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# ABSTRACT

### Kendra Quinn

# A Bioarchaeological Study of the Impact of Mobility on the Transmission of Tuberculosis in Roman Britain

Tuberculosis (TB) is an infectious disease mainly transmitted to humans by the inhalation of infected droplets (produced when an infected person coughs or sneezes). It is caused by bacteria within the *Mycobacterium tuberculosis* complex. In the early 1990s, the World Health Organisation (WHO) declared TB a global emergency and this continues to be the case today, and the increase in global travel, including migration, is thought to be exacerbating its spread.

This research tests the hypothesis that people buried in Roman Britain who were infected with TB had been mobile at some point in their lives, by the application of stable isotopic analysis (C, N, Sr, O) to their skeletons to establish if their childhoods were local or non-local to their burial locations.

The study uses bone and dental samples from skeletons who were buried on chalk geology and with bone changes suggesting possible TB and/or a positive TB ancient DNA result. Collagen was successfully extracted from bone for 19 out of 21 individuals. Carbon and nitrogen isotope analysis revealed that all but three of these people ate a diet based on  $C_3$  terrestrial ecosystems with limited aquatic food intake, and they were similar to other people buried in the same or other contemporary cemeteries. Enamel from the teeth of all 21 individuals was subject to strontium and oxygen isotope analysis, which identified six people as not having been brought up in the area where they were buried. The remaining 15 people were possibly raised locally, although other places of origin have been considered.

It was concluded that linking mobility, as identified using stable isotope analysis, with transmission of infectious disease evidence in the skeleton is very challenging, particularly because there is no way of knowing how long people had been infected with the disease before or after they were mobile.

# A BIOARCHAEOLOGICAL STUDY OF THE IMPACT OF MOBILITY ON THE TRANSMISSION OF TUBERCULOSIS IN ROMAN BRITAIN

KENDRA QUINN PhD

DEPARTMENT OF ARCHAEOLOGY DURHAM UNIVERSITY

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#### **Chapter 1: Introduction**

The research within this thesis examines the potential relationship of the transmission of tuberculosis (TB) due to mobility of people in Roman Britain, namely within the area of modern England. It tests the hypothesis that people buried in Roman Britain (modern England), who were infected with TB at the time of their death, originated from an area some distance from where they were eventually buried. The research uses techniques of isotope analysis to look for differences in diet (using carbon and nitrogen isotope systems) and for differences in place of upbringing (using strontium and oxygen isotope analysis) in order to establish if the TB infected individuals were local to their place of interment.

Tuberculosis is an infectious disease caused by *Mycobacterium tuberculosis* complex bacteria. It typically affects the lungs (pulmonary TB) but can also affect other sites in the body (extrapulmonary TB). TB remains a considerable cause of ill-health and death worldwide in the 21<sup>st</sup> century (WHO 2015), which makes it a particularly interesting disease to study. It is briefly introduced in this chapter and considered in much more detail in Chapter 2.

Although TB can be transmitted via the ingestion of infected meat and milk, generally, the disease is spread from human to human in the air by droplets of saliva exhaled from the lungs containing *Mycobacterium tuberculosis* complex bacteria (see Table 2.1). These infected droplets are transmitted when a person coughs, or even speaks, and individuals in contact with them inhale the droplets. Fortunately, a relatively small proportion (approximately 5 to 15%) of people infected by *Mycobacterium tuberculosis* complex bacteria will go on to develop TB disease (WHO 2015). However, the probability of developing TB disease is much higher in people today who are already infected with Human Immunodeficiency Virus (HIV) due to these individuals having a weakened immune system. TB in humans is most commonly caused by *Mycobacterium tuberculosis* or *Mycobacterium bovis* and the disease is more common in men than in women,

and it tends to affect more adults than children, particularly those adults in the most economically productive age groups (Ibid. 2015).

Today, the most common method for diagnosing TB worldwide is still the sputum smear microscopy method developed more than 100 years ago (WHO 2015). This technique requires sputum samples to be stained and then examined under a microscope. The *Mycobacterium tuberculosis* complex bacteria are, however, often quite difficult to see and could be missed. Developments in TB diagnostics in the last five years mean the use of rapid molecular tests to diagnose both TB and drug- resistant TB are increasing. Some countries are now even phasing out the use of sputum smear microscopy for initial diagnosis, although in countries with good laboratory facilities cases of TB are also diagnosed via culture methods. This is the current reference standard although it has the disadvantage of taking up to six weeks to culture the bacteria (Ibid. 2015).

Up until the 1940s, there were no effective drug treatments for TB, and without treatment the death rate is high; today approximately 70% of untreated people with positive sputum smears die of pulmonary TB within 10 years (Tiemersma et al. 2011). Effective drug treatment of TB started in 1946 with the introduction of the antimicrobial drug, streptomycin (Mitchison and Davies 2012:724) and the most effective first-line anti-TB drug, rifampicin, became available in the 1960s (WHO 2015). The currently recommended treatment for new cases of drug-susceptible TB is a six-month regimen of four first-line drugs, namely isoniazid, rifampicin, ethambutol and pyrazinaminde (Ibid. 2015). Treatment success rates of 85% or more are reported from the World Health Organisation (WHO) from those patients who correctly follow this treatment regime (Ibid. 2015).

Multi-drug resistant TB (MDR-TB) is defined as being caused by bacteria that are resistant to isoniazid and rifampicin, which are the two most powerful anti-TB drugs (WHO 2015). Treatment for patients infected with MDR-TB takes longer and is more expensive than for drug-susceptible TB, and requires the use of more toxic drugs. Treatment for 20 months for these patients is currently recommended by

the WHO. The emergence of Extensively Drug Resistant TB (XDR-TB) has also recently emerged and was first described in 2006 (WHO 2006). XDR-TB is defined as MDR-TB plus resistance to one fluoroquinalone drug and also to a second-line injectable antibiotic (WHO 2015). Unfortunately, success rates for these treatment regimes are much lower than that for the sensitive strains of the bacteria (Ibid. 2015). However, new drugs with novel modes of action are currently undergoing extensive research and clinical trials. Several vaccines are also undergoing trials, although these are primarily aimed at protecting children; a vaccine that is effective in preventing TB in adults is proving difficult to develop (Ibid. 2015).

There are many risk factors which predispose people to contracting TB and these will be discussed in more detail in Chapter 2. However, this research focuses on one risk factor in particular, which is mobility. Mobility, including the migration of people for various reasons, is known to be a major factor in the spread of TB around the world today. Reasons for this appear to be rather complex (Blumberg et al. 2010, Zammarchi et al. 2014) with second-generation immigrants continuing to be at higher risk of contracting the disease than locally born and raised individuals (Zammarchi et al. 2014, Marx et al. 2015). This could be due to frequent contact and transmission within migrant communities, which is not picked up by current screening methods for immigrants themselves (Marx et al. 2015:2).

The current PhD arose due to two previous projects examining the aDNA of MTBC bacteria. These previous projects were both NERC (the Natural Environment Research Council) funded and run collaboratively between the Universities of Manchester and Durham. The first of these, entitled "Biomolecular archaeology of ancient TB in Britain and Europe", ran from 2007 to 2011 (Roberts et al. 2011). It aimed to study the origin and evolution of the causative agents of TB in Britain and other parts of Europe by analysing samples of bones from skeletons of different dates and from a range of archaeological sites of different periods (Müller et al. 2014a). The second project, entitled "Palaeopopulation genomics of *Mycobacterium tuberculosis*", exploits the outcomes of the first project and ran from September 2013 to September 2016 (Brown et al. 2016, Müller et al. 2014b,

2014c, 2016). During this research, 491 archaeological skeletons from across Britain and Europe dating back to the Roman period were screened for the presence of *Mycobacterium tuberculosis* complex aDNA. The samples containing the best-preserved aDNA were subject to polymerase chain reaction (PCR) which was used to type a small number of single nucleotide polymorphisms (SNPs) whose identities enable strains to be placed into broad population groupings recognised in modern genomic work on the *Mycobacterium tuberculosis* complex (Brown et al. 2016, Müller et al. 2014b, 2014c, 2016). The project also established that next generation sequencing (NGS) can be used to type substantially greater numbers of SNPs in *Mycobacterium tuberculosis* complex aDNA than is possible by PCR, and that genotypes resulting from NGS allow detailed examination of evolutionary relationships between historic and extant types of TB.

The specific objective of the second aDNA project was to test the hypothesis that hybridisation capture and NGS of *Mycobacterium tuberculosis* aDNA can provide enough genotype data from enough archaeological skeletons for palaeopopulation genomics of TB to become a reality. To test this hypothesis, the researchers attempted to obtain genotype information from samples taken from British and European skeletons.

As a result of these two previous NERC projects and skeletal samples available from them, the current research focuses on 21 individuals who were likely to have been infected with TB during their lifetimes (detected by aDNA and/or osteological analyses) and who were buried at sites in Roman Britain. This era was chosen as the incidence of the evidence of skeletal TB had greatly increased from the Iron Age. This was thought to be likely to be due to some alterations in lifestyle, of which increased mobility and urbanisation were two changes typifying the Roman Era in Britain. Mobility was chosen for investigation as it is known to be a major factor in the spread of TB today and isotope analysis has been used previously to identify mobility in the past, but it has not been widley linked with identifying the origins and spread of infectious diseases.

It was also expected that strain data for the MTBC aDNA would be available from the NERC funded work. This would have been used to further inform of origin of the infecting strains of TB and to lend weight to the argument that mobility was mainly responsible for the increase in TB in Roman Britain. Alas as the current mobility project progressed, it became clear that strain data would not be available (discussed in Chapter 7) and so the hypothesis of the research was adjusted slightly to compare mobility of individuals with TB with those buried nearby who were probably not infected with the disease.

Bones and teeth from the sample of 21 individuals selected were subjected to a multi-isotope analysis to explore if they were born and brought up, or lived the last decades of their lives, in the area in which they were buried. Strontium and oxygen isotope analysis was chosen as these are the widely used indicators of mobility, but carbon and nitrogen isotope analysis was also included to inform of any differences in diet consumed by the individuals with TB compared with others buried in the same area. For example, the consumption of plants grown in warmer areas (C<sub>4</sub> plants) could indicate the people had migrated from warmer areas as these types of plants were not grown or widely available in Roman Britain. The research did not result in further destruction of skeletal collections because remnant dental and bone samples remaining from the aDNA projects described above were available for use. This eliminated any ethical issues about unnecessary destructive analysis.

The hypothesis proposed in the current research is that the mass movement of people that characterised the Roman period was responsible for the spread of TB into Britain from the rest of the Roman Empire, and that individuals from Roman Britain who were infected with TB were less likely to be local to the area in which they were finally buried than other individuals buried in Roman Britain who had no skeletal evidence of TB.

The current project is important because it will help to explore how TB was transmitted in the past. This disease continues to be a "global emergency" today

(WHO 2015), when human migration has reached massive proportions with around one in every seven people in the world being migrants (Blumberg et al. 2010), and there are multi-drug and extensively-drug resistant emergent strains of TB. Hence the project may also confirm that migration has been a longstanding risk factor for TB transmission and may be something that should be considered as a major risk factor if migration continues at current rates or indeed if it increases in future.

The format of this thesis shall now be described. There are four background chapters to provide information relevant to the research on TB and mobility. Chapter 2 considers TB today and in the past in more detail by examining risk factors for transmission of the disease, diagnosis, treatment and current research into prevention of transmission of infection and/or development of the disease. Although much of the current treatment would not have been available in the Roman era, many of the risk factors for transmission that were experienced in the past are still relevant today.

In Chapter 3, previous stable isotope analyses of Roman skeletons will be discussed; this is important to understand how the current research fits with extant data, and because some of the published work is used as "control" comparisons for the data in this study. In Chapter 4, evidence for mobility in the Roman Empire is explored, and in particular within Europe and Britain. This will help to clarify how difficult or easy it was to be mobile in the Roman period. Finally, in Chapter 5, the impact of mobility on transmission of infectious diseases will be reviewed, and specifically for tuberculosis. This will show how mobility today has a considerable effect on how rapidly tuberculosis (and other diseases) can move around the world. Chapter 6 discusses materials and methods used in the project, including a description of the burials and skeletons which were sampled. Chapter 7 describes the results from isotope analysis of the skeletons, Chapter 8 discusses the results to establish if isotope analysis can help to identify locals and non-locals in relation to the research hypothesis and Chapter 9 draws together the conclusions from the

research and suggests some ideas for further research to support and strengthen the findings of this project.

### Chapter 2: Tuberculosis today and in the past.

This chapter is divided up into two main sections; TB today and then TB in history. The nature of tuberculosis will be explored. This will include its epidemiology, pathogenesis, signs and symptoms, diagnosis, treatment, current global frequency, and bone changes associated with the disease. The chapter will provide a base from which a more nuanced picture of tuberculosis in the past can be understood.

Firstly, in section 2.1, the problems caused by tuberculosis today will be examined. This is important to show that the disease, which became increasingly common in Britain during the Roman era, is still causing serious health issues today, not least because many strains of the disease are now resistant to the antimicrobial drugs available in the 21<sup>st</sup> century. Of course, these drugs were not available in the Roman era, and hence current treatments of the disease are becoming as severely limited as they were in the Roman period. In light of the problems with treatment of the disease, prevention of spread becomes more important. Perhaps transmission methods and patterns of the disease in the past could help predict pathways the disease will take today, bearing in mind that public transport is so much more rapid than it was in the Roman era these issues of transmission due to mobility will probably arise far quicker today than they did in the past.

# 2.1 Tuberculosis today

2.1.1 What is TB?

Tuberculosis (TB) is an infectious disease caused by a group of gram-positive bacteria of the genus *Mycobacterium:* the "*Mycobacterium tuberculosis* complex". This consists of several species of bacteria that are capable of causing TB in humans (see Table 2.1). These are closely related species of which *M*.

*tuberculosis* and *M. bovis* are particularly pathogenic to humans (Goering et al. 2013:230, Grange 2008:66).

Name of bacterium	Usual host	References
Mycobacterium	Humans	Bouakaze et al. (2010)
tuberculosis		
Mycobacterium bovis	Cattle, badgers	Bouakaze et al. (2010)
Mycobacterium microti	Voles	Bouakaze et al. (2010)
Mycobacterium	Humans (in Africa)	Bouakaze et al. (2010)
africanum		
Mycobacterium avium	Birds, soil	Koirala + Bronze (2015)
Mycobacterium	Seals	Bos et al. (2014)
pinnipedii		
Mycobacterium cannetti	Humans (in Africa)	Somoskovi et al. (2009)
Mycobacterium caprae	Deer, cattle, pigs, wild	Rodríguez et al. (2011)
	boar	

Table 2.1 Causative organisms of TB in humans

Tuberculosis in humans typically affects the lungs (pulmonary TB) but can also affect other sites in the body (extrapulmonary TB). The disease is spread between humans through exhalation and inhalation of bacteria-laden droplets caused by sneezing and coughing (CDC 2016a). The high quantity of droplets produced in a single sneeze are shown in Figure 2.1:



Fig. 2.1 Droplets produced during a sneeze. (Pappas 2010)

When the bacteria from these infected droplets reach the alveoli of the lungs they multiply rapidly and infection begins (Goering et al. 2013:231). Airborne transmission of TB is very efficient because infected people cough up enormous numbers of bacteria whose waxy coat prevents desiccation and therefore they can survive for "long periods of time" in the air and in house dust (Ibid. 2013:231) although it is not clear if long periods of time covers months, years, decades or even millennia.

TB can also be contracted from working with infected animal products (such as skins via the tanning industry) or through the consumption of milk and meat infected with *M. bovis* (O'Reilly and Daborn 1995:1, Stone et al. 2012:164-165, Waters et al. 2014:115). This method of transmission was probably more common in Roman times due to more use of leather and animal products then than today, now manufactured alternatives (eg. use of synthetic materials in shoemaking) are available. Milk during Roman times would have been "raw" (that is unpasteurised) and thus bacteria present would survive in it to be ingested by people consuming it. Distribution of raw milk today is illegal in Scotland and strictly limited and controlled in England and Wales (Raw Milk 2017), so presumably risks of contracting *M.bovis* are far lower today than they were in the past.

*M. bovis* is the organism which most commonly causes TB in animals. It can be found in wild and domestic animals and has been of particular concern in the UK over recent years, due to the risk of infections passing to humans from cattle with the disease. For example, dairy cows tend to have higher rates of TB than beef cattle and this is thought to be because they live in more crowded conditions (Morris et al. 1994:157, Waters et al. 2014:115). Movement of infected animals and fence-line contact with other herds is a common means of transferring the disease between herds and regions (Waters et al. 2014:115). International trade in livestock is also significantly increasing risks for the spread of bovine TB over large distances (Ibid. 2014:115). TB in cattle causes a cough, weight loss and shortness of breath as it also does in humans, and in dairy cows it results in a decline in milk production. Obviously, infected animals should not be contributing to the food chain, and thus the increase in TB in UK cattle, and indeed across the world, is causing concern (Smith et al. 2012:1).

The European badger, which is found throughout the UK, except in the far north of Scotland, is of special interest because the Government suggests that these animals are a reservoir of TB and that a cull will decrease its incidence (TB Free England 2013). However, infection in badgers probably presents a relatively low risk for domestic livestock and the effectiveness of a mass cull is questionable, particularly as there is no test for TB infection in live badgers; testing for TB for research purposes occurs on road killed animals, so killing would be indiscriminate (Atkins and Robinson 2013). In addition, badger culling appears to increase the risk of transmission of bovine TB to cattle because surviving badgers in culling areas show expanded ranging behaviour. This leads to each infected badger coming into contact with more cattle herds and other badgers in neighbouring areas, despite overall badger density being reduced (Vial and Donnelly 2012:50-53).

# 2.1.2 Contracting TB



*Fig. 2.2 Second World War poster showing how diseases can be spread on public transport.* (Posters of War 1940)

The methods by which TB bacteria can be transmitted have been examined (section 2.1.1 and Figures 2.1 and 2.2), and now the route taken by resulting infection, should it occur, will be discussed. Primary infection occurs in individuals encountering TB for the first time. The *Mycobacteria* are engulfed by macrophages in the alveoli of the lungs, in which they survive and multiply. Other macrophages are attracted to the site where they also ingest invading bacteria and carry them via the lymphatic system to lymph nodes where the immune response is stimulated. This is detectable four to six weeks after infection via the TB skin test in which purified protein derivative (PPD) of *Mycobacterium tuberculosis* is introduced into the skin. A positive result occurs 48 to 72 hours later when signs of local erythema can be observed (Goering et al. 2013:231). Primary TB is mild and asymptomatic and in around 90% of cases it does not proceed further. However, clinical disease develops in the remaining 10% (Ibid. 2013:232). This happens when some bacteria escape to set up foci of infection in other body sites.

macrophages and increase their ability to destroy the microorganisms. The body then reacts to contain the bacteria within "tubercles", or small granulomas consisting of epithelioid cells and giant cells. The tubercles may heal spontaneously, become fibrotic or calcified, and they persist as such for life while the person remains otherwise healthy (Russell 2006:39). These tubercles would be seen on radiographs as radiopaque nodules. However, in a small percentage of people with primary infection, particularly those who are immunocompromised, the bacteria are not contained within tubercles and instead invade the bloodstream, causing disseminated disease, or "miliary" TB (Goering et al. 2013: 231, Leung et al. 2014, Russell 2006:40).

Secondary TB is due to the reactivation of dormant *Mycobacteria* (caused by some change in the host immune system) or due to reinfection. However, reactivation is usually linked to an impaired immune function caused by poverty, malnutrition, infection such as that caused by HIV, chemotherapy for treatment of malignancy, or corticosteroids for the treatment of inflammatory diseases (Galagan 2014: 307, Goering et al. 2013: 231). The secondary stage starts when the immune response that occurs during the acute and/or chronic phases fails and allows *M. tuberculosis* infection to become clinically apparent. This most commonly presents as pulmonary signs and symptoms, but sometimes as extra-pulmonary disease (Boom 2004:105).

#### 2.1.3 Signs and Symptoms of TB

Signs and symptoms help with the diagnosis of a particular disease. Signs of the disease are features that are visible (eg. pallor) and measurable or objective (eg. weight loss). Symptoms describe what is felt by the patient who is suffering from the disease (eg. tiredness or chest pain). Symptoms of pulmonary TB include extreme fatigue and chest pain. Signs of pulmonary TB include a persistent cough that may be bloody, weight loss, a high temperature (in excess of 38<sup>0</sup> C) and night sweats. The classic signs of the pulmonary form of the disease are fever, cough

and weight loss (Schluger 2008:79). Extrapulmonary TB causes signs and symptoms specific to the area of the body infected. For example, urinary tract TB can cause abdominal pain and persistently swollen glands (NHS 2016a).

