance, phytoliths produced in the inflorescence of wheat are different from phytoliths produced in its stem, and these in turn do not share any similarities with oak phytoliths (Piperno 2006a). Therefore, it is possible to distinguish different plant taxa at the Genus and Family level, with clear benefits for the interpretation of plant use at an archaeological site



(Weiner 2010). Hereafter we use the term “phytolith” to refer to the opaline form.

Starch is the most common carbohydrate in human diet. It is produced by most green plants as an energy store and consists of a large number of glucose units joined by glycosidic bonds. Starch molecules are almost entirely composed of amylose and amylopectin and the proportion of these two polymers depends on the botanical origin.

The starches of many plant show concentric layers called lamellae visible on the surface of some kinds of starches that are growth bands formed during the development of the granules. The lamellae are deposited around the hilum, which is the botanical center of the granule that can be placed in centric or eccentric position. Starch grains are named compound when there are two or more hila present. Due to this type of growth, the molecules are radially arranged, and this characteristic allows seeing the extinction cross in crossed-polarized light.

Starch originates almost exclusively in the plastids, and it can be divided into two categories: transitory starches, formed in the chloroplasts, which are present in leaves and green stems, and reserve starches, formed in the amyloplasts and detected within roots, rhizomes, tubers, and seeds (Wright 2010). Generally starches found in the seeds, roots and tubers are more abundant and diagnostic (Esau 1985) and the size of the granules vary from 1  $\mu\text{m}$  up to 100  $\mu\text{m}$ . Despite various years of research, some question such as the size distribution of crystalline areas of organization of mixed large and small type granules and the distribution of ordered and unordered areas in the granule remain unresolved. In some cereals such as barley and wheat large and small granules are present, whose biosynthesis occurs at two distinct stages of development. However, there is no explanation from biosynthesis studies about the origin of this dual population (Buleón *et al.* 1998).

The criteria used to describe and identify the granules of starch can be observed with a compound light microscope and include: 1) identification if the granule is single or compound; 2) size and shape such as spherical, oval or other shape; 3) form and position of the hilum; 4) number of pressure facets; 5) appearance and features of fissures that are internal cracks indentify on the hila of some starch granules produced when the granule begins to grow outward from the hilum; 6) presence/absence of lamellae (if visible); and 7) features of the extinction cross such as the angle of the arms (Piperno 2006b).

Starch grains may be the most fragile micro-botanical remains. Different studies have shown that changes in temperature are responsible for the degradation of granules. Boiling of starch grains causes breakdown of crystalline layers, loss of distinctive features, and can induce the gelatinization of starch or the formation of a viscous complex with water, thus making the granules unrecognizable (Henry *et al.* 2009, Reichert 1913).

Starch grains begin to swell at temperatures generally between 45° and 55°, more or less, according to their origin. Lippmann in 1861 observed that swelling of barley starch starts at 37.5°, whilst its gelatinization starts at 57.5°, becoming complete at 62.5°. Whympfer in 1909 also studied the temperature of swelling and gelatinization and noted that the values differed in most cases from those of previous observers, and that these changes depended from the state of maturity of the grains (Reichert 1913). Recently, studies conducted on degradation of cooked starch grains (Henry *et al.* 2009) have shown that longer boiling times caused more extreme swelling and loss of their characteristics. The exact nature and timing of this swelling changes among different species of plants. For example, granules of *Panicum miliaceum* (a grass) start to change their characteristics after only one minute of boiling; legumes instead undergo damages more quickly than grasses (Henry *et al.* 2009).

The damaged granule is very different from the undamaged one. Although starch grains are too damaged to identify the species, it is important to

understand whether they are cooked or not in order to describe human behaviour and practices. Indeed, the degradation of starches recovered from archaeological contexts may help to explain past methods of plant processing (Barton 2007). In this sense, we can take into account that food processing generates starchy remains at many stages, starting with the discard of inedible parts of the plant and continuing with peeling, cooking, slicing, and other food preparation techniques (Babot 2006).

The nutritional importance of starch has been enormous for Mediterranean populations since the Neolithic and its main source were cereals (Zohary and Hopf 2000). Cereals are members of the *Poaceae* grass family that grow in open land and complete their life cycle in less than a year. Their cultivation is based on sowing the seed in the tilled field, reaping the mature spikes or panicles, and threshing the grains. Wheat and barley have been the traditional staples of Europe and west Asia. The first definite signs of their cultivation appeared in the Near Eastern fertile crescent in the second half of the 8th millennium BC. They were also the main domesticates that made possible the explosive expansion of Neolithic agriculture to the territories of Asia, Europe and North Africa.

In the Roman period the most important crop was wheat (*Triticum aestivum*). However, after the crisis of the 3th century AD, the cultivation of cereals of lower quality such as rye, spelt, oat, millet, emmer wheat and sorghum was favored because they are more resistant and productive than “*frumentum*” (Montanari 2012). Some of them were employed marginally. For example, rye was the outcome of recent selection and for this reason not well appreciated. Romans deemed rye as a bad grass. Pliny the Elder, in his *Naturalis Historia*, wrote: “*Secale Tauri sub Alpibus asiam vocant, deterrimum et tantum ad arcendam famem, fecunda, sed gracili stipula, nigritia triste, pondere praecipuum. Admiscetur huic far, ut mitiget amaritudinem eius, et tamen sic quoque ingratisimum ventri est. Nascitur qualicumque solo cum centesimo grano, ipsumque pro laetamine est*” (XVIII, 40). Therefore, Pliny confirms that

rye is bitter and very unpleasant to the stomach but gives us an important information: rye can grow upon any soil, yields a hundred times more than wheat, and it is employed also as manure. Indeed, rye will be the most cultivated cereal in the European continent until the 10th -11th c. AD due to its robustness and ability to grow everywhere (Montanari 2012). Emmer wheat and spelt were only sown to make fodder for cattle, and are known by the name of "farrago", or mixed grain (Pliny the Elder, *Naturalis Historia*, XVIII, 10). Pliny reports also valuable information regarding the "low-quality" cereals. For instance, he writes that Campania is particularly prolific for millet, and that a fine white porridge is made from it; millet makes a bread of remarkable sweetness (*Naturalis Historia*, XVIII, 24); barley is one of the most ancient aliments of man and Greeks prefer it to everything else for making *pulmenta* (*Naturalis Historia*, XVIII, 14). Indeed barley, even though is not considered as important as wheat, played a crucial role in antiquity. Romans used barley in some recipes such as "*tisanam et alicam*" and "*tisanam sic facies*" mentioned in the *De Re Coquinaria*, the most important gastronomy work written in Latin (Carazzali, 2013).

With the economic crisis of the late Roman period and the fall of the Roman Empire, some changes took place in the diet of European populations. In particular, the consumption of lower cereals, stronger and easily cultivable, increased considerably. The different types of bread mark a contrast within social classes: on one side white bread (*T. aestivum*) is a luxury product consumed by wealthy people; on the other side, black bread (spelt, rye or mixed cereals) is the basic food of poor people. Frequently, for the lower classes bread was missing and lower cereals such as oat, barley and millet became fundamental for *pulmenta* production, along with vegetables and legumes (Montanari 2012).

The aim of this work is to describe and to recognize micro-remains such as starch grains, phytoliths and fibres from the most used plants in the ancient Mediterranean context, in order to obtain original reference images for

comparison with micro-remains extracted from archaeological dental human calculus.

#### 4.3.1.2 Material and methods

Seeds belonging to domesticated Old World cereal species and the acorns of a *Quercus ilex* (holly oak) were chosen for this study (table 4.3.1.1). They were analyzed to distinguish the dominant micro-components such as phytoliths, starch grains and fibres found in processed and non-processed flour. We decided to study the seeds of plants because they are used for the preparation of several types of food and because they have an high starch content. The cereals used in this study are cultivated in the Mediterranean basin, Africa and Asia and were extensively used by humans before the discovery of the Americas. For this reason cereals such as maize were excluded.

Table 4.3.1.1: plant species analyzed

Family	Subfamily	Tribe	Genus	Species	Common name
Poaceae	Pooideae	Triticeae	<i>Triticum</i>	<i>Aestivum</i>	Wheat, bread wheat
Poaceae	Pooideae	Triticeae	<i>Triticum</i>	<i>Spelta</i>	Spelt
Poaceae	Pooideae	Triticeae	<i>Triticum</i>	<i>Dicoccum</i>	Emmet wheat, Farro
Poaceae	Pooideae	Triticeae	<i>Hordeum</i>	<i>Vulgare</i>	Barley
Poaceae	Pooideae	Triticeae	<i>Secale</i>	<i>Cereale</i>	Rye
Poaceae	Pooideae	Aveneae	<i>Avena</i>	<i>Sativa</i>	Oat
Poaceae	Panicoideae	Andropogoneae	<i>Sorghum</i>	<i>Bicolor</i>	Sorghum
Poaceae	Panicoideae	Paniceae	<i>Panicum</i>	<i>Miliaceum</i>	Proso Millet
Fagaceae	Fagoideae	Querceae	<i>Quercus</i>	<i>Ilex</i>	Holly Oak

Seeds were first ground in an agate mortar to powder them into flour that was collected in 0.5 ml tubes. To analyze the degradation of starch grains, seeds were boiled in water on a combined hot plate/magnetic stirrer device. Seeds of each plant were boiled for 20 or 60 minutes at constant temperature (100° C). Only in one case (*T. spelta*) seeds were also roasted. A small aliquot of the

sample was transferred to a microscope slide, adding water and glycerin in a 1:1 ratio. Each sample was examined using an optical microscope Zeiss Axio Lab A1 (at magnifications ranging 200 - 400x) with an AxioCam ERc5s camera, linked to a PC running the Axio Vision software.

The photomicrographs were analyzed using the ImageJ software and samples were described by noting the morphologies of starch grains and seed tissues, and the characteristics of the degradation of boiled/roasted samples. Individual starch granules were described according to standard morphological and metric variables such as length and width, shape, description of the extinction cross, hilum position and presence of lamellae and fissures (Babot 2007, Cortella and Pochettino 1994, Henry and Piperno 2008, Reichert 1913). Other remains were also described.

#### **4.3.1.3 Results**

The micro-remains of the 10 main Mediterranean crops have been analyzed, with special attention to starch grains and tissues. For sake of clarity, they are described separately.

##### **Wheat (*Triticum*)**

Wheat is the cereal grain that grows in most parts of the world, from near-arctic to near-equatorial latitudes. It is the most important crop among the cereals by area planted and is followed in importance by corn, barley and sorghum. The protein and caloric content of wheat is greater than that of any other food crop (FAO 2009). Its grains contain 60-80% (wet weight) carbohydrates and significant amounts of proteins (8-14%, wet weight) (Zohary and Hopf 2000). Wheat can be easily cultivated and its harvest can be preserved for a long time. Wheat is one of the first cereals to have been domesticated (around 10,000 BC)

and the archaeological record suggests that it occurred in the region known as the Fertile Crescent (Colledge and Conolly 2007, Lev-Yadun *et al.* 2000).

The modern classification of cultivated wheat is based on cytogenetic criteria and it depends on the number of chromosomes. Some species are diploid (two sets of chromosomes) and others are polyploids (with four - tetraploid - or six - hexaploid - series of chromosomes). For this study, *Triticum aestivum* (bread wheat, hexaploid), *Triticum spelta* (spelt or hulled wheat, hexaploid, which is considered a subspecies of *Triticum aestivum*) and *Triticum dicoccum* (emmer wheat or farro, tetraploid) are used.

### **Wheat, bread wheat (*Triticum aestivum*)**

The analysis of starch grains of *T. aestivum* shows simple grains. Usually, there are two types of granules: large and small (fig. 4.3.1.1 A, C and D). The minimum diameter of the granules is 3  $\mu\text{m}$ , whereas the maximum is 35  $\mu\text{m}$ . Grains are generally spherical and oval (fig. 4.3.1.1 B), sometimes with one pressure facet. Lamellae are visible in some cases, with a few rings near the border of the granule. No fissures are visible. The hilum is centric and sometimes elongated. Often, the extinction cross has arms wider towards the ends such as in a Maltese cross, and the arms create different types of angles (right, obtuse and acute). Some grains exhibit an elongated central portion of the extinction cross, whereas the arms are very short compared to the central part. The shape of this type of granules is an elongated oval (fig. 4.3.1.1 B, white arrows) and the diameter varies between 16  $\mu\text{m}$  and 35  $\mu\text{m}$ .

After boiling the seed for 60 minutes a gelatinous mass was formed (fig. 4.3.1.2 A). Henry *et al.* 2009 have shown that *T. aestivum* does not reach full gelatinization until after 10 minutes. Some starch grains survive as single grains (fig. 4.3.1.2 B), but they lose some features, such as the presence of the extinction cross, and they appear damaged, generally with a cracked border. In the gelatinous mass, no granules are distinguishable, whereas when they appear as single damaged granules it is possible to identify them.

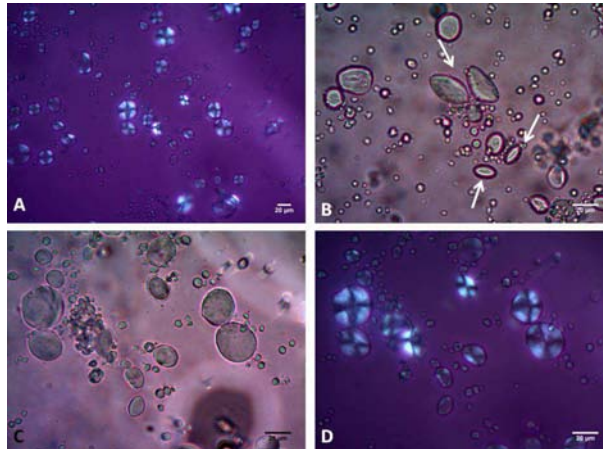


Figure 4.3.1.1: wheat starch grains under polarized light (A and D), wheat starch grains under regular light (B and C), the white arrows show the elongated oval grains (C)

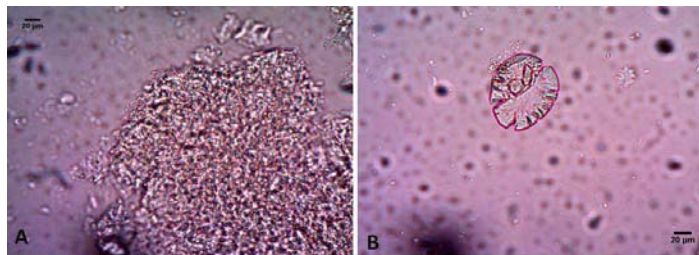


Figure 4.3.1.2: gelatinous starch mass (A), a damaged wheat starch grain (B)

### **Spelt (*Triticum spelta*)**

Starch granules of *T. spelta* are simple and two typologies, one large and one small, are present (fig. 4.3.1.3 B, C and D). The smaller grains exhibit a diameter of 3.5 µm whereas the bigger 29 µm. Regarding size, the grains of spelt are very similar to those of wheat.

The majority of granules are spherical, but some of them are oval in shape. Several have a pressure facet. The hilum is central and its appearance is similar to a point or a line. Lamellae are visible near the border and the arms of extinction cross developed various types of angles. The extinction cross exhibits



wider arms towards the ends, and sometimes a broken arm is present (fig. 4.3.1.3 D). Occasionally, in the oval-shaped granules the central part of the extinction cross is elongated and the arms are very short compared to the central part of the cross.

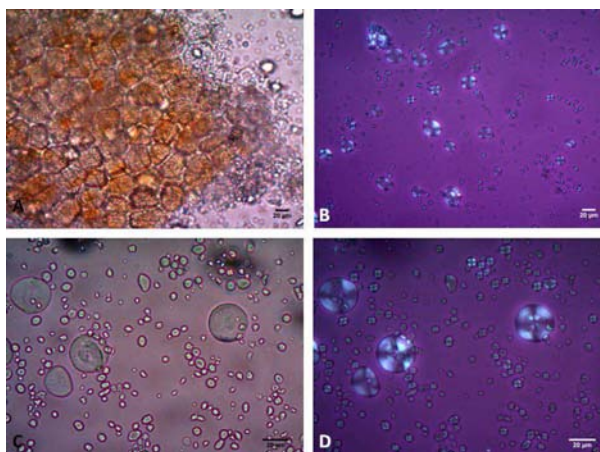


Figure 4.3.1.3: seed coat of spelt before cooking (A), spelt grains under polarized light (B and D), spelt grains under regular light (C)

Figures 4.3.1.3 A and 4.3.1.4 A show the spelt seed coat before and after 60 minutes boiling. After boiling a group of raphides (fig. 4.3.1.4 B), which are needle-shaped crystals found in plants, and only a few well-preserved granules can be observed. The majority of grains are completely gelatinized.

In the case of spelt, seeds were also roasted to ascertain whether there is variation between the two different methods of cooking. In general, the roasted granules (fig. 4.3.1.5 B) are better preserved than the boiled ones (fig. 4.3.1.4 C and D). Some of the roasted granules are well preserved, whereas other are more damaged but generally recognizable (fig. 4.3.1.5 B). Many of the damaged granules are broken, without characteristic features such as a clear extinction cross, although they preserve a definite border with some interruption.

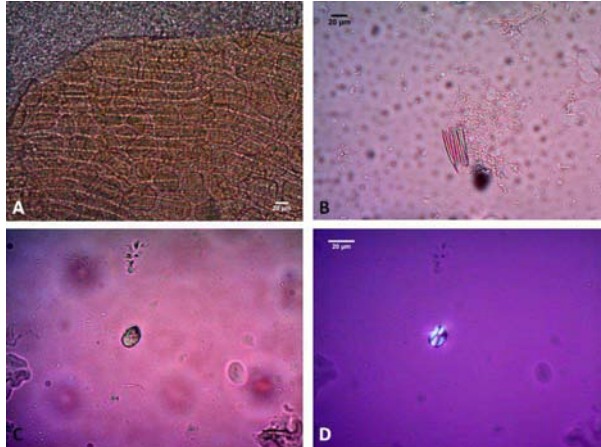


Figure 4.3.1.4: seed coat of spelt after boiling (A), a group of raphides (B), a well preserved starch granule which diameter is 15  $\mu\text{m}$  (C and D)

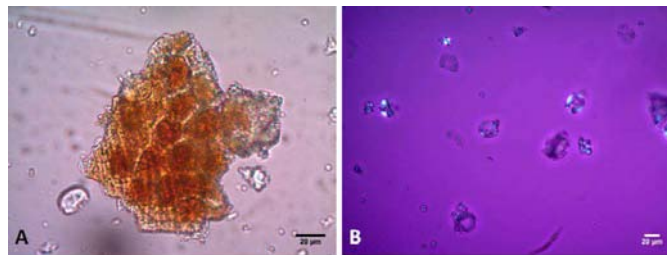


Figure 4.3.1.5: a fragment of spelt seed coat after roasting (A), a group of spelt grains after roasting (B)

### **Emmer wheat or farro (*Triticum dicoccum*)**

The diameter of the granules of emmer wheat ranges from 2  $\mu\text{m}$  to 34  $\mu\text{m}$ . Grains are divided into two categories: small and large (fig. 4.3.1.6 B). The small granules are more spherical with a radially symmetrical and right-angle cross, whereas the bigger grains are oval-shaped and exhibit a different shape of cross, similar to that of pea starch (fig. 4.3.1.6 B, white arrows). The central part of the extinction cross is elongated and the arms are short compared to the central part of the cross.

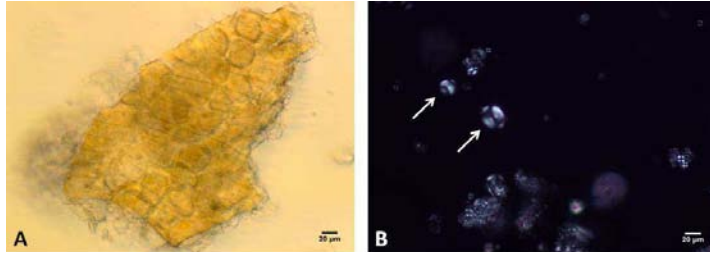


Figure 4.3.1.6: a fragment of emmer wheat coat seed (A), emmer starches under polarized light, the white arrows shown the particular shape of the extinction cross (B)

### **Barley (*Hordeum vulgare*)**

Cultivated barley was, together the wheat, one of the main cereals in the Mediterranean area. Compared to wheat, barley is considered a lower staple and therefore it has been the main cereal of the paupers. However, barley has other qualities, such as the ability to withstand drier conditions and poor soils. Furthermore, it was the main cereal used for beer fermentation in Old World (Zohary and Hopf 2000). The wild ancestor of *Hordeum vulgare* is *Hordeum spontaneum*, which grows in the Near East. This area coincides with the archaeological sites of the earliest finds of barley cultivations.