(i) Risk factors for infection with TB

The risk factors for contracting TB can be classified into intrinsic, or host factors, and extrinsic, or "environmental" factors. Intrinsic factors which increase the risk of becoming infected with TB include ethnicity. For example, "black" Caribbeans are more susceptible to infection than other ethnic groups. Genetics have been shown to have a key role in host defence and susceptibility to TB. Interleukin-12 (IIL-12), interferon gamma (IFN- $\gamma$ ) and tumor necrosis factor (TNF) have a critical role in the formation and functioning of the tuberculosis granuloma. Recent research has shown that variants of the cytokine-inducible SRC homology 2 domain protein Chromogenic in Situ Hybridisation (CISH) allele (which is necessary for essential IL-12 signalling) have been associated with an increased risk of TB infection (Ponnuswamy 2014:131-2). Families with polymorphisms of IL-12, IFN- $\gamma$ , TNF or their receptors also have a very increased susceptibility to TB infection (Ibid. 2014:132). Liked with ethnicity, increased genetic susceptibility to TB has also been demonstrated in Asians with polymorphisms of the SLC11A1 gene, and also among Sudanese people who exhibited common TLR4 polymorphism (Ibid. 2014:132). Another risk factor is age group, with older people being more susceptible to infection (Cohen and Dye 2014:23, Ponnuswamy 2014:131). Sex is also a risk factor, with males being more likely to become infected than females (WHO 2015).

TB is classed as a "disease of poverty", and many factors can predispose people to becoming poor. If people are poor, then their immune systems do not function as well as they should, possibly because of inferior nutrition and living conditions, and this makes people more liable to contract TB (Ponnuswamy 2014:131). Extrinsic factors increasing the risk of TB infection include being in close contact with anyone who has the disease, and high population density enhances this

prospect. This is because TB is transmitted by infected droplets, and therefore the likelihood of infection increases with duration and intensity of exposure (Ibid. 2014:131). This association has been demonstrated for individuals living in crowded housing and has also been noticed during long-haul air travel (Abubakar 2010). TB bacteria in droplets are killed by exposure to ultra-violet radiation from sunlight, but some cramped, confined conditions, such as in work situations in mines, being a person in a prison, (all conditions that people in the Roman era could have endured), do not favour UV light exposure and can increase the risk of infection as a result (Ibid. 2014:131).

Other extrinsic factors which can predispose people to infection with TB include previous infection with HIV and other diseases which also impair immune system function. Whilst HIV did not exist in the Roman era, other malfunctions of the immune system would have been prevalent. For example, malnutrition results in reduced cellular responses to invading pathogen antigens and also causes a reduction in immunoglobulin. This reduced immunity increases the likelihood of TB infection leading to TB disease. Excessive alcohol consumption inhibits the function of alveolar macrophages and also results in a decrease in immunoglobulin if liver disease occurs as a result of the alcoholism (Ponnuswamy 2014:131). Alcohol was certainly consumed during the Roman era, although it is not possible to conclude if this was in "excessive" amounts. Recreational drug users have an increased risk of becoming infected by TB, as have smokers or people exposed to cigarette or other smoke, for example from open fires using biomass fuels (Ibid. 2014:131), the latter of which will have applied to people in the Roman era. Linked with this is exposure to inhaled dust, (which could have also been relevant in the past), and subsequent silicosis, asbestosis (inhaled asbestos particles), and anthracosis (anthracite inhalation), all associated with increased rates of TB infection (Ibid. 2014:131). Further examples of extrinsic risk factors for TB include malignancy (especially head and or neck and haematological cancers), renal disease, a previous gastric by-pass, coeliac disease, diabetes and people with vitamin D deficiency. People in health care professions and individuals with occupational contact with high-risk groups, for example persons working in

homeless shelters or with substance abusers, are also at risk (Cohen and Dye 2014:23, Ponnuswamy 2014:131-2).

In conclusion, it can be seen that the risk factors for TB, both today and in the past, are many and complex, and some may have synergistic effects, for example, the darker skin of some ethnicities may lead to vitamin D deficiency, which can predispose these individuals to infection with TB. Now that risk factors which increase the risk of becoming infected with TB have been discussed, methods of diagnosing the disease today will be examined.

#### (ii) Diagnosis of TB

The sputum smear microscopy method for the diagnosis of TB was discovered around 100 years ago and is still the most widely used tool for TB diagnosis, particularly in low and middle income countries (WHO 2015:69). The staining of smears and the observation of the resulting slide using a microscope is a relatively straightforward and inexpensive method to perform. However, in patients with a low load of the tuberculosis bacilli in their sputum, for example HIV positive people and children, positive results can be missed. Also, the test provides no information as to the antibiotic sensitivity and resistance profile of the bacteria (Ibid. 2015:70).

Bacteriological culture is still considered to be the reference standard for detecting TB. However, it takes at least six weeks to obtain results (WHO 2015:70) during which time the patient is not receiving treatment and could possibly be infecting other individuals. Culture also requires a well-equipped laboratory with trained staff. Rapid transport is further required to transfer patients' samples from the clinician to the laboratory in order to ensure the viability of the bacteria in the sample. Culture itself also does not provide information on the antibiotic sensitivity profile of the bacteria. This is established through further testing, with Phenotypic Drug Sensitivity Testing being the conventional method used to detect resistance to first and second line TB drugs, although faster liquid culture systems are now

available. However it is proving costly and slow to provide adequate culture capacity in many countries with a high burden of TB (WHO 2015:70). Recently a small number of quicker and more sensitive diagnostic tests have been introduced. These test samples for the presence of drug sensitive and drug resistant TB based on molecular methods. These rapid testing methods include the Xpert MTB/RIF (Mycobacterium tuberculosis / Rifampicin resistance), and lineprobe assays (LPAs) (Ibid. 2015:70). The WHO reports an increase in the use of the rapid molecular test, the Xpert MTB/RIF, since 2010 when it was first recommended. By 2015, 69% of countries recommended using the Xpert MTB/RIF test as an initial diagnostic tool for people at risk of being infected with drugresistant TB, and 60% recommend it as an initial diagnostic test for people with HIV (WHO 2015:2), who may have a low bacterial load that is difficult to detect using the sputum smear technique. However, despite the advantages of using these new molecular diagnostic tests, the conventional microscopy and culture methods still remain necessary for monitoring the response of patients to their treatment regimens (WHO 2015:70). In addition, culture-based drug sensitivity testing methods are currently the only diagnostic techniques available for accurately testing bacterial sensitivity to second-line drugs (Ibid. 2015:70). In June 2015, the WHO started to review the evidence on the use of the urine lateral flow lipoarbinomannan (LF-LAM) assay for the detection of TB in patients with HIV. This test works by detecting a lipoarbinomannan (LAM) antigen, which is only present in people with active TB disease. LAM is a lipopolysaccharide present in the cell walls of *Mycobacteria*. It is released from metabolically active or degenerating bacterial cells, and hence the presence of LAM in a patient's urine indicates active TB. Other advantages of the use of this method are that the urine samples are usually easier to collect than sputum samples (WHO 2015:70).

Throughout 2016, WHO were evaluating and reviewing the use of several other diagnostic technologies for TB testing. These include LPAs for the detection of resistance to first and second line anti-TB drugs, and the use of DNA sequencing for the detection of drug resistance-conferring gene mutations. Other methods under review were the Xpert Ultra assay and the GeneXpert Omni, and several

rapid and sensitive diagnostic tests that will be used in reference laboratories, as well as by clinicians in contact with the patient (WHO 2015:71). In order to reduce transmission of TB and to improve treatment, it will be of great advantage for clinicians to have a positive diagnosis more quickly than when using the current culture and traditional antibiotic sensitivity testing methods. This rapid diagnosis will enable them to put patients onto a suitable treatment regimen almost immediately. Of course, no rapid diagnostic tests were available to Roman people, so anyone infected probably did not initially realise they were ill and could pass on the infection to their close contacts.

#### (iii) Treatment of TB

Tuberculosis has been a disease of humans for many millennia (Roberts and Buikstra 2014:3). During this time, prior to the discovery of suitable antibiotics in the 1940s, many "cures" for the disease were attempted: 'the human desire to help the sick prompted the most fantastic remedies for tuberculosis' (Webb 1936:136). A good example of one of these was blood letting in order to rid the body of blood containing the cause of infection. This method was used from the 5<sup>th</sup> century BC and the time of Hippocrates right up into the 20<sup>th</sup> century (Roberts and Buikstra 2003:225). Bloodletting is still used as a cure in some parts of the world today (Ibid. 2003:225). Leeches were also used in an attempt to drain fluid from swollen tuberculosis infected joints (Smith 1988:43). Other remedies included the recommendation of any methods that were suggested to be able to cleanse the body of infection, for example, sweating, urinating and defaecating (Daniel 1997:42).

Classical physicians had opinions on how TB should be treated. Galen suggested that the disease was at best difficult to cure, and that it was possibly incurable (Pease 1940). On the other hand, Celsus (1<sup>st</sup> century BC to 1<sup>st</sup> century AD) suggested rest and breathing exercises, which was a treatment well ahead of his time. Pliny thought that the application of grease to the shoulders and chest whilst the patient was in a pinewood should cure the disease. Hippocrates suggested

wine, liquid foods and gruel were an effective remedy, while Aelinanus (AD170 – 235) recommended consumption of the blood of a bull, and Tertullian (AD160 – 240) thought consuming butter boiled with honey would be a successful cure. Whilst probably unpleasant, these suggestions were undoubtedly preferable to the drinking of pitch and resin prescribed by Galen, Pliny and Dioscorides (Roberts and Buikstra 2003:225, and references therein).

Pliny was probably the only one of the ancient physicians to suggest a remedy which may possibly have offered some protection against initial infection with TB. He recommended the drinking of milk and the inhalation of smoke from a fire of burning dung (Roberts and Buikstra 2003:225, and references therein). However, exposure to the TB bacillus in this manner would have to be undertaken for a long period in order for resistance to the disease to be built up. It is doubtful that it would have any effect as a cure for an established case of the disease. As these were the only "cures" for tuberculosis that were around in the Roman period, it is clear that the individuals studied in the current research would have had no effective method of treatment other than their own immune systems attempting to combat the disease.

In historical times, in Italy in 1699, the burning of clothes and possessions of people with TB was performed in order to attempt to prevent transmission of infection (Warring 1981:179), although the precise nature of the infective agent of the disease was not yet known. It probably was not until Robert Koch discovered the tubercle bacillus in 1882 that widespread worldwide attempts to control the disease began (Roberts and Buikstra 2003:226; 2014:12). In 1886, France was the first country to introduce laws banning spitting in public places (Dormandy 1999:137), and by 1897 it was well understood that TB was transmitted by infected droplets (Roberts and Buikstra 2014:12).

In 1898 in Britain, the National Association for the Prevention of Consumption and Other Forms of Tuberculosis was formed. This body began work in order to educate the public in avoiding the spread of TB infection and to eradicate the

disease in cattle to prevent its spread to humans. They also created open institutions for the treatment of TB which were accessible to all (Roberts and Buikstra 2003:226). In 1902 in Europe, the International Union Against Tuberculosis was founded to control the disease using methods such as contact tracing and the provision of sanatoria (Evans 1998:13). Arthur Ransome was the first person in Britain to concern himself extensively in the decline of TB (Worboys 2010:150). Ransome claimed that TB declined because of an 'unconscious campaign' of sanitary reform which took place in the 1830s and 1840s. In 1896, he acknowledged the similarities of the disease with leprosy in an article for the Lancet. He stated that leprosy was brought under control by the implementation of improved sanitary methods whilst being largely unaffected by methods to directly prevent the spread of the disease (Ibid. 2010:150). Ransome was an advocate for improving water supply, sewerage systems and better housing with good ventilation (Ibid. 2010:150), which undoubtedly led to better living conditions and improvement of general population health. This could have resulted in fewer people having weakened immune systems and thus succumbing to TB. By 1913, compulsory notification of TB began in England but had already been in place for almost ten years in Scotland (Evans 1998:14). Between the wars, research into TB increased, but it was not until after the Second World War that treatment for TB really improved. Mass radiography became available along with national benefits for people with TB and their families. National rehabilitation schemes commenced with BCG vaccinations and pasteurisation for milk being introduced (Roberts and Buikstra 2003:227).

The use of sanatoria became popular in England around 1840 and treatment in these institutions was one of the main weapons in the fight against TB for around a century. However, there is no scientific evidence that sanatoria had a significant effect on the reduction of the disease (Evans 1998:13). In the UK, although access was available to sanatoria for people of all classes and backgrounds, beds were often scarce and not all patients stayed for the full course of their treatment (Gleissberg 1999:400). It was probably also difficult to recruit nursing staff to work in these establishments due to stigma associated with TB. However, sanatoria did
promote a healthier lifestyle for TB patients. This included exposure to fresh, clean air and sunlight (Roberts and Buikstra 2003:228), and no drinking or smoking was allowed (Ibid. 2003:229). By the late 1950s, sanatoria were being phased out and replaced by drug treatments and vaccines (Warring 1981:184), which resulted in the disease going into further decline. By the late 1980s, TB had become so uncommon that it rarely featured on a school of nursing syllabus and so the level of knowledge and understanding of the disease among UK nurses was probably similar to that of the average lay person (Gleissberg 1999:400-401). The nurses probably also shared misconceptions about the disease with the general populace. The situation changed by the mid-1990s when TB had experienced an upturn and the British Thoracic Society recommended that there should be appropriate numbers of tuberculosis nurses available, according to the number of notifications of the disease within a given area of the UK (Gleissberg 1999:401).

## (iv) Drug treatments for TB

As has been discussed (in section (iii) Treatment of TB), in the Roman era, effective drug treatment would not have been available for TB and other infectious diseases, so reliance was upon the ability of the immune system to overcome infections. While we now have antibiotics, we shall see that infections are becoming increasingly antibiotic resistant (below, in this section), and so humans are returning to the reliance upon their immune systems to rid their bodies of disease. It is necessary to discuss some drug treatments that have been and are being used against TB in modern times in order to learn now progress in treatment has been made and to observe why concerns about mobility spreading the disease are still relevant in today's world.

Effective drug treatment for TB started in 1946 when streptomycin was introduced. Streptomycin, when given alone, caused a large reduction in mortality, and also striking improvements in chest radiology and sputum smear and culture results (Mitchison and Davies 2012:725). Unfortunately, however, after monitoring for a

period of five years, it was discovered that patients treated with streptomycin died in the same proportion and with similar speed to patients not receiving the drug. This was due to the frequent emergence of streptomycin resistance (Fox et al. 1954:347). However, treatment using streptomycin and para-aminosalicylic acid was found to greatly reduce the incidence of streptomycin resistance (Medical Research Council 1950, Fox and Sutherland 1956). In 1952, isoniazid was introduced. This drug was found to be effective in low concentrations and had the added advantage of having a low toxicity (Mitchison and Davies 2012:725). The activity of isoniazid was tested on its own or in combination with streptomycin and para-aminosalicylic acid (Medical Research Council 1952, 1953). Shortly after these trials, drug resistance research discovered that resistant strains of the tubercle bacillus were almost always resistant to only one of the three available drugs (Fox et al. 1957, Mitchison and Selkon 1957). This discovery led to exploring a treatment regimen using the three drugs so that almost any resistant strain of the bacteria would be sensitive to the other two drugs. However, the disadvantage of this "triple" treatment was that it required a prolonged treatment period of at least one year in hospital, and thus was a very expensive method that could only be used in richer countries (Mitchison and Davies 2012:725).

Between the 1960s and up until 1986, further drug development found that the far cheaper drug, thioacetazone, could be substituted for para-aminosalicylic acid (East African/British Medical Research Council 1970). By 1960, it was discovered that chemotherapy at home was as effective as treatment programmes undertaken in expensive hospitals or sanatoria (Tuberculosis Chemotherapy Centre 1959, Andrews et al. 1960). However, this then raised the issue of ensuring patients followed their treatment regimes correctly over the course of the year which was required to cure them (Fox 1958). This issue eventually led to the Directly Observed Treatment, Short-course (DOTS) strategy which was introduced by the WHO and concentrated on the necessity of shortening the treatment period in order to increase patient compliance with the regimen (Mitchison and Davies 2012:725). During the 1950s and 1960s, experiments on mice discovered that pyrazinamide was highly successful at killing tubercle bacilli, which were persistent

following treatment with isoniazid and streptomycin (McCure et al. 1956:763). Later experiments on mice established that rifampicin could accelerate the killing of the bacillus (Grumbach 1970:392). These discoveries led to a new drug regimen being tested that added rifampicin or pyrazinamide to a basic six month treatment programme using streptomycin and isoniazid. This regimen was found to radically reduce the rate of relapse (East African/British Medical Research Council 1972), and thus became the basis of the modern short course treatment. Various forms of this treatment continued to be used until it was found, with the advent of AIDS (Acquired Immune Deficiency Syndrome, the disease caused by HIV), that HIV infection led to an increase in the toxicity of thioacetazone so this drug could no longer be used. Instead, ethambutol was given to replace thioacetazone (Mitchison and Davies 2012:726). At the same time it was discovered that a six month duration of treatment was much more effective than the previously used eight month regimen with a two month continuation phase. The WHO now recommends this six-month regimen (Mitchison and Davies 2012:726, WHO 2015).

Standard WHO recommended treatment for people with drug susceptible TB now consists of a two-month induction phase with, at least, isoniazid, rifampicin and pyrazinamide, followed by a four month consolidation phase with, at a minimum, isoniazid and rifampicin. During the first two months, if therapy is effective, viable bacteria in sputum reduce in two different ways with one sub-population of bacteria being rapidly killed and the other responding more slowly (Horsburgh et al. 2015:2149). The second sub- population has been described as "persistent". These persistent bacteria are thought to be in a metabolic state that makes them less susceptible to being killed by drugs. These differences in susceptibility could be due to local variation in an environmental factor (eg. abundance of bacteria) or to genetic variation of the pathogen (Horsburgh et al. 2015:2149). This would be relevant in the past as much as it is today, although not in relation to drug susceptibility in the past, but to pathogenicity of the disease being higher in some bacterial sub-populations. Thus making it more likely that people in a particular

location could become infected if a sub-population of the bacteria is more virulent than in other areas.

It has recently been discovered that *Mycobacterium tuberculosis* complex organisms are successful pathogens because they have a remarkable ability to exploit cellular host factors to enable their own survival and persistence. The bacteria have been found to establish infection by targeting macrophages in the lungs and to actively evade host defences by modulating host protein transcription and translation (Guler and Brombacher 2015:748). Once inside macrophage cells, *M. tuberculosis* prevents antigen processing and can escape the phagosome. This ability to escape the phagosome determines the bacterial virulence (Ibid. 2015:748). Through further research into the methods that *M. tuberculosis* uses to influence the host immune system, new drugs may prevent this influence. Owing to increased resistance to current drugs, new treatments for TB are urgently required. Some existing drugs also appear to have some potential for TB treatment. For example, the tyrosine kinase inhibitor, imatrnib, promotes maturation of phagosomes, and vitamin D promotes bacteriocidal defence mechanisms, such as the induction of antimicrobial peptides (Guler and Brombacher 2015:748). In addition, it has been discovered that *M. tuberculosis* may use host cholesterol as a nutrient. The cholesterol-lowering drugs, statins, could possibly give increased host protection with reduced bacterial burden, reduced dissemination and decreased pulmonary histopathology (Parihar et al. 2014:754). If people in the past were lacking in vitamin D (likely in England in winter), and if they consumed a diet that was high in cholesterol (evidence from the isotope analysis results suggests the 21 people in the current project all ate animal products which are the largest source of dietary cholesterol - WebMD 2017), it seems that they could have been at increased risk of developing TB.

People today with multiple drug resistant TB (MDR-TB) are known to have infections resistant to rifampicin and isoniazid, and frequently to other drugs (WHO 2015:4), and treatment is complicated (Horsburgh 2015:2152). WHO recommends an initial regimen for treatment of MDR-TB to include four drugs to which the

patient's infection is susceptible, plus pyrazinamide. This treatment phase should last six to eight months (Ibid. 2015). Globally, approximately 3.3% of new TB cases and 20% of previously treated ones have MDR-TB and, in 2014, around 190, 000 people died of MDR- TB worldwide (Ibid. 2015:2). In 2014, more people than in previous years were tested for resistant TB globally. This improvement is largely attributed to the increased availability of rapid molecular testing methods, but the WHO suggests that if all people with TB notified in 2014 had been tested for antibiotic resistance, an estimated 300,000 would have been found to have MDR-TB; 54% of these new cases occurred in India, China and Russia (Ibid. 2015:2). However, globally, the most common diagnostic test is still the sputum smear microscopy method that was developed more than 100 years ago and, as previously discussed, this gives no information as to the antibiotic sensitivity pattern of any Mycobacterium tuberculosis complex infection identified. In countries with better access to laboratory facilities, TB diagnosis is carried out by bacterial culture, the current reference standard (Ibid. 2015:4), which also provides no information about antibiotic resistance of identified strains of the tubercle bacillus. This resistance knowledge is critical because MDR-TB is increasing, strains of Extensively Drug Resistant TB (XDR-TB) are now appearing, and appropriate treatment for people with these strains needs to be given. XDR-TB is defined as MDR-TB plus resistance to at least one fluoroquinolone and also to a second line injectable antibiotic (Ibid. 2015:58). This strain had been reported by 105 countries by 2015, with an estimated 9.7% of people having XDR-TB (Ibid. 2015:2).

In summary, a better understanding of how and where drugs penetrate the tissues, along with the optimum dosages, is needed in order to achieve effective drug levels in the infected tissues of the body (Horsburgh 2015:2157). As has been discussed, the current recommended treatment for new cases of people with drug-susceptible TB is a six-month regimen of four first-line drugs: isoniazid, rifampicin, ethambutol and pyrazinamide. This treatment has a success rate of 85% or more (WHO 2015:4) but, with increasing rates of resistant strains of TB causing infection, it is imperative that new diagnostic tests and novel treatments are

developed and used carefully. Methods of transmission also need to be fully investigated in order to decrease transmission rates and thus the numbers of new cases of the disease arising each year. Between 1995 and 2011, 51 million people were successfully treated for TB. However, while progress in treatment of multi-drug resistant TB (MDR-TB) remains slow, innovations in diagnosis have moved on with the Xpert MTB/FIF, a rapid molecular test that can diagnose TB and rifampicin resistance within 100 minutes (Ibid. 2012). This will hopefully replace the sputum microscopy smear and notoriously difficult culture methods currently used in diagnosis in the near future. Clinical and pharmaceutical research has also lead to the development of new drugs and vaccines which are in the developmental and clinical trial stages (Ibid. 2012, Schluger 2008) and are needed to achieve the targets set by WHO in the End TB Strategy (England et al. 2008:499).

#### (v) Vaccinations against TB

As has been previously discussed, TB is a complicated disease. It is an infection whose pathogenesis is characterised by a period of subclinical, asymptomatic infection which may continue for varying periods of time, from weeks to decades. As a result, a large reservoir of infected people exists and new cases of active TB disease may arise from this group at any time (Rangaka et al. 2015:2344). Added to this problem is the increasing emergence of antibiotic resistant strains of the TB bacteria, namely MDR-TB and XDR-TB, which take a long time to cure and are challenging and expensive. It is therefore becoming increasingly important that new and effective vaccines are developed to prevent infection in the first place, or are used to stop a latent TB infection from developing into an active disease if there is any hope of reduction and eradication of the disease in the future. The only vaccine that is currently available is that of Bacillus Calmette-Guérin (BCG), which has been in use for a century in many countries (Tang et al. 2016:30). However, the main problem with this vaccine is that it does not protect adults against pulmonary TB although it is reasonably effective in protecting

children (Rangaka et al. 2015:2345, Tang et al. 2016:469), particularly from tuberculosis related meningitis (Tyagi et al. 2011:469). Another problem with the BCG vaccine is that prior exposure to environmental mycobacteria interferes with the BCG efficacy, and that efficacy is variable, ranging from 0% in Southern India up to about 80% in the UK (Tang et al. 2016:33). In addition, BCG does not induce long lasting immunological memory and hence its efficacy against TB in adults is questionable (Tyagi et al. 2011:469, Rangaka et al. 2015:2345, Tang et al. 2016:30). The aims of a new vaccine would be to prevent new TB infections or to stop latent TB infection from developing into TB disease, or to eliminate *Mycobacterium tuberculosis* infection completely (Ibid. 2016:33).

Currently, most vaccines that are being developed target the prevention of new infections and the reactivation of latent TB (Tang et al. 2016: 33). Some of these vaccines that are in trial presently are already proving to be more successful than the BCG in offering protection (Ibid. 2016:33). However, despite the promising trial results of these new vaccines, none of them have demonstrated the ability to completely prevent and eradicate *Mycobacterium tuberculosis* infection in humans or other animals (Ibid. 2016:37).

In summary, whilst vaccine development is moving on, and several different types of vaccine with differing modes of action are currently undergoing trials, it still remains necessary to discover novel TB vaccines which are more effective. This will necessitate further research into the correlation of immunity with effective protection against the disease, and also a deeper understanding of the pathogenesis of *Mycobacterium tuberculosis* and its interaction with the host defence system (Tang et al. 2016:37). In this respect, little progress has been made since 2011 when Tyagi et al. (2011:477) concluded that, although many TB vaccines developed in various countries had shown superior protection against TB compared with BCG in animal models, some of these vaccines were still in clinical trials. Obviously, clinical trials take a long time, but, in the meantime, it is important that researchers continue to work on new types of vaccine if worldwide eradication of TB is ever to be achieved.

#### 2.1.4 TB: a re-emerging disease

As has been discussed, due to new drugs and improved treatment regimens, TB was thought to have been under control by the late 1980s (Smith 1988:2), but it had begun to rise again by the early 1990s, particularly in relation to HIV infection (Pozniak 2008:343). The WHO Global Tuberculosis Report 2012 stated that in 2011 there were an estimated 8.7 million new cases of TB globally, 13% of these being in people who also had HIV infection (WHO 2012). In 2012 an estimated 8.6 million people developed TB and 1.3 million died from the disease, with 320,000 of these deaths being in HIV-positive people (WHO 2013). By 2013, nine million people worldwide were estimated to have developed TB, 1.1 million (13%) of these people developing TB in 2013 were estimated to be HIV positive. HIV increases the risk that a latent TB infection will reactivate and that new infection or re-infection with TB will progress to active disease. Progression from infection to disease in a patient with HIV usually occurs within three months (Goldfield and Ellner 2007:526). The increased susceptibility of HIV patients to TB increases the incidence of TB in the community, and atypical features of the disease associated with TB infection, such as sputum smear negativity and isolated extra-pulmonary disease, make delays in diagnosis highly probable (Ibid. 2007:527). This means that during that time the person would be untreated and could be transmitting the infection. In addition to HIV causing acceleration from latent TB infection to active TB disease, TB also accelerates the course of HIV by activating viral replication and increasing the rate of decline of CD4 T cell counts (Ibid. 2007:527). Along with these issues are the problems that HIV/TB co-infected people have with drug interactions and drug toxicities that do not affect TB patients without HIV (Ibid. 2007:527). It should be noted that the prevalence of TB/HIV co-infection is highest in low-income countries such as those in sub-Saharan Africa (Ibid. 2007:529).

In 1993 the World Health Organisation (WHO) declared TB a global emergency (Squire 2008:288), and launched the *Stop TB Strategy*, with specific targets to eradicate TB by 2050 (England et al. 2008:499). In 2013, there were reported to be 1.5 million deaths from TB (WHO 2014). The latest WHO figures (WHO 2015)

illustrate that the policies for global reduction of TB are having some effect; TB mortality has decreased by 47% since 1990. This has been attributed to effective diagnosis and treatment, which has saved around 43 million lives between 2000 and 2014; TB incidence has declined globally by 1.5% per year since 2000. This means the incidence is now 18% lower than it was in 2000 (WHO 2015:1). However, in 2014, TB still killed 1.5 million people, which is similar to the figures for 2013. Of these, 1.1 million people were HIV negative and 0.4 million were HIV positive; 890,000 were men, 480,000 were women and 140,000 were children. TB now ranks alongside HIV as a leading cause of death worldwide (Ibid. 2015:1), and therefore despite reductions in mortality and incidence, it is still a major global challenge. The aims of WHO are now to end the TB epidemic (in other words, to have an incidence of fewer than 100 cases per million people) by 2035 (Ibid. 2015:x).

Figure 2.3 shows the latest (2014) WHO figures for the global incidence of TB:



Estimated TB incidence rates, 2014

Fig. 2.3. Global incidence of TB (WHO 2015)

In the UK in 2014 (the latest figures available from WHO in the report WHO 2015), there were a total of 7,077 new TB cases notified. This is a slight decrease from the number of new and relapse cases notified in 2013, which WHO reported was 7,293 (WHO 2015) but Public Health England (PHE) reported as 7,892 for the same period (PHE 2014:7), with 73% of these people being born outside the UK, and stating India, Pakistan and Somalia being the most common countries of origin (PHE 2014:13) and only 15% being recent migrants (that is, within the last two years) (Ibid. 2014:7). PHE state that 7,892 cases equates to an incidence of 12.3 per 100,000, with overall TB cases having declined in the two years prior to 2013, due to a small decline in the numbers and rates in the non-UK born population (Ibid. 2014:7). This decline is likely to have been influenced by a number of factors including changes in migration patterns, with less migrants entering the UK from high and very high TB burden countries (Ibid. 2014:50). It is likely that the UK-based and global interventions to improve the control of TB. such as pre-entry screening for migrants from all high incidence countries (Ibid. 2014:17), could also be responsible for the decline in cases. As in previous years, London accounted for the highest proportion of TB cases in 2013 (37.8%) which is a rate of 35.5 cases per 100,000 (Ibid. 2014:9). The main burden of disease countrywide was concentrated in other large urban areas (Ibid. 2014:50).

Of the new cases reported in the UK in 2014, 63 were confirmed as being infected with MDR-TB (WHO 2015), and PHE confirmed this has remained constant over the past three years with 87.3% of these cases being born outside the UK (PHE 2014:25). Among all culture-confirmed cases of TB in notified in the UK in 2013, 97.5% were caused by *Mycobacterium tuberculosis*, 0.6% by *M. bovis*, 1.4% by *M. africanum* and 0.5% with *M. tuberculosis* complex bacteria which were not further differentiated (Ibid. 2014:25). 82% of these culture-confirmed cases were strain typed and there was found to be considerable variation in lineage by country of birth, with 35% of cases being of Euro-American lineage, 25.7% were Central Asian strain, 12.2% were East African Indian, 5.3% were Beijing strain, 0.8% were *M. africanum*, 0.5% were *M. bovis* and 19.1% had no lineage assigned to them (Ibid. 2014:40).

Globally, of the 9.6 million new cases of TB notified in 2014, 58% were in the South East Asia and Western Pacific regions (WHO 2015). The African region had 28% of the World's TB cases in 2014 and the most severe burden relative to the population size, with 281 cases for every 100,000 people. This is more than double the current global average of 133 cases per 100,000 people. Meanwhile, India, Indonesia and China had the largest number of TB cases at 23%, 10% and 10% of the global total, respectively (Ibid. 2015:2).

While only 5 to 15% of the estimated global 2 to 3 billion people infected with *Mycobacterium tuberculosis* complex go on to develop the actual disease, the WHO state that this figure is probably much higher in people infected with HIV as has previously been discussed. Additionally, death rates in untreated individuals are high; without treatment, the death rate is approximately 70% within 10 years from diagnosis for sputum smear positive pulmonary TB and 20% for people with culture positive but smear negative pulmonary TB (WHO 2015:4).

Many risk factors have been identified that can predispose people to TB infection. Some of these are similar today to those in the past (eg. poor diet, overcrowding and travel/migration), but the single and most important factor from past to present appears to be poverty. There appears to be a higher incidence of HIV and TB co-infection in Africa, (WHO 2015) where high levels of poverty could lead to impaired immunity in the population. High-income countries (where there is less poverty) have a lower incidence of TB, while frequency and death rates are lower in all areas of the world except African countries. This is partly because of the ease of access to, and use of, antibiotics in developed countries (Goering 2013:232), although TB is a complex disease and other poverty-related factors, such as malnutrition, can also lead to compromised immune systems and therefore higher incidences of infection. It is argued here that dealing with poverty around the world would drastically reduce the problem of TB, but mobility also has a large effect on the incidence of the disease.

It is increasingly being recognised that there is a specific link between poor health and migrant populations, and it is well documented that there is a higher risk of TB in travellers and in people who migrate to other countries (WHO 2010, Albert and Davies 2008:367). For example, there are studies exploring the link between air travel and the transmission of TB, and recommendations for the reduction of these risks (eg. Dowdall et al. 2010, Neilson and Mayer 2010). Travel permits the introduction of TB into previously unexposed populations. In addition to this, the travellers themselves may be more vulnerable because they are adapting to changes in their environment, and this process places stress upon their immune systems and makes them more prone to infectious diseases or activation of a dormant primary TB infection. Moreover, as the WHO (2015) suggests, immigrants often experience poor living conditions as they travel and when they arrive in new countries. Although the Roman world would have been very different from the modern world. Hall suggests that the working classes and craftsmen of Roman London would have been migrants to the town with some of the more specialist craftsmen being brought to London from abroad (Hall 2005:125). These people were likely to have lived in recently excavated Mediterranean-style buildings with timber frames and no foundations, which, due to problems with rising damp, would have needed replacting every five to ten years (Ibid. 2005:125). Such damp living conditions would certainly have predisposed the craft-workers and shopkeepers living there to TB. Hall also describes people as living in close proximity to each other (Ibid. 2005:127), another factor making the transmission of TB more likely.

That immigrants experience poor living conditions whilst travelling and sometimes on arrival at their destination was therefore probably true in the Roman era and would certainly be the case for people involved with the 18<sup>th</sup> and 19<sup>th</sup> century slave trade when Africans were forced to migrate to the Americas (eg. Merbs 1992), but also for refugees in the past and political asylum seekers today (Figueroa-Munoz and Ramon-Pardo 2008:733). One of the most frequent migration pathways is possibly from rural to more urban environments in the pursuit of work. However, in the past and also in developing countries today, migration could mean exposure to overcrowded living conditions, unsanitary situations and an increased risk of

contracting TB and other infectious diseases. In addition, people become exposed to new diseases when they travel, while carrying pathogens with them that could cause infection in the local population. Furthermore, the food that people eat can transmit pathogens, including *M. bovis* (Stone et al. 2012), so poor food preparation and storage methods encountered whilst travelling could lead to increased exposure to TB.

## (i) Eradication of TB

The global vision of the End TB Strategy (2016 – 2035) is for zero deaths, disease and suffering due to TB (WHO 2015:7). This aim is building upon the previous achievements of targets set in the Millennium Development Goals which stated that, by 2015, world TB incidence should fall and TB prevalence and mortality rates should be halved compared with their 1990 levels (Ibid.2015:7). The Stop TB Strategy was developed for the period 2006 – 2015 in order to try to achieve these aims. In addition to the 2015 target, the Stop TB Strategy's ultimate aim was for the elimination of TB as a public health problem (defined as less than one case per million population per year) by 2050 (Ibid. 2015:6). This is a continued aim of the End TB Strategy, which has the ultimate goal to end the global TB epidemic. Although a date is not set for when this is to be achieved, it is assumed that as the End TB Strategy continues until 2035, this goal is intended to be achieved by then.

Theron et al. (2015) have suggested that ending the global TB epidemic will require a shift from strategies which focus on control of the disease to those which actually focus on elimination of it (Ibid. 2015:2324). In order to achieve this shift, they suggest a three-step process is required;

- improved collection and use of existing TB notification data,
- collection of data additional to notification of new TB cases (eg. drug resistance, risk factors and precise geographical location of new cases) in order to develop tailored responses in treatment and control,

targeted collection of novel data (for example, sequencing data and contact investigations).

The researchers hope this expanded data collection will result in the improvement of the understanding of the dynamics of TB transmission (Theron et al. 2015:2324). It has been these sorts of responses that have been instrumental in eliminating other infectious diseases such as smallpox (Fenner et al. 1988), and all such successful methods have involved two components, namely the systematic reporting of every case of the disease, and successful identification of disease clusters at the local level (Theron et al. 2015:2324).

Local TB epidemics have been found to differ in intensity, "drivers" and key characteristics. Approaches to control that work in some hotspots (eq. urban areas) may not work in others, for example prisons or rural villages with poor access to care (Theron et al. 2015:2324). Efficient collection of data at a local level is therefore suggested to be key to the development of effective control strategies. Unfortunately, routinely collected data for TB vary substantially in scope and detail between countries and, although WHO recommends the use of secure and selfcontained electronic systems, paper forms are still predominantly used, with delays thus occurring for this information to be entered into databases (Ibid. 2015:2325). The researchers also suggest that because TB in children is currently under detected, there should be more of a focus on childhood TB; this is of particular importance for interrupting transmission of the disease. Additional data collection to include would be the reporting of the geographical location of newly notified cases of TB, to assist with community-based follow-ups or transmission hotspot mapping. Drug-resistance patterns for predicting region-specific drug susceptibility, and documenting risk factors, such as diabetes, smoking, previous hospitalisation or imprisonment, in order to inform local screening strategies should also be included (Ibid. 2015:2326). Other suggestions for enhancing data systems included investment in surveillance systems for TB, including WHOsupported electronic data collection systems, in order to improve local control of TB (Ibid. 2015:2328). While this will undoubtedly be easier in some areas of the

world than others, they suggest implementation of flexible systems for a locally tailored TB response. This is especially important in high burden countries with resource limitations, little political support and the highest need for such systems, but it is acknowledged that this will present great challenges.

Having considered current issues concerned with TB diagnosis, treatment and possible future eradication, TB in the archaeological record is now discussed, starting with skeletal diagnosis of the disease in skeletal remains.

# 2.2 Tuberculosis in the past: recognition of the skeletal changes and their differential diagnosis

In the archaeological record, osteological examination of the skeleton and diagnosis of disease is needed in order to learn what we can about morbidity and mortality in the past. However, not all illnesses will leave their mark upon the skeletal tissue, perhaps because these diseases affected only the soft tissues, or perhaps because the person died before the infection, in the case of infectious diseases such as TB, could cause bone changes (Ortner 2003:51). Fortunately, TB does sometimes leave traces on the skeleton, but some of these are non-specific changes that could also be caused by other diseases. However some, such as Pott's disease of the spine, are accepted as being strongly likely to indicate the presence of TB disease. These specific and non-specific bone changes associated with TB will now be detailed and discussed.

The skeleton can be affected by TB when tubercle bacilli, which are circulating in the bloodstream or lymphatic system of an infected individual, begin to invade the bones. This occurs largely in bones with large amounts of haemopoietic (red) marrow, which is essentially cancellous bone, rather than the cortex or medullary cavity (Resnick and Niwayama 1995:2462). Within the long bones of adults, this means the metaphyses and epiphyses are primarily affected, and also the spine, with the spine being implicated in 50% of adult cases with bone involvement

(Ormerod 2008:166). In infants and young children, there is more haemopoietic marrow in the skeleton, and hence tuberculous foci often occur in tubular bones of the hands and feet and in ossification centres of tarsal and carpal bones, in addition to lesions in the long bones (Resnick and Niwayama 1995:2477). Figure 2.4 shows some of the more common sites of the skeleton that can be affected in TB:



Fig. 2.4 The most common sites of skeletal TB infection (Steinbock 1976:178), coloured black, although any bone or joint could be affected by TB, as is shown by the grey shaded areas.

*M. tuberculosis* and *M. bovis* can both produce similar skeletal changes. Left untreated, *M. bovis* causes bone and joint TB in approximately 20% of all cases (Resnick and Niwayama 1995:2462), and this is particularly thought to be the case in children (Myers 1951). Unfortunately for palaeopathology, studies suggest only 3-5% of infected and untreated people with TB may display skeletal involvement (Peto et al. 2009:1350) with some more conservative estimates putting this figure

for untreated infections today as low as 1 to 3% (Rankin and Tuli 2010:161). These bone changes tend to present three to five years following initial respiratory infection, after the bacteria have lain dormant. The low frequency of skeletal pathology of course cannot be taken as representative of the total number of people infected in the past, as aDNA studies have recently proven, because skeletons with no bone change have been analysed and have shown that the person had TB when alive (Müller et al. 2014a).

There are three types of TB in humans, namely primary, secondary and miliary. Primary TB from a site of initial infection is the most common form of TB in children (Leung et al. 1992:87), with secondary TB resulting from reactivation of latent infection and usually occurring in adults and adolescents (Marais et al. 2004:394). However, miliary TB results from the extension of primary caseating lesions into pulmonary blood vessels, leading to haematogenous or lymphatic spread of infection to lungs and distant sites. This is a frequent complication in children (Shingadia and Novelli 2003:635). Miliary TB and meningitis can occur one to three months after exposure to infection, with spinal and joint changes becoming evident after one to three years in children under five years of age (Wallgren 1948:245).

Congenital TB is considered to be rare, i.e. transmission from mother to child via the placenta or by the foetus ingesting bacteria through infected amniotic fluid (Hakim and Grossman 1995:119, Lorin 1983:333). In children aged over 10 years old, the most common manifestations of skeletal TB are spondylitis (spinal lesions), with joint involvement and osteomyelitis being more common in younger children (Teo and Peh 2004:853). Lewis (2011) examined some of the children at one of the study sites in the current research, namely Poundbury in Dorset and she points out that much of what is known about the prevalence and progression of TB in children is based on post-1950s data where the natural progression of the disease, and even how it is expressed in the skeleton, is affected by chemotherapy. In the pre-antibiotic era there was likely to have been a more

severe expression of the disease resulting from the lack of suitable treatment to reduce numbers of bacteria present in the individual (Lewis 2011:13).