Starch granules of barley are simple. Some are extremely small ( $< 2 \mu\text{m}$ ) whereas other range from 5 to 30  $\mu\text{m}$  (fig. 4.3.1.7 C). Generally, the biggest grains are spherical, oval or reniform. (fig. 4.3.1.7 A, B). The hilum is central and elongated and the arms of the extinction cross create various types of angle (right, obtuse and acute).

After boiling, the sample was in part gelatinized (fig. 4.3.1.8 A). Within the starch mass, just a few granules survive the boiling process. Those preserved are round, with a visible right-angle extinction cross. The diameter of granules is 8.4  $\mu\text{m}$ . In this sample, multicellular structure wavy long cell phytoliths (fig. 4.3.1.8 B) (Hart 2011, Wu *et al.* 2014) and plant hair (fig. 4.3.1.7 D) were also found.

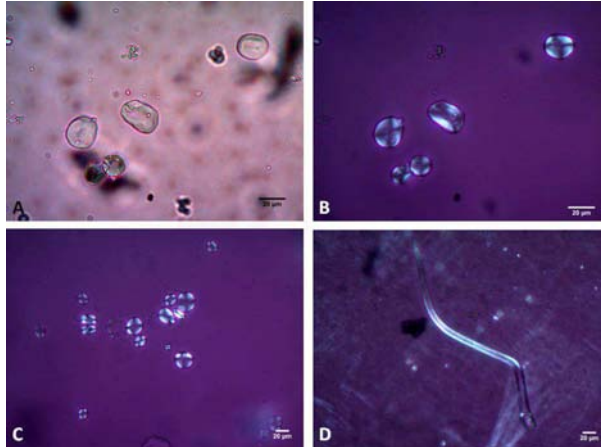


Figure 4.3.1.7: barley starch grains under regular light (A), starches under polarized light (B and C), plant hair of barley (D)

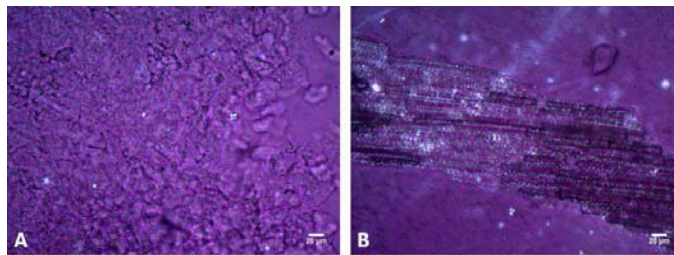


Figure 4.3.1.8: gelatinous starch mass (A), multicellular structure wavy long cells (B)

### **Common oat (*Avena sativa*)**

Common oat is one of the major crops in Old World agriculture and its nutritive value is very high. The grains contain about 15-16% protein and 8% fat. In northeastern Europe, common oat is one of the principal grain crops because it grows well in moist climates of temperate latitudes. The archaeological evidence shows that oat was probably a weed infesting other crops. Only much later, it was domesticated by humans for food consumption (Zohary and Hopf 2000).

In the analyzed sample, the diameter of granules is generally smaller than 10  $\mu\text{m}$  (from 3 to 7  $\mu\text{m}$ ) (fig. 4.3.1.9 A, B and C). Some grains are single, whereas other are compound (fig. 4.3.1.9 D, white arrow). Most of them show a polyhedral shape. Figure 4.3.1.9 D (black arrow) shows a 15  $\mu\text{m}$  granule presenting a visible fissure. The arms of the extinction cross, visible under crossed-polarized light, are not at a right angle. Some crosses are similar to those of emmer wheat.

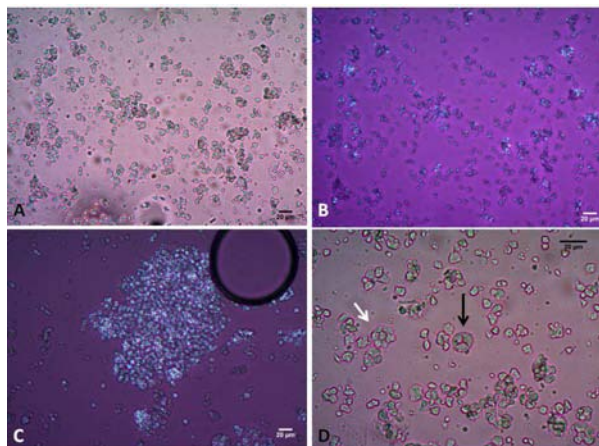


Figure 4.3.1.9: oat starch grains under regular light (A), oat starch grains under polarized light (B and C), oat granules single and compound (white arrow) and in one of them a fissure is visible (black arrow) (D)

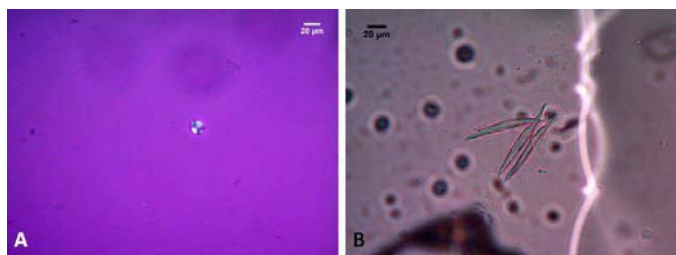


Figure 4.3.1.10: well-preserved oval shaped oat starch of 16  $\mu\text{m}$  (A), a group of raphides (B)

Boiled seed shows a complete gelatinization of starch and just a few well-preserved granules were observed. The granule observed in fig. 4.3.1.10 A has a diameter of 16  $\mu\text{m}$  and it is oval in shape. The extinction cross is similar to that of emmer wheat large starch grains: the central part of the cross is elongated, the arms are symmetrical but shorter compared to the central part of the cross. Well-preserved raphides are present (fig. 4.3.1.10 B), although the temperature was 100 ° C.

### **Rye (*Secale cereale*)**

Rye is the grain of the temperate areas of the Old World. The earliest remains of this cereal were found in Epi-Palaeolithic sites in northern Syria and date back to 8500-7500 BC (Zohary and Hopf 2000). Pliny the Elder, in the *Naturalis Historia*, wrote that *secale* was a poor-quality grain. It was employed to avert famine and generally mixed with spelt. He reported that the people of *Taurinum*, present-day Turin, gave *secale* the name of “asia” (XVIII, 40). In current world production rye is used to make bread, rye whisky and as animal fodder.

Rye starch grains are simple, spherical or oval-shaped and some exhibit pressure facets and fissures (fig. 4.3.1.11 B, C and D). The diameter of individual grains ranges from 2 to 35  $\mu\text{m}$ . These grains are the biggest among the cereal samples. The hilum is central and is point- or line-shaped. Often, the extinction cross shows large arms, wider towards the end, and the crosses create different types of angles. Some granules such as emmer wheat and oat show an elongated central part of the cross and the arms are shorter compared to the central part (fig 4.3.1.11 A).

Rye seeds were boiled for 60 minutes at a constant temperature (100° C). After boiling, a gelatinous starch mass is observed (fig. 4.3.1.12 A, D) although multiple granules survived the process, keeping their original morphology (fig. 4.3.1.12 C, D). Other granules lost their characteristic features (fig. 4.3.1.12 B).



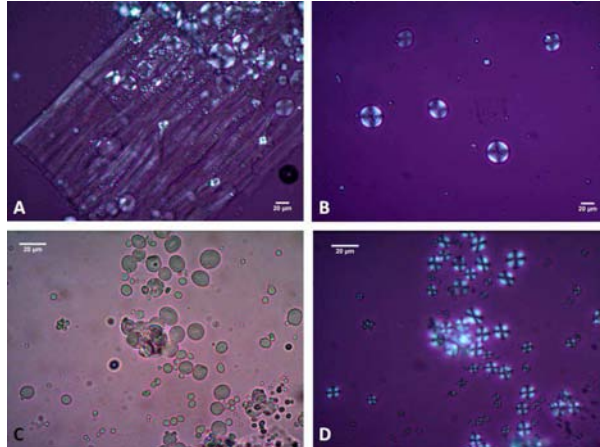


Figure 4.3.1.11: rye starch grains under polarized light (A, B and D), granules under regular light (C)

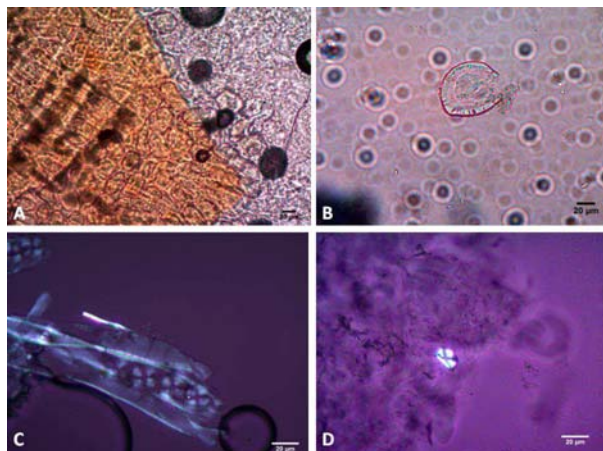


Figure 4.3.1.12: rye seed coat after boiling (A), a damaged starch (B), a group of starches inside an organic tissue (C), a boiled starch granule (D)

Figure 4.3.1.12 C shows that all the well-preserved starch grains are encased by organic tissue. The diameter of these granules is ca. 18  $\mu\text{m}$  and they are spherical. The extinction cross exhibits wider arms towards the end such as those of wheat and spelt, and the hilum is central.

### **Common or proso millet (*Panicum miliaceum*)**

Millet is a warm season plant, very sensitive to cold temperature and its life cycle is short (60-90 days), growing in areas with short rainy seasons. *Panicum miliaceum* is the millet of classical times (the Roman *milium*) and the grains are sufficiently rich in proteins (10-11%) for human consumption (Zohary and Hopf 2000). The archaeological evidence shows that millet appeared in Europe between the 5th and 4th millennia BC.

### **White proso millet**

Starch grains are simple and range from 2 to 13  $\mu\text{m}$  (fig 4.3.1.13 A, B). Granules are spherical or polyhedral and some of them show pressure facets. The hilum is central and the arms of the extinction cross create different types of angles (right, obtuse and acute). No lamellae or fissures were observed.

After boiling, groups of raphides (fig. 4.3.1.13 C) and of well-preserved starch grains (fig. 4.3.1.13 D) were identified. The diameter of these granules ranges from 5 to 8  $\mu\text{m}$  and they are oval and polyhedral in shape.

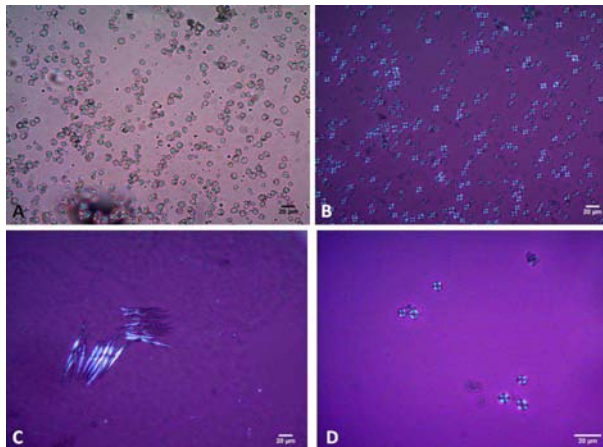


Figure 4.3.1.13: white proso millet starch grains under regular light (A), granules under polarized light (B), a group of raphides (C), a group of starches after boiling (D)



### Red proso millet

All the grains, except for one, have a diameter less than 10  $\mu\text{m}$  and more than 4  $\mu\text{m}$ . The shape is spherical or polyhedral (fig. 4.3.1.14 A, B, C) and the majority of the grains exhibit right-angle extinction crosses. No lamellae or fissures were observed. Figure 4.3.1.14 C shows a group of grains within a multicellular polyhedral structure (Albert and Cabanes 2008). After boiling, a group of well-preserved grains were observed, whereas another aliquot of the sample underwent a gelatinization process (fig. 4.3.1.14 D). The diameter of the grains is ca. 5-7  $\mu\text{m}$ . A proper description of the characteristic features of each grain was hampered by the occurrence of clusters of superimposed grains.

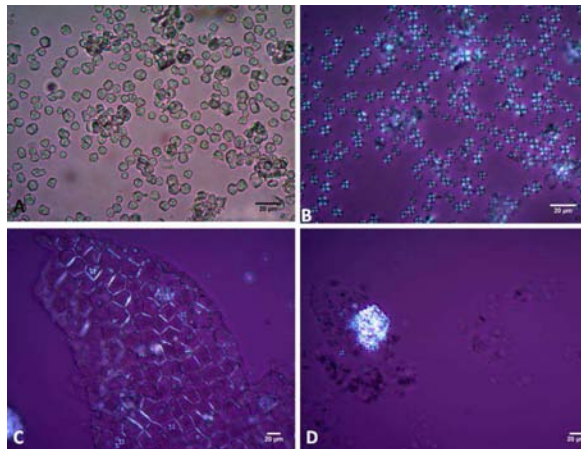


Figure 4.3.1.14: red proso millet granules under regular light (A), granules under polarized light (B), granules within a multicellular polyhedral structure (C), a group of starch grains after boiling (D)

### Sorghum (*Sorghum bicolor*)

Sorghum is not native to the Near East and the Mediterranean basin. It reached this area as an already fully-developed crop only during the Greek and Roman periods. Sorghum is a cereal that grows in Africa, south-west Asia and the Indian subcontinent. The earliest archaeological evidence of sorghum

cultivation was found in the Indian subcontinent and dates back to the second millennium BC (Zohary and Hopf 2000).

Granules are single and large. The diameter ranges between 12 to 21  $\mu\text{m}$ , although grains of 5 and 7  $\mu\text{m}$  are also present (fig 4.3.1.15 A and B). Grains are spherical or polyhedral and the hilum is central. In some cases lamellae, fissures and lines radiate from the center towards the edges. Often, the extinction cross exhibits large arms, wider towards the end, and the arms show different types of angles (right, obtuse and acute) (fig. 4.3.1.15 C). After boiling, no recognizable starch grains are found. Only a large gelatinous mass is visible (fig. 4.3.1.16 A and B).

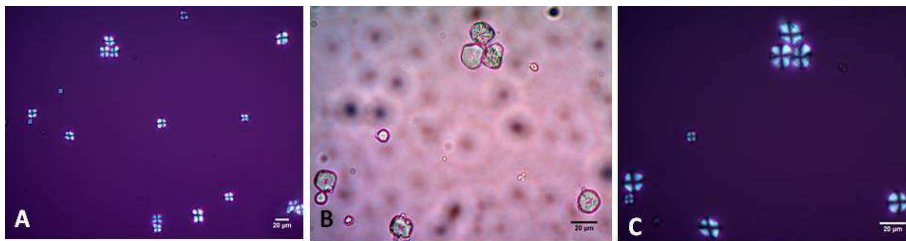


Figure 4.3.1.15: sorghum starch grains under polarized light (A and C), granules under regular light (B)

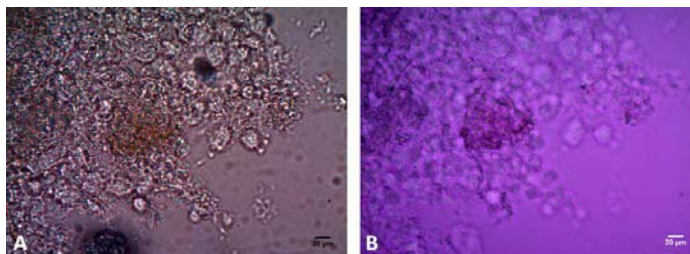


Figure 4.3.1.16: gelatinous starch mass under regular (A) and polarized light (B)

### Acorn of holly oak (*Quercus ilex*)

The acorn used for this study comes from *Quercus ilex*, or holly oak. It is a large evergreen oak native to the Mediterranean region, which replaced deciduous trees due to increasing intensity of human-driven disturbances (especially fire) throughout the Neolithic period. The principal use of holly oak forests by humans includes nomadic grazing, acorn harvesting and fire-wood exploitation. In the south-west of the Iberian Peninsula, starting from the early Middle Ages, holly oak forests turn into lands devoted to extensive livestock rearing (Pulido *et al.* 2001).

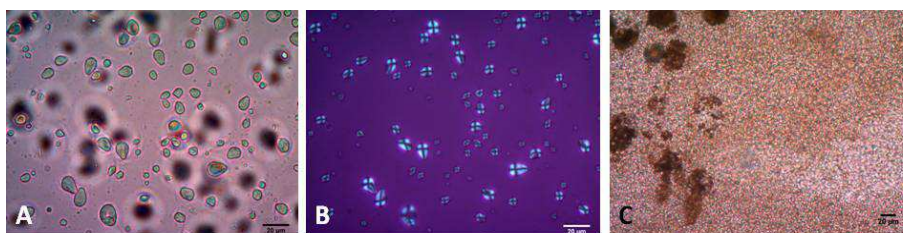


Figure 4.3.1.17: acorn starches under regular (A) and polarized light (B), gelatinous starch mass (C)

Figures 4.3.1.17 A and B show grains of acorn with a particular shape. Generally, acorn granules can be divided into two groups: granules with a circular or oval appearance, and those that are “pear-” or “drop-” shaped. The hilum is not central, rather proximal to the rounded extremity. Some lamellae are visible in the largest grains, in particular near the border. No fissures were observed. The diameter of the granules ranges from 3 to 14  $\mu\text{m}$ . The arms of the extinction cross are large and show different types of angles. After boiling, no granules were identified. Only a large gelatinized mass was visible (fig. 4.3.1.17 C).

#### 4.3.1.4 Discussion

In spite of the difficulties encountered in identifying starch granules of cereals, some features such as shape, size and position of the hilum could be helpful in differentiating them. In our samples, all the starch grains are simple, with the exception of oat grains, which are also compound. Regarding size (table 4.3.1.2), the largest grains are found in rye, emmer and wheat. Also granules of barley, spelt and sorghum show a remarkable size. The grains of the remaining species are smaller than 20  $\mu\text{m}$ . Lamellae are visible in some grains of wheat, spelt and acorn, whereas fissures occur only in oat, rye and sorghum. The hila of starch granules are usually central, point- or line-shaped, whereas in acorn granules the hila could be located in central or not central position due to its drop-shaped structure.

Table 4.3.1.2: starch grain diameter in modern cereals and into an acorn of *Quercus Ilex*




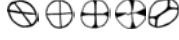
















<b>Species</b>	<b>Minimum size</b>	<b>Maximum size</b>	<b>Mean length</b>
<i>T. aestivum</i>	3 $\mu\text{m}$	35 $\mu\text{m}$	17.5 $\mu\text{m}$
<i>T. spelta</i>	3 $\mu\text{m}$	29 $\mu\text{m}$	16 $\mu\text{m}$
<i>T. dicoccum</i>	2 $\mu\text{m}$	34 $\mu\text{m}$	18 $\mu\text{m}$
<i>H. vulgare</i>	<2 $\mu\text{m}$	30 $\mu\text{m}$	15 $\mu\text{m}$
<i>S. cereale</i>	2 $\mu\text{m}$	35 $\mu\text{m}$	18.5 $\mu\text{m}$
<i>A. sativa</i>	3 $\mu\text{m}$	17 $\mu\text{m}$	10 $\mu\text{m}$
<i>S. bicolor</i>	5 $\mu\text{m}$	21 $\mu\text{m}$	13 $\mu\text{m}$
<i>P. miliaceum</i> (white)	2 $\mu\text{m}$	13 $\mu\text{m}$	7.5 $\mu\text{m}$
<i>P. miliaceum</i> (red)	>4 $\mu\text{m}$	<10 $\mu\text{m}$	7 $\mu\text{m}$
<i>Quercus ilex</i>	3 $\mu\text{m}$	14 $\mu\text{m}$	8.5 $\mu\text{m}$














In all species the extinction cross (table 4.3.1.3) exhibits different types of angles such as right, obtuse and acute. For this reason, this feature is not useful in identifying a species. In species such as, wheat, spelt, emmer, oat, and rye the particular type of extinction cross could be more helpful, with its elongated central part and short arms. Other traits are less common, such as the presence of one broken arm, found only in spelt, or the appearance of large arms,

characteristic of rye and acorn. The Maltese cross (arms wider towards the ends) is observed in wheat, spelt, rye and sorghum.

Within the samples, some exceptions to the main characteristics can be observed. For instance, sorghum has an average diameter of 13  $\mu\text{m}$  and round shape, but some isolated large granules can occur (47  $\mu\text{m}$ ). They are oval and exhibit a hilum displaced from the center.

Table 4.3.1.3: shape of the granules and characteristics of the extinction cross

Species	Shape	Extinction cross
<i>T. aestivum</i>		
<i>T. spelta</i>		
<i>T. dicoccum</i>		
<i>H. vulgare</i>		
<i>S. cereale</i>		
<i>A. sativa</i>		
<i>S. bicolor</i>		
<i>P. miliaceum</i> (white)		
<i>P. miliaceum</i> (red)		
<i>Quercus ilex</i>		

Legend: shape: 1)  spherical, 2)  oval, 3)  pressure facet, 4)  reniform, 5)  polyhedral, 6)  "pear" or "drop". Extinction cross: a)  obtuse and acute angle, b)  right-angle, c)  arms widen towards the ends, d)  large arms widen towards the ends, e)  broken arm, f)  elongated central part and shorts arms, g)  eccentric hilum

In relation to food processing, all the boiled samples reached the gelatinization stage and starch grains, in most cases, formed a mass in which it is impossible to discern single grains and their features. In sorghum and acorn samples well-preserved grains are not visible, whereas in spelt, barley and oat only a few granules have been observed. Rye and millet grains survived well the boiling process. It is interesting to note that after boiling only a few granules were

preserved in spelt, whereas a larger amount of granules survives in roasted seeds.

In some samples of spelt, oat and millet groups of raphides were also identified. The length of spelt raphides is ca. 73-76  $\mu\text{m}$  and the width ca. 3.5  $\mu\text{m}$ . The length of millet raphides is ca. 50-56  $\mu\text{m}$  whereas the width is the same as that measured in spelt. Oat raphides are slightly larger compared to other samples: the length is ca. 84  $\mu\text{m}$  and the width ca. 5-8  $\mu\text{m}$ . The morphology is the same for all the raphides: they are elongated, narrow and sharp-pointed.

In barley wavy long cell and plant hair phytoliths were also identified. Depending on the strength and duration of grinding, partial vegetal structures can survive and it is not uncommon to find fragments of tissues or fibres of different sizes.

#### **4.3.1.5 Conclusions**

In general, we conclude that in spite of the large degree of similarity among starch grains, the comprehensive interpretation of the characteristics of all micro-residues from plant food could provide a useful tool for the identification of the species exploited by ancient populations for food consumption.

## **4.3.2 The significance of microremains recovered from dental calculus for anthropological analysis: the case study of Can Reiners**

### **4.3.2.1 Introduction**

The study of diet is one of the most important anthropological contributions to the knowledge of ancient population. Food plays an important role in everyday life: food has to nourish and build the body and it is helpful for mental and physical wellness. The ancient Greek physician Hippocrates, in the 5th century BC, wrote “Let food be your medicine and medicine be your food”. Moreover, food procurement occupied much of an individual’s day in ancient times determining his/her life: activity, movements, health, leisure time, etc. In physical anthropology the study of oral and skeletal pathologies and stable isotopic compositions are fundamental for the reconstruction of paleonutrition and human health. In the last decade, dental calculus analysis has contributed to increasing the information about ancient diet, primarily regarding its micro-remains (Afonso-Vargas *et al.* 2015, Blatt *et al.* 2011, Hardy *et al.* 2009, Hardy *et al.* 2012, Henry and Piperno 2008, Musaubach 2012). The microfossils that could be observed in dental calculus are starch grains, vegetal and animal fibres, phytoliths, pollen, spores and other organic or inorganic micro-remains. These findings are helpful also for environmental reconstructions.

The main goal of this work is to analyse the micro-remains that could be observed in the dental calculus of a small sample of Can Reiners, in order to reconstruct diet and lifestyle of these individuals and to understand the dynamics of feeding in the population. Generally, publications of dental calculus analysis have been focused on maize and potato starch grains from New World diet (Mickleburgh *et al.* 2012, Piperno *et al.* 2009, Rosewig *et al.* 2014). For this reason, a study of the micro botanical remains observed in seeds of some Old World cereals and an acorn of a holly oak were previously studied (see section 4.3.1). The information recovered is useful for a proper description

and identification of the granules that could be observed in an ancient European context. Furthermore, other types of microfossils such as pollen, phytoliths and fibres have been considered.

#### 4.3.2.2 Material and methods

Dental calculus was collected from 17 teeth that belong to 12 adult individuals of the Can Reiners necropolis (table 4.3.2.1). The individual, the tooth and the position of dental calculus were selected randomly. Figure 4.3.2.1 shows the general dental calculus distribution of the samples. In the individual Cr 77.1, the dental calculus of each available tooth was studied. The choice of studying all the calculi of this individual is due to the presence of an uncommon non-alimentary dental wear on its incisors teeth. This infrequent finding is observed only in upper and lower anterior teeth.

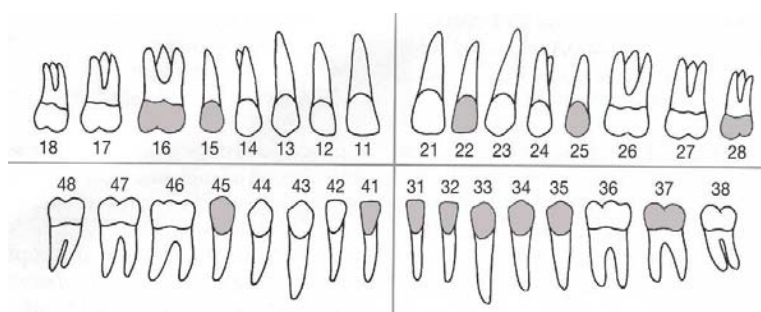


Figure 4.3.2.1: distribution of dental calculus of the recovered samples

To avoid contamination, some precautions were taken into account (Crowter *et al.* 2014, Wesolowski *et al.* 2010). The samples were separated from other skeletal remains and transferred into a laboratory at the Biological Anthropology Unit, Department of Animal and Plant Biology and Ecology, Autonomous University of Barcelona. During the analysis, the door and



windows of the laboratory were always kept closed. The samples were treated under a fume hood and the work area was cleaned with 70% ethanol before and after each analysis to prevent contamination between samples. Powder-free latex gloves were used to handle the samples and they were changed after each analysis. Every tool was cleaned before and after each utilization. Teeth were washed with a toothbrush and distilled water in order to remove the soil pollution present on the surface. After that, dental calculus was removed from teeth with a dental probe. The removed material was collected in a filter paper and immediately transferred in a microcentrifuge tube (0.5 ml). Then acid was added to each tube and supervised until calculus was completely dissolved. Three different acids (hydrochloric, glacial acetic and EDTA) were employed to test if there were differences between methods. Although the majority of methodologies to dissolve dental calculus use hydrochloric acid, a standard operating procedure has not yet been developed (Blatt *et al.* 2011, Boyadjian *et al.* 2007, Henry and Piperno 2008, Wesolowsky *et al.* 2010). Hardy (2007) suggests using also EDTA (ethylenediaminetetraacetic acid) to dissolve the calculus. EDTA is a less fast method, although it is as effective as hydrochloric acid.

Acids were randomly selected and time exposure depended on the nature and composition of dental calculus. Samples were dissolved in acids, and constantly supervised for a minimum of one hour to a maximum of 25 hours. When samples did not achieve complete dissolution, calculus fragments were powdered into an agate mortar to increase the particle surface area exposed to acids. In some samples, to assess whether acids are too aggressive and some important data could be lost, no chemical treatments were employed (Cr 33.1.a, Cr 94.2, Cr 119.1 and Cr 131.1). Therefore, dental calculus was powdered and mounted directly onto slides.

After the chemical treatment, tubes with powder were centrifuged for 3 minutes at 1000 RPM in a Heraeus Biofuge Fresco centrifuge. After this procedure, dental calculus forms a pellet at the bottom of the tubes. The solution was

removed using a pipette. Tubes were then filled with distilled water and centrifuged at the same conditions described above. The supernatant was removed and tubes were filled again with water. The rinsing procedure was repeated at least three times. Afterwards, a few drops of 70% ethanol were added to cover each sample.

Finally, samples were mounted on a slide and examined using an optical microscope Zeiss Axio Lab A1 (at magnifications ranging from 200x to 400x) equipped with an AxioCam ERc5s camera, and linked to a PC running the Axio Vision software. The photomicrographs were elaborated using ImageJ.

Table 4.3.2.1: code, tooth number, calculus position and chemical treatment of Can Reiners dental samples

<b>CR code</b>	<b>Tooth</b>	<b>Calculus Position</b>	<b>Chemical treatment</b>
<b>17.1</b>	34	Buccal	5% HCL for 1 hour, EDTA for 17 hours
<b>33.1.a</b>	32	Lingual	No chemical treatment
<b>33.1.b</b>	32	Lingual	Glacial acetic acid for 1 hour
<b>33.1.c</b>	32	Lingual	10% HCL for 21 hours
<b>70.1</b>	35	Distal	EDTA for 17 hours
<b>76.1.a</b>	15	Buccal	10% HCL for 1 hour
<b>76.1.b</b>	16	Mesial	10% HCL for 1 hour
<b>77.1.a</b>	37	Lingual	5% HCL for 1 hour, 10% HCL for 2 hours
<b>77.1.b</b>	33	Lingual	5% HCL for 1 hour, 10% HCL for 2 hours
<b>77.1.c</b>	28	Lingual	5% HCL for 1 hour, 10% HCL for 2 hours
<b>77.1.d</b>	25	Lingual	5% HCL for 1 hour, 10% HCL for 2 hours
<b>77.1.e</b>	28	Buccal	5% HCL for 1 hour
<b>77.1.f</b>	22	Distal	5% HCL for 1 hour
<b>94.2</b>	41	Lingual	No chemical treatment
<b>119.1</b>	41	Lingual	No chemical treatment
<b>131.1</b>	31	Lingual	No chemical treatment
<b>132.1</b>	45	Buccal	5% HCL for 18 hours
<b>144.1.a</b>	15	Mesial	5% HCL for 18 hours
<b>144.1.b</b>	15	Mesial	EDTA for 25 hours
<b>160.1.a</b>	33	Buccal	EDTA for 17 hours
<b>160.1.b</b>	45	Buccal	5% HCL for 18 hours
<b>169.1</b>	45	Buccal	5% HCL for 1 hour, EDTA for 17 hours

### 4.3.2.3 Results

Several microremains were observed within the dental calculus of Can Reiners individuals. Starch grains are the most common finding, followed by fibres, pollen, phytoliths, spores and other organic materials. For sake of clarity, samples were described separately depending on the method employed. Table 4.3.2.2 shows the main findings divided by individuals and teeth.

Table 4.3.2.2: descriptions of the micro botanical remains observed into the dental calculus of the individuals of Can Reiners

CR	Tooth	Description
17.1	34	Starch grains (sorghum, rye), pollen ( <i>Pinaceae</i> ), spores into an organic matrix, black carbonized organic matter
33.1.a	32	Phytoliths, undetermined vegetal fibres
33.1.b	32	Undetermined starch grain and vegetal fibres
33.1.c	32	Starch grains (wheat, spelt or rye)
70.1	35	Starch grains (sorghum, rye), cotton fibre, libriform fibre of tracheid, fragment of spiral microstructure of a tracheid
76.1.a	15	Starch grains (wheat, spelt)
76.1.b	16	Undetermined starch grains
77.1.a	37	Starch grains (cooked, undetermined, rye, sorghum, spelt and wheat), fragments of fibres (wool), raphides and tracheids.
77.1.b	33	Starch grains (rye, sorghum), fragments of vegetal fibres, raphides, tracheids and fragment of a multicellular structure polyhedral
77.1.c	28	Starch grains (wheat, spelt, rye and sorghum) and large undetermined grains, fragment of fibres (linen and undetermined), raphides, and undetermined vegetal remains
77.1.d	25	Starch grains (wheat, spelt, rye, sorghum), agglomeration of starches, uncommon compound starch grains, fibres, vessels and bordered pits, raphides
77.1.e	28	Starch grains (rye, sorghum, uncommon large grains), fibres and vegetal remains
77.1.f	22	Starch grains (wheat, rye, sorghum, <i>fabaceae</i> ), spores, tracheids
94.2	41	Starch grains, human hair
119.1	41	Fibres
131.1	31	Fibres, a fragment of a nematode
132.1	45	Undetermined starch grains, a multicellular structure polyhedral, tracheids and vessels, vegetal fibres
144.1.a	15	Starch grains (rye, sorghum), fibres, a vegetal tissue
144.1.b	15	No micro botanical remains
160.1.a	33	Starch grains (sorghum, rye), hair phytoliths, undetermined organic matters, pollen ( <i>Pinaceae</i> ), fibres
160.1.b	45	Conglomeration of starches (oat, farro), isolated grains (wheat, spelt, rye, sorghum), fibres (linen, jute), tracheids, spores, sporangium annulus of fern
169.1	45	Black carbonized organic matters, pollen ( <i>Pinaceae</i> )

Some samples, due to the lack of results, were excluded from this report. Generally, the samples mounted on the slide without chemical treatment were difficult to analyse due to the presence of thick calculus fragments that hampered proper identification of several micro-remains.

### Cr 17.1

The individual buried in grave 17.1 corresponds to an adult male and his age at death was between 45 and 59 years old. The general state of preservation is good. Regarding pathologies, a fracture is present in the fourth and fifth metacarpals. Malocclusion (Class III) is present in the left side of the dental arch; the right side is not preserved. No overjet, overbite or other buccal anomalies were observed. Dental calculus is present only in the mandibular left first premolar (tooth 34). The sample was treated with 5% hydrochloric acid for one hour. After rinsing, the sample was covered with EDTA for 17 hours. After this period of time, the calculus was completely dissolved.

Some starch grains, which show similar features, were observed within dental calculus. One of them (fig. 4.3.2.2 A) is polyhedral in shape and its diameter is ca. 14  $\mu\text{m}$ . The extinction cross exhibits large arms wider towards the ends (Maltese cross). The granule features are similar to those of sorghum and rye grains, which were described in the previous chapter.

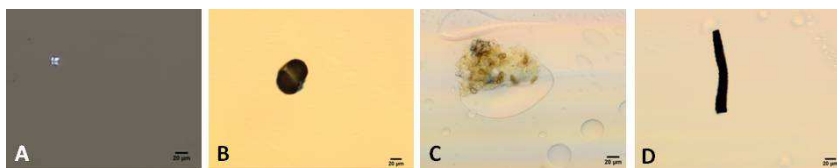


Figure 4.3.2.2: calculus micro-remains from Cr 17.1: starch grain under polarized light (A), *Pinaceae* pollen grain (B), spores into an organic matrix (C), carbonized organic element (D).  
Scale bar 20  $\mu\text{m}$

Figure 4.3.2.2 B shows a pollen granule of the *Pinaceae* family (Moore and Webb 1978). The pollen grain is bisaccate (or vesiculate) and it is composed of a body with two laterally-placed bladders (sacca, vesicles). Its maximum length is 64  $\mu\text{m}$ . Figure 4.3.2.2 C exhibits an organic matrix with embedded oval granules. These are probably spores. Figure 4.3.2.2 D shows one of the two black carbonized organic components observed in this sample. Both of them are elongated, with a rectangular shape and echinate border.

### Cr 33.1

Individual 33.1 is an adult male and his age at death can be placed between 30 and 40 years old. The state of preservation is good. Dental calculus was removed from the mandibular left second incisive (tooth 32) and divided in three samples.

In sample 33.1.a, (not subject to chemical treatments) different botanical elements were found: a hair cell phytolith (fig. 4.3.2.3 A), some undetermined vegetal fibres (fig. 4.3.2.3 B), a parallelepiped elongate phytolith with a facetate edge (fig. 4.3.2.3 C) similar to those of a tracheid or a dicotyledon fruit observed in Piperno (2006a) or in Henry *et al.* (2012), or a probable facetate terminal tracheid phytolith (Piperno 2006a) (fig. 4.3.2.3 D).



Figure 4.3.2.3: calculus micro remains from Cr 33.1a: an hair cell phytolith (A), an undetermined vegetal fibres (B), a parallelepiped elongate phytolith (C), a facetate terminal tracheid phytolith (D). Scale bar 50  $\mu\text{m}$

Sample 33.1.b was treated with glacial acetic acid for one hour. After that, dental calculus was powdered in an agate mortar. Only a few starch grains and vegetal fibres, similar to those observed in the first sample, were observed. The diameter of the grains are ca. 12  $\mu\text{m}$  and their hila are central.

Sample 33.1.c was dissolved in 10% hydrochloric acid for 21 hours. Figure 4.3.2.4 shows some well-preserved starch grains embedded within a matrix.

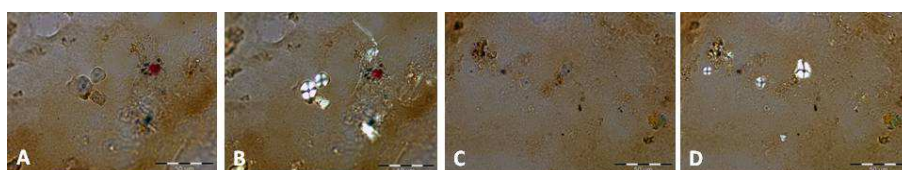


Figure 4.3.2.4: calculus microfossils from Cr 33.1c: starch grains under regular light (A and C), starch grains under polarized light (B and D). Scale bar 50  $\mu\text{m}$

In figure 4.3.2.4 A and B two spherical starch grains which diameters are 15  $\mu\text{m}$  and 13  $\mu\text{m}$ , respectively, can be observed. In both granules, the hila are central and point-shaped. The extinction crosses exhibit arms wider towards the ends (Maltese cross). Considering the size and the characteristics of the granules, they are similar to those of wheat, spelt or rye. Figures 4.3.2.4 C and D show three starch grains embedded within a matrix. The first one is spherical with a central hilum. The extinction cross has arms wider towards the ends and its diameter is ca. 7.4  $\mu\text{m}$ , and is similar to those of figures 4.3.2.4 A and B. The other two are probably deformed. One of them is 9.5  $\mu\text{m}$  long, whereas the largest one is 16  $\mu\text{m}$  long and has a central hilum.

### Cr 70.1

Grave 70.1 contained the skeletal remains of an adult female between 25 and 35 years old. The skeleton shows different biomechanical stress markers such as Schmorl's nodules, accessory facets in the tibiae (squatting facets) and in the

coxo-iliac articulation. Dental calculus was extracted from the mandibular left second premolar (tooth 35) and this is the only tooth covered by calculus. The sample was treated with EDTA for 17 hours. After this period of time the calculus was completely dissolved. Regarding starches, some granules were observed and their diameter ranges between 9 and 15  $\mu\text{m}$ . Four of them are spherical (fig. 4.3.2.5 A and C) whereas one is polyhedral. The arms of the extinction cross are large and wider towards the ends (fig. 4.3.2.5 B and D). In general the granules, if compared with those studied in the previous chapter, are more similar to sorghum and rye. Other elements found in this sample were fibres and vessel elements. In figure 4.3.2.5 E a blue fibre is visible, which is probably from cotton textile. Its width is ca. 14  $\mu\text{m}$ , which is similar to the average width of modern Egyptian cotton fibres (16  $\mu\text{m}$ ) analyzed at the United States Department of Agriculture (Von Bergen and Krauss 1942). Figure 4.3.2.5 F shows another fragment of fibre of a tracheid (Pujana et al. 2008). Figure 4.3.2.5 G exhibits a fragment of a spiral microstructure (tracheid) from the xylem of a plant.