A wide range of sites in the skeleton can be affected by TB (Figure 2.5). These are now discussed, starting with the most common site: the spine.



*Fig. 2.5 The distribution and frequency of skeletal tuberculosis. (Aufderheide and Rodríguez-Martin 1998:134). These data were derived from pooling the results of clinical studies by Fraser (1914), Johansson (1926), Sorrel and Sorrel-Dejerine (1932), Sevastikoglou and Wernerheim (1953), LaFond (1958), Somerville and Wilkinson (1965), Tull (1975), Martini et al. (1986), and Martini (1988).* 

## 2.2.1 The Spine

Bone changes in the spine are the most common and most characteristic skeletal lesions which can be caused by TB (Ortner 2003:230). In the past, the disease usually began in childhood but today, older individuals are often affected (Resnick and Niwayama 1995:2464). Spinal change is the most commonly reported skeletal change associated with TB in bioarchaeology (Ibid. 1995:2462) and accounts for between 25 to 60% of all cases of bone and joint TB (Ibid. 1995:2463). The most common part of the spine to be affected by TB is the first lumbar vertebra with the lower spine being the primary focus for skeletal TB for people of all ages (Ortner 2003:231, Moon 1997:1791, Rajeshwari and Sharma 1994:1214). Frequency of involvement of other lumbar vertebrae decreases with distance from this site (Resnick and Niwayama 1995:2436), however cervical spine infection may occur in individuals with absence of any other vertebral involvement (Mathew and Bais 1997:899). Today, spinal damage has been estimated in 25 to 50% of people with untreated skeletal TB (Resnick and Niwayama 1995:2462). However, this figure appears to be dependent on the population studied, with other researchers suggesting 40 to 50% of all people with skeletal TB are estimated to have involvement of the spine (Moon 1997:1791).

Resnick and Niwayama (1995:2464) suggest infection of the spinal vertebrae first takes place in the anterior part of the vertebral body. Even after extensive destruction of several adjacent vertebral bodies, it is uncommon for the damage to extend to the vertebral arches and the spinous processes are almost never destroyed (Ortner 2003:231). The damage to the vertebral body can lead to collapse and this will result in kyphosis of the spine (Resnick and Niwayama 1995:2472). This can be seen in Figure 2.6 and is known as Pott's disease after the 19<sup>th</sup> century physician, Sir Percival Pott, who first named the condition (Luk 1999:338).



Fig. 2.6 Pott's disease of the spine. (Courtesy of C.A. Roberts)

However, other diseases can cause similar spinal changes. These diseases include brucellosis, fungal infections, pyogenic osteomyelitis, vertebral fractures and neoplasms (Évinger et al. 2011:165) and therefore careful differential diagnosis is required to eliminate these alternative causes in skeletal remains. Bone lesions caused by TB are reported as occurring in other locations of the skeleton, but most frequently at major weight-bearing joints such as the hip and knee where the typical lesion is destruction of the articular surfaces of the joint (Resnick & Niwayama 1995:2480). Therefore, looking at the location of changes in the spine and within affected vertebrae, and examining a skeleton for lesions in weight-bearing joints may help the osteologist to make a differential diagnosis for TB. Roberts and Buikstra (2003:96) suggest that half of the people with spinal damage caused by TB will also have signs of bone infection elsewhere in their bodies, which would substantiate any diagnosis of TB. These other bone changes shall now be examined, starting with the hip joint.

## 2.2.2 The hip Joint



*Fig. 2.7 Destructive lesions of the hip joint (circled), and loss of head of the femur, possibly caused by TB* (Courtesy of C.A.Roberts).

The hip joint is the second most frequent part of the skeleton affected, making up approximately 20% of the cases of skeletal involvement (Aufderheide and Rodríguez-Martin 1998:139). It tends to occur in childhood (most commonly between three and 10 years of age) with an onset after 25 years of age being rare (Ibid. 1998:139). The lesions which form are destructive (see Figure 2.7) and can lead to total bone destruction and partial or complete dislocation of the remains of the femoral head or neck (Schinz et al. 1953, in Aufderheide and Rodríguez-Martin 1998:139). Diagnosis may be confused with septic (non-tuberculous) arthritis but this tends to have much more limited bone destruction than TB, in addition to some bone formation, which is unlikely to occur in TB. Growth deficit may also be observed (Aufderheide and Rodríguez-Martin 1998:139).

## 2.2.3.The knee joint



Fig. 2.8 Bone destruction in the knee joint, possibly caused by TB. (Courtesy of C.A. Roberts).

TB of the knee joint is not uncommon, being seen in approximately 16% of cases of skeletal TB (Aufderheide and Rodríguez-Martin 1998:139), with the majority of cases beginning before the age of five years and with an equal distribution between the sexes. The infection usually starts in the synovium and can extend along the capsular insertions of the femur and tibia, and along the cruciate ligament attachments. Destruction of the articular surface then occurs (See Figure 2.8). Tuberculosis of the knee is more often unilateral than bilateral, and dislocation with associated deformity can occur in severe cases, particularly in children. Generally, adult cases show less bone destruction than infant cases (Martini and Ouahes 1988:861).

# 2.2.4 Other sites of skeletal TB

Although the spine, hips and knee are the most common sites, any bone or joint can be affected by TB (Resnick and Niwayama 1995:2462) as tuberculous osteomyeltitis can remain localised to bone or it can spread to include adjacent joints (Ibid. 1995:2474). Some other areas of skeletal involvement will now be briefly considered.

# (i) The ribs

Bone changes in the ribs are more non-specific in nature and could be caused by chronic chest infections and conditions other than TB. However rib lesions need to be discussed, due to the high number of individuals (17 out of 21) in the current project sample having them.

The middle and lower rib cage is more often affected by bone changes than the upper region (Eyler et al. 1996:926) and infection arrives here from a peripheral lung focus of TB infection which disseminates directly to affect the pleura and ultimately the visceral/internal surfaces of the ribs (Roberts et al. 1994:169). Pulmonary TB may be the cause of empyema in the pleural cavity, which may initiate inflammatory changes on the ribs (Ibid. 1994:169). Rib lesions are described in some detail now, as previously mentioned, 17 out of 21 people used in this project had rib lesions described as being likely to indicate TB infection. The typical rib lesions discussed are shown in Figures 2.9 and 2.10 and are characterised mainly by new bone formation;



*Fig. 2.9 Ribs showing new bone formation (grey - circled) on the visceral surfaces, Skeleton 189 from Cirencester (sample CIRE189 used in this study), Roman Gloucestershire (*Courtesy of C.A. Roberts)



*Fig. 2.10 The effects of TB as additional bone growth on the visceral surfaces of the ribs (circled).* (Courtesy of C.A Roberts)

In 1984, Kelley and Micozzi presented what they stated as being 'a new diagnostic approach to tuberculosis' (Kelley and Micozzi 1984:381). They researched the link between a mild form of periostis (periosteal new bone) on the internal surface of the ribs and pulmonary TB. The start of "rib lesion" research focused on the Hamann Todd documented skeletal collection in the United States (Ibid.1984).

This collection is curated in the Cleveland Museum of Natural History and comprises individuals who died in the early 20<sup>th</sup> century: these people were white and black adult Americans from the lower socio-economic classes of greater Cleveland, Ohio, USA. Information documented for each individual includes name, age, sex, "race", height, weight, cause of death and whether an autopsy was performed or not (Ibid.1984:381-2). Kelley and Micozzi concluded that, of the 39 people found to have rib lesions, 31 had died of pulmonary TB, and seven were listed as dying from "tuberculosis" but the location of this infection was unspecified. One person died of meningeal TB. In addition to these 39, only two people who had died from pneumonia had any rib changes and these were very slight (Ibid. 1984:382). However, this study was weakened by the lack of two additional control groups of skeletons; those who had died of a lung disease that was not pneumonia and those who had not died from any form of lung disease to see if other lung diseases and uninfected individuals also had any lesions on these bones. Without these controls, it is difficult to conclude how valid it is to record rib lesions as being strongly indicative of TB in skeletons when other diseases could cause similar bone changes. These issues were examined in later work.

The documented Robert J. Terry Collection, curated in the Department of Anthropology, National Museum of Natural History, Washington DC, USA was the focus of the next study (Roberts et al. 1994). Individuals in this collection had dates of birth between 1822-1943 (Hunt 2016). There were 414 of 1718 individuals (24%) who displayed bone formation on one or more ribs, and 62% of the individuals with lesions had died of TB, or TB alongside another pulmonary disease. These findings emphasise that lung diseases other than TB can also cause rib lesions. It was also found that 15% of individuals with rib lesions had died from non-pulmonary causes. However, overall, individuals with rib lesions were more likely to have had TB or TB and another lung disease as their cause of death.

More recent work on rib lesions in a palaeopathological context which supported the findings that rib lesions may be indicative of TB infection was carried out on

the Coimbra Identified Skeletal Collection curated at the Department of Anthropology, University of Coimbra in Portugal (Santos and Roberts 2006). This collection includes individuals with dates of birth ranging between 1826-1922, and dates of death between 1904-1938. Of the individuals with rib lesions, 85.7% (69/81) had TB (pulmonary or extrapulmonary) stated as their cause of death compared with 17.8% (16/90) with rib lesions and a non-TB cause of death (Ibid. 2006:38).

These findings support those of a clinical study into rib lesions by Eyler et al. (1996), who studied radiographs of living people who were categorised into four groups. Group I consisted of those with chronic pleural disease, group II consisted of those who had had TB for more than five years, and group III was made up of patients with empyema (thoracic cavity abscess). The data on rib enlargement (presumably due to new bone formation) from these three groups were all compared with a control group without any lung diseases to see whether any group had a higher frequency of rib enlargement (Eyler et al. 1996:921). The researchers found more asymmetry between the thickness of ribs on the diseased and non-diseased sides of the thorax of patients with chronic lung infections than in the control group patients (Ibid. 1996:924). The most common condition associated with rib enlargement was TB. This was concluded not to be skeletal TB *per se* because there was no evidence of bone destruction, and the enlargement of the rib remained stable over time (Ibid. 1996:925).

The conclusions to be drawn from these studies suggest that rib enlargement/ changes may be seen in patients with chronic pleural disease, and the disease most commonly associated with these rib changes appears to be TB (Eyler et al. 1996:926). However, research focusing on rib lesions in skeletal remains appears to be mainly confined to palaeopathology rather than clinical medicine (Roberts 1999:312) and thus clinicians are likely to be unaware that they can occur. Further research along the lines of Eyler et al.'s work is needed, but with a far larger sample size of patients known to be suffering from tuberculosis and patients with other chronic lung diseases, as well as patients without any form of lung disease

whatsoever. This would establish how likely it is for new bone formation on the ribs (leading to their enlargement) to be indicative of pulmonary TB. Until such time, new bone growth and thickening of the ribs of skeletons found in archaeological contexts may be recorded only as indicating a chronic pulmonary condition. These bone changes appear to be more likely to be caused by TB than any other lung disease, but until further work has been done in clinical contexts they can still only be classed as a non-specific indicators of the infection.

(ii) The trochanter of the femur.



*Fig. 2.11 Bone destruction of the trochanter of the femur (circled), possibly caused by TB.* (Courtesy of C.A. Roberts)

Destruction of the greater trochanter of the femur can result from complications of spinal TB. Chronic TB of this part of the femur is not common but when it does occur, it most commonly tends to affect individuals between 10 and 40 years of age, with the lesion remaining localised in the trochanteric area, which is

progressively destroyed (see Figure 2.11), (Ortner 2003:239, Resnick and Niwayama 1995:2465).

## (iii) The sacroiliac joint

Fig. 2.12 Bone destruction (circled) associated with possible TB. (Courtesy of C.A. Roberts).

TB of the gastrointestinal tract may extend to the pelvic bones. The sacrum, ilia and sacroiliac joint may be affected if this situation occurs. The sacroiliac joint is involved in TB in approximately 2% of cases, and is one of the few joints that, if infected, shows bilateral involvement. This is because most of the cases are linked with spinal involvement, with the sacroiliac joint becoming affected as a result of direct extension of the infection. As a result of this taking time to occur, sacroiliac lesions are rarely seen until very late in childhood/ young adulthood (Aufderheide and Rodríguez-Martin 1998:139). The majority of lesions are destructive to the bones (Ortner and Putschar 1985:149 in Aufderheide and Rodríguez-Martin 1998:139) as can be seen in Figure 2.12.

## (iv) The ankle and feet

TB involvement of the short, tubular bones of the hands and feet is known as tuberculous dactylitis. This is especially common in children with the condition increasing in frequency after the age of five years, and becoming rare after ten years of age, before increasing again in frequency in an adult (Resnick and Niwayama 1995:2477). Ankle involvement in TB is also most frequently observed in children (Ortner 2003:241). One of the most frequent sites of skeletal TB in infants and young children is often multiple involvement of metacarpals, metatarsals and phalanges, known as tuberculous dactylitis, as previously mentioned, or *spina ventosa* (Resnick and Niwayama1995:2477). If the child does not die from TB located elsewhere, these lesions often heal. However, destruction of the growth plate in metacarpals and metatarsals (and less frequently of the phalanges) may lead to shortening of the affected digits on healing (Ibid. 1995:2477). In children, tuberculous dactylitis does not affect the phalangeal joints, whereas in adults the phalanges may occasionally be affected, with the lesion extending into the joint (Ortner 2003:242).

## (v) The shoulder, elbow and wrist

The shoulder, elbow and wrist joints can also be affected by TB. Adults are more frequently affected by lesions in the shoulder than children, although these can occur at any age, with males being affected more frequently than females. If it does occur in children, shoulder TB may heal, although extensive destruction of the humeral head and of the glenoid fossa is common in adults, with the occasional involvement of the acromium and clavicle (Ortner 2003:242). Elbow TB most commonly affects individuals up to 20 years of age, with the most frequent osseous involvement being that of the distal humerus followed by the proximal ulna, being least common in the proximal radius (Ibid. 2003:243). TB of the wrist can occur equally in children or adults, but involvement of different joints is characteristic of different age groups. The metacarpophalangeal joints are most commonly affected in children, with the radiocarpal joint remaining unaffected. In

adults, lesions usually begin in the radiocarpal joint from where they spread rapidly throughout the joint compartments of the wrist (Ortner 2003:243). Involvement of the shafts of the long bones is uncommon, with estimated frequency of occurrence in less than 1% of cases of TB although, for reasons unstated, this is somewhat greater in Chinese patients (Resnick and Niwayama 1995:2474). When tuberculous osteomyelitis of the long bones does occur, it is almost exclusively observed in children and then usually as a manifestation of multiple skeletal foci, particularly spina ventosa (Ibid. 1995:2477). The highest frequency of involvement is seen in the tibia, followed respectively by the ulna, radius, humerus, femur and fibula (Ibid. 1995:2477, Ortner 2003:245).



(vi) The skull

*Fig. 2.13 Bone destruction of the skull (circled) possibly caused by TB. This is a cast of the original skull.* (Courtesy of C.A. Roberts).

The skull is rarely involved in skeletal tuberculosis except in young children, who account for up to 50% of cases that occur. However, the lack of cancellous bone in infant skulls accounts for the limited number of cases in infants aged under one

year old (Malhotra et al. 1993:1119). In children, it is unlikely to be the only focus of TB (Malhotra et al. 1993:1119, Rajeshwari and Sharma 1994:1214). The cranial vault is the most common site of cranial TB and this is characterised by numerous small, round areas of destruction up to 2cm in diameter (see Figure 2.13), which perforate the inner and outer tables (Ortner 2003:248). Tuberculosis meningitis occurs in up to 50% of cases of untreated military TB in children (Cruz and Starke 2007:107). These children are usually aged up to four years old (Walls and Shingadia 2004:13). Infection reaches both the cranial vault and meninges through the haematogenous/ lymphatic routes (Malhotra et al. 1993:1119, Rajeshwari and Sharma 1994:1214).

#### 2.2.5 Summary

As has been discussed, if TB is not treated, it can spread via haematogenous and lymphatic routes to other parts of the body, including the skeleton. Identification of TB in the skeleton therefore primarily relies on the recognition of characteristic alterations to the bones of the spine, hip and knee joints, although there is evidence to show that rib lesions could also be useful in diagnosis of the disease in skeletal remains. As has already been mentioned, TB can, less commonly, affect any bone or joint in the body. However, diseases other than TB could leave similar signatures and therefore care must be taken to look for as many skeletal indicators as possible in order to draw a confident conclusion about the nature of the infection. In conclusion, it should now be apparent that there are limitations to the diagnosis of disease from the examination of skeletal remains. It is therefore important to consider the osteological paradox when making diagnoses of past disease from skeletons. To this end Wood et al. (1992:343-4) state that a 'common sense approach' to estimating the prevalence of a disease in the past would suggest that there is an association between the frequency of skeletal lesions typical of that disease and the number of people in that population suffering this disease. Although it must be noted that the exact number of people affected in a given population may never be established. Wood et al. also suggest that the interpretation of these sorts of data is more difficult than it appears due to

failure to address three important conceptual issues, namely demographic nonstationarity, selective mortality and hidden heterogeneity in risks.

Demographic non-stationarity refers to the departure of a population from the stationary state. This is due to migration, age-specific fertility, mortality and population growth (Wood et al. 1992:344). Selective mortality brings to notice the sample of skeletons that is observed are the dead part of the population: we can never be sure of the number of people in that population at risk from a disease such as TB or who suffered from the disease, but who did not die from it. Hence, the presence or absence of skeletal lesions demonstrates a data bias because it is not known how the percentage affected relates to the number of people in the whole population who suffered TB (Ibid. 1992:344). An additional factor to be considered is whether the frequency of people affected, if it could possibly be estimated, reflects the frequency of disease in the once living population. Finally, the problem of hidden heterogeneity means that the variation in susceptibility to diseases such as TB and to death from them is an unknown variable (Ibid. 1992:345).

Taking these points into consideration, it is impossible to obtain direct estimates of the demographic profile, or the prevalence, of a disease in a given population from archaeological skeletal remains. However, it is argued that the introduction of new biochemical techniques such as the previously mentioned aDNA analysis could be used to address some of these challenges. For instance, as will be discussed later in the chapter, the identification of aDNA of *Mycobacterium tuberculosis* complex in skeletons not exhibiting any bone changes indicative of TB have led scientists to conclude that the prevalence of the disease, which was until recently based upon osteological analysis alone, has been previously underestimated.

# 2.3 Bioarchaeological and historical studies of TB

## 2.3.1 The emergence of TB as a human pathogen

Tuberculosis has existed alongside humans and caused infection for thousands of years. Until relatively recently, prior to the advance of biochemical techniques, literary and artistic depictions were the only sources available to inform us of this ancient association, alongside visual or macroscopic examination of human skeletal remains. However, as only 3 to 5% of untreated people infected with TB go on to develop these bone changes (Peto et al. 2009:1350, Holloway et al. 2013:2), there were obvious limitations and gaps in knowledge.

It is now possible to identify and isolate Mycobacterial ancient DNA (aDNA) from human remains, and the development of genetic techniques, such as the use of PCR amplification and DNA sequencing (Galagan 2014:307), are helping us to clarify our understanding of this ancient pathogen, and are suggesting that the disease was more common than we may have previously thought from osteological evidence alone. It appears *Mycobacterium tuberculosis* complex (MTBC) aDNA can sometimes be detected in skeletons showing no bone changes indicative of the disease (Müller et al. 2014a:178). Furthermore, genetic studies have lead to the identification of variable genomic regions that are present in some, but not all, MTBC strains. The distribution of these regions has enabled scientists to construct a phylogenetic tree of the organisms making up the MTBC species and this has led to an overturning of long-held beliefs about the origin of tuberculosis as a human infection (Brosch et al. 2002:3684, Galagan 2014:308). The evidence for TB in the past is now discussed in more detail.

The earliest documented evidence detailing the signs of TB comes from India and Egypt, approximately 4,000 years old (Keers 1981:91), and from medical texts from China dated to approximately 3,000 BC (Brothwell and Sandison 1967:48). It is also mentioned in the Old Testament of the Bible, approximately 1,300 to 400 BC (Daniel and Daniel 1999:1557) and in the writings of Hippocrates,

approximately 460 BC (Hippocrates and Chadwick 1950, Roberts and Buikstra 2003:8). These texts refer to a disease often called schachepheth (Hebrew), sosha (Sanskrit), yaksmā or xoy (Hindi) or phthisis (Greek). Phthisis translates as "wasting away", which describes one of the signs of TB. These signs also gave rise to the common nineteenth century name used in Britain: "consumption" (Galagan 2014:308), although it must be noted that several diseases other than TB cause loss of appetite and thus "wasting away", so these descriptions are not necessarily referring to TB in every instance.