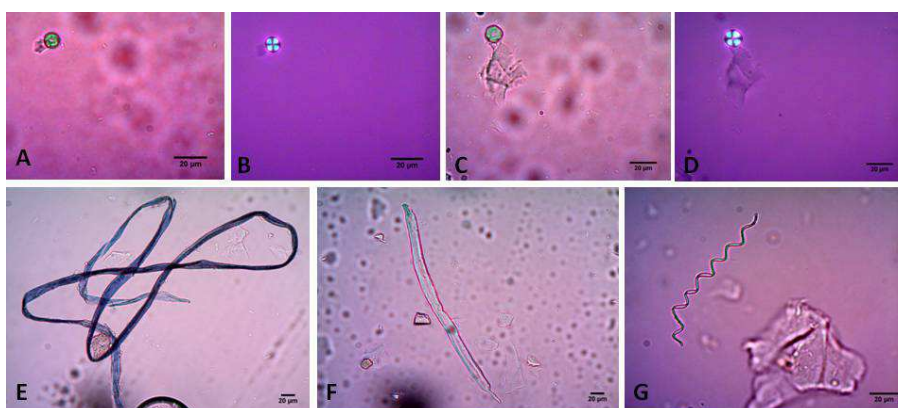


Figure 4.3.2.5: calculus micro-remains from Cr 70.1: starch grains under regular light (A and C), starch grains under polarized light (B and D), a textile fibre, probably cotton (E), an undetermined vegetal fibre (F), a spiral microstructure from a plant xylem (G). Scale bar 20  $\mu\text{m}$

## Cr 76.1

Individual 76.1 is an adult female, between 35 and 45 years old. The state of preservation is moderately good, although the skeleton is incomplete and fragmented. Dental calculus was removed from maxillary right second premolar (tooth 15) and from maxillary right first molar (tooth 16). The samples were treated with 10% hydrochloric acid for one hour. Some granules of starch were observed in both samples. In sample 76.1.a starch grains (tooth 15, fig. 4.3.2.6 A) were found embedded within a matrix.

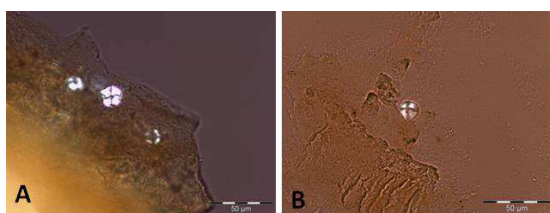


Figure 4.3.2.6: Calculus microfossils from Cr 76.1.a and 76.1.b: granules embedded into a matrix (A), an undetermined starch grain (B). Scale bar 50 µm

As observed, the better-preserved starch is pentagonal in shape with a central hilum. The arms of the extinction cross are thin and probably wider towards the ends. The diameter of the granule is ca. 17 µm whereas the diameter of the other two grains is ca. 12 µm. In the same sample, other granules between 12 and 17 µm in diameter were observed. Due to the size, the shape and the characteristics of the extinction cross, the granules are similar to those of wheat and spelt. In Cr 76.1.b (tooth 16), only a few starch grains were observed. Figure 4.3.2.6 B shows a granule with a central hilum and the arms of the extinction cross are large. Its diameter is ca. 13 µm.



## Cr 77.1

Within grave 77.1 was buried an adult male between 35 and 40 years old. The individual shows anomalous dental erosion in the anterior teeth of the maxilla (fig. 4.3.2.7). The left dental arch showed a malocclusion (Class II) and an important periodontitis. Dental calculus was analysed in every tooth.

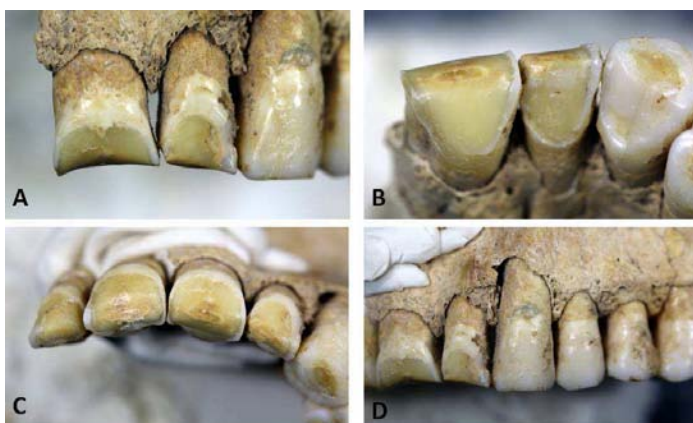


Figure 4.3.2.7: The anomalous erosion of the maxillary teeth of individual Cr 77.1: anterior view of the incisors (A), posterior view of the incisors (B), superior view of the anterior teeth, general view of the upper teeth (D)

Samples 77.1.a to 77.1.d were treated with 5% hydrochloric acid for one hour. Afterwards, they were rinsed and dissolved again with 10% hydrochloric acid for two hours and rinsed again. Samples 77.1.e and 77.1.f were dissolved in 5% hydrochloric acid over one hour.

**Sample 77.1.a:** dental calculus was extracted from the mandibular left second molar (tooth 37), marked by an important occlusal wear. Starch grains were found within this sample. Some of these granules was certainly cooked (fig. 4.3.2.8 A) whereas other starch (fig. 4.3.2.8 B) has different features compared to all the grains described in the previous chapter and in the other individuals of this section. Its diameter is ca. 78  $\mu\text{m}$  and the hilum is not central, with a small

fissure above it. This large granule is not easy to identify and probably it has come from a tuber (Hardy *et al.* 2009). Similar features were observed in *Dioscorea* (yam) and *Solanum tuberosum* (white potato) starch grains (Wesolowski *et al.* 2010, Babot 2006).

Furthermore, there are grains that share some features such as polyhedral shape, fissures and similar diameter (ca. 20  $\mu\text{m}$ ). These characteristics are similar to those of the grains of rye and sorghum. The last group of granules is relatively large and they are embedded within a dental calculus matrix (fig. 4.3.2.8 C and D). Some of the granules show features in common such as a spherical and oval shape and a central hilum. The only difference is the size, indeed grains can be divided in “small granules” whose diameter ranges from 5.5 to 9  $\mu\text{m}$ , and “large granules” whose diameter ranges from 19 to 23.8  $\mu\text{m}$ . These grains are similar to those of wheat and spelt. Within the sample, different types of fibres are present, and at least one of them is similar to those of wool. Moreover, tracheids and raphides are present.

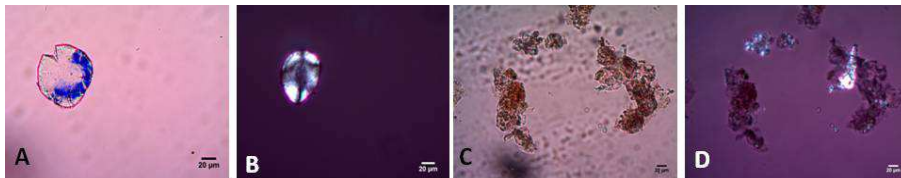


Figure 4.3.2.8: calculus microfossils from Cr 77.1.a: a cooked starch under regular light (A), an isolated large starch grain under polarized light (B), a group of starch grains under regular and polarized light (C, D). Scale bar 20  $\mu\text{m}$

**Sample 77.1.b:** dental calculus was extracted from the mandibular left canine (tooth 33). Different starch grains were found within this sample. The majority are spherical with a central hilum and the arms of the extinction cross are large and wider towards the ends (fig. 4.3.2.9 A). The diameters range from 6 to 28  $\mu\text{m}$  and they are similar to those of rye and sorghum. Other granules are oval in

shape, with a pressure facet and its diameter is 9.5  $\mu\text{m}$ . One granule shows several fractures and therefore it was probably cooked. Its diameter is ca. 39  $\mu\text{m}$  (fig. 4.3.2.9 B). Raphides, a fragment of a multicellular structure polyhedral from leaves or fruits of dicotyledonous plants (fig. 4.3.2.9 C), microstructures of xylem such as vegetal fibres and tracheids (fig. 4.3.2.9 D) are also observed (Albert and Cabanes 2007).

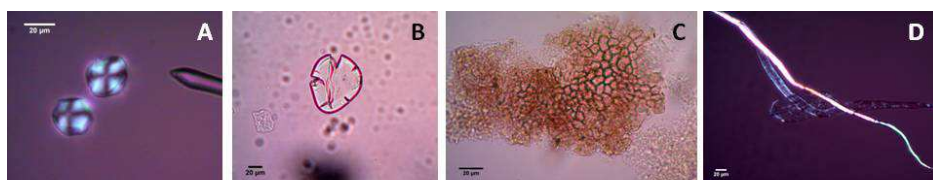


Figure 4.3.2.9: calculus micro remains from Cr 77.1.b: starch grains under polarized light (A), a cooked starch under regular light (B), a fragment of a multicellular structure polyhedral (C), fibres and tracheids under polarized light (D). Scale bar 20  $\mu\text{m}$

**Sample 77.1.c:** calculus was removed from the maxillary left third molar (tooth 28). Starch grains were found within this sample, Some of them are spherical, with a central hilum, arms of the extinction cross are wider towards the ends and Some exhibit broken arms. The diameter ranges from 4  $\mu\text{m}$  to 14.5  $\mu\text{m}$ . These type of grains are similar to those of wheat, spelt, rye and sorghum. Other granules are polyhedral and their diameter is ca. 16  $\mu\text{m}$ . The remaining starches are large (ca. 47  $\mu\text{m}$ ), oval shaped with a non-central hilum (fig. 4.3.2.10 A). A similar large starch grain was also observed in sample 77.1.a, however the extinction cross is different compared to the cross of the previous grain. In this case, the grain is similar, for example, to those of white potato and tropical plants such as *Metroxylum sagu* of the *Arecaceae* family (Babot 2006, Torrence *et al.* 2004). Other remains observed in this sample are raphides, undetermined vegetal remains and fibres (fig. 4.3.2.10 B and C). In figure 4.3.2.10 D two possible flax fibres (linen) were observed.

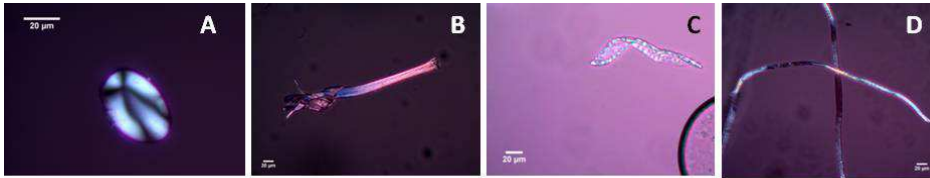

































Figure 4.3.2.10: calculus microfossils from Cr 77.1.c: a large starch grain under polarized light (A), an undetermined vegetal fibre (B), a undetermined vegetal remain (C), possible flax fibre (D). Scale bar 20 µm

**Sample 77.1.d:** dental calculus was removed from the maxillary left second premolar (tooth 25). Single and agglomeration of starch grains were found within this sample. The granules are described in table 4.3.2.3.

The aggregate granules overlap (fig. 4.3.2.11 F) and therefore their features were difficult to observe. Some of these grains have a spherical shape, pressure facet and central hilum. All of them are less than 20 µm in diameter. Among the isolated starch grains occur granules that exhibit features in common with those of sorghum, rye, wheat and spelt, however some of them show features that were not observed in other samples. These grains, illustrated in figure 4.3.2.11 A, B, C and D, are similar to those analysed from the edible terrestrial fern *Angiopteris yunnanensis* (Yang *et al.* 2013). Probably they come from other plants that were eaten for medical or other purposes. This sample also contains fragments of fibres, raphides, vessels with bordered pits (fig. 4.3.2.11 G and H) and undetermined vegetal remains.

Table 4.3.2.3: description of a sample of starch grains found in 77.1.d

Starch code	Diameter	Shape	Extinction cross	Other features	Cooked
1	58.9 $\mu\text{m}$			Not central hilum, damaged	+
2	29 $\mu\text{m}$			Central hilum, Fissure	-
3	16.2 $\mu\text{m}$			Central hilum, Fissure	-
4	14.5 $\mu\text{m}$			Central hilum	-
5	38 $\mu\text{m}$			Not central hilum, damaged	-
6	14.7 $\mu\text{m}$			Central hilum, pressure facet	-
7	17 $\mu\text{m}$			Central hilum, pressure facet	-
8	30.7 $\mu\text{m}$			Central hilum	-
9	17.7 $\mu\text{m}$			Central hilum	-
10	16.5 $\mu\text{m}$			Central hilum	-
11	10.3 $\mu\text{m}$			Central hilum, pressure facet	-
12	8.9 $\mu\text{m}$			Central hilum, pressure facet	-
13	13.6 $\mu\text{m}$			Central hilum	-
14	14.9 $\mu\text{m}$			Central hilum	-
15	8.5 $\mu\text{m}$			Central hilum	-
16	7.7 $\mu\text{m}$			Central hilum	-
17	24.1 $\mu\text{m}$			Central hilum	-

Legend: + = presence, - = absence

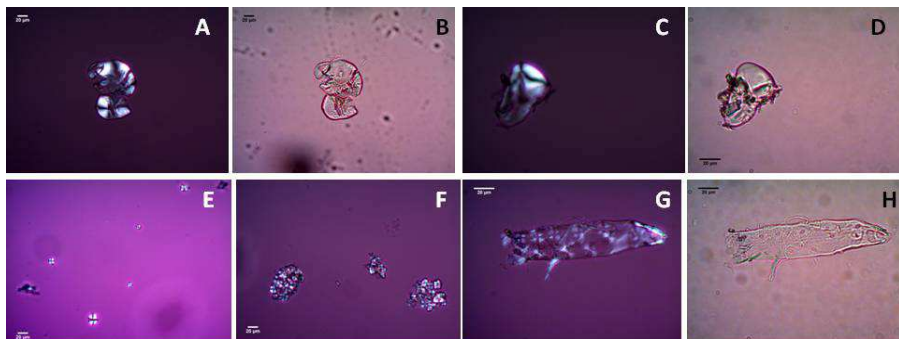


















Figure 4.3.2.11: calculus microfossils from Cr 77.1.d: damaged compound starch grains under polarized and regular light (A and B), damaged starch grains under polarized and regular light (C and D), various single starches (E), three groups of starch grains under polarized light (F), vessel and bordered pits under polarized and regular light (G and H). Scale bar 20  $\mu\text{m}$

**Sample 77.1.e:** calculus was removed from the maxillary left third molar (tooth 28). Starch grains were found within this sample. Some of them are very large (40 and 46  $\mu\text{m}$ ) with a spherical and oval shape and non-central hilum. The other granules exhibit a central hilum. Two types of shape were observed: a) some grains are spherical with the arms of the extinction wider towards the ends. The largest grain (20.7  $\mu\text{m}$ ) has a fissure above the hilum, other grains exhibit diameters of 9  $\mu\text{m}$ , 14.7  $\mu\text{m}$  and 11.8  $\mu\text{m}$ ; b) the other grains (19.8 and 17.6  $\mu\text{m}$ ) were oval and polyhedral with pressure facets. Some granules are similar to those of rye and sorghum whereas the largest grains shared features in common with the large grains described in the previous samples. Fibres and other undetermined vegetal remains were also observed within this sample..

**Sample 77.1.f:** calculus was removed from the maxillary left second incisor (tooth 22). In general, starch grains described in table 4.3.2.4 are similar to those of rye, sorghum and wheat. The grain shown in figure 4.3.2.12 A and B is similar to those of the *Fabaceae* family, such as *Pisum sativum*, *Vicia faba*, *Cicer arietinum* and *Phaseolus vulgaris* (Aceituno and Lalinde 2011, Henry 2012, Henry and Piperno 2008, Hart 2011). We also observed a similarity with some elongated grains of the wheat, analysed in the previous chapter.

Table 4.3.2.4: description of the 77.1 F starch grains

Starch code	Diameter	Shape	Extinction cross	Other features	Cooked
1	15.2 $\mu\text{m}$				-
2	12.7 $\mu\text{m}$				-
3	16.7 $\mu\text{m}$				-
4	17.7 $\mu\text{m}$				-
5	11.4 $\mu\text{m}$				-
6	28.8 $\mu\text{m}$				-
7	5 $\mu\text{m}$				-
8	40.2 $\mu\text{m}$			Damaged	+

Legend: + = presence, - = absence

Regarding other findings, a spore (fig. 4.3.2.12 C) and a fragment of tracheid (fig. 4.3.2.12 D) were also observed. The conidium was ovoid in shape and its size is 24 x 15 µm with 3 transverse septa. This is a spore fungus of the *Pleosporaceae* family, which could belong to the genera *Alternaria*, *Drechslera* or *Ulocladium* (Ellis 1971). They are among the most common allergenic fungi found in air. Other genera such as *Torula*, *Epicoccum* and *Puccinia* are excluded due to their size and morphology, whereas other species, such as *Curvularia* are excluded due to their geographical distribution.

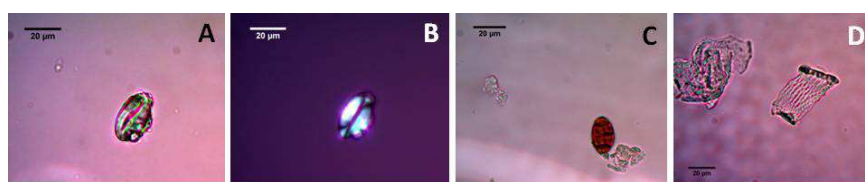


Figure 4.3.2.12: calculus micro remains from Cr 77.1.f: a probable legume (*Fabaceae*) starch under regular and polarized light (A, B), a spore (C), a fragment of tracheid (D). Scale bar 20 µm

## Cr 94.2

The individual buried in grave 94.2 is an adult male, between 26 and 45 years old, without particular pathological condition, except for a Schmörl's nodule on a thoracic vertebra. Dental calculus was removed from the mandibular right first incise (tooth 41) and mounted on a slide without chemical treatment. Only a few starch grains and a fragment of a possible human hair were observed. In figures 4.3.2.13 A and B a single starch is observed. Its diameter is 15.8 µm and the hilum is central and point-shaped. The extinction cross has thin arms, and one of them is broken or forked. In figures 4.3.2.13 C and D a fragment of an animal hair is observed. The shaft diameter is between 30 and 41 µm and the total length is more than 200 µm. The fragment observed in figure 4.3.2.13 D is probably a root. The general morphology is similar to those of human hair and

usually the overall shaft diameter of human hair ranges between 40-50  $\mu\text{m}$  to 110-120  $\mu\text{m}$  (Hicks 1977).

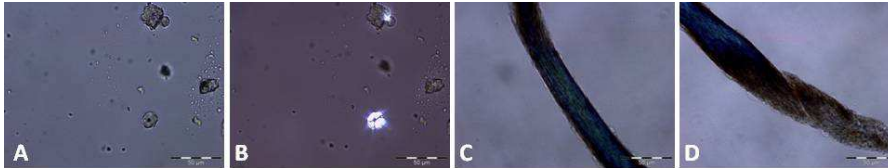


Figure 4.3.2.13: calculus microfossils from Cr 94.2: a starch grain under regular and polarized light (A and B), a fragment of a human hair (C and D). Scale bar 50  $\mu\text{m}$

### **Cr 119.1**

The individual buried in grave Cr 119.1 is an adult male, between 33 and 45 years old. Dental calculus was removed from the mandibular right first incisor (tooth 41) and mounted on slides without chemical treatment. Unfortunately, no micro-botanical components, except for some fibres, were found.

### **Cr 131.1**

Grave 131.1 contains an adult male, between 24 and 32 years old. The skeletal remains are well-preserved. No pathologies were observed. Dental calculus was removed from the mandibular left first incisor (tooth 31) and mounted on a slide without chemical treatment. Within the sample, a fragment of a nematode and some undetermined fibres were observed. Figure 4.3.2.14 A and B show a larva with a finely striated cuticle and a posterior non-sharpened extremity. Its length is ca. 900  $\mu\text{m}$ .

Table 4.3.2.5 shows some features of the intestinal parasites that can affect humans. All of them belong to the phylum Nematoda.



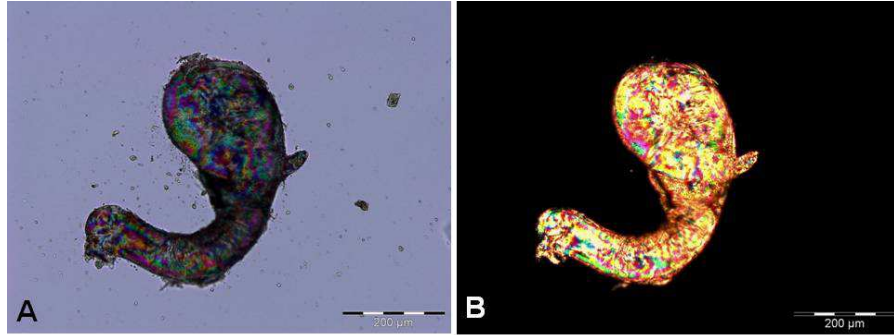


Figure 4.3.2.14: calculus microfossils from Cr 131.1: a fragment of a nematode (A and B). Scale bar 200  $\mu\text{m}$

Table 4.3.2.5: some features of the intestinal human parasites

	Size infest larva	Posterior extremity morphology	Place of the complete larva
<i>Strongyloides</i>	L3 Length: 400-700 $\mu\text{m}$	V sharpened	Intestine
<i>Ancylostoma</i>	L3 Length: 500-700 $\mu\text{m}$	Conical and sharpened	Intestine
<i>Trichostrongylus</i>	L3 Length: 500-700 $\mu\text{m}$	Sharpened	Intestine

In this study, the differential diagnosis has been made with other nematode species that can parasitize humans. The image allows excluding nematodes of the genera *Strongyloides*, *Trichostrongylus* or *Ancylostoma* since their larvae in the infective stage found in humans are all smaller, and the posterior morphology is sharpened (table 4.3.2.5). In addition, the location of the larvae is not compatible with the cycle of all species considered as *Strongyloides*, *Ancylostoma* and *Trichostrongylus*, which complete the life cycle in humans with the presence of the adult form evolved from larvae intestine (Grove 1989, Anderson 2000).

Therefore, the morphological and morphometric data and the location of the larva suggests that it could be a *Larva Migrans* of a roundworm, probably belonging to the genus *Toxocara*, although its location in the oral cavity has not been reported previously. Human toxocariasis is a major neglected disease caused by *Toxocara canis* and *Toxocara cati*, which are nematodes that often affect dogs and cats, respectively. These monoxenous nematodes complete their life cycles in the hosts mentioned. While they may also affect humans, in humans they cannot reach their full development, causing a larval parasitism known as *larva migrans* (Kłapeć and Borecka 2012, Rodriguez-Caballero *et al.* 2015). The human infection occurs when embryonated eggs that can contaminate surfaces, soil, water and vegetables are accidentally eaten. Ingestion of eggs and their hatching in the guts allows the spread of *Toxocara* larvae. These larvae exhibit a small diameter allowing their migration through capillaries and surrounding tissues until they reach their final location. Larvae will grow and develop at this location (0.50 to 1.24 mm in larvae 10 days) surrounded by a granulomatous reaction in which the host attempts to isolate it. The locations may be different, including the liver, lungs, brain and frequently the eyes (Ranasuriya *et al.* 2014, Anderson 2000).

### **Cr 132.1**

Grave 132.1 contains an adult male, between 25 and 30 years old. The skeletal remains are fragmented and incomplete. The dental arch shows a malocclusion (incomplete Class III) on the right-hand side. The left-hand side is fragmented. The dental calculus was extracted from the mandibular right second premolar (tooth 45) and treated with 5% hydrochloric acid for 18 hours. Only a few starch grains with similar diameter (12-16  $\mu\text{m}$  ) were observed, and corresponded to three morphologies. The first morphotype corresponds to grains that are embedded into a calculus matrix with spherical shape and central hilum. The second morphotype, probably damaged, is polyhedral. The third morphotype is oval. The border of the third morphotype is broken and this feature is probably due to a cooking process. The characteristics of these three

morphotypes do not help the identification of a particular cereal or plant. Figure 4.3.2.15 shows other vegetal remains observed within this sample. In figure 4.3.2.15 A a fragment of a multicellular structure polyhedral from leaves or fruits of dicotyledonous plants was observed (Albert and Cabanes 2007). Figure 4.3.2.15 B exhibits well-preserved tracheary elements: a tracheid and a vessel. The tracheid is an elongated cell present in the xylem of vascular plants that serves in the transport of water and mineral salts, whereas the vessel elements are the building blocks of the water transportation system of the plants. The main difference between tracheids and vessels is that vessels display perforations at the end plates, which make them a tube-like, long structure, whereas tracheids do not have end plates. In addition, vessels show larger diameters compared to tracheids. Vessel elements are typically found in flowering plants (angiosperms) (Friis *et al.* 2011). Figure 4.3.2.15 B shows a fragment of a tracheid with a diameter of 15.5  $\mu\text{m}$ . Pits are visible along the walls. The diameter of the vessel element is about 65  $\mu\text{m}$  and the length is about 247  $\mu\text{m}$ . A simple perforation plate and alternate pits are visible. There are many families of plants that show similar characteristics, such as *Malvaceae* and *Lythraceae*, in which the vessels show simple perforation plates. In some species of *Lythraceae*, vessel diameter ranges from 50 to 100  $\mu\text{m}$ , and the intervessel pits are round and arranged in opposite position (Schweingruber *et al.* 2012). These features are similar to those illustrated in figure 4.3.2.15 B. Figure 4.3.2.15 C shows some braided undetermined vegetal fibres.



Figure 4.3.2.15: calculus microfossils from 132.1: a fragment of a multicellular structure polyhedral (A), fragments of a vessel member and tracheid (B), braided vegetal fibres (C). Scale bar 20  $\mu\text{m}$

## Cr 144.1

Grave 144.1 contained skeletal remains that belong to an adult male, between 30 and 40 years old. The dental arch shows a malocclusion (Class II incomplete) in the left-hand side, whereas the right-hand side is fragmented. Dental calculus was extracted from the maxillary right second premolar (tooth 15) and divided in two sample. Sample 144.1.a was treated with 5% hydrochloric acid for one hour whereas sample 144.1.b was treated with EDTA for 25 hours. In sample 144.1.b no micro botanical remains were observed whereas in sample 144.1.a several starch grains and different types of fibres were observed. Some starch grains are spherical and exhibit a pressure facet. Their diameter is ca. 21  $\mu\text{m}$  and the extinction cross has large arms, wider towards the ends. The second typology of grain (12.5  $\mu\text{m}$ ) is spherical and the extinction cross has arms wider towards the ends. Another granule shares similar features, such as a polyhedral shape, a central hilum and the extinction crosses exhibit arms wider towards the ends. Their diameter is ca. 16  $\mu\text{m}$ . The features of these granules are similar to those of rye and sorghum.

The fibres observed in this sample are different from one another. Some display a diameter of 9.5  $\mu\text{m}$  and broken extremities (fig. 4.3.2.16 A). Figure 4.3.2.16 B shows a group of flat fibres and figure 4.3.2.16 C shows a fragment of an undetermined vegetal tissue and a cotton fibre with a diameter of ca. 19  $\mu\text{m}$ .

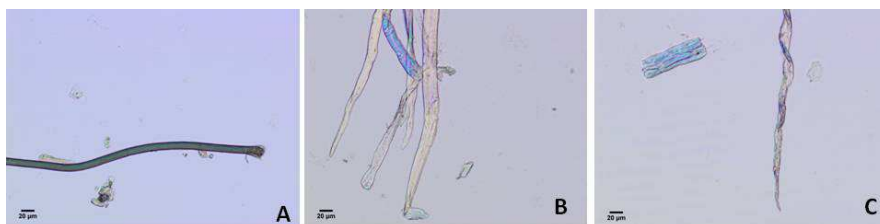


Figure 4.3.2.16: calculus micro remains from Cr 144.1.a: a undetermined fibre (A), a group of flat fibres (B), a cotton fibre (C). Scale bar 20  $\mu\text{m}$

## Cr 160.1

The individual buried in the grave 160.1 is an adult male between 25 and 35 years old. The skeleton is incomplete and therefore some information was lost. Dental calculus was extracted from two teeth and two different methodologies were applied. In sample 160.1.a, calculus was extracted from the mandibular left canine (tooth 33) and treated with EDTA for 17 hours. After this period of time the calculus was completely dissolved. In this sample starch grains, a possible hair cell phytolith, an undetermined organic material, pollen and some fibres were observed.

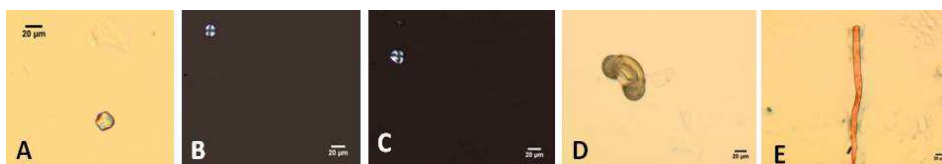


Figure 4.3.2.17: calculus microfossils from Cr 160.1.a: a starch grain under regular light (A), a starch grain under polarized light (B), a starch grain under polarized light (C), a pollen grain (D), an hair cell phytolith (E). Scale bar 20 µm

The first type of starch grain is polyhedral (fig. 4.3.2.17 A). The hilum is central and the extinction cross exhibits large arms, wider towards the ends. The diameter is ca. 18 µm. The second type (fig. 4.3.2.17 B) is oval with a pressure facet; its diameter is ca. 16 µm. The hilum is central and the extinction cross shows the same characteristics of the first grain. The third grain type (fig. 4.3.2.17 C) is polyhedral, shows a central hilum and its diameter is ca. 20 µm. The extinction cross exhibits broken arms. All three starches are well preserved and they are probably similar to those of sorghum and rye. Figure 4.3.2.17 D shows a pollen grain of the *Pinaceae* family (Moore and Webb 1978) and figure 4.3.2.17 E exhibits a hair cell phytolith. The pollen grain is bisaccate (or vesiculate) and is composed of a body with two laterally-placed bladders (sacca, vesicles). Its maximum length is 86 µm.

Dental calculus was extracted from the mandibular right second premolar (tooth 45) of sample 160.1.b and treated with 5% hydrochloric acid for 18 hours. The results are remarkable. Several micro botanical remains were identified in this sample. Simple starch grains and an aggregation of different granules were observed (fig. 4.3.2.18 C). Generally, these grains are oval and exhibit an elongated central portion of the extinction cross, whereas the arms are very short compared to the central part. Their diameters ranges from 5  $\mu\text{m}$  to 10  $\mu\text{m}$ . These features are similar to those of oat and farro starches, described in the previous chapter.

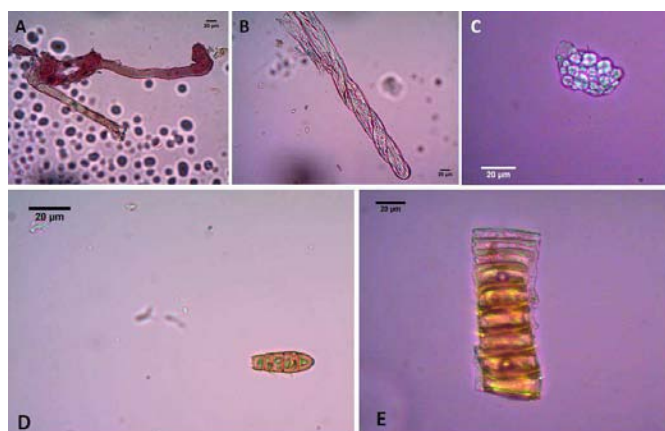




























Figure 4.3.2.18: calculus micro remains from Cr 160.1.b: a vegetal fibre (A), an undetermined twisted fibres (B), starch grains aggregation (C), a spore fungi of dreschlera or alternaria (D), a fragment of a fern annulus (E). Scale bar 20  $\mu\text{m}$

The characteristics of the other single granules are described in table 4.3.2.6. Granules 3, 5, 11, 12 and 13 exhibit some features in common such as polyhedral fissures, and diameter that range from 12  $\mu\text{m}$  to 22.7  $\mu\text{m}$ . They are probably similar to those of sorghum. On the other hand grains 1, 6, 7, 8 and 9 are spherical and oval, the arms of the extinction cross are wider toward the ends and their diameters ranges from 7.7  $\mu\text{m}$  to 27  $\mu\text{m}$ . These features are similar to those of wheat, spelt, and rye granules. Grains 2, 4 and 10 are unrecognizable due to their undiagnostic features. We conclude that this sample

includes starch grains from different plant species. Starches were divided in three groups: the first includes the aggregation of starches that are similar to those of oat and farro cereals; the second group includes granules similar to those of sorghum; the third group is similar to starch of wheat, spelt and rye. It is interesting to note that there are 6 out of 13 grains with fissures. In the previous chapter, fissures were observed only in rye, oat and sorghum granules. Two starches were probably exposed to high temperature because they show fissures on the border.

Table 4.3.2.6: description of the 160.1.b starch grains

Starch code	Diameter	Shape	Extinction cross	Other features	Cooked
1	23.5 µm			Fissure	+
2	10.9 µm			Deformed	-
3	22.5 µm			Fissure	-
4	9.4 µm				-
5	22.7 µm			Fissure	-
6	14.6 µm				-
7	17.7 µm			Fissure , pressure facet, broken border	+
8	27.3 µm			Damaged, broken border	+
9	7.7 µm				-
10	11.3 µm			Deformed	+
11	19.3 µm			Fissure, deformed	-
12	20.6 µm			Fissure, deformed	-
13	12 µm			Deformed	-

Legend: + =presence, - = absence

Some fibres were observed in sample 160.1.b. Figure 4.3.2.18 A shows a possible linen or jute fibre whereas figure 4.3.2.18 B shows an undetermined twisted textile fibre. Fragments of spiral tracheids were also observed in this sample. A conidium from a fungus spore of the *Pleosporaceae* family (fig. 4.3.2.18 D) was found. It could belong to the genera *Alternaria*, *Cladosporium* or *Drechslera* (Ellis 1971). The spore size is 29 x 11 µm with 3-4 transversal septa. No longitudinal septa were observed.

Figure 4.3.2.18 E shows a fragment of a fern sporangium annulus. A sporangium is the reproductive structure of a fern and resembles a capsule in which spores are formed, protected and finally expelled. Aggregates of sporangia are called sori. A sporangium is composed by annulus and spores. The annulus is a ring of cells (basals and indurated) around the sporangia of many ferns that splits and allows the liberation of spores. The geographic position and climate of Mallorca and the absence of the annulus in some species of ferns, point to families such as *Adiantaceae*, *Hypolepidaceae*, *Polypodiaceae*, *Aspleniaceae*, *Athyriaceae* and *Dryopteridaceae* (<http://www.uib.cat/depart/dba/botanica/herbari/alfabetica/A.html>, Marchetti 2003, Murtaza *et al.* 2006). For instance, the *Blechnaceae* family is present in the northern regions of the Iberian Peninsula (Quintanilla *et al.* 2007). The most interesting species present in Mallorca Island are *A. Balearicum* and *Scolopendrium* of the *Aspleriaceae* family, *P. Cambricum* of the *Polypodiaceae* family and *D. Thyrrena* and *Pallida* of the *Dryopteridaceae* family (fig. 4.3.2.19). *D. Thyrrena* is a western Mediterranean endemism and its current distribution leads to hypothesise a Hiber-Provenzal-Thyrranian origin. In Spain, it is present only in Mallorca and in Sierra Nevada. Probably, the species is a relic of the Tertiary flora of the Mediterranean mountain (Magrini 2012). More interesting is *Polypodium Cambricum*, that has medicinal properties and it is known since the 1<sup>st</sup> century AD, when Pedanius Dioscorides reported this plant in the *Materia Medica*. He wrote that Polypodium, identified with the simple Lonchitis or *P. cambricum* (Lev and Amar 2008), could be served boiled



as a purgative. Dioscorides also reported that if the Polypodium is dried, powdered and sprinkled into honey and water, it expels phlegm and bile. The root is helpful for cleaning, and for cuts between the fingers (Gunther and Goodyer 1959). Dioscorides mentioned also the “*scolopendrium*”, called by the Romans “*filicula licitalis*”, which is probably the name of the *Asplenium* or *Phyllitis scolopendrium* (deer’s tongue or hart’s tongue), a species of the *Aspleniaceae* family used in antiquity as a medicament. In the past, people who lived in rural settings, especially in England, attributed to ferns, and in particular to the hart’s tongue, magical properties (Page 1997).



Figure 4.3.2.19: *Polypodium Cambricum* and *Phyllitis scolopendrium* geographical distribution, modified from Page 1997

### Cr 169.1

Grave 169.1 contained an adult male, between 25 and 35 years old. The skeletal remains showed several pathologies such as a gonarthrosis of the right knee, a fracture of the left radio and ulna, a fracture of the fifth right metacarpal and eburnation on the right scaphoid. Dental calculus was removed from the mandibular right second premolar (tooth 45) and treated with 5% hydrochloric acid for one hour. After rinsing it was dissolved with EDTA for 17 hours. Within the dental calculus occur carbonized organic matter, the same of

individual 17.1, and three bisaccate pollen grains with diameters of ca. 58  $\mu\text{m}$  from the *Pinaceae* family (Moore and Webb 1978).

#### 4.3.2.4 Discussion

Dental calculus was collected from 17 teeth that belong to 12 adult individuals of Can Reiners. The sample is composed of 10 males and 2 females and the age at death ranges from 25 to 59 years old. The individuals were buried in simple pits in different areas of the necropolis. The location of dental calculus is generally in the lower incisor and in the lower left dental arch especially in buccal (35%) and lingual (41%) position.

Under the microscope, samples without chemical treatment were difficult to analyse due to the presence of thick calculus fragments that hampered proper identification of several micro-remains. Therefore the procedure adopted includes rinsing the calculus fragment with distilled water and grinding to a powder using an agate mortar and pestle. As a result, only a few samples were observed without the aid of acid treatments. Nevertheless, untreated samples show different types of remains. For instance, a fragment of a nematode was observed in one of these samples (Cr 131.1), whereas in the samples treated with acids, no animal remains were found. These may have been destroyed by the acid step in the case they were present in the original assemblage. The fragment of a roundworm could be a *Larva Migrans*, probably belonging to the genus *Toxocara*. Human Toxocariasis is a major neglected disease caused by *Toxocara canis* and *Toxocara cati*, which are nematodes that often affect dogs and cats, respectively. This is a unique discovery in dental calculus analysis. Publications about ancient intestinal parasites are more frequent in relation to archaeological contexts (Bouchet *et al.* 2002, Dittmar *et al.* 2012, Florenzano *et al.* 2012), whereas reports regarding intestinal parasites in dental calculus are unusual. Charlier *et al.* (2013), for example, have identified a *Schistosoma mansoni* egg of 100  $\mu\text{m}$  maximal length in the dental calculus of a 9th c. AD

individual from France. To the best of our knowledge, no fragments of roundworms have been found to date. Therefore, this finding is exceptional.

Regarding samples treated with acids, no differences were observed among the various methods. Starch grains, for example, are not present in all the samples. Five samples lack starch (Cr 33.1, Cr 119, Cr 131, Cr 144 B, Cr 169.1) and were treated with HCl, EDTA, and water. We left samples in HCl from 1 to 21 hours and in EDTA from 17 to 25 hours and the majority of microfossils was still present at the end of the procedure. Therefore, we conclude that the duration of the acid step does not influence the results and it does not change the morphology of the grains. It was observed that EDTA created a gelatinous mass that complicates the extraction of dental calculus. For this reason, it was not used for a large number of samples. Glacial acetic acid was used in some samples but it was noted that it does not dissolve the calculus. Therefore, we opted for HCl.