Artistic evidence of the disease is interpreted from illustrations and sculptures of people with kyphotic spines, and also of pale, thin people (Morse et al. 1964:526). However, these representations must be interpreted with care as they could be depicting medical conditions other than TB; for example, anaemia causes paleness. Additionally, Mitchell (2011:82) brings attention to the fact that illustrations in medical texts were often made by non-medically trained artists who may not have seen people with the disease they were asked to illustrate, which leads to further problems in interpreting artistic depictions. A further complication of using past documentary and artistic evidence to provide an indication of disease in the past is that genetic changes in the microbial agents of those diseases may have led to different expressions of the infection (Mitchell 2011:84). Signs and symptoms of TB in the past may not have been as they are understood today. Additionally, patients described and depicted in ancient texts and pictures, respectively, could have been affected by more than one disease, the combination of which could produce signs and symptoms similar to TB today. Another confounding issue is that while microorganisms can mutate to become more pathogenic or resistant to antibiotics in use today, they could also mutate to be less or even non-pathogenic and thus disappear from medical history (Mitchell 2011:84). Some of these "extinct diseases" could have caused signs and symptoms similar to those related to TB and thus lead to inaccurate interpretations of old texts and pictures.

In terms of palaeopathological evidence of TB in the Old World, the majority is in Europe with few countries, for example Belgium and Iceland, having very little evidence and other countries, for example the UK and Hungary, having much more (Roberts 2015:S118). All of the Old World evidence for TB is found in the Northern Hemisphere (Ibid. 2015:S118). The earliest skeletal evidence of TB is from Israel and is dated to 7250 – 6160 BC (Hershkoviz et al. 2008:3), although controversial (Wilbur et al 2009). This is followed by early Egyptian evidence (4500BC) (Morse et al. 1964:526), and evidence from German (5400-4800 BC) (Nicklisch et al. 2012:391), Italian (approximately 5,500 BC) (Canci et al. 1996:487), and Hungarian skeletons (approximately 5,000 BC) (Köhler et al. 2012:697, Masson et al. 2015:13). However, most evidence for TB in Europe comes from the Roman and later periods, especially the early and later Medieval eras, dating to around 5<sup>th</sup> to 15<sup>th</sup> centuries AD (Roberts 2015:119). The lack of evidence in areas such as most of Africa could be due to lack of survival of skeletal remains because of the manner of funerary practices (eg. cremation), poor preservation of skeletal remains (for instance, due to acidic soils), and a lack of excavation of cemeteries and bioarchaeological training and research, rather than to lack of the presence of the disease itself (Roberts 2012:439-440). Perhaps it could be the case that there is not any evidence of the disease, or it could be found in the future as the scope of archaeological work and paleopathological training extends.

In addition to this osteological evidence, aDNA analysis has successfully taken place on skeletons and mummies with and without evidence of TB in their tissues from Britain, Egypt, Hungary and Lithuania and to a lesser extent from the Czech Republic, France, Germany, Hungary, Spain and Sweden in Europe, and Israel, Japan and Siberia outside of Europe (Roberts 2015:119).
## (i) Tuberculosis in the New World

For many years, it was thought that TB had not existed in the Americas before Columbus brought it to the New World in 1492 (Hrdička 1909, Morse 1967:250). However, this has been proven to be an incorrect assumption. Within North America, for example, there are two main clusters of pre-Columbian TB evidence (Roberts and Buikstra 2003:190). These were both areas of major population density during late prehistory and are located in the midcontinent and in the Southern USA (Roberts and Buikstra. 2003:190). In Mesoamerica, there were even larger centres of population concentration than those found in North America prior to Columbian contact (Ibid. 2003:191). However, there is very little evidence of TB in this region, which may be partly explained by the poor preservation conditions of many Central American regions (Ibid. 2003:191). However, large skeletal samples have been found from post-Columbian contexts, such as in the Valley of Mexico at Teotihacán, but no evidence of TB was reported (Sempkowski and Spence 1994). This could be due to lack of palaeopathological training on the part of bioarchaeologists who have worked on these remains, and therefore further re-examination of the skeletons could reveal some evidence for the disease. However, it could be that there indeed is not any evidence to be discovered.

The earliest convincing cases of TB in the New World have been found in South America. Evidence comes from Peru (Buikstra and Williams 1991, Bos et al. 2014), Venezuela (Requena 1945), Chile (Allison et al. 1981) and Columbia (Arregoces 1989). It is suggested that New World TB developed in South America approximately 1,500 years ago and the spread to North America by AD1000 (Roberts and Buikstra 2003:193). However, this does not account for the lack of TB in Mesoamerica if migration and trade followed overland routes. However, sea routes could have been mainly used for trade and this may explain why the spread of TB appears to have "missed out" Mesoamerica.

A recent aDNA study suggested that as the majority of *Mycobacterium tuberculosis* genetic strain diversity exists in Africa, the pathogen probably spread worldwide via human movements out of Africa during the Pleistocene period (Bos et al. 2014:494). However, this research proved another transmission route appears to have happened for people who lived in the New World. The researchers screened 68 skeletal samples from pre- and post-Columbian contact sites in the New World (Ibid. 2014:494). All of these skeletons had some form of bone changes indicative of TB infection, but only three of the samples showed convincing evidence of aDNA preservation (Ibid. 2014:495). Coincidentally, genetic analysis for these samples made an interesting discovery; the DNA from these Peruvian remains was not closely related to human Mycobacterial strains, but were very closely related to *M. pinnipedii* strains which have only been isolated from seal and sea lion species found in the Southern Hemisphere (Ibid. 2014:495). Bos et al. suggested that seals probably contracted TB from a host species in Africa and migrated across the oceans to South America where human exploitation of the marine mammals allowed a zoonotic transfer some time within the first millennium AD (Ibid. 2014:495). It was suggested that TB reached North America around AD 900 via a similar zoonotic route (Ibid. 2014:495), which would also explain the lack of evidence of TB in Mesoamerica.

#### 2.3.2 Biomolecular studies of TB

It is now important to examine some of the research that has taken place on isolating *Mycobacterium tuberculosis* genetic material in the form of DNA, from archaeological skeletons of people who suffered from TB in their lifetimes. This is necessary as the current isotope research required bone and tooth enamel samples from skeletons positively identified as having had TB. This identification has been done by traditional osteology and also by the examination of the bones for the presence of the bacterial DNA. Although the osteology and DNA research was not undertaken by the current author, and the results were somewhat disappointing, some familiarity with the methods is required.

As previously discussed, TB usually initially infects the lungs and can spread to the bones via the bloodstream and lymphatic systems. *Mycobacterium* 

tuberculosis complex bacteria are present in the bone marrow of patients suffering this infection (Brown and Brown 2011:245). When the infected person dies, genetic information in the form of DNA from infecting *M. tuberculosis* bacterial cells may remain in affected bones, such as the vertebrae in Pott's disease, or in the visceral surface new bone of the ribs (Brown and Brown 2011:246, Galagan 2014:308, Donoghue et al. 2004:585), although it must be noted that it is not certain where the bacterial DNA is actually located within the rib bones; it is not necessarily confined to the areas of new bone formation (Roberts 2016 pers. comm.). However, not all pathogenic bacteria leave traces of their aDNA in this manner: the successes with isolating *M. tuberculosis* complex DNA is largely due to the infecting bacteria sometimes finding their way into the bones, which tend to be better preserved in archaeological contexts than ancient soft tissues (Brown and Brown 2011: 245). Hence, reported survival of pathogen aDNA within the skeleton is restricted to a small number of microorganisms of which Yersina pestis. the causative organism of plague (Bos et al. 2011) is one example, and Mycobacterium leprae, the bacterium that causes leprosy (Rafi et al. 1994), is another.

A recently developed clinical technique used in DNA analysis is also showing much potential in archaeological studies. Next generation sequencing (NGS) is a newer DNA sequencing technology which has revolutionised genomic research. It is faster than the older Sanger sequencing technology that took over a decade to sequence the human genome. In contrast, NGS can sequence the entire human genome within a day (Behajati and Tarpey 2013:236). There are a number of different NGS sequencing technologies, but all of them involve sequencing millions of small fragments of DNA in parallel. Bioinformatics analyses are then used to piece together these fragments by mapping the individual readings and comparing them with a reference genome (Ibid. 2013:236).

In clinical practice, NGS is used to capture a broader spectrum of mutations than can be achieved with Sanger sequencing (Behajati and Tarpey 2013:237). In microbiology, the main use of NGS is to replace conventional characterisation of

pathogens through looking at morphology, their staining properties, and the metabolic criteria with a genomic definition that can provide information about drug sensitivity and can also inform the evolutionary relationship of different pathogens with each other. The information provided by NGS can also be used to trace the source of infection outbreaks (Ibid. 2013:237). Limitations of NGS include requiring trained staff and the use of high specification computers. In clinical settings this is leading to developing the required infrastructure required to analyse and interpret the data produced (such as computer capacity and personnel training) which add to initial expenses. However, the actual cost of NGS is far lower than that of traditional Sanger sequencing (Ibid. 2013:238).

In terms of use of NGS in TB research, the technique is used successfully to more effectively detect antibiotic resistance in clinical samples (Merker et al. 2013, Feuerriegel et al. 2015, Mokrousov et al. 2016). Mokrousov et al. (2016:1127) suggest that NGS technology is becoming more affordable and so it is increasingly used for high-resolution molecular epidemiology of tuberculosis. For example, in this study the researchers used spoligotyping (a classical method of M. tuberculosis genotyping) and they were particularly interested in spoligotype international type (SIT) 266. This genotype constitutes a large proportion of M. tuberculosis found in patients in Belarus in Eastern Europe. It has also been described sporadically in neighbouring provinces in northwest and central Russia and Latvia (Ibid. 2016:1128). Of particular concern is the fact the genotype is multidrug resistant (MDR) and very likely to be extensively drug resistant (XDR). A recent study found SIT266 in 25 out of 163 strains tested. All 25 of these were MDR (Zalutskaya et al. 2013). This contrasts with the apparently parental type, SIT264, which differs from SIT266 in a single spacer 8 in its DNA. SIT264 is more widespread across Eastern Europe but it is in low prevalence and not associated with MDR. Both SIT266 and SIT264 isolates have been assigned to the Latin American-Mediterranean lineage of the *M. tuberculosis* complex on the basis of genetic similarities (Mokrousov et al. 2014).

While this NGS technique is of great value in clinical microbiology, in order to update *M. tuberculosis* lineages and to understand the global spread of MDR genotypes, the technique could also be used to extend the current project to provide even more detailed strain data than that attempted by Müller et al. (2014b). This information could then be used to assess to what extent mobility played a part in the transmission of TB in the Roman period. aDNA analysis can therefore also be used to study pathogen evolution. This is of particular interest because the evolutionary pathway of disease-causing microorganisms could possibly help microbiologists today to predict the route of future evolution (Brown and Brown 2011:246, Galagan 2014:307) and antibiotic resistance (Merker et al. 2013:9, Feuerriegel et al. 2015:1908, Mokrousov et al. 2016:1833). Furthermore, the resulting data from this type of study could be used for exploring possible routes of transmission of TB, and hence new prevention methods and treatments for antibiotic resistant strains already in existence, and to predict a pattern for those new strains likely to emerge in the future.

Fortunately, for the purposes of the current research, TB is the disease most extensively studied by biomolecular archaeologists (Brown and Brown 2011:250). This is in part due to the re-emergence, persistence and widespread nature of the disease today, leading people looking for patterns in the past to perhaps inform us of why this disease is so successful at evading eradication. The interest in TB is also due to the disease causing typical identifiable bone changes, albeit in a small percentage of skeletons, thus pinpointing individuals to whom biomolecular analysis, such as aDNA analysis, can be applied (Brown and Brown 2011:250, Donoghue et al. 2004:584). In addition, studies suggest that TB DNA survives well for long periods of time in some archaeological contexts so it lends itself to genetic study (Spigelman and Lemma 1993:137-138, Fletcher et al. 2003:143).

However, in view of the low frequency of bone changes associated with TB actually occurring in a skeletal population, there have been debates about the accuracy of estimates of the prevalence of the disease in past populations that are represented in the bioarchaeological record (Brown and Brown 2011:251). aDNA

analysis is therefore providing a potential means of confirming osteological diagnosis and identifying TB in individuals not displaying the bone changes typical of the infection, for example Pott's disease, although it cannot prove a direct association between a positive aDNA result and the bone changes observed (Müller et al. 2014a). If aDNA survives in all archaeological skeletons, and all archaeological skeletons are then analysed for the presence of TB aDNA, this could lead to more accurate estimates of the prevalence of TB infection in past populations (Brown and Brown 2011: 251, Galagan 2014:308, Formicola et al. 1987:1, Fusegawa et al. 2003:146, Hershkovitz et al. 2008:1, Nicklisch et al. 2012:391, Sager et al. 1972:176). However, this would make the assumption that aDNA of TB survives equally well in every skeleton, regardless of date of burial, age of the person at death and burial environment conditions. This would appear to be an unlikely scenario. The expense of performing aDNA analysis, and the time required, is also likely to rule out the analysis of every skeleton on whom a palaeopathological and osteological report has to be made.

Biomolecular and aDNA analysis of TB are also relevant to this project. Although in-depth description and discussion of biomolecular methods of detection of ancient tuberculosis are largely outside the remit of this study, it is necessary to briefly mention the use of mycolic acids and proteomics. It should be recognised that in the 1980s, a decade prior to the use of DNA analysis in identifying biomolecules related to TB, mycolic acids were discovered as a tool to identify different species of Mycobacteria (Minnikin et al. 1984). Mycolic acids are high molecular weight 3-hydroxy-2-alkyl-branched fatty acids found in all strains of Mycobacteria within their cell walls (Minnikin et al. 1984:225). Mycobacterial mycolic acids have between 60 and 90 carbon atoms and the different structural types of mycolic acid include  $\alpha$  -, keto-and methoxy-mycolates. These can be separated and identified by thin-layer chromatography, and several types of mycolic acid patterns have been found to be particularly prevalent (Minnikin et al. 1984:225). The mycolates of *Mycobacterium tuberculosis* consist of  $\alpha$  -mycolates, ketomycolates and methoxymycolates are one example of a pattern, with other species of mycobacteria showing differing patterns (Ibid. 1984:226). Mycolic acid

patterns appear to be stable under standardised growth conditions (Ibid. 1984:228). The patterning of mycolic acids was the only biomolecular method available to identify *Mycobacterium tuberculosis* prior to the introduction of aDNA analysis, and considerably more work has been done on the use of this technique since 1984, including a comparison of the usefulness of mycolic acid and aDNA analysis in confirming the presence of TB in archaeological skeletons, which is now briefly discussed.

In 2001, Gernaey et al. tested the capacity of two biomarkers to confirm osteological diagnoses of TB. These were Mycobacterium tuberculosis complex mycolic acids, and DNA targeting the mobile genetic element, IS6110, which is discussed in more detail in the following section, (Gernaey et al. 2001:259). In order to do this, the researchers examined three archaeological skeletons that were approximately 1000 years old. One of these individuals had Pott's disease, one had rib lesions, and one had no bone changes indicative of infection with TB (Ibid. 2001:260). Rib samples from all three of these individuals were examined for the presence of *M. tuberculosis* mycolic acids and Mycobacterial aDNA. Only the individual with Pott's disease tested positive for the presence of *M. tuberculosis* aDNA. Two ribs from this individual also tested positive for mycolic acids. The individual without any lesions indicative of TB tested negative for both M. tuberculosis aDNA and mycolic acids. Finally, the individual with rib lesions tested negative for *M. tuberculosis* aDNA, and had traces of mycolic acids but these were insufficient in quantity to be confirmed as being of *M. tuberculosis* complex origin (Ibid. 2001:261-262). The conclusions drawn by the researchers were that M. tuberculosis complex mycolic acids survive undegraded in the archaeological record for periods of at least 1000 years, and that their use in diagnosing TB in archaeological skeletons complements aDNA and osteological analysis. However, further discussion of these techniques is beyond the scope of this study, but Mycobacterial related protein analysis is now discussed.

In 2011, Boros-Major et al. reported on their research into ancient Mycobacterial proteins to detect the presence of TB in archaeological skeletons. Their methods

utilised the techniques of direct sequencing and peptide mass sequencing of several proteins. However, the study did not make clear whether modern proteins were used as comparisons for the ancient proteins, or whether these proteins were specific to pathogenic bacteria in the MTBC or to the genus *Mycobacteria* in general. Other problems with the use of proteomics appeared to be linked to the possibilities of proteins becoming denatured and thus changing in structure and amino acid composition. This could occur as a result of exposure to the burial environment, for example, the presence of possibly protein-hydrolysing acidic soils, or the action of decomposing microbes within the soil, whose enzymes may digest the proteins into shorter polypeptides and thus change their primary structures. In addition, it has been acknowledged by other researchers examining the aDNA of TB that mutations have taken place. These have led to the development of new strains of TB (Müller et al. 2014b) and also to the emergence of antibiotic resistance (Galgan 2014, Guler and Brombacher 2015, Horsburgh et al. 2015). The nature of DNA mutations means that the amino acid sequence of the resulting protein will be changed. Sometimes, the mutation can have farreaching consequences and will lead to a non-functional form of the protein being synthesised. Boros-Major et al. (2011) did not discuss how these issues could affect their research. However, their method would only be useful in the detection of ancient TB if it was used in conjunction with mycolic acid and aDNA analysis. The researchers did allude to this requirement without discussing the shortcomings of their own methods (Ibid. 2011:197-198).

Some very recent work on identification of TB using proteomics has attempted to replicate the work of Boros-Major et al. (2011) - Hendy et al. (2016). However, the new research confirmed some of the issues the current author has highlighted above, namely that the three proteins Boros et al identified were not specific to *Mycobacterium tuberculosis,* although one was similar to proteins found in other bacteria of the MTBC (Hendy et al. 2016:148). These proteins were however also very similar to human collagen, which will obviously be present in most archaeological samples, as it is frequently preserved. Boros-Major et al. had failed to notice or comment upon this major issue. Another problem was that there has

not been sufficient research done on the effect of preservation on proteomic profiles (Ibid. 2016:148), which also confirms what is stated above. Further issues to confound the proteomics method include the discovery of many other bacterial proteins in all seven of the samples tested. These were thought to be environmental contaminants, probably introduced from the burial soil. This lead to concerns about the unknown effect of these upon other proteins in the samples (Ibid. 2016:149). Hendy et al. recommend that, before any more research is published using proteomics, detection methods should be made more sensitive and that all archaeological journals should insist on accepted minimum standards for reporting protein mass spectrometry data (Ibid. 2016:152).

As previously mentioned, the current research leads on from a joint Durham and Manchester Universities NERC funded project which isolated MTBC aDNA from the remains of British and other European skeletons (Müller et al. 2014a, Müller et al. 2014b, Müller et al. 2014c, Müller et al. 2016). The skeletal samples used for the NERC project were shared with the current research project. In the following sections, a brief summary of the history of study of MTBC aDNA in terms of detection, and its use in diagnosis and phylogenetic reconstructions are discussed. It is important to consider these here because it is a method that can be used to identify TB in skeletal remains and this project has its basis in the Durham/Manchester projects that have aDNA analysis.

The first studies of TB aDNA were carried out in the mid 1990s and were aimed at identification of *M. tuberculosis* in archaeological human remains (Spigelman and Lemma 1993, Salo et al. 1994) and the complete genome of *Mycobacterium tuberculosis* was sequenced in 1998 by Cole et al. The early aDNA research projects focused on the identification of two insertion sequences present on the *M. tuberculosis* genome. These sequences are mobile, can move about within the bacterial genome, and can also transfer between bacteria, taking genes such as those for antibiotic resistance with them (Brown and Brown 2011:251, Cole et al. 1998:537). The insertion sequences may have up to 20 copies in each bacterial cell, thus aiding the chances of detection (Brown and Brown 2011:252, Cole et al.

1998:537). A point of consideration of this technique is that, when attempts to identify the presence of TB aDNA are made, it is essential that polymerase chain reactions (PCRs) and primers specific for *M. tuberculosis* are used or, alternatively, it must be known which other species of Mycobacteria may also give a positive result. *M. tuberculosis* is only one species of the genus *Mycobacteria*, all members of which have very similar genomes (Müller et al. 2014c). Some of these bacteria also cause human TB (eg. *M. bovis*), but some are soil-dwelling contaminants (Konomi et al. 2002:4740, Müller et al. 2016). These discoveries have called into question the accuracy of the use of aDNA analysis in identification of ancient tuberculosis.