Phytoliths were observed in 4 individuals and they consist of hair cells, phytoliths belonging to tracheids or dicotyledonous fruits, and polyhedral multicellular microstructures from leaves or fruits of dicotyledonous plants (Albert and Cabanes 2007, Piperno 2006). Some undetermined phytoliths were also found.

Fibres are difficult to recognise and generally are categorised as undetermined (Cr 33.1, Cr 70.1, Cr 77.1, Cr 119.1, Cr 131.1, Cr 132.1, Cr 144.1). Some vegetal fibres show similar features to those of libriform fibres (Cr 70.1), linen (Cr 77.1 C and 160.1) and cotton (Cr 70.1 and 144.1). In one case, a possible wool fibre was observed (Cr 77.1 A). The cotton fibre found in the adult female Cr. 70.1 seems to be blue in colour. In sample Cr 94.2 a possible human hair was observed.

Xylem elements, such as tracheids and vessels, were observed in many samples (Cr 70.1, Cr 77.1 A, Cr 77.1 B, Cr 77.1 D, Cr 77.1 F, Cr 132.1, Cr 160.1 B), whereas various fragments of vegetal remains were not identified.

In 2 individuals fragments of carbonized organic matter were found (Cr 17.1, Cr 169.1), whereas in 3 individuals (Cr 17.1, Cr 160.1, Cr 169.1) pollen grains that belong to the *Pinaceae* family were observed.

Fungal spores were observed in two samples and they could belong to the genera *Alternaria*, *Drechslera*, *Cladosporium* and *Ulocladium*. The species of this genus are plant pathogens and in the genus *Ulocladium* they contain both plant pathogens and food spoilage. Spores are helpful because they can reveal the relationship between population and environmental features, such as climatic conditions and geographical changes (Ma *et al.* 2000).

Regarding starch grains, we observed that some of them were not identifiable due to their undiagnostic features. However, the identified starch grains could belong to grass seeds including wheat (Cr 33.1, Cr 76.1, Cr 77.1, Cr 160.1), spelt (Cr 33.1, Cr 76.1, Cr 77.1, Cr 160.1), rye (Cr 17.1, Cr 33.1, Cr 70.1, Cr 77.1, Cr 144.1, Cr 160.1) and sorghum (Cr 17.1, Cr 70.1, Cr 77.1, Cr 144.1, Cr 160.1). Conglomerations of starches were observed in individuals Cr 77.1 and Cr 160.1. In the latter case starches could belong to oat and farro cereals. Only in one case, a starch grain of the *Fabaceae* family was observed (Cr 77.1 F). Some of the recovered starch grains that belong to the adult male Cr 77.1 have provided evidence of damage consistent with cooking and processing food. These starch grains display alterations in morphometric characteristics such as several fractures and the loss of the extinction cross. The different samples of individual CR 77.1 showed the existence of particular types of starches. These granules are different compared to all the grains described in the previous chapter and the other samples examined. It is possible that some of these unidentified starches come from tubers (Babot 2006, Hardy *et al.* 2009, Wesolowski *et al.* 2010) or in the case of compound grains, from an edible terrestrial fern (Yang *et al.* 2013). However, this is currently speculative as we do not yet have modern reference starch collections of plants taxa with respect to the island of Mallorca. In any case, it is intriguing that this is the only

individual that shows these particular types of starch grains, together with other starch grains types and a special type of wear.

Another fascinating finding is the discovery of a fragment of a fern sporangium annulus in the dental calculus of the adult male 160.1. The annulus releases the spores into the air while it is attached to the leaves. For this reason, it is possible to suppose that the individual ate a part of a fern. Ferns have edible leaves and since the Roman period they were used for several medicinal purposes and believed to have magical properties (Gunther and Goodyer 1959, Lev and Amar 2008, Page 1997). We find that species of fern such as *Polypodium cambricum*, *Asplenium* or *Phyllitis scolopendrium* used in antiquity as a medicament could be present in the island of Mallorca during late Roman and medieval period.

In summary, we may be seeing evidence of a rural group relying on a variety of cultivated cereals (wheat, spelt, rye, sorghum and probably oat and farro) for their own nutrition. However, it is unclear what other plants made up the majority of the diet of this small group of individuals analysed in the present study, and whereas we can speculate about other issues. We are probably dealing with a community that lived close to domestic animals and could be infected by their parasites (Toxocariasis) and therefore may have used the same type of medication popular in Roman times (i.e. consumption of fern leaves). Also the presence of textile fibres, some of which are coloured, provides information on other aspects of the life of this group, as it could represent manual labour with the help of the teeth.

Finally, it is interesting to note the high concentration of different starch grains, fibres, and vegetal remains that characterize the adult male buried in grave 77.1. This is one of the three graves of the necropolis that contain coins. Near the hands a small bronze of *Probus* (276-282) was found (Arribas and Tarradell 1987). Furthermore, the individual shows anomalous non-alimentary dental erosion on the incisive teeth of jaw and mandible. All these pieces of information could point out that this individual came from another place, that he

conducted a particular activity or that he played a different role within this community.

#### **4.3.2.5 Conclusions**

The most common microfossils detected in Can Reiners dental calculi are starch grains, fibres and phytoliths. However, also other remains such as spores, pollen and even nematodes were observed. The analysis of starches pointed out the use of wheat, spelt, rye, sorghum and probably oat and farro in the diet of the individuals analysed, probably cooked as indicated by the gelatinous substance that embedded grains in some samples and by the presence of carbonised elements. In addition, it is possible to demonstrate the presence of some parasites and the presence of some plants with medicinal uses. Our results showed that dental calculus is a powerful tool for palaeonutrition, paleopathology and paleoenvironment analyses

## **5.GENERAL DISCUSSION**





## **5.1 The population of Can Reiners: demography and life conditions on the Mallorca island in the 7th c. AD**

The bioanthropological study of the necropolis of Can Reiners has a fundamental role on the reconstruction of the biological profile of the ancient populations that inhabited the island of Mallorca. Can Reiners is one of the three large cemeterial assemblages from the Balearic Islands that were entirely analysed.

The studies conducted in the Talayotic necropoles of S' Illot des Porros (Alesan *et al.* 1992, Malgosa 1992) and Son Real (Font 1977) provided a useful basis to analyze the evolution of the population of the Balearic Islands.

The individuals of the necropolis of Can Reiners were buried in the area of the ancient forum of the Roman city of *Pollentia*. The archaeological data provided information about the general skeletal position, the grave types and the presence of grave goods. Unfortunately, the photographic material was not complete and consequently we could not obtain additional information. The description of the general positions of individual (supine, with inferior limbs extended and upper limbs situated in different position) allowed identifying the burials as primary depositions. There were four graves typologies: 1-simple pit dug into the ground or within the bases of monuments or other structures, 2- pit covered by stone slab, 3- stone lined grave covered by stone slabs (cist), 4- mound tomb or tomb with banquet. Generally, no grave goods or other material were found in the burials, except in three cases in which three coins were recovered.

We observed that the general state of preservation of the skeletal material was good (72.7%), and superior and inferior limbs were better preserved than other skeletal remains. In addition, males were better preserved than females. Most authors acknowledge bone size and strength as an important factor for enhanced preservation; therefore male skeletons are, in general, better preserved.

Generally, all the skeletal remains were better preserved in the simple pit burials. No correlation was found between grave types and sex and age at death. Regarding the number of individuals contained in each burial we observed that the stone lined grave covered by stone slabs and mound tomb or tomb with banquet were frequently re-used. It is possible to hypothesize that they were employed as family burials like current pantheons.

A total of 216 individuals were analyzed in the present study. Among them we identified 138 adults (63.9%) and 78 non-adults (36.1%). Regarding adult groups, we observed that there were few mature individuals whereas no senile were found. The lacking of the senile category could be due to methodological errors, although we suggest that the individuals of Can Reiners did not reach, due to hard life conditions, the senile age. The lack of senile individuals was corroborated by the low values of diseases related to the age at death, such as osteoarthritis. The mortality was not balanced between sexes, being the sex ratio (80:55) favourable to males. We observed in fact that the probability of death of adult males was twice the probability of death of adult females, whereas for mature individuals the ratio was balanced (13:14). Where are females? We ruled out a methodological error because the skeletal remains of Can Reiners showed a remarkable sexual dimorphism. Therefore, we suppose that the reduced number of females is due to the fact that they were buried in a different area of the necropolis or that the community of Can Reiners had a higher male presence due to the military function assumed by *Pollentia* since the 5<sup>th</sup> century. The former possibility arises from the large extension of the necropolis in contrast to the partial sector studied in this thesis. The latter hypothesis is suggested by the archaeological finds: the area of the forum, indeed, was previously fortified and the city of *Pollentia* was occupied by Vandals and Byzantines respectively in the 5th and 6th century. However, this hypothesis contrasts with the lack of evidence of violent traumatism. In addition, among the possible explanations, we should note that in early

medieval Italian cemeteries, the same sexual dimorphism has been interpreted as the result of infanticide of female newborns (Giovannini 2001).

The frequency of non-adult mortality is 36.1% and from birth to first year is ca. 7%. Generally, the expected values for the first year of life should have been between 30% and 50% (Angel 1969, Brothwell 1986-87). Therefore, as expected, there was a sub-representation of immature individuals. We suggest to take into account that if there was a sub-representation of females, lower frequencies of subadults are justified.

The life tables and the mortality curve provided others useful data: the highest peak of mortality for individuals of Can Reiners is between 35 and 40 years, the life expectancy at birth is ca. 26.6 years of age and the crude mortality rate is 37.59%. We can conclude that the life conditions in this community were hard and similar to those of the protohistorical populations of S' Illot des Porros and Son Real that lived in the same geographical area.

Regarding the estimation of height, we observed significant differences between the four methodologies employed in this study. The mean of the stature of males ranged from 165.2 (Pearson1899) to 168.5 (Trotter and Gleser 1952, 1977), and for females ranged from 152.5 (Pearson1899) to 156.6 (Trotter and Gleser 1952, 1977). The estimation of height was compared with some populations that lived in Spain, France and Italy in different chronological periods. We observed that the male stature was similar to the general trends of the other populations, whereas the values of females were rather lower in Can Reiners than in the comparative sample, except for Roman females. The mean of the female stature seems to maintain the same values of the Roman period. Female values in Can Reiners were lower than those of the prehistoric inhabitants of the same geographical frame. Being that stature is considered one indicator of health conditions, the population of Can Reiners seems to be faced to hard living conditions.

To characterize more in particular our sample, the paleopathological analyses played a fundamental role. We observed that few diseases related to age at death were observed. There were few cases of osteoarthritis in the skeleton and the study of degenerative vertebral changes revealed that the onset of osteoarthritis in the thoracic vertebrae and the formation of the Schmorl's nodes in all the vertebral column were related with the mechanical stress that the individuals were exposed to. Traumas were present in few individuals, similarly to S'Illot des Porros, which is consistent with the lack of evidences of lesions suspected to be due to violence/war actions. The incidence of tumours and infections among the population was less than 2%. The results of the oral pathologies were compared with two protohistoric populations of Mallorca: S'Aigua Dolça and S'Illot de Porros. Results showed an increase of dental decay, *ante mortem* tooth loss, fistulas and enamel hypoplasia in the Can Reiners individuals. Nevertheless, the frequency of oral pathologies in Can Reiners was similar to other necropolis of the same historical period (Ortega *et al.* 2003). *Cribra orbitalia* was present in 64.5% of the individuals, with no differences among sexes. The findings of the study of *cribra orbitalia* seem to reflect the poor life conditions of the inhabitants of this area and matches with the demographical analysis.

A striking finding that we observed in the analysis of dental calculus was the presence of a nematode. The roundworm, identified as a *larva migrans*, probably belonging to the genus *Toxocara*, indicated the possible presence of intestinal parasites that could be responsible of anaemia and infections in the community.

## **5.2 The study of dental occlusion: a new approach based on dental clinical practice**

The analysis of dental occlusion could only be applied to 33 individuals, all of them with permanent dentition. A good preservation of archaeological skeletal

material is not usual and few studies concerning dental occlusion analysis have been hitherto done. Therefore we decided to elaborate a methodology based on clinical dental practice suitable for fragmented skeletal remains (Fiorin *et al.* 2014). Generally, authors studying ancient well-preserved skeletal material state that prehistoric populations exhibited a correct or ideal dental occlusion, whereas with the passing of the centuries and the change of diet, there was an increase of malocclusion (Evensen and Øgaard 2007). This tendency is too simplistic, because multiple factors could be involved in the development of the maxillary system structure. Genetics and environmental factors played an essential role in dental position variation. Recent studies, indeed, demonstrated that malocclusion was present, with high frequencies, also in prehistoric populations and authors suggested that malocclusion is due to genetic factors instead of an excessive tooth size and environmental change (Mockers *et al.* 2004). In this framework we decided to develop a straightforward lab sheet in which the main methodologies used by orthodontists were employed and to apply the methodology to the ancient skeletal remains.

The aim of the study was to determine if in a sample of the ca. 7th c. AD the dental variation features were more related to prehistoric populations or to modern ones. The results showed that normocclusion was present in ca. 70% of the samples, whereas some values, such molar relationship, were not very different to the modern ones. We observed statistically significant correlation between canine and molar relationships (Angle's classification) and between molar relationship and inferior molars and premolars dental wear. Therefore we suppose that dental wear could be the influencing factor of the onset of molar malocclusion (Class III) in Can Reiners sample. Diet and habits, such the use of the mouth as a tool, thus may influence the occlusion patterns. Regarding variables such as spacing characteristics and some occlusal features such as overjet, overbite and openbite we observed that they were not frequent. Dental crowding, for instance, was present only in four individuals and did not affect their occlusion. Pathologies such as dental decay and tooth loss were not very

frequent in this small sample of the population and affected only adults and mature individuals.

Our sample exhibited characteristics that could be observed on ancient (such as high dental wear and molar relationship Class III) and modern (crowding, crossbite, among others) skeletal material. Therefore, Can Reiners neither supports nor confirms the theory that malocclusion is higher in modern than in ancient populations. We compared the values of molar relationship with ancient and modern samples and we observed that usually in modern populations the values of Class II increase at the expense of Class III, whereas in ancient samples the opposite occurs. By contrast, in our population these percentages are rather similar (right dental arch for Class III and left dental arch for Class II) and the values of Class III are not as high as in the other ancient samples.

Regarding transversal and sagittal measurements, modern studies observed that usually they are dependent on each other (Lindsten *et al.* 2012), whereas in Can Reiners no correlation was observed between the arch width and the arch depth measurements.

We observed that dental wear, in addition to molar relationship, also influences some variables such as the curve of Spee, the superior length, and the inferior intermolar width. Regarding the curve of Spee, references on ancient skeletal remains are scarce. Sengupta *et al.* (1999) examined the effect of dental wear on the curve of Spee in a medieval archaeological population with a high rate of dental wear. Their data suggested that there was no straightforward relation between the Spee curve and occlusal wear. By contrast, our findings showed a significant association between Spee curve and dental wear, involving in particular the upper premolars and molars, and also the teeth of the right mandibular dental arch. This association is not present in the modern sample due to the absence, even in the mature individuals, of heavy dental wear.

Generally the population of Can Reiners did not show a marked dental malocclusion. Few cases of anomalies such as dental crowding, overjet and

openbite were present. Concerning malocclusion, the situation of our sample concurs with its chronological position, between the prehistoric and modern patterns.

### **5.3 Dental calculus analysis: the creation of an atlas and the case study of Can Reiners**

The intention to create an atlas of microphotographs of micro-botanical remains, and in particular of starch grains, arises from the necessity to have reference to recognize the microremains in the samples of dental calculus. Regarding phytoliths, fibres, pollen grains and other microremains there are several atlases of references. In the last decades several studies regarding starches have been published, although they have different chronologies and geographical location from our sample (Mickleburgh *et al.* 2012, Piperno *et al.* 2009, Rosewig *et al.* 2014). Consequently, the characterization of starch grains of cereals or other types of plants of the Mediterranean area lacks in the literature.

The atlas initially focused on the cataloguing of the most diffuse cereals in the ancient Mediterranean area. The palynological and carpological studies carried out in the Balearic Islands demonstrated the presence of some types of plants, useful for the reconstruction of the diet of a population, since prehistoric periods. From 4000 BP and 2000 BP, for instance, the Mediterranean predominant trees in Mallorca and in Minorca were *Olea* and *Quercus* species (Hernández-Gasch *et al.* 2002). Carpological studies documented the presence of barley, oats, figs, olives, blackberry, raspberry and beetroot for the prehistoric period in Minorca and Mallorca (Lull *et al.* 1999). In the cemetery of S'Illot de Porros, presence of caryopses of common wheat, grape seeds, juniper berry and cotyledons of acorns was observed in levels dated from 4th century to 2th c. BC (Hernández-Gasch *et al.* 2002). Regarding the Roman period the most important crop was wheat (*Triticum aestivum*). However, after

the crisis of the 3th century AD, the cultivation of cereals of lower quality such as rye, spelt, oat, millet, emmer wheat and sorghum was favoured because they are more resistant and productive than “*frumentum*” (Montanari 2012). Being aware of the hypothetical cultivation of different types of cereals after 3th c. AD and the presence of the holly oak in Mallorca, we decided to analyze the starch grains contained inside cereal seeds and fruits of holly oaks. To recognize the features of processed food, we boiled some samples and we observed the characteristics of the cooked starch grains.

Our findings showed several difficulties about the identification of the types of cereals based on starch grains. However, features such as shape, size and position of the hilum could be helpful in differentiating some of them. Generally, the analyzed samples exhibited simple starch grains, with the exception of oat grains, which are also compound. We also observed that some granules of spelt, barley and holly oak exhibited a particular shape that may be useful for their identification. The reniform shape was present only in spelt and barley, while several granules of holly oak displayed a “pear or drop” shape. Spherical shape was present in all the samples with the exception of the granules of oat, which were mainly polyhedral in shape.

Concerning size, the largest grains were observed in rye, emmer and wheat. Also granules of barley, spelt and sorghum showed a remarkable size. The grains of the remaining species were smaller than 20  $\mu\text{m}$ . Lamellae were visible in some granules of wheat, spelt and acorn, whereas fissures occurred only in oat, rye and sorghum. The hila of starch granules are usually central, point- or line-shaped, whereas in acorn granules the hila could be located in central or not central position due to its drop-shaped structure. In all the samples the extinction cross exhibited different types of angles and we considered that this feature was not useful for identifying species. We observed that the presence of one broken arm was characteristic of the spelt granules and the appearance of large arms was characteristic of rye and acorn grains. The Maltese cross (arms wider towards the ends) was observed in wheat, spelt, rye and sorghum.



Within the samples, some exceptions to the main characteristics could be observed. For instance, sorghum has an average diameter of 13  $\mu\text{m}$  and round shape, but some isolated large granules can occur (47  $\mu\text{m}$ ). They are oval and exhibit a hilum displaced from the centre.

In relation to food processing, all the boiled samples reached the gelatinization stage and the granules, in most cases, formed a mass in which it was impossible to discern single grains and their features. In spelt, barley and oat only a few starch grains have been observed. By contrast, rye and millet grains survived well the boiling process.

This first approach of the study of starch from Mediterranean cereals provided a reference atlas. In general and despite some problems related to some features of the granules, this atlas offers the main characteristics of the micro-residues from plant food, providing a useful tool for the identification of the species exploited by ancient populations for food consumption.

The study of dental calculus extracted from the teeth of some individuals of Can Reiners provided several interesting findings. Dental calculus was collected from 17 teeth that belonged to 12 adult individuals (10 males and 2 females). The location of dental calculus was generally in the lower incisors and in the lower left dental arch especially in buccal (35%) and lingual (41%) position.

Due to the lack of a standardized methodology, three different acids (hydrochloric, glacial acetic and EDTA) were tested. Samples treated with acids did not shown differences in the microremains composition among them. The duration of the acid step did not influence the results and it did not change the morphology of the grains. However, it was observed that EDTA created a gelatinous mass that complicates the extraction of dental calculus, whereas glacial acetic acid did not dissolve the largest pieces of calculus. Regarding samples without chemical treatment, we observed that they were difficult to analyse due to the presence of thick calculus fragments that hampered proper

identification of several micro-remains. Therefore, we suggest to use hydrochloridric acid.