A distinction between the DNA and therefore the organisms, *M. tuberculosis* and *M. bovis*, became possible with the identification of a single nucleotide polymorphism (SNP) in the *oxy*R pseudogene, where the presence of nucleotide base A indicates *M. bovis*, and the presence of G indicates one of the other members of the *Mycobacterium tuberculosis* complex. Shortly after this discovery, two SNPs were discovered in the *kat*G and *gyr*A genes, which, in combination, achieved the same result. In recent years, other diagnostic SNPs have been discovered, thus making distinctions between the two species much easier (Biet et al. 2012:264, Cagneux 2013:1, Brown and Brown 2011: 252). These early studies of the use of aDNA are now considered in more detail in the following section.

#### (i) The history of studies of the aDNA of TB

Spigelman and Lemma (1993) considered that collagen can be preserved in bones for thousands of years despite the actions of bacterial decay, and so they hypothesised that other organic substances may also be preserved and detectable. Thus, if a bone was infected with a micro-organism during life, it could be possible to detect that organism using DNA analysis many years after death and burial (Spigelman and Lemma 1993:137). However, it is necessary for the pathogen to have been present in the bone or tooth being analysed (and be preserved for analysis) at the time of death of the individual, and for the bacteria to be present in large enough numbers to contain detectable amounts of aDNA (Brown and Brown 2011:245). The complete genome of modern *M. tuberculosis* was sequenced and published in 1998 (Cole et al. 1998), and has thus been used for comparison with aDNA studies. PCR had been developed in 1987 (Mullis and Faloona 1987) and the importance of PCR lies in the ability to amplify, in just a few hours, small traces of DNA, such as may be found in archaeological bones. Spigelman and Lemma proposed that PCR might contribute to our knowledge of disease in the past by allowing pathological bones with suspected disease to have a diagnosis confirmed or refuted (Spigelman and Lemma1993:138).

Bacteriologists had already made primers for Mycobacterium tuberculosis (i.e. short stretches of single-stranded DNA complementary to known sequences of DNA in the target organism). Spigelman and Lemma described these primers, possibly optimistically, as being so specific that they would not give false positive results for other Mycobacterial species (Spigelman and Lemma 1993:138). On testing their hypothesis on some archaeological bones sourced from several sites around the world, namely Cyprus, Egypt, England, Scotland and Turkey. These skeletons were from different time periods ranging from Romano-British (approximately AD 43 to AD 410) up to and including the 18<sup>th</sup> century AD. It was found that aDNA of TB was found in four skeletons, two of which had skeletal signs of the disease in the form of Potts disease, and two that had no skeletal indicators of TB (Ibid. 1993:140 and 142). The researchers concluded that more knowledge of the degradation of DNA over time was needed, and the possibility of contamination with foreign DNA after death or during excavation needed to be considered. In order to mitigate against the introduction of possible contaminant DNA using their methods, they suggested their further work would be on diseases no longer found in Britain, such as the plague (Ibid. 1993:142). However, they remained very optimistic that huge advances in the diagnosis and confirmation of the presence of ancient disease had been made through their research, although further research called this into question due to the lack of detail given about procedures used, and the lack of DNA sequencing used to confirm the aDNA that

was discovered as being that from *Mycobacterium tuberculosis* (Spigelman et al. 2002:393).

In some clinical DNA research in 1990, Thierry et al. (1990:188) had reported finding a "mobile genetic element" (IS6110), which they reported as having been found in *M. tuberculosis* and *M. bovis* but not in any other MTBC complex species. They also suggested IS6110 could be used as a probe for the identification of organisms from the *M. tuberculosis* complex (Thierry et al. 1990:188). Salo et al. (1994) made use of this clinical research by applying it to archaeology. Salo et al. (1994:2091) reported the recovery of DNA "unique to" *M. tuberculosis* from a lung lesion of a spontaneously mummified adult body in southern Peru. No evidence of skeletal change could be identified in the spine, ribs or anywhere else in this individual (Ibid. 1994:2091). The lung lesion was suggested to provide good evidence for the pre- Columbian presence of human TB in the New World. The mummy analysed was a 40 to 45 year old woman found entombed in a burial site used by the Chiribaya. These people were a largely agricultural population that occupied the lower Osmore Valley near the southern Peruvian coastal community of the IIo about A.D. 1000-1300. <sup>14</sup>C dating of liver tissue from the mummified woman yielded a date of  $1040 \pm 44$  years B.P. (Salo et al. 1994:2091).

Although the lungs of the deceased individual had collapsed, the right upper lobe of the right lung was found adhering to the chest wall and contained a small, calcified nodule. DNA was extracted from samples of the lung lesions and subjected to PCR, which was targeted at a segment of DNA unique to *M. tuberculosis*, i.e. the insertion sequence mentioned previously (IS6110). Although the first round of analysis was negative for TB a DNA, the lung nodule and a lymph node were re-examined using nested PCR. The authors justify this method by stating that nested PCR 'can greatly reduce background and increase sensitivity' (Ibid.1994: 2092). This time, the results from one of the mummy's lesions indicated the presence of *M. tuberculosis* (Ibid.1994: 2093). The partial sequence of the 97 bp (base pair) nested PCR target of IS6110 was stated to be identical to modern *M. tuberculosis* (Ibid.1994: 2093). However, these methods only tested the soft

tissues of this individual; at no point did the researchers extend the study to include repeating this PCR analysis on the bones of the mummy, and thus did not test for the survival and possible identification of *M. tuberculosis* in the skeletal tissues. It is possible that aDNA preservation in the skeletal tissues may have been different to that of mummified soft tissues.

Baron et al. (1996) acknowledged the work of Salo et al. (1994), but were more interested in trying to follow Spigelman and Lemma's (1993) lead in amplifying DNA from ancient bone. They also chose to experiment with aDNA of TB. As TB can affect the bone, the authors expected aDNA to possibly be present in bones of affected individuals (Baron et al. 1996:667). They were careful to choose primers specific to Mycobacterium tuberculosis and also to run negative controls with DNA extracted from soil-dwelling Mycobacterial species for comparison (Ibid. 1996:668). Skeletal remains for analysis were sourced from a historical pathological collection curated in the Institute of Anthropology, University of Göttingen, Germany. This collection had been developed at the end of the 19<sup>th</sup> century up to the beginning of World War II. Two femora and one skull were selected for analysis from individuals who were documented as having suffered from bone tuberculosis (Ibid. 1996:668). The results showed that the 123 bp Mycobacterium tuberculosis specific DNA sequence could be amplified from all three of the bones (Ibid. 1996:669). Baron et al. claimed that an important part of their study was to test if the PCR assay they used was specific for the M. tuberculosis complex, and could be applied to skeletal remains that had been excavated from an archaeological site. The soil-dwelling species of Mycobacteria used as controls did not give any specific amplification product, and thus it was concluded that it was possible to use PCR for the detection of TB aDNA in archaeological remains (Baron et al. 1996:670), a statement which has recently been called into question (Müller et al. 2014c, 2016). The only limitations of the method that the researchers acknowledged were that there could be a possibility of false-negative results and the impossibility of the method to distinguish between silent, dormant primary TB infection and acute TB disease (Baron et al. 1996:671).

Additionally, during 1996, and again in 1999, Taylor et al. used PCR to identify the presence of *M. tuberculosis* in mediaeval human skeletal remains, which were dated to between AD 1350 and 1538. They acknowledged that a positive result with PCR can confirm the presence of a bacterial DNA sequence in ancient human remains, but a negative result cannot rule out the possibility of a negative result occurring when the individual had a prior healed infection, or in whom there was poor DNA preservation. To resolve some of these issues, Taylor et al. used a slightly different, and supposedly more sensitive, nested PCR method (also used by Salo et al. 1994), to examine the Mediaeval bones with lesions suggestive of the presence of TB (Taylor et al. 1996:790). Taylor et al. also amplified IS6110 and part of the  $\beta$  subunit of RNA polymerase to identify MTBC complex DNA (Ibid. 1999:899).

The skeletal remains used for these analyses came from the large cemetery of the Abbey of St Mary Graces, overlying part of the 1348/9 Black Death cemetery on the site of the old Royal Mint in London. This burial ground had been associated with the last Cistercian foundation in England, which was founded in 1350 and lasted until the dissolution in 1538 (Ibid. 1996:791). The bones studied included a fused wrist and two adjacent lumbar vertebrae from the skeleton of a man aged at least 45 at the time of his death. Two further lumbar vertebrae were sampled from a man aged between 15 and 25 years old (Ibid. 1996:791). The nested PCR results showed the presence of TB in both of these skeletons. However, Taylor et al. highlighted that a negative result would not necessarily rule out an infection because the bacteria may have died before the death of the sufferer. They also mentioned their concern that bones without obvious lesions indicative of the disease may produce a positive result. It was suggested that individuals with this sort of result could have been exposed to the disease without developing obvious clinical symptoms or bone damage by the time of their deaths (Ibid. 1996:798).

However, it is argued that not all people with active TB develop bone lesions, perhaps because they die before the lesions have time to develop (as per Wood et al. 1992), and therefore a positive PCR result could be indicative of such an individual. Therefore, this is not considered to be a negative point, and could possibly help provide a more accurate picture of the number of people infected with TB in the past than using osteological analysis alone.

In the 1999 study, which used bones from the same individuals as their 1996 research, the authors stated that 'each of the samples had osteological evidence of TB' (Taylor et al. 1999:900). However, this evidence was not detailed in the 1999 paper, the bones having been described in depth in the previous paper (1996). None of these osteological changes actually provide the firm 'osteological evidence of TB' described (Taylor et al. 1999:899) as the bone changes were non-specific and could equally have been caused by any number of other infective agents. The results showed the presence of IS6110 via PCR, which was performed once on each of the three samples (Ibid. 1999:902), and an *M. bovis* PCR was performed twice on two of the extracts and was negative (Ibid. 1999:902).

By 2000, Haas et al. were also using aDNA analysis, this time to examine apparent different stages of TB in archaeological bone from Hungary (Haas et al. 2000). These skeletons dated from the 7<sup>th</sup> to 8<sup>th</sup> centuries and 17<sup>th</sup> century A.D. Three of the skeletons showed typical bone changes associated with TB whereas six had bone changes probably caused by TB. Five individuals had evidence of skeletal pathology but these were all atypical for TB, four having minor pitting of the vertebral bodies indicating a possible inflammatory response, and the final one having anklosing spondylitis (Haas et al. 2000:301). Baker (1999:301) suggested pits on the ventral aspects of thoracic and lumbar vertebral bodies, accompanied by rib lesions, may indicate TB infection. Once again, DNA was extracted from the bones and PCR was used to amplify the DNA. Fourteen samples were processed and, of these, eight were unambiguously positive for the presence of *Mycobacterium tuberculosis* complex. This was demonstrated by amplification of the IS6110 sequence (Ibid. 2000:293). However, the presence of another PCR product suggested that other Mycobacterial species from the soil had caused

contamination (Ibid. 2000:293), which casts doubt on the accuracy of previous studies where "foreign DNA" contamination from the soil was not accounted for.

A study of mycobacterial DNA in Andean mummies from South America focused on samples of preserved dried soft tissue from 'histologically confirmed skin samples in the pelvic region', (that is, confirmed TB positive), of 12 mummies curated in the American Museum of Natural History Collection in New York (Konomi et al. 2002:4738). Radiocarbon dating places the date for the mummies to between A.D. 140 and 1200 (Ibid. 2002:4738). The authors do not discuss the nature of the histological confirmation of TB in the skin samples, so it is assumed microscopy was used to identify the presence of Mycobacterial species. The results showed that *M. tuberculosis* was detected in two of 12 of the samples and Mycobacteria other than *M. tuberculosis* (MOTB) in seven samples. However, this latter group of Mycobacteria is present in soil and water and probably does not equate to a clinical infection (Konomi et al. 2002:4740). *M. tuberculosis* was confirmed in the two samples using PCR with primers specific for insertion sequence IS6110 and unique to the *M. tuberculosis* complex (Ibid. 2002:4739).

Taylor et al. (2007) used aDNA analysis to report what is the first discovery of *M. bovis* causing Pott's disease in archaeological human remains. In their research, they examined five Iron Age individuals with spinal lesions buried in the cemetery of Aymyrlyg in Southern Siberia radiocarbon dated to between 1761 and 2199 years B.P (Taylor et al. 2007:1243). The skeletons were chosen because three of them showed 'infective lesions that are considered to be indicative of tuberculosis' (Ibid. 2007:245). These were lytic lesions of eight or nine vertebrae (in two individuals), in addition to lytic lesions in the knee joint (in one individual) and new bone formation on one or more of the lumbar vertebrae in two individuals (Ibid. 2007:1244). Mycobacterial DNA was successfully amplified and genotyped from four out of five of the individuals sampled, and was identified as *M. bovis* (Ibid. 2007:1245-1246). However this first evidence for *M. bovis* may not be too surprising since Brosch et al.'s research in 2002 into the evolution of the MTBC suggests that *Mycobacterium tuberculosis* strains are direct descendants of

tubercle bacilli that existed before the *M. africanum* – *M. bovis* lineage separated from the *M. tuberculosis* lineage (Brosch et al. 2002:3684). However, whether or not the progenitor of currently existing MTBC complex was already a pathogen when the *M. africanum* – *M. bovis* lineage separation occurred is unknown, but they believed this to be the case (Ibid. 2002:3688). They also believed most cases of tuberculosis in the archaeological record had been caused by *M. tuberculosis* and not *M. bovis*, which could account for the paucity of examples of *M. bovis* proven to be causing infection in archaeological human remains. This therefore supports this first evidence of *M. bovis* causing a human infection.

Recent advances in biomolecular archaeological techniques have allowed the mapping of genotypes of a historic strain of *Mycobacterium tuberculosis* (Fletcher et al. 2003, Bouwman et al. 2012). For example, the genotype of TB was determined by analysing the rib of a 19<sup>th</sup> century adolescent female buried in St George's Crypt in Leeds, West Yorkshire, England. The rib displayed surface new bone formation that may indicate pulmonary TB. The study concluded that the person suffered from a strain of *M. tuberculosis* that is uncommon worldwide today but was known to be present in North America in the early 20<sup>th</sup> century (Ibid. 2012). However, the scope of the study did not include establishing, by stable isotope analysis, if the infected individual had originated from Britain or if they may have migrated from (or visited) North America, although this research is currently ongoing.

Previous to this work, aDNA analysis of the TB bacteria in paleopathology had been restricted mainly to confirmation of TB in individuals with identified skeletal lesions. However, Bouwman et al.'s work increased the potential of aDNA analysis. Mapping the genotype of TB had, until recently, required the performing of different PCRs (polymerase chain reactions) for each gene locus being studied (Spigelman and Lemma 1993). The advent of NGS has meant that it is now possible to generate genome-wide sequence data from small amounts of aDNA starting material. The technique has also been used by Schuenemann (2013) to compare genomes of medieval and modern *Mycobacterium leprae*. This genome

knowledge can inform us how pathogenic species and strains have altered over time, in order to see if different strains are currently causing infection compared to those causing infections in the past. The genome sequencing can also help us to understand how pathogenicity of a disease may have increased or even decreased since archaeological/historical times. However, gaining a complete genome sequence for *Mycobacterium tuberculosis* is complicated by the presence of other Mycobacteria that may be in the soil of the burial (Wilbur et al. 2009:1990). Nevertheless, this problem can be avoided by using a more directed NGS approach, using hybridisation capture directed at specific polymorphic regions of the *M. tuberculosis* genome (Bouwman et al. 2012:2).

Müller et al. (2014a) analysed some of the same skeletal remains utilised in the current study for the presence of aDNA of TB. There has been concern about the high success rates of earlier researchers in identifying the presence of aDNA of TB in archaeological skeletons (Müller et al. 2014a:186). The study aimed to apply a rigorous analytical regime to the detection of MTBC DNA in 77 bone and tooth samples from 70 individuals from Britain and Continental Europe, dating from the 1<sup>st</sup> to the 19<sup>th</sup> century A.D. (Ibid: 2014a:178). Most (27) of the samples analysed were taken from ribs with new bone formation, or non-pathological long bones. Only a few sampled skeletal elements were of vertebrae showing signs of TB infection. Nineteen samples were taken from skeletal elements without signs of any pathology, and these included teeth from six individuals, and fourteen samples were taken from skeletons without any visible bone changes suggestive of TB infection (Ibid. 2014a:182). The results of the study illustrated that 12 samples had definite evidence of MTBC DNA. Twenty-two further samples were classified as probably or possibly containing MTBC DNA (Müller et al. 2014a:178). Interestingly, compared with other previous studies discussed, Müller et al. did not find "definite" MTBC DNA in any vertebrae analysed. Instead, eight positive identifications of MTBC DNA came from ribs with new bone formation on their visceral surfaces, one was from a tooth of an individual with rib lesions, one was from an individual with endocranial lesions, one from an individual with sacral and sacroiliac joint lesions, and the final individual had no lesions indicative of TB.

The rigorous techniques discussed and used in Müller et al.'s study included the use of dedicated aDNA laboratories at the University of Manchester and at the Complutense University of Madrid, Spain. Four key principles were followed to ensure accuracy and reliability of results. Firstly, the laboratories used were aDNA dedicated facilities with filtered air systems. This was deemed necessary to eliminate the risk of amplicon cross-contamination which could potentially lead to false positive results for MTBC DNA (Müller et al. 2014a:185). Secondly, positive PCRs were not scored just from the generation of amplicons of the expected size. Instead, all IS6110 amplicons were checked by sequencing of cloned products (Ibid. 2014a:184). The requirement for this rigorous approach was confirmed by the discovery of several amplicons of the correct size, which were found to not match the IS6110 reference sequence. Thirdly, replicates of all PCRs were carried out using fresh extracts with a set of samples from an independent laboratory (Ibid. 2014a:185). Fourthly, samples were not concluded to be "positive" or "negative" for the presence of MTBC aDNA. Instead, samples were classified as "probable" or "possible" based on the results of individual PCRs.

The conclusions reached, despite taking all of these precautions, were that the results are probably not entirely accurate. This was due to false positives resulting from contamination with MTBC aDNA from unknown and unaccounted for sources eg. environmental Mycobacteria. The paper also concluded that the vertebrae, whilst being an obvious target for aDNA analysis due to them frequently displaying typical bone changes of TB, are not a good skeletal element to use for this type of study. None of the 11 vertebrae analysed in the study were definitely positive for MTBC aDNA (Ibid. 2014a:184). It was suggested that vertebral body surfaces can be very porous and can be infiltrated by ground water from the burial environment. Indeed, Baker suggested TB may actually cause pitting of vertebrae (Baker 1999:301), which, if present, would allow more water ingress. This may result in aDNA degradation or the introduction of PCR inhibitors into the bone, and four of the five bone samples that were found to contain PCR inhibitors were vertebrae. However, as only 11 vertebrae were tested, this was probably too small a sample to make damning conclusions about the use of these skeletal elements in genetic

analysis. After all, of the 27 ribs with lesions tested, only eight of them yielded definite MTBC aDNA, which is also a low success rate, but no such negative conclusions were drawn by the authors about the use of ribs in future studies.

The Müller et al. (2014a) study suggests that the reported prevalence of TB during the past in Britain may be underestimated, and this is particularly true for Roman Britain. This was based on finding that MTBC aDNA was 'probably present' in three individuals of a possible 33 analysed from Roman Britain who did not display any TB related bone changes (Ibid. 2014a:184). A final point to consider was that the low number of individuals testing positive for the presence of MTBC aDNA was not reflected in previous work by other research teams (Ibid. 2014a:186), some of which are discussed above. This suggests that the rigorous approach utilised by Müller et al. may have eliminated contaminants, which could have caused false positive results.

Müller et al. extended their research by genotyping ancient Mycobacterium tuberculosis to find out more about different strains and genetic diversity in these bacteria in the past (Müller et al. 2014b). The researchers used aDNA sequencing to type 11 SNPs and two large sequence polymorphisms in the MTBC strains present in 10 archaeological samples from the skeletons already published (Müller et al. 2014a). The new research found evidence of a mixed strain infection in an individual from a Roman site at Ashchurch in south-west England. They ruled out that this result had been caused by any errors due to contamination, or being due to a miscoding of the DNA (Müller et al. 2014b:3). They also pointed out that mixed infection has been reported in patients today, either as a result of concurrent infections (multiple MTBC strains), or as reinfection with a different strain (Ibid. 2014b:3). Therefore, the Roman individual probably was unfortunate to contract a "double strain" infection due to one of these scenarios. Of further interest is that this information proved that two strains of TB (namely PCG 2 and PCG 3) co-existed in south-west Britain during the 2<sup>nd</sup> to 4<sup>th</sup> centuries AD. The use of isotope analysis to help assess the origin of the people from the Müller et al. projects, and to explore if they were migrants, was identified as the next logical

phase of research into understanding TB in the past and is, of course, the subject matter of the current project. It would be interesting, however, to extend the current project to include the Ashchurch individual in order to learn more about the origin and mobility of this person to establish the origins of these strains.

However, Müller et al.'s later work (2016) suggested some important limitations and considerations to be made with aDNA analysis of MTBC from archaeological contexts. This is namely that extracts from the ancient skeletons may contain DNA from the burial environment from 'mycobacteria other than tuberculosis' (MOTT). These bacteria are common in the environment but are not normally associated with clinical TB. By 2016, Müller et al. were not convinced of the specificity of PCRs to MTBC alone, and suggested that PCR products could actually pertain to MOTT rather than MTBC. Furthermore, the identification of such errors is difficult if not impossible (Müller et al. 2016:5). This must be taken into consideration especially when skeletons display no bone changes that may indicate a possible clinical infection with TB – the aDNA analysis may be providing a false positive in such cases.

Another study of MTBC aDNA research, with very novel findings, was also published in 2014 (Bos et al.). Instead of concentrating on the identification and isolation of MTBC aDNA as a guide to prevalence of infection in individuals with or without the bone changes, the study looked for the source of TB causing infections in the New World (Bos et al. 2014:494). This was briefly mentioned in the section above entitled Tuberculosis in the New World, and is considered here in more detail, because it is one of the newer uses of genetics in TB research. Modern strains of *M. tuberculosis* from the Americas are genetically closely related to the strains from Europe, so it has always been assumed that TB was introduced to the Americas post-Columbian contact; that is after Columbus landed in America in October 1492. Firstly, it was shown that a member of the *M. tuberculosis* complex had caused TB in humans prior to Columbian contact. Secondly, it was found that the ancient strains of TB were most closely related to strains isolated from seals

and sea lions. It was concluded that these sea mammals had a role in transmitting the disease to humans in these parts of the world (Ibid. 2014:494).

Bos et al. screened 68 skeletal samples from skeletons from New World sites dated to pre- and post- contact with Europe. All of these individuals displayed bone changes associated with TB (Ibid. 2014:494). Unfortunately, no details of what types of bone changes present were given. Only three of the 68 samples showed convincing evidence of MTBC aDNA presence and preservation. Radiocarbon dating revealed a date range of between AD 1028 and AD 1280, thus predating European contact (Ibid. 2014:495). All the MTBC aDNA which had been sequenced from three 1,000 year-old Mycobacterium genomes from bacteria isolated from the three Peruvian skeletons was compared to that of identified ancestral strains of MTBC. This comparison showed that the lineage tree reconstructions of the Peruvian genomes did not cluster with other human strains. They were found to be more closely related to the non-human lineage, and particularly to *M. pinnipedii*, a strain that had previously only been isolated from seal species in the Southern Hemisphere (Ibid. 2014). Hence it was suggested that, within the past 2,500 years, pinnipeds (seals and sea lions) probably contracted TB from an African host species and then carried the disease across the oceans. The disease was then contracted, via zoonotic transfer from these seals, by the coastal peoples of South America around the first millennium AD (700 and 1000) Bos et al. (2014:496). The contact between humans and pinnipeds which resulted in the disease being spread from animal to human presumably happened as a result of human exploitation of, and thus close contact with, these marine mammals, although the nature and extent of this human-pinniped contact is not understood. The appearance of skeletal lesions suggestive of TB infection in North America later in time (around AD 900) is consistent with either a transcontinental spread of the pathogen via established trade routes, or as a later and independent introduction of TB from a different source (Ibid. 2014: 496). The Bos et al. (2014) study drew two new conclusions about TB that had not previously been considered. Firstly, it was shown that a member of the *M. tuberculosis* complex had caused TB in humans prior to Columbian contact, and secondly, that

the ancient strains of TB were most closely related to strains isolated from seals and sea lions.

## 2.3.3 Phylogenetic studies of TB

In recent years, considerable research has been undertaken in clinical contexts which involves analysing DNA and amino acid sequences of proteins in order to understand how closely related other microbial species are to each other (Hocking et al. 2008:142). This work includes research to understand the sequence variations between the species of MTBC and also between different strains of these species (Brosch et al. 2002, Fletcher et al. 2003, Müller et al. 2014b).

The complete genome of modern *M. tuberculosis* was sequenced and published in 1998 (Cole et al. 1998), as discussed above, thus enabling phylogenetic research to take place. This resulted in an evolutionary tree demonstrating that *M. bovis* was not an ancestor of *M. tuberculosis* - indeed the converse was thought more likely to be true; that *M. bovis* probably derived from human-adapted *M. tuberculosis* (Brosch et al. 2002:3684). It was previously thought that TB started in humans when they contracted it as a zoonosis from their cattle soon after domestication. This was suggested to have taken place in southwest Asia (Ibid. 2002:3684). However, aDNA research found that *M. bovis* is probably descended from *M. africanum*, which in turn descended from an ancestor of *M. tuberculosis* (Ibid. 2002:3684, Brown and Brown 2011: 254, Mostowy and Behr 2005:207). Of course, more recent work by Bos et al. (2014) has introduced the idea of human TB originating from different sources, such as *M. pinnipedii*, and then being spread zoonotically to humans from other animals, in this case seals. This has certainly proven true for TB in the New World.

Further studies of modern strain varieties of organisms of the *M. tuberculosis* complex have confirmed that the species probably originated two to three million years ago in East Africa, and has been a human pathogen since infecting early

hominids in this region (Brown and Brown 2011: 254, Comas et al. 2013:1176). By 2011, 875 strains of *M. tuberculosis* had been analysed from 80 countries and this work has allowed the species to be divided into six main lineages and 15 sublineages, with each of the main lineages being associated with a specific geographical area. All six of these main lineages are found in Africa, which supports the hypothesis that the disease spread out of Africa and through Europe with the first migrations of *Homo erectus* two million years ago, and again when *Homo sapiens* left Africa 70,000 years ago (Brown and Brown 2011: 255, Mostowy and Behr 2005:208). A diagram of the phylogenetic lineages of the *Mycobacterial species* is shown in Figure 2.14;



Fig. 2.14 The phylogenetic lineages of Mycobacterial species. (Rue-Albrecht et al. 2014)

Brown and Brown (2011:255) suggest a point to consider is that, although phylogenetic studies show human TB did not derive from bovine TB, it cannot be concluded that all TB in the archaeological record must have been caused by *M. tuberculosis.* There were quite probably localised transfers of bovine TB to humans from their animals in the past, and, as Bos et al. (2014) show, from TB in other animal species, although it is thought that human-to-human transmission of *M. bovis* would probably be very limited.

In summary, in this chapter, literature pertaining to the clinical, bioarchaeological and genetic aspects of TB has been reviewed and discussed. The preceding aDNA analysis projects (Müller et al. 2014a, 2014b, 2014c, 2016), from which the current research project skeletal samples derive, have now been considered, and their results discussed. As the current research focuses on stable isotope analysis to examine the proposed origins and likely mobility histories of TB sufferers who were buried in Roman Britain, it is appropriate to now explore these techniques and their recent uses in exploring mobility.

## Chapter 3: Stable isotope analysis

#### 3.1. Introduction

Stable isotope analysis of archaeological human remains may be utilised to examine diet in the past, but also to explore evidence for possible places of origin and mobility during a person's life. Over the past few decades, isotope data have been increasingly used in archaeology to study ancient diets and to learn about movements of groups of people or individuals, including those from Romano-British contexts. For this study, collagen was extracted from bones for carbon and nitrogen isotopic analysis, and tooth enamel was used for strontium and oxygen analysis, so it is appropriate at this point to consider the formation and composition of bones and teeth before the isotope systems are discussed.

Dry bone is about 70% mineral and 30% organic, with the greatest part of the organic component being collagen (Roberts et al. 1993:179). Collagen is a fibrous protein and the fibres of which it is composed are long (see Figure 3.1). The crystals that make up the mineral portion of the bone are embedded within a matrix of these collagen fibres (Mays 2010:1). The main component of bone mineral, hydroxyapatite (a form of calcium phosphate), gives the bone its rigidity (Roberts et al. 1993:179). The organic component gives the bone slight flexibility and hence strength. It is the organic part which degrades after death, and this process accounts for the brittle nature of archaeological bone (Mays 2010:1).

The bone component essential in carbon and nitrogen isotope analysis is collagen (see Figure 3.1). Collagen is a protein found within bones and bone is renewed (turned over) throughout life, so carbon and nitrogen isotope analysis of collagen (if preserved), will potentially provide information about the diet of the sampled individual in his or her later years of life, and not their full life history. However, Hedges et al. suggest that human femoral bone collagen isotopically reflects a diet over a much longer period than 10 years and can include a substantial portion of collagen synthesised during adolescence (Hedges et al. 2007:815). Bone turnover rates are poorly documented, and vary for different bones, but complete replacement takes approximately between 10 and 30 years in an adult (Ambrose 1993:59). For an adult skeleton, collagen stable isotope ratios give a fairly longterm indication of diet of the individual, but probably not for the full life span as previously mentioned. The carbon incorporated into collagen derives mainly from dietary protein as the majority of the amino acids in collagen cannot be synthesised from other sources. However, if a diet is particularly low in protein, contributions from other food groups may occur (Mays 2000:426, Fernandes et al. 2012:298, Craig et al. 2013:346). It can be concluded therefore, that collagen  $\delta$ <sup>13</sup>C mainly reflects dietary protein (Ambrose and Norr 1993:1, Tieszen and Fagre 1993:121), and virtually all of the nitrogen taken in in the diet comes from protein and therefore nitrogen stable isotope values provide information about dietary protein (Mays 2000:426).



Fig. 3.1 The biochemical structure of the protein, collagen. (Harbor Medtech 2017)

Collagen, if preserved and intact, will contain *in vivo* isotope ratios. During diagenesis, a collagen-like amino acid profile (suggesting that collagen remains intact) is preserved until about 5% of the original protein content remains (Stafford et al. 1991:35). Other researchers suggest that amino acid elemental (C:N) composition,  $\delta^{13}$ C and  $\delta^{15}$ N change very little until 99% of the collagen is lost

(Dobberstein et al. 2009:31). However, dental enamel is far less subject to diagenetic change than bone mineral or dentine. This can be attributed to the fact that dental enamel has larger crystals than bone mineral (Koch et al. 1997:417, Ayliffe et al. 1994:5291). Enamel also contains very little organic material and hence, unlike bone, enamel generally is not degraded by soil micro-organisms (Hackett 1981:243).

## 3.2 Bone

## 3.2.1 Gross structure of bone

There are two different types of bone tissue: cortical and trabecular bone. Cortical bone is the solid, dense outer layer. It is thickest in the diaphysis (shafts) of the long bones (see Figure 3.2), and forms a thin layer round the long-bone ends and around flat bones, for example ribs, and irregular bones, such as the vertebrae (White and Folkens 2005:40, Mays 2010:2). Trabecular bone is less dense and has a honeycomb structure. This is located within the ends (epiphyses) of long bones and in the interior of the flat and irregular bones (Roberts et al. 1993:180). In living bone, most of the outer surface is surrounded by a thin membrane; the periosteum (White and Folkens 2005:41, Mays 2010:2) (see Figure 3.2). The internal walls of the medullary cavities of long bones are lined with another membrane, the endosteum. This also lines the network of tiny cavities in trabecular bone (Roberts et al. 1993:181, White and Folkens 2005:40, Mays 2010:3).



Fig. 3.2 A longitudinal section of a long bone (Nasir 2014)

Bone is living tissue, permeated with nerves and blood vessels and is continually being formed and broken down. This also means it can repair damage after disease or injury (Mays 2010:4-5). Relevant to this project, this reparative process has actually been observed by bioarchaeologists studying the skeletons that have provided the samples for this project, with inflammatory related new bone growth on the ribs observed for most (17 out of 21) of the individuals.

## 3.2.2 Microscopic structure of bone

Bone is initially laid down as woven or primary bone. This is replaced gradually by mature, lamellar bone. Woven bone forms the foetal skeleton, but this has been replaced by lamellar bone by the age of one year. Woven bone may also be produced in the adult skeleton in response to disease or injury (White and Folkens

2005:48, Mays 2010:6), as has been seen on the ribs of some of the individuals in this study, as a result of their possible response to infection with *M. tuberculosis* (See Figures 2.10 and 2.11). Woven bone is distinguishable from lamellar bone with the naked eye as the former is coarser and more porous (Mays 2010:6). Lamellar bone is composed of a series of microscopic layers (lamellae) about four to twelve microns thick (one micron ( $\mu$  m) = 1/1000 mm). Lamellar bone is stronger than woven bone (Mays 2010:6).

In adults, both trabecular and cortical bone have a lamellar structure, but they differ in the way they are organised. Cortical bone is permeated by many interconnected channels known as Haversian systems, or canals (see Figure 3.3) (Roberts et al. 1993:181). The bone relies on these for its blood supply. Bony lamellae are arranged concentrically around Haversian canals (See Figure 3.3, below). There are normally around 4 to 20 lamellae around each Haversian canal and this unit of bone organisation is called an osteon (White and Folkens 2005:43). Trabecular bone does not have a Haversian system because its fine honeycomb structure allows it to receive sufficient nutrients and oxygen from the plentiful supply of blood vessels which run through this open structure (Mays 2010:7).



Fig. 3.3 The structure of mature bone (Teach Me Anatomy 2016)

## 3.2.3 Bone cells

Bone renewal and replacement occurs due to the activity of bone cells. There are three main types of bone cells, namely osteoblasts (responsible for formation of new bone), osteocytes (responsible for maintenance of bone as a living tissue) and osteoclasts (responsible for resorption of bone) (Roberts et al. 1993:181, Mays 2010:7, White and Folkens 2005:43). Osteoblasts are concentrated on bone surfaces, for example beneath the periosteum, where they produce the organic matrix, osteoid, which then mineralises to form bone. Osteocytes are found within lacunae (spaces) in bone and receive nutrients via canaliculi (small channels), which connect osteocyte lacunae with neighbouring lamellae. Osteoclasts which are actively resorbing bone are found in depressions on bone surfaces known as Howship's lacunae (White and Folkens 2005:43).

# 3.3 Teeth

Humans possess four different types of teeth: incisors (chisel-like for cutting food), canines (conical for puncturing and tearing), premolars and molars (broad, flattened surfaces for crushing and grinding) (Berkovitz et al. 1986:18 Hillson 1996:7). Humans have had one set of teeth (deciduous) that are replaced by another (permanent) by the time they reach adulthood (see Figure 3.4). The deciduous (milk) teeth are smaller than the permanent adult teeth and appear during infancy and early childhood. The deciduous dentition is composed of 20 teeth. Both the maxilla (upper jaw) and mandible (lower jaw) each contain four incisors, two canines and four molars. There are no deciduous premolars (Berkovitz et al. 1986:18, Hillson 1996:9). The deciduous teeth are all replaced from middle childhood onwards with all of the permanent teeth having erupted by about 18 years of age. The third molars (wisdom teeth) are the last to emerge. The adult permanent dentition is normally comprised of 32 teeth with each jaw containing four incisors, two canines, four premolars and six molars (Berkovitz et al. 1986:7), as shown in Figure 3.4;



*Fig. 3.4 Human dentition. Deciduous dentition (centre) and permanent dention (top and bottom). Ages of tooth formation (in brackets) shown as months (mo) and years (yr) of age.* (Open Stax 2013)

## 3.3.1 Tooth structure

Each tooth consists of a crown (which projects above the gum) and one or more roots which taper into sockets (known as alveoli) within the jaw (see Figure 3.5). The junction between crown and root is termed the neck or cervix of the tooth (Hillson 1996:8, Scott 2008:267). Teeth consist of three hard tissues; enamel, cementum and dentine. These enclose the dental pulp (soft tissue made up of nerves and blood vessels), which is located in the pulp cavity and the root canal. The dental hard tissues do not possess a blood supply and, unlike bone, are not being continuously renewed. Once formed, they are unable to repair themselves in response to damage of disease (Scott 2008:267, Mays 2010:11).



Fig. 3.5 The structure of a tooth (IGCSE 2016)

Enamel is mostly made up of inorganic matter with a chemical composition similar to that of bone mineral (hydroxyapatite). Enamel is arranged in rods or prisms and, because it is not a living tissue, it lacks a cellular structure. Dentine consists of about 70% inorganic material by dry-weight, mainly hydroxyapatite, along with an organic component, which is mainly collagen (Berkovitz et al. 1986:79). Odontoblast cells are only found on the inner surface where they line the pulp cavity. Cementum coats the roots of the teeth and anchors them into their sockets. It has a composition similar to bone with a degree of cellular structure (Hillson 1986:6, Hillson1996:8, Mays 2010:11).

# 3.3.2 Tooth formation and development

Dental hard tissues start to form during the 15<sup>th</sup> week of foetal development. Dentine forms first. Odontoblasts secrete an organic matrix which then becomes mineralised to form dentine (Berkovitz et al. 1986:177). Enamel formation (amelogenesis) consists of three phases; the formation of an organic matrix, mineralisation of the matrix, and a maturation phase when enamel loses most of its organic component. The enamel-forming cells are known as ameloblasts. Enamel formation begins near the tip of the crown and the crown grows back towards the cervix until it is complete (Ibid. 1986:176). Cementum, like the other hard dental tissues, results from the laying down of an organic matrix followed by its mineralisation. The cementum layer increases in thickness with age because it continues to be formed throughout life (Mays 2010:13). The development of permanent tooth crowns takes place in three phases (the roots continue to form after the crowns are completed – see also Figure 3.4):

1. Incisors, canines and first molars initiate development during the first year after birth (or just before birth) and the crowns of these teeth are completed between three and seven years of age (Berkovitz et al. 1986:176, Smith 1991:144).

 Premolars and second molars start formation during the second and third years after birth and are completed between four and eight years (Berkovitz et al. 1986:176, Smith 1991:144).

3. Third molars begin formation between seven and 12 years and are completed between 10 and 18 years of age (Smith 1991:145), with some variation for completion and eruption (Fanning and Moorrees 1969:999). In terms of the teeth used for the current research, the author only had permission for access to samples of bones and teeth remaining unused following the collaborative Durham and Manchester Universities aDNA TB project (see Chapter 1). This means that some of the teeth were formed during the years an individual would have been breastfeeding, and hence interpretation of these results need to take this breastfeeding effect into account. Ideally, second premolars, second molars or third molars would be used for isotope analysis, as these teeth tend to be formed after the period where breastfeeding would impact on their composition and thus may inform more about the history of the mother of the sampled individual rather than of about the individual themselves. These limitations of the study are discussed in depth in Chapter 8 Discussion, and Chapter 9 Conclusion, and so will not be mentioned in more detail here.

Having discussed bone and tooth composition and development, stable isotope analysis of these skeletal elements are now detailed.

## 3.4 Isotope analysis

Stable isotope ratios of carbon and nitrogen vary in different foods. These differences are reflected in the tissues (including the skeleton) of the consumer. This makes these skeletal tissues useful for reconstructing past diets for the individuals in this study, and informing us if the sample population being studied was eating similar foods to those available from the diet in the locality in which they were buried, and if the sample population were consuming the same foods as the rest of the cemetery population where they were interred. Interpretation of these results can also provide evidence of possible migration into the country, i.e. if they consumed a different diet to local individuals, they are likely to be immigrants to the area from somewhere with different food availability and dietary customs.
Isotope analysis provides researchers with information on who was moving in a specific population and can suggest where they may have come from (Prowse 2016:205). Oxygen and strontium isotope analyses of dental enamel are used in this study to determine if the study sample individuals spent their childhood in the same area where they were eventually buried. Isotope ratios of strontium differ according to local geology and oxygen isotopes in rainwater vary with local climate, meaning that isotopic ratios of these elements differ with respect to locality. Strontium and oxygen isotope ratios are incorporated into bones and teeth via foods and drinking water, respectively. Assuming people in the past sourced their foods and drinking water locally, skeletal strontium and oxygen ratios may be used to indicate the area where people lived at the time their bones and teeth were mineralising (eg. see Mays 2010:265, Brown and Brown 2011:79, Müldner et al. 2007, 2011, Eckardt et al. 2009, Chenery 2010, 2011 and Prowse 2016).