Within the samples we observed different types of microremains. Phytoliths were found only in 4 individuals and they consist of hair cells, fragments of tracheids or dicotyledonous fruits, and polyhedral multicellular microstructures from leaves or fruits of dicotyledonous plants (Albert and Cabanes 2007, Piperno 2006). Fibres were difficult to recognise and generally were categorised as undetermined. Nevertheless, we recognized vegetal fibres similar to those of linen and cotton. In two case cases, a possible wool fibre and a fragment of human hair were observed. Xylem elements, such as tracheids and vessels, were abundant, whereas various fragments of vegetal remains were not identified. In 2 individuals fragments of carbonized organic matter were found, and in 3 individuals pollen grains that belonged to the *Pinaceae* family were observed.

Regarding starch grains, some of them were not identifiable due to the lack of diagnostic features. The identified starch grains could belong to grass seeds including wheat, spelt, rye and sorghum. Conglomerations of starches were also observed and they were similar to those found in farro and oat cereals. Only in one case, a starch grain of the *Fabaceae* family was observed. The starch grains of the adult male Cr. 77.1 showed particular features: some grains had provided evidence of damage consistent with cooking and processing food and others were different from all the grains of the rest of samples. It is possible that some of these unidentified starches came from tubers (Babot 2006, Hardy *et al.* 2009, Wesolowski *et al.* 2010) or in the case of compound grains, from an edible terrestrial fern (Yang *et al.* 2013). However, this is currently speculative as we do not yet have modern reference starch collections of plants taxa with respect to the island of Mallorca. In any case, it is intriguing that this is the only individual that shows these particular types of starch grains, together with a unique dental wear.

Fungal spores were observed in two samples and they could belong to the genera *Alternaria*, *Drechslera*, *Cladosporium* or *Ulocladium*. The species of this genus are plant pathogens and in the genus *Ulocladium* they contain both plant pathogens and food spoilage. Afonso-Vargas *et al.* (2015) detected in the dental calculus of two individuals from the church “La Concepción” (18th c. AD, Tenerife, Canary Islands) fungal spores of *Ustilago maydis*, a corn (*Zea mays*) parasite. Their results demonstrated the consumption of corn in the Canary Islands and the consumption of plants contaminated by the so-called “Corn smut” (*U. maydis*). These findings are important for the reconstruction of the diet and the environment of the individuals.

Another fascinating finding was the discovery of a fragment of a fern sporangium annulus in the dental calculus of the adult male 160.1. Ferns were used since the Roman period as edible plants and people believed that they had therapeutic and magical properties (Gunther and Goodyer 1959, Lev and Amar 2008, Page 1997). The annulus releases the spores into the air while it is attached to the leaves. For this reason, it is possible to suppose that the individual ate a part of a fern. We observed that species of fern such as *Polypodium cambricum*, *Asplenium* or *Phyllitis scolopendrium*, used in antiquity as a medicament, were possibly present in the island of Mallorca during late Roman and medieval period.

The most intriguing result was the discovering of a nematode within the dental calculus of an adult male of ca. 28 years of age (Cr 131.1). The roundworm could be a *larva migrans*, probably belonging to the genus *Toxocara*. Human *toxocariasis* is a major neglected disease caused by *Toxocara canis* and *Toxocara cati*, which are nematodes that often affect domestic animals. This is a unique discovery in dental calculus analysis because reports regarding *larvae* of intestinal parasites are unusual (Charlier *et al.* 2013). To the best of our knowledge, no fragments of roundworms have been found to date. Therefore, this finding is exceptional.

Regarding other studies performed on dental calculus in individuals from Mallorca, we report below the results observed in S'Illot de Porros (Pla et al. 2003) and in S'Aigua Dolça (Hernández-Gasch et al. 2002). Pla and coauthors in 2003 briefly described some microremains observed within the dental calculus of the individuals of S'Illot d Porros (Mallorca, 6th-2th c. BC). Their results showed the presence of phytoliths, pollen grains, starch grains, fibres and algae. The phytoliths were not frequent. By contrast, they found various starch grains of cereals (wheat and barley) and of the genera *Quercus* and *Vicia*. Some of them were cooked and others were not. In two individuals they observed fragments of algae (*Chrysophyceae*). The samples of S'Aigua Dolça (ca. 2000 BP) exhibited starch grains that belonged to the family of *Fagaceae*. They interpreted these data as consumption of edible nuts of the *Quercus* species (Hernández-Gasch et al. 2002). In comparison to the findings of S'Illot de Porros and S'Aigua Dolça, a principal difference was observed: we did not find, in the samples of Can Reiners, starch grains ascribable to the *Quercus* species. We hypothesize that the individuals that we studied did not eat fruits of *Quercus* species or that there are problems with the differentiation of these grains from others attributed to cereals. Another problem was that we used an optical microscope, and better outcomes would be obtained if an electron microscope could be also employed. Pla *et al.* (2003), indeed, found fragments of algae (*Chrysophyceae*) exclusively with the electron microscope.

Comparing the results of the analysis of dental calculus and the analysis conducted by Garcia and Subirá (2003) with trace elements and by García *et al.* (2004) with stable isotopes, some analogies were observed. The results of stables isotopes exhibited a diet based on animal proteins, obtained from herbivorous such as cows, goats and sheep and their milk, and on vegetal proteins obtained from plants such as wheat, rice, green beans and legumes. The percentage of marine proteins exhibited lower values. Female diet was richer in cereals and legumes than male diet. The results of trace elements were rather similar because they exhibited a diet basically composed by vegetables and the

marine food contribution was poor. Once again, the female diet was a little bit different from male diet, with a higher supply of vegetables, cereals and dried fruit. In the dental calculus analysis we excluded sex differentiation because the samples were prevalently composed by males. Generally, within the dental calculus, we observed great amounts of plant remains such as vegetal fibres, xylem elements and starch grains, whereas few phytoliths were found. We observed various starch grains of wheat, spelt, rye, sorghum and probably oat and farro, and in some cases we found also starch from legumes (*Fabaceae*). No evidence of marine plants and dried fruits (except for fragment of a multicellular polyhedral structure from leaves or fruits of dicotyledonous plants) were found. Therefore, our findings agree with the previous chemical analysis, although we observed that males displayed a great quantity of plant and cereal microremains.



## **6. CONCLUSIONS**





1- The bioanthropological study of the individuals of Can Reiners provided an image of the human community that shares features with late antique and early medieval populations. The demographic (few infants, absence of senile individuals, high mortality in the adult age, low life expectancy at birth), morphological (low stature) and pathological results (higher frequencies of oral pathologies than in older necropolis of Mallorca, high presence of *cribra orbitalia*, evidences of parasites) show a picture of the hard life conditions in Can Reiners population around the 7th century AD. Even though within a radical transformation frame, the dimension of the necropolis attests the continuity of *Pollentia* between the Late Antiquity and the Early Middle Ages. Certain anthropological data (few infants and young adult women, and unbalanced sex proportion) could suggest that the main use of the site maintained a military function in this period.

2- The application of a new method that evaluated the dental occlusion on ancient skeletal remains reveals important information about oral conditions of the individuals of Can Reiners. The findings of the study show that normocclusion is present in ca. 70% of the samples, and openbite, overjet and dental crowding is almost absent. We observed that despite dental wear was not marked, it is the most influencing factor regarding the onset of Class III malocclusion. In addition, dental wear influences others variables such as the curve of Spee, the superior length, and the inferior intermolar width. Therefore, in the population of Can Reiners dental wear rather than genetics was responsible for the onset of malocclusion features. Thereafter, diet and habits, such the use of the mouth as a tool, influence the occlusion patterns.

3- Remarkable results were obtained from the dental calculus analysis and thus this method is suggested to be valuable for the identification of consumed food and habits of the ancient inhabitants of Mallorca. The individuals of Can

Reiners employed different cereal crops, including wheat, spelt, sorghum and probably oat and farro. We also found starch grains belonging to legumes. The great amounts of fibres that we observed within samples suggests a diet rich in vegetables (xylem elements) and the use of the hand as a tool (cotton and linen fibres). The extraordinary finding of a *larva migrans* indicates the presence of intestinal parasites, which reveals that the hygienically conditions were probably critical. Infections and parasitic diseases was maybe be the most frequent cause of death. The discovery of a fern sporangium annulus enables us to hypothesize that plants with presumed magical and medical properties were used in this population.

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## **APPENDIX**





# Study of Dental Occlusion in Ancient Human Remains: A Methodological Approach

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## ABSTRACT

*The anthropological dental and maxillary study in human skeletal remains usually refers to alterations or conditions of the oral cavity. These alterations could have repercussions on life style, dietary habits and diseases. In this particular context, dental occlusion is not often analyzed due to the fragmented condition of the remains, and especially due to the lack of methodology adapted to study ancient remains. The aim of this study is to propose an anthropological method based on clinical dental practice. In the method presented in this work, odontological parameters such as overjet, overbite, and Angle's Classification of Malocclusion, are evaluated.*

**Key words:** malocclusion, paleopathology, paleodontology, reconstruction of paleodiet, life style

## Introduction

There are various research projects on ancient populations that are focused on the reconstruction of dietary habits by the study of teeth and maxillary bones. In this particular context oral pathology, dental wear, dental calculi and their composition and other oral conditions must be taken into account, as well as skeletal markers of metabolic diseases and nutritional stress. Moreover, physical and chemical techniques are used in many studies, in order to determine the type of food consumed in the past. Nevertheless, the lack of studies based on dental occlusion should be highlighted. Thus, the authors propose to study the factors that could lead to malocclusion as well as other diagnostic features for the interpretation of health and lifestyle in an anthropological context.

Dental occlusion is the relationship between both dental arches, where the maximum number of contacts between upper and lower teeth are achieved. Occlusion is dependent on the morphology and function of the different stomatognathic system components<sup>1</sup> (bones, muscles and teeth). Therefore, in skeletal remains, occlusion usually refers to upper and lower teeth articulation. A correct dental occlusion »refers to how well the teeth are arranged individually and one-to-another within and bet-

ween the dental arches<sup>2</sup>. Conversely, malocclusion is considered a discrepancy between teeth that can modify the maxillary shape, sometimes affecting mastication.

Classifying the dental occlusion of ancient remains in normal (normocclusion) or altered (malocclusion) is not the only purpose of the study, but also to define the occlusion characteristics of a population when only skeletal remains are available. It is important to highlight that a slight variation in dental positioning is not enough to be considered malocclusion. Thus, discriminatory criteria must be applied in order to distinguish a slight variation of dental position from a true malocclusion.

The most influencing factors in dental position variation are genetic and environmental – mainly diet and dental wear. In physical anthropology, dental occlusion is not often included in maxillary analysis due to the fragmented and incomplete bone material, and in particular, due to the lack of suitable methodology for the study of ancient remains.

The aim of the present study is the development of a guide for dental occlusion analysis suitable for maxillary remains. The proposed methodology is a contribution to complete the dental data collection in anthropological studies.

## Methods

Archaeological remains do not always include complete skeletal elements, and they may not even be in a good preservation state. This disadvantage is really important in the study of dental occlusion, because sometimes maxilla or mandible are lost, dental arches are incomplete, or some teeth are missing. In addition to this limitation, there is a lack of soft tissue, which complicates the reconstruction of the movement between the dental arches. Even so, it is possible to gather relevant information about mastication and its interpretation.

In the clinical study of dental occlusion, different factors that guide the mandible movement, with or without dental contact, are evaluated, such as articular joints, teeth, arches and muscles. Conversely, in skeletal remains it is only possible to analyze teeth, dental arches and some aspects of the temporomandibular joint (TMJ). Hence, the study of the occlusion in skeletal remains may only be referred to only as »static« occlusion, which means the study of the jaws with maximum dental contact and maximum intercuspidation.

In this work, a lab sheet has been developed with dental and maxillary parameters that would be recommended to evaluate occlusion and its anthropological interest. Different biological traits, such as age, sex and preservation status have also been considered.

### The lab sheet

Due to the limitations of the preservation status of skeletal remains, the evaluation of dental occlusion requires an accurate analysis of the maxillary and dental position, as well as other variables. To achieve a consensus in this analysis, differences between intra- and inter-observer must be previously calibrated. The lab sheet (Figure S1) includes different types of information grouped into five main sections: anthropological information, dental features, dental arches features, occlusal features and articulation characteristics.

### Anthropological information

The individual's information is fundamental for the evaluation of dental occlusion, because dental parameters are influenced by biological and social status, and occupational habits. Moreover, this information must be temporally and geographically situated in order to understand it.

The lab sheet includes different levels of information. The archaeological data and biological profile are collected, including all potential information about the remains and their conservation (a maxillary fragment or a complete skull and mandible). Thus, the origin of the remains, its dating, the individual reference, as well as sex and estimated age at death, must be collected. The section of sex information is divided in four categories: masculine, feminine, ambiguous or undetermined. The age at death categories have been defined according to Vallois<sup>3</sup> and they are divided into six different groups: Infantile I (0–6 years old), Infantile II (7–12 years old), Juve-

nile (13–20 years old), Adult (21–40 years old), Adult Mature (41–60 years old), senile (more than 60 years old). The maxillary and mandible preservation status is separated into hemi-arch and it specifies the presence or absence of condyles, which are basic to analyze certain situations that can be observed in occlusal alterations.

### Dental features

Dental and oral pathology analysis: Number of teeth, size, shape and position, as well as the associated pathology, have an important effect in causing an abnormal functional occlusion, in order to avoid pain or improve the masticatory function. Each dental status and the possible alterations that can be found in the teeth are collected in the dental sheet. The FDI system (World Dental Federation notation, ISO-3950 notation) has been used for teeth enumeration. Using that system, the presence or absence of the teeth is registered. In the case of teeth absence, it specifies if its loss took place *ante-mortem* (when alveolar cavity has been obliterated due to bone healing) or *post-mortem* (when an empty alveolar cavity can be observed). The registration of *ante-mortem* teeth loss is relevant because it can modify the dental arch morphology. For instance, a first molar loss can cause the mesial migration of the second and third molars, modifying not only the arch morphology, but also the occlusion type. Isolated teeth are not included in this study. Nevertheless, they are registered and described in order to complete the individual's dental information.

Regarding oral pathology, dental decay (its location and degree of dental involvement), root fragments, possible abscesses and fractures are described according to Chimenos et al. criteria<sup>4</sup>. For decay, description number and letter are used. The numbers indicate the location of the decay: 0 absent, 1 present, 2 occlusal, 3 crown, 4 necks, 5 root and 6 others. The letters, associated with the numbers, describe the degree of the lesion: a – enamel, b – dentin, and c – pulp. Other variables that influence dental occlusion are also evaluated, such as dental crowding, rotation, and medial, distal, buccal and lingual dental movements. Moreover, abnormal dental eruption such as ectopic eruption, agenesis, supernumerary teeth and other conditions are also evaluated.

Dental wear: As mentioned, dental aspects are basic in the study of dental occlusion. Nevertheless, dental wear requires an independent evaluation as first evidence of the relationship between teeth, as well as evidence of the masticatory and para-masticatory functions. Its analysis is necessary to understand the position of the teeth in occlusion. In the proposed dental sheet, occlusal wear is evaluated according to Brothwell<sup>5</sup> and Lovejoy<sup>6</sup> (Figures 1 and 2). In this study the dental arch is divided in sextants, in order to achieve an accurate analysis of central and lateral dental wear (right and left). Furthermore the degree of dental wear is classified into 5 categories: absence (0), point of enamel wear (1), islands of visible dentine (2), confluence of islands (3), and absence of enamel (4).



Fig. 1. Categories of dental occlusal wear in anterior teeth. Modified from Brothwell (1981) and Lovejoy (1985).



Fig. 2. Categories of dental occlusal wear in posterior teeth. Modified from Brothwell (1981) and Lovejoy (1985).

Spacing characteristics: Dental arch size, as well as teeth presence, position and size, can often explain the articulation between dental arch and teeth. Conditions of space are evaluated by the presence of a diastema or dental crowding. Diastema is the space separating two adjacent teeth. The arch length is higher than the sum of mesiodistal diameters of the teeth; hence there is an excess of space and some teeth are not in contact with the adjacent one. In archaeological studies central diastema (inter-incisal space in the upper arch midline), as well as between other teeth, are analyzed. Diastema is classified into 4 categories: absence of diastema (0), 2–3 mm diastema (1),  $\geq 4$  mm diastema (2), and un-recordable (9).

On the other hand, dental crowding is caused by the lack of space for proper teeth alignment in the dental arch, which can influence dental occlusion. It produces possible inclination, and position or rotation anomalies of erupted teeth. In the sheet, dental crowding is classified into 4 categories: lack of crowding (0), 2–3 mm crowding (1),  $\geq 4$  mm crowding (2), and un-recordable (9). Measurements of dental crowding are taken between the contact points of the two crowded teeth by using a sickle probe.

### Dental arches features

Dental arch morphology: From an evolutionary point of view, dental arch morphology is highly influenced by the different masticatory movements, which are related to diet. The shape and size of the human dental arch has been modified through time, in association with a higher processed food intake. Dental arch morphology is evaluated according to Molnar and Molnar<sup>7</sup>. It is classified into: 1) parabolic, where the anterior curve is wide and divergent ramus, 2) oval, with wide anterior curve and convergent ramus in the last molars, and 3) squared, with nearly plane anterior curve and posterior ramus nearly straight<sup>1,8,9</sup>.

The analysis of symmetry<sup>8</sup> is performed by the evaluation of dental arch morphology with a symmetrograph. This is a transparent squared pattern that is superimposed on the arch, where dental position as regards the midline can be observed; metric data are also collected. Asymmetry can be caused by different pathogenic mechanisms, and whether it is congenital or acquired (traumatic for instance) must be registered.

Dental arch measurements analysis provides the quantification of different parts of the masticatory system related to teeth. This analysis consists of an objective evaluation of each dental arch that can be compared between different individuals or populations. Measurements are sagittal (longitudinal), and transversal (in width)<sup>1,10</sup>.

Sagittal measurements: Maxillary or superior depth (Figure. 3A): It is defined as the anterior-posterior distance taken in maxillary bone, from the inter-incisal point to the plane conformed by the tangent of first upper molars mesial contact point. In case of lack of one of the incisors the measurement is taken in the interproximal wearing facet, and in case of lack of both incisors the reference point is the most anterior part of inter-incisal alveolar crest. If there is a molar missing, the reference point is the inter-proximal distal wearing facet of the second upper premolar. If there are no molars or premolars, the reference point is central area of alveolar bone crest. Maxillary Length (Figure. 3B): It is defined as the anterior-posterior distance taken in maxillary bone, from the inter-incisal point to the plane conformed by the tangent of first upper molars distal contact point. Values are taken in the same way, and by applying the same criteria as in maxillary depth.

Mandible depth (or inferior depth) (Figure 3C): it is the anterior-posterior distance taken from the midpoint of the inter-incisal point to the plane conformed by the tangent of first lower molars mesial contact point. Values are taken in the same way, and by applying the same criteria as in maxillary depth. Mandible length (Figure 3D): It is defined as the anterior-posterior distance taken in maxillary bone, from the inter-incisal point to the plane conformed by the tangent of first upper molars distal contact point. Values are taken in the same way, and by applying the same criteria as in maxillary length.

**Transversal Measurements:** Superior and inferior intercanine width (Figure 3E): it is the distance between both canines (superior or inferior), measured from cuspid to cuspid; in case of marked wearing the reference point is the centroid (central point of incisal surface, located in the intersection of mediobuccal diameter with buccal-lingual diameter) or alveolar central point in case of lack of teeth. Superior and inferior intermolar width (Figure 3F): It is defined as the distance between the two centroid points of upper or lower first molars. In case of absence of molars, the alveolar central point can be used as reference.

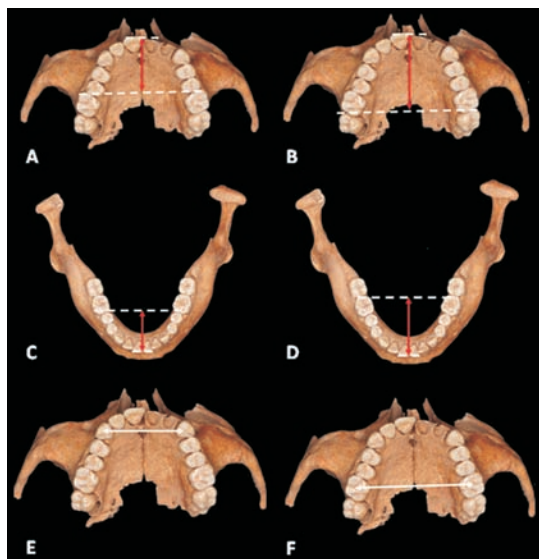


Fig. 3. Reference points of maxillary depth (A) and length (B); reference points of mandible depth (C) and length (D); Reference points of intercanine (E) and intermolar (F) distance.

### Occlusal features

Occlusion is also analyzed through the inter-maxillary relationship in the three spatial planes (sagittal, vertical and transversal). The inter-maxillary relationship influences the maxillary movements, because they are directly related to oral activities, depending on whether they are masticatory or extra-masticatory. Its proper evaluation enables to hypothesize about diet and occupational habits, as regards the analysis of dental arch applied force, and type of movements done.

Overbite (Figure 4) consists of the vertical covering of lower incisors by upper incisors, which is considered normal when that covering is about 1/3<sup>11</sup>. So that the ranges used in the proposed dental sheet are: 1/3 of dental crown of lower teeth covered (0), from 1/3 to 2/3 of dental crown of lower teeth covered (1), more that 2/3 of dental crown of lower teeth covered (2), un-recordable (9).

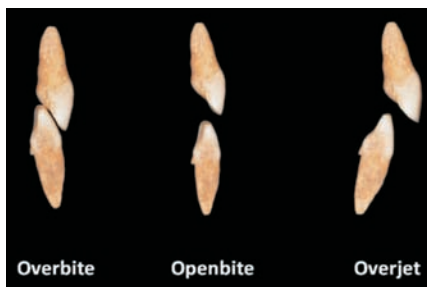


Fig. 4. Overbite, Open bite and Overjet.

Open Bite (Figure 4) is the occlusion characterized by the presence of a space between upper and lower anterior teeth incisal edges when jaws are closed. So that there is a lack of contact between upper incisor’s lingual surface and lower teeth incisal edges. The following criteria are considered: absent (0), present (1), and un-recordable (9).

In the same way, posterior open bite is caused by a lack of upper and lower posterior teeth contact. The criteria used are: no open bite (0), presence of open bite, indicating the number of non-contacting molars (1-2-3), un-recordable (9).

Overjet (Figure 4) quantifies the distance between lower incisor buccal surface and upper incisor incisal edge measured parallel to the occlusal plane considering the starting point of the measurement at the superior incisor. A measurement of +2 mm with  $\pm 2$  mm of deviation is considered normal<sup>12,13</sup>. Overjet is classified into four categories: overjet from 0 to 4 mm (0), 4 to 8 mm overjet (1), more than 8 mm overjet (2), and un-recordable (9).

Anterior cross bite is a malocclusion by a mandible or maxillary bone sagittal displacement. Anterior cross bite can originate from a dental mal-position, dental hypoplasia, or diminished maxillary growth, which implicates maxillary retrusion, as well as prognathism or advanced anterior mandibular, which causes upper incisors positioned behind lower incisors. It is classified into: absent (0), affecting just one tooth (1), affecting two teeth (2), affecting three teeth (3), and un-recordable (9).

Midline displacement can be observed by the asymmetry between the dental arch in the incisor area. The intercisor point, which is defined as the mesial contact point between both central incisors, must coincide with palatine midplane. Its possible displacement as regards to the midline is evaluated taking the palatine suture as reference. The possible values registered in the dental chart are: absent (0), present 2–3 mm (1), present  $\geq 4$  mm (2) and un-recordable (9).

Canine relation had been in the same way as registered for molar relation, Angle’s classification (I, II and III) are defined for the canine relationships. Class I or normocclusion is established when the upper canine cuspid is located between the lower canine and lower first premolar, when dental arches are occluding. The possible values for this variable are: Class I (0), Class II (1), Class III (3), and un-recordable (9).



Molar relationship: Angle's classification<sup>14</sup> has been used to describe molar relationships, distinguishing: class I, class II and class III (Figure 5). Class I. Upper first molar mesiovestibular cuspid occludes in the lower first molar buccal sulcus. Class II. Lower first molar buccal sulcus is located distally as regards the upper first molar mesiovestibular cuspid. The whole maxillary arch is displaced forward, or mandibular arch is displaced backwards as regards the maxillary bone. Moreover, Class II is divided into Complete or Uncompleted Class II, according to the intensity of sagittal deviation. Complete Class II is considered when the upper first molar distovestibular cuspid is at the level of lower first molar buccal sulcus. Whereas Incomplete Class II is considered as a lower degree of this type of malocclusion, a cuspid to cuspid relationship exists when the mesial surface of both upper and lower first molars are in the same vertical plane. Class III. Lower first molar buccal sulcus is located mesially as regards to upper first molar mesiovestibular cuspid. The mandibular bone is displaced forward or maxillary bone is displaced backwards regards to the mandible. As in class II, there is a distinction between complete class III and incomplete class III, according to the degree of affectation. Thus anteroposterior molar relationships (or Angle classes) are registered as: Class I or normocclusion (0), Incomplete Class II (1), Complete Class II (2), Incomplete Class III (3), Complete Class III (4), and un-recordable (9).



Fig. 5. Angle's Occlusion classification.

Posterior cross bite: Lingual posterior cross bite is caused by a transversal malocclusion, where the upper molar and premolar buccal cuspids occlude in the lower molar and premolar pits. This item can be classified into three categories: absent (0), present (1), and un-recordable (9).

Buccal posterior cross bite occurs when upper molar and premolar palatine cuspids contact with lower molar and premolar buccal cuspids. The different values of classification are: absent (0), present (1), and un-recordable (9).

Occlusal plane: Different combined factors determine inter maxillary teeth contact while masticating. Because mandible and teeth generally show a good degree of preservation in skeletal remains, occlusal plane can be studied through Spee curve and Wilson curve evaluations, as well as dental arch morphology.

The Spee curve<sup>1,15</sup> is a line defined by occlusal surfaces of the teeth of the mandibular hemiarch, joining anterior teeth incisal edges with posterior teeth buccal cuspids, this line draws a superior concavity curve. Its

evaluation is performed with a thin rigid and slight scale (occlusal plane), estimating whether the concavity is normal (upside), plane or reversed. This curve permits the evaluation of proper dental root distribution in maxillary bones. The Spee curve is due to anteroposterior mandible movements (protrusion and retrusion). When an increased Spee curve is observed, there is root crowding, with a convexity in its curve. Consequently, there is a decrease in bone mass between those dental roots, which must properly support force and loading.

Wilson curve<sup>1,15</sup> is a transversal curvature in occlusal plane in frontal view. It is influenced by lower posterior teeth lingual inclination. This curve changes from the first to the third molars, and it also changes with occlusal dental wear. Mandibles where teeth showed a great dental wear have concave Wilson's curve and it evolves to convex as occlusal wear increases. The measurement is taken at lower first molars level using a flexible thin acetate layer, which lays on the lower first molar occlusal surface (right and left), observing the shape that the layer adopts: concave (or normal), plane, convex (or reversed). This curve originated from the requirement of the height cuspid difference compensation, due to buccal cuspids being higher than lingual ones. According to that curve, harmonious intercuspids displacement can occur with lateral movements. Thus, the significance of the Wilson curve evolution is based on lateral movements of the mandible.

### Articulation

Temporo-mandibular joint (TMJ) is examined in order to determine the presence or absence of condyle pathology that could induce an occlusion anomaly, such as asymmetries or arthropathies. It can be classified as: absent (0), present (1), and un-recordable (9).

### Discussion

The study of malocclusion arises from the practice amongst the current population of adjusting incorrect alignment of teeth that could cause many mastication and aesthetic problems. Indeed, in modern populations the predominance of malocclusions is about 40 to 80%<sup>16</sup>. The main aetiological factors of malocclusion are of genetic and environmental origin. Some of the genetic factors are the evolutionary reduction in jaw and tooth size, or defects of embryological development<sup>17</sup>, whereas some examples of environmental factors are trauma, habits, anomalies of postnatal development, as well as physical agents and malnutrition. Different methodologies to describe, measure and classify various typologies of malocclusion have been developed to understand the problem and to apply correct treatment. Any method including quantitative and qualitative analysis, has to be universally accepted, applicable to distinct populations, and should allow inter-population comparison.

There is a lack of studies on dental occlusion in the anthropological literature. In general, orthodontic methodologies are not applied on skeletal remains, especially

on archaeological material. Works usually conducted on ancient skeletal material agree that prehistoric populations exhibited a correct or ideal, dental occlusion but, with the passing of centuries and changing of diet, there is an increase of malocclusion<sup>16</sup>. This tendency is too simplistic, because multiple factors are involved in the development of the maxillary system structure.

Dental wear, for instance, plays an essential role in explaining the evolution of occlusion changes. In the 1950's, Begg hypothesized that the human teeth are »designed« in order to cope with extensive tooth wear, and consequently have developed compensation mechanisms<sup>18</sup>. This theory coincides, in part, with the studies conducted on prehistoric populations and on the modern hunter/gatherers. These groups were and are vulnerable to the heavy wear of the surfaces caused by an abrasive diet and the use of teeth as a tool. Starting from this assumption, it is accepted that the modern population has inherited the same dental model. Hence the different use of teeth has generated, together with the change of eating habits and the decrease of the dental arches, a proliferation of problems related to dental misalignments<sup>18</sup>. It is important to highlight that some points of Begg's theory are criticized by anthropologists and orthodontists. In particular the absence of dental wear in modern population does not mean that an increase of malocclusion and attritional occlusion is not a treatment model for contemporary dentistry<sup>18,19</sup>.

A study recently carried out on a Copper Age population<sup>20</sup> has underlined the presence of dental crowding in 100% of lower jaws analysed. The results contrast with the proposed trend, and suggest that malocclusion is due to genetic factors instead of excessive tooth size and environmental change. This conclusion is supported by several data: mesiodistal diameters of the lower teeth are similar to modern equivalents, dental wear is comparable to other prehistoric populations, and generally the third molar is on the occlusal plane or absent<sup>20</sup>. Another research study performed on skulls of Xia dynasty dated 4000 years ago shows that the malocclusion is 27.6%<sup>21</sup>. Considering these, and other articles about malocclusion, some questions arise, such as: is dental crowding a genetic problem or are more factors involved? Is the important change of malocclusion related to industrial society, or also with the passage of the hunter/gatherer world towards a farmer society? Does the decrease in jaw and dental size have an important role in malocclusion? The increased study in this area could partly change the current view suggesting other hypotheses in relation to the presence or the absence of the malocclusion in ancient populations.

In this framework, it is proposed to create a straightforward lab sheet in which the main methodologies used by orthodontists are employed. The lab sheet was created taking into account some of the classifications used in epidemiological studies, like the Angle's classification<sup>14</sup>, the basic method for recording occlusal traits<sup>22</sup> and the method for epidemiological registration of malocclusion<sup>23</sup>. This is a multidisciplinary work adapted to the needs and the peculiar characteristics of anthropological observation.

Firstly, it is essential to contextualize the skeletal remains in order to evaluate the genetic and environmental factors. Therefore, the lab sheet includes data related with geographical and historical background. Information about sex and age at the death of the individual has also been included. To begin the dental report, the alveolar status is described. Often the material analyzed presents alterations like *ante mortem* (during life) and *post mortem* (after death) tooth loss. Another very significant element is the description of dental wear. In this work, modified methodologies of Brothwell<sup>5</sup> and Lovejoy<sup>6</sup> have been used. Tables are created to be able to observe the five typologies of dental wear. Usually, in archaeology or in forensic anthropology it is normal to find a fragment of dental arch; for this reason dental wear in the lab sheet is studied, dividing the arch into the anterior and posterior sextants.

A complete description of the state and pathologies of teeth is provided in the proposal of Chimenos et al.<sup>4</sup>. For this reason, pathologies are absent in the present lab sheet, with the exception of those that can interfere with or be explained by dental occlusion. Also, fractures are recorded only when they developed during the life of the individual.

It is also fundamental to collect data concerning possible pathologies affecting dental arches, for example, mandibular torus or an asymmetry of the superior arch that could indicate a congenital cranial asymmetry or a probable fracture. Both cases may affect the position or morphology of teeth and arches, and also the typology of occlusion. Also a temporo-mandibular joint disorder and bruxism could include an occlusion alteration. A correct position of the arches is also necessary to ascertain whether dental wear is unusual or not, and if it is related to occlusion or with a non-alimentary use of the teeth, as in the case of chipping marks or notches. Once the cause is detected, it is possible to collect more information, for instance, the correct orientation of an instrument used for a particular activity.

In the sections related to the characteristics of arches, occlusion, occlusal plane and temporo-mandibular joints, current odontological methodologies adapted to skeletal remains have been used. To assist in the identification of occlusal variables, a score classification is applied for each item. Also, a space has been reserved to specify diastema, crowding, overjet, cross bite, overbite and open bite measurements in millimeters. Some of the principal measurements of dental arches are also included, because some occlusion analyses require them, for instance for discerning sexual dimorphism and changes of molar class occlusion<sup>24</sup>. These measurements are also of interest in human masticatory evolution. Generally, the hypothesis, in which the transition from a diet composed of food that is hard to chew to more mild and elaborate food has contributed to the decrease in teeth and jaw dimensions, is accepted.

The study of dental occlusion in skeletal remains is also important in forensic contexts. In this field, reconstruction of maxillary occlusion has different applica-

tions, for example to recreate the face of a person, or to characterize deceased individuals, as well as historical characters by using known defects or dentistry cures. Although different disciplines (medicine, anthropology, art) collaborate together in this field, they can run into various problems. The main difficulties are due to the fragmentation of bones. The lack of the condyles, for instance, certainly complicates the analysis of the type of occlusion. Therefore physiognomy reconstruction may differ radically if arches are assembled in class II or III. For this reason orthodontic methodologies adapted for skeletal remains generate improved information regarding occlusion.

## Conclusions

Good occlusion is necessary for a correct mastication, health and diet. Occlusion is relevant to the acquisition

of food and has changed during evolution, in relation to the adjustment to the availability of the food. In spite of this, occlusion is often overlooked in anthropological studies (evolutionary, historical and forensics). This entails the loss of many data that could be useful for the reconstruction of the occlusal characteristics of an individual or a population, both proto-historical or pre- or post-industrial. Furthermore a proper evaluation of dental occlusion could be employed in forensic areas to identify a minor feature that could be fundamental for the reconstruction of the face of a recently deceased individual or a historical character.

## Acknowledgements

This study has been partially supported by MICIIN (CGL2008-00800/BOS) and Generalitat de Catalunya (SRG 2009-566)

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## STUDIJA DENTALNE OKLUZIJE NA DREVNIM LJUDSKIM OSTACIMA: METODOLOŠKI PRISTUP

## SAŽETAK

Antropološka stomatološka i maksilarna studij ljudskih skeletnih ostataka obično se odnosi na promjene ili stanje usne šupljine. Promjene mogu biti posljedice životnog stila, prehrambenih navika i bolesti. U ovom kontekstu, stomatološka okluzija nije često analizirana zbog različite očuvanosti skeletnih ostataka, a posebno s obzirom na nedostatak metodologije prilagođene studijama drevnih ostataka. Cilj ovog rada je predložiti antropološku metodu koja je temeljena na kliničkoj stomatološkoj praksi. U metodi prikazanoj u ovom radu, vrednuju se stomatološki parametri, kao što su zagriz, pregriz i Angleova klasifikacija malokluzije.

**DENTAL OCCLUSION**

**ANTHROPOLOGICAL INFORMATION**  
 Burial/Skeleton number: ..... Site Name: .....

Sex: M  F  Ambiguous  Ind.  Age: Inf. I  Inf. II  J  A  M  S  Ind.

State of Conservation: R Maxilla  L Maxilla  R Mandible  L Mandible

TMJ: Complete  Incomplete: Bilateral  R  L

**DENTAL FEATURES**

															55	54	53	52	51	61	62	63	64	65																					
18	17	16	15	14	13	12	11	21	22	23	24	25	26	27	28																														
															48	47	46	45	44	43	42	41	31	32	33	34	35	36	37	38															
															85	84	83	82	81	71	72	73	74	75																					

D= Decay      X= Postmortem tooth loss      L/B= Lingual/buccal      ■= Incomplete alveolus or partially missing  
 GR= Granuloma      O= Antemortem tooth loss      D/M= Distal/Mesial  
 RR= Root remains      V= No eruption/no visible      R= Rotation  
 F= Fracture      Ag= Agenesis      C= Crowding

**Teeth wear:**

0= Absence  
 1= Point of enamel wear  
 2= Islands of visible dentine  
 3= Confluence of islands  
 4= Absence of enamel

(55-54) (18-14)	(53-63) (13-23)	(64-65) (24-28)
(85-84) (48-44)	(83-73) (43-33)	(74-75) (34-38)

**Spacing characteristics:**

Sup. Inf.      Diastema: .....mm      Sup. Inf.      Crowding: .....mm

     0= Absent             0= Absent  
 1= Present 2-3 mm      1= Present 2-3 mm  
 2= Present ≥ 4 mm      2= Present ≥ 4 mm  
 9= Un-recordable      9= Un-recordable

**DENTAL ARCHES FEATURES**

Parabolic Oval Squared      Yes Not

Arch morphology: Sup.         Symmetry: Sup.    
 Inf.         Inf.

Sagittal measurements: Depth: Sup. .... Inf. ....  
 Length: Sup. .... Inf. ....  
 Transversal measurements: Intercanine: Sup. .... Inf. ....  
 Inter-molar: Sup. .... Inf. ....

**OCCLUSAL FEATURES**

<p><input type="checkbox"/> <b>Overbite:</b> .....mm                  0= Up to 1/3                  1= From 1/3 mm to 2/3                  2= &gt; 2/3                  9= Un-recordable</p> <p><input type="checkbox"/> <b>Open bite:</b> .....mm                  0= Absent                  1= Present                  9= Un-recordable</p> <p><input type="checkbox"/> <b>Overjet:</b> .....mm                  0= Up to 4mm                  1= From 4mm to 8mm                  2= &gt;8 mm                  9= Un-recordable</p> <p><input type="checkbox"/> <b>Anterior Cross bite:</b> .....mm                  0= Absent                  1= 1 Tooth                  2= 2 Teeth                  3= 3 Teeth                  9= Un-recordable</p> <p><input type="checkbox"/> <b>Midline Displacement:</b>                  0= Absent                  1= Present 2-3 mm                  2= Present ≥ 4 mm                  9= Un-recordable</p>	<p style="text-align: center;">Right    Left</p> <p><input type="checkbox"/> <input type="checkbox"/> <b>Canine relationship:</b>                  0= Class I                  1= Class II                  2= Class III                  9= Un-recordable</p> <p><input type="checkbox"/> <input type="checkbox"/> <b>Molar Relationship:</b>                  0= Normocclusion Class I                  1= Incomplete Class II                  2= Complete Class II                  3= Incomplete Class III                  4= Complete Class III                  9= Un-recordable</p> <p><input type="checkbox"/> <input type="checkbox"/> <b>Posterior Open bite:</b>                  0= Absent                  1= Present                  9= Un-recordable</p> <p><input type="checkbox"/> <input type="checkbox"/> <b>Lingual Posterior Cross bite:</b>                  0= Absent                  1= Present                  9= Un-recordable</p> <p><input type="checkbox"/> <input type="checkbox"/> <b>Buccal Posterior Cross bite:</b>                  0= Absent                  1= Present                  9= Un-recordable</p>
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**Occlusal planes:**

Normal Flat Inverted

Spee Curve:

Wilson Curve:

**ARTICULATION**

**TMJ Pathology:**  
 0= Absent  
 1= Present  
 9= Un-recordable

Figure S1. The Lab sheet