Isotopes of elements have the same chemical properties but differ in their atomic masses due to having different numbers of neutrons in the nucleus of their atoms. Most elements exist as two or more isotopes. Some isotopes are radioactive; these steadily decay and break down into other elements. Other isotopes are stable; they are not radioactive and do not change proportion in the natural environment over time (Norris and Corfield 1998:2, Mays 2010:265, Brown and Brown 2011:80, Prowse 2016:205). To illustrate this point, the element carbon occurs both as a radioactive isotope (<sup>14</sup>C) and as the more abundant stable isotopes, <sup>12</sup>C and <sup>13</sup>C with <sup>12</sup>C constituting 98.99% of stable carbon and <sup>13</sup>C making up the remaining 1.11%. Organism metabolism and many physical processes, such as evaporation, discriminate between different isotopes and their relative abundances in biological tissues. This is due to lighter isotopes (the ones with fewer neutrons in their atomic nuclei) often being more reactive than heavier isotopes because the bonds of the lighter isotopes are more easily broken, thus leading to easier evaporation or easier incorporation into organic tissues than that occurring with the heavier isotopes (Prowse 2016:205). For instance, clouds are more enriched in <sup>16</sup>O compared with surface waters because  $H_2^{16}O$  evaporates more easily than  $H_2^{18}O$ . This results in surface water, rain, snow and ice being

enriched in <sup>16</sup>O, compared with the seawater from which they originally evaporated (Norris and Corfield 1998:2). This is called isotope fractionation, a term which is used to describe a change in the relative proportions of the isotopes in the products of a reaction compared to the proportions in the original substrate (Coplen 1994:274, Brown and Brown 2011:81).

Stable isotope ratio data are not reported in SI units; instead the results for carbon, nitrogen and oxygen isotopes are recorded as a series of  $\delta$  numbers with positive or negative values relative to an international standard (Coplen 1994:275, Coplen 2011:2540) and the variation in these ratios are so small they are recorded as parts per thousand ("per mil" or ‰) (Prowse 2016:206). Strontium isotope results are not recorded as  $\delta$  numbers, but instead are noted as a ratio of <sup>87</sup>Sr/<sup>86</sup>Sr (Ibid. 2016:206).

The equation used to describe and calculate the  $\delta$  difference is shown as:

$$δ^{13}C = \frac{(^{13}C/^{12}C)_{sample}}{(^{13}C/^{12}C)_{standard}} - 1$$

The  $\delta$  notation is used because we are dealing with such small variations in isotopic ratios compared with a standard (Coplen 1994:275, Mays 2010:266). A positive  $\delta$  <sup>13</sup>C value would therefore mean that the <sup>13</sup>C content of the sample being tested is enriched compared with the standard, or it could conversely mean that the <sup>12</sup>C is depleted compared with the standard. A negative  $\delta$  <sup>13</sup>C indicates a depletion in <sup>13</sup>C and an enrichment in <sup>12</sup>C compared with the standard.

A variety of standards are used as comparisons in different isotope systems. Since the early days of isotope research, these have been updated because confusion had existed in the reporting of stable hydrogen, carbon and oxygen isotopic results. This reporting was taking place on non-corresponding scales because, for carbon, the standard was originally a marine limestone called Pee Dee Belemnite

(PDB). The original belemnite material is now all used up and secondary standards (notably the marble, NBS-19) are used. The differences between NBS-19 and PDB are accurately known and so results can be reported relative to a notional standard called Vienna PDB (VPDB) (Norris and Corfield 1998:2, Coplen 1994:275, American Society of Limnology and Oceanography 1995:1182). Oxygen ratios can also be measured against VPDB or against Vienna "standard mean ocean water" (VSMOW). The original standard was SMOW (Standard Mean Ocean Water), which was a hypothetical water sample with abundances of stable hydrogen and oxygen isotopes similar to those of average ocean water. However, there were three versions of SMOW in use; the U.S. National Bureau of Standards version, the H. Taylor and S. Epstein (California Institute of Technology) version, and the International Atomic Energy Agency (IAEA) version, all of which were different. So VSMOW was introduced and superseded all three of these versions in order to provide one comparable standard reference material (Coplen 1994:275). Atmospheric air is usually used as the standard for nitrogen isotope measurements (Coplen 2011:2543). These standards are thus used for the measurement of relative differences of isotope ratios, and the standard materials are assigned reference delta values of zero by agreement (lbid. 2011:2543).

### 3.4.1 The study of palaeodiet using carbon and nitrogen stable isotopes

As previously discussed, carbon occurs as two stable isotopes, the lighter <sup>12</sup>C and the heavier <sup>13</sup>C (Prowse 2016:206). The relative abundances of these are about 98.99% and 1.11%, respectively (Norris and Corfield 1998:2). Nitrogen also has two stable isotopes; these are <sup>14</sup>N and the heavier <sup>15</sup>N and they show relative abundances of approximately 99.6% and 0.4%, respectively (Mays 2010:265). Most palaeodietary studies have concentrated on carbon and nitrogen in bone collagen, which is the method used here, and approximately 90-200mg of bone is required for the process. In order to analyse stable isotopes in collagen, the collagen is first extracted from the bone and then purified. The resulting material is

burned and the gases produced are analysed using a mass spectrometer. This gives the relative abundance of the different isotopes present.

(i) Carbon stable isotopes

Carbon stable isotope ratios in plants vary according to the photosynthetic pathway the plant utilises in order to produce carbohydrates from atmospheric carbon dioxide in the presence of sunlight. There are two major pathways and these are known as the  $C_3$  (Calvin cycle), eg. in wheat and barley (Roberts et al. 1993:303), and the C<sub>4</sub> (Hatch-Slack), eq. in millet, mechanisms (Norris and Corfield 1998:102, O'Leary 1988:330, Roberts et al. 1993:304, Mays 2010:266). There is a third type of pathway; that of the crassulacean acid metabolism (CAM) used by plants which live in arid environments such as deserts. These also use a photosynthetic pathway similar to that utilised by C<sub>4</sub> plants (Roberts et al. 1993:304). The C<sub>3</sub> pathway is so-called because it involves the production of a three-carbon compound in its first step. Plants using this pathway are from temperate zones and have mean  $\delta^{13}$ C values of around -27 to -28‰, whereas some grasses adapted to living in tropical and sub-tropical zones with high temperatures and light intensities use the C<sub>4</sub> pathway, resulting in the production of a four-carbon compound in its first step. Examples of important dietary C<sub>4</sub> plants include maize, millet, sorghum and sugar cane, none of which were grown in Britain in the Roman era. The  $\delta^{13}$ C values of C<sub>4</sub> plants cluster round a mean of around -14‰ (O'Leary 1988:330). There are slight disagreements about the range of these values, with Saunders and Katzenberg (1992:112) stating that C<sub>4</sub> plants have  $\delta^{13}$ C of -9 to -14‰ and C<sub>3</sub> plants have  $\delta^{13}$ C of -20 to -35‰. Although the photosynthetic pathway has the main effect on the  $\delta^{13}$ C value of a plant, water and nutrient availability, light intensity and altitude also have an effect on the precise figure (O'Leary 1981:553, Tieszen 1991:227). If plants grow on the forest floor beneath a dense canopy of higher shrubs and trees, they may produce unexpectedly negative delta values caused by taking in carbon dioxide which has

been released from rotting vegetation and trapped beneath the canopy. This is known as the canopy effect (Vogel 1978).

Heaton et al. (2009:2224) found that there are also differences in  ${}^{13}C/{}^{12}C$  ratios of plants grown in different environments and thus different geographical locations. This needs to be considered when interpreting results, as they concluded that because the <sup>13</sup>C/<sup>12</sup>C ratios of plants depends upon the composition of atmospheric CO<sub>2</sub> this could vary with different climates and soil types. However, interpretation of this is far from straightforward; they also pointed out that differences in environmental conditions could occur over time as well as in plants from different locations. For example, this would result in wheat grain grown on different sites in different years displaying varied <sup>13</sup>C/<sup>12</sup>C ratios. Further confounding interpretations is that differences in  $\delta^{13}$ C of different wheat species/variety were discovered (Ibid. 2009:2228). Their research found that in some years, emmer wheat had  $~\delta~^{13}C$ values of 1.0 to 1.4‰ higher than those of einkorn wheat. However, in other years there was little difference observed between the emmer and einkorn wheat varieties. Heaton et al. discovered that different wheat species can show large differences in  $\delta^{13}$ C even when grown under the same conditions, and thus observed differences may not actually indicate different years and/or locations (Ibid. 2009:2229).

Plants growing in marine environments function slightly differently from terrestrial plants and thus it must be noted that carbon in marine environments derives largely from dissolved bicarbonate, which is enriched in <sup>13</sup>C compared with atmospheric carbon dioxide (Smith and Epstein 1971:380). Marine plants and plankton, which mainly utilise the C<sub>3</sub> pathway for photosynthesis, have less negative  $\delta$  <sup>13</sup>C values than terrestrial C<sub>3</sub> plants (Chisholm et al. 1982:1131, Tan 1989). Marine mammals and fish have a delta value of around -17 to -18‰ in their flesh (Chisholm et al. 1982:1131).

In relation to the above discussion, the influence of diet on the distribution of carbon isotopes in animals has also been investigated (eq. DeNiro and Epstein 1978). Animals were fed a diet composed of a constant isotopic composition. It was found that the isotopic composition of the whole body of an animal reflects the isotopic composition of its diet, but that the animal is only enriched in  $\delta^{13}$ C by an average of 1‰ relative to its diet (Tieszen et al. 1983). The <sup>13</sup>C enrichment relative to diet of the whole body was found to be balanced by a <sup>13</sup>C depletion of the respired CO<sub>2</sub>. The relationship between  ${}^{13}C/{}^{12}C$  ratio of a tissue and  ${}^{13}C/{}^{12}C$  ratio of the diet depends on the nature of the tissue and of the diet (DeNiro and Epstein 1978:495). This is due to the difference in tissue composition and turnover time, secondary fractionation effects and synthesis from different constituents of the diet (Lee-Thorp et al. 1989:586). By 1989 it was already established that the  $\delta^{13}$ C of bone collagen was 3-6‰ more positive than the animal's diet (Lee-Thorp et al. 1989). Estimations for humans include +5.1‰ (van der Merwe and Vogel 1978) and +6.1‰ (Chisholm et al. 1982), with an average of +4.5‰ proposed by Kennedy (1988). However, the figure may vary with dietary quality. It has been suggested that the contributions of different nutrients in the diet to the  $\delta^{13}$ C value is unknown. There is debate about how much collagen carbon derives from dietary proteins and/or carbohydrates and different mixes of these foods in the diet may affect these relationships (Lee-Thorp et al. 1989:557, Craig et al. 2013:345, Prowse 2016:206).

In summary, as has been discussed above, Tieszen et al. (1983) argued that the differences in  $\delta^{13}$ C values between diet and the flesh of a consumer is small (around +1‰) which means that, in a given environment,  $\delta^{13}$ C values of animal and plant foods are similar (Tieszen et al. 1983:32). The use of  $\delta^{13}$ C measurements from the bone collagen of the skeletal samples in this study will determine if the individuals consumed a diet containing marine foods, which was uncommon for people in Britain during the Iron Age (Müldner and Richards 2007:690) but may have become more acceptable in some populations by the Roman period. In regions such as Britain where there were no C<sub>4</sub> plants grown

during the Roman era, people consuming a local terrestrial diet would have a  $\delta$  <sup>13</sup>C of -19‰ or less. Bearing in mind  $\delta$  <sup>13</sup>C data for people with a large intake or marine resources is -14.5 to -16‰ (Saunders and Katzenberg 1992:112), it should be possible to establish if individuals in this study ate mainly C<sub>3</sub> plants, and if they had any input from marine resources. The isotope analysis may also show these people ate C<sub>4</sub> plants, which would have been largely unavailable in Britain during the Roman period. As C<sub>4</sub> plants only grew in warmer climates than experienced in Britain, this would indicate that these people are very likely to have moved into the country, unless they were able to source imported foods in a large enough quantity to register in their  $\delta$  <sup>13</sup>C values; this would appear to be very unlikely from consumption of imported food alone.

Another cause of enrichment of  $\delta^{13}$ C values for the individuals would be the consumption of wheat and other grains grown in warmer, drier areas than Britain, for example the Mediterreanean, or more likely from North Africa where much Roman wheat was cultivated. Field studies have shown that  $\delta^{13}$ C values for plants become higher with decreasing aridity, due to the closure of stomata in order for the plants to prevent water loss by transpiration and therefore a decrease in carbon dioxide diffusion leading to lower carbon isotope fractionation (Goude and Fontugne 2016:118). Isotopic variability in plants is the primary determinant of the carbon isotope signature in the food eaten by humans with human bone  $\delta$  <sup>13</sup>C values ranging from -23.3% to -18.2% and  $\delta^{15}$ N values ranging from 5.5% to 13.2‰ at the southern latitudes found in the South of France (Ibid. 2016:122). van Klinken also found that climatic differences occur in regional patterning across Europe in the  $\delta^{13}$ C values of plants and he described this as being caused by the influences of temperature and/or relative humidity on the photosynthetic processes of plants (van Klinken 2000:42). In addition to the closure of stomata being responsible for this climatic variation in plants, van Klinken also cites differences in the actual fixation of carbon as being affected by temperature and partial pressure of CO<sub>2</sub> (Ibid. 2000:42). van Klinken examined Holocene charcoal samples and suggested that a  $\delta^{13}$ C value of -25.7‰ would be typical for Britain, whereas  $\delta^{13}$ C

values of Holocene charcoal samples from Italy would be around -24.3‰ and from North Africa, they would be nearer to -23.0‰ (Ibid. 2000:43). This enrichment effect, if observed in humans, needs to be considered as another indicator of mobility in that they may have originated from areas of the world which are warmer and drier than Britain, or they may have consumed significant quantities of grain imported from these areas.

#### (ii) Nitrogen stable isotopes

Most plants cannot use atmospheric nitrogen to make their proteins. Instead they take it in from the soil after it has been 'fixed' into nitrate by the actions of nitrifying bacteria present in the soil. The exceptions are plants known as legumes, such as peas and beans and also clover (Roberts et al. 1993:307), which have nitrogen-fixing bacteria in their root nodules and therefore can use atmospheric nitrogen as well as nitrates absorbed from the soil (Ambrose 1991:296, Roberts et al. 1993:307, Hocking et al. 2008:202). This biological fixing of atmospheric nitrogen leads to <sup>15</sup>N depletion of the soil. However, de-nitrifying bacteria (also found in soils) reverse the nitrogen fixing process (Roberts et al. 1993:307), and thus tend to increase soil  $\delta$  <sup>15</sup>N values. In most environments the net contributions of nitrification and de-nitrification leads to an increase of soil  $\delta$  <sup>15</sup>N values. However, soil nitrogen fixation is inhibited by arid soils and high temperatures, and therefore in warm and arid environments incorporation of atmospheric nitrogen into the soil is low and  $\delta$  <sup>15</sup>N values are high (Ambrose 1991:296).

 $\delta^{15}$ N values for leguminous plants (eg. peas, beans and clover), lie between 0‰ and +4‰ and for non-leguminous plants, they are around +5‰ (DeNiro and Hastorf 1985:97). Factors that affect the precise  $\delta^{15}$ N values of plants include temperature, altitude, rainfall and salinity of the soil in which they grow (Ambrose 1991:297) together with the addition of fertilisers to the soil (Ambrose 1991:297, Bogaard et al. 2007:335). Higher rates of nitrogen fixation have been observed in moist grasslands compared to arid ones (Maasdorp 1987:7), and cool, moist forest soils tend to have higher nitrogen fixation and lower  $\delta^{15}$ N than hot, dry savannah areas. Clay soils also tend to have higher  $\delta^{15}$ N values than sands and silts (Ambrose 1991:296), probably because clays retain moisture while sands and silts drain rapidly.

In mammals, insufficient protein (Gannes et al. 1998:728, Fuller et al. 2005) and water taken in with the diet was previously thought to result in elevated  $\delta$  <sup>15</sup>N (Ambrose 1991:293). This would mean that animals and humans living in arid climates would have high delta values (Schoeninger and DeNiro 1984:625) as would those undergoing starvation or protein deficiency. However, the reasons behind these elevated nitrogen isotope values compared to animals and humans from similar trophic levels in more temperate climates are complex and debated. The difference may be the result of a physiological response by animals to heat and to water stress, (Ambrose 1991) but alternatively, diet may be predominantly responsible for this difference due to the positive correlation between rainfall and the  $\delta$  <sup>15</sup>N values of plants and herbivores (Hartman 2011). In the case of animals and humans consuming insufficient protein in their diet, starving animals can "live on their own meat", meaning that the tissues of animals at high tropic levels become enriched in <sup>15</sup>N (Gannes et al. 1998:728). Starving or protein-deprived animals deaminate tissue proteins to use as energy and a source of amino acids for protein synthesis. As <sup>14</sup>N is excreted in preference to <sup>15</sup>N, the nitrogen which remains in animal proteins becomes <sup>15</sup>N enriched. This increase in <sup>15</sup>N in animal tissues can be used as an indicator of body condition (Gannes et al. 1998:728). It is also argued that loss of appetite during illness is not unusual, and therefore it would not be entirely unexpected to see people who are ill with TB exhibiting elevated <sup>15</sup>N in their body tissues for these reasons. Animal tissues have a higher  $\delta^{15}$ N value than their diets, and therefore  $\delta^{15}$ N increases at higher trophic levels in a food chain. This trophic level effect is making it possible to distinguish vegan from omnivore diets, although this can be difficult in practice. The "trophic level enrichment" effect between diet and body tissues results in an overall increase in nitrogen values as the food chain is ascended, as discussed. The archaeological

literature assumes this increase to be about 3–6‰ (Bocherens and Drucker 2003:46). This means that, while broad-scale changes in diet are observable in human isotopic values (Vogel and van der Merwe 1977:238, Richards et al. 2003:366), our lack of knowledge of the exact  $\delta$  <sup>15</sup>N<sub>diet-body</sub> value and also of factors which may influence this value mean that we can not identify small dietary changes in humans (O'Connell et al. 2012:426).

Whilst a large number of controlled diet feeding studies have been carried out on animals, this is more complex to do for humans; this is because the choice of tissue used for isotope analysis needs to be considered alongside its comparability with bone collagen. Additionally, collagen, hair keratin and blood proteins isotopically reflect medium or long-term diet, and while it is possible to raise an animal and keep it on an isotopically controlled diet for the duration of its life span, this is obviously not ethically possible for humans (O'Connell et al. 2012:427). For example, O'Connell et al. used a controlled diet for humans with known isotope values but with similar foods to that normally eaten by humans so as not to implement any short-term drastic changes. The researchers were then faced with the problem of how their data would translate to a  $\delta^{15}N_{diet-collagen}$  value. However, by comparing the data with tissue offset data from controlled dietary studies in animals, it was concluded that the  $\delta^{15}N_{diet-collagen}$  offset in the sample group was approximately +6‰ (O'Connell et al. 2012:431); this is at the higher end of the range of 3–6‰ assumed in the archaeological literature (Bocherens and Drucker 2003:46).

While the trophic level enrichment effect for body tissues in humans has been discussed above, breast milk must also be considered. This is because some of the individuals in this study were children and so their bone collagen would still show traces of the breast feeding effect due to them not having had chance to renew and replace this collagen before they died. Lactating mammals secrete milk that is enriched in  $\delta$  <sup>15</sup>N by approximately 3–6‰ when compared to their diet (Steele and Daniel 1978:7), which is the same as the trophic level effect. In studies

of breastfeeding practices in the past it was also discovered that pre-weaning age children have elevated  $\delta^{15}$ N values in their collagen compared to those of the mother. This is because a breastfeeding child is effectively one trophic level higher than its mother due to it consuming a product of its mother's tissues. This is reflected in elevated  $\delta^{15}$ N tissue values of the child (Schurr 1997:920, Millard 2000:5).

Marine food chains are enriched in  $\delta^{15}$ N compared with atmospheric and soil nitrogen. This means marine plants have a higher delta value than terrestrial plants, and marine animals have higher delta values than terrestrial animals at corresponding trophic levels. Marine fish have values of about +11% to +16% with marine mammals having values of +11‰ to +23‰. Shellfish  $\delta^{15}$ N values are lower than this at around +7% to +8% (Sealy et al. 1987:2702). Nitrogen isotopes are also used to distinguish a marine from a terrestrial diet if there are C<sub>4</sub> plants in the diet; examination of the nitrogen isotope data of coastal people versus marine resource users would generally show higher  $\delta^{15}$ N than inland people (Saunders and Katzenberg 1992:112). Consumption of inland freshwater fish may, however, result in higher  $\delta$  <sup>15</sup>N bone collagen values in the consumers (van Klinken et al. 2000:56). van Klinken et al. (2000:56) stated that other factors affecting  $\delta$  <sup>15</sup>N of plants can include human agricultural practices such as manuring. Modern plants subject to manuring can have  $\delta^{15}$ N as high as 8‰. Research on the impact of manuring on  $\delta^{15}$ N values of modern cereals found manuring significantly increases  $\delta^{15}$ N in cereal grain and chaff (Bogaard et al. 2007:335). The researchers also commented that, depending on the frequency of manuring, humans who have had a large dietary input of manured cereal grains could be interpreted as having eaten a largely animal-based or mixed plant/animal diet when this may not, in fact, have been the case (Bogaard et al. 2007:335). van Klinken et al. (2000:56) also analysed <sup>15</sup>N data but did not find any climate-related patterning across Europe, concluding that variations seem to be more linked to non-climatic effects, for example soil nutrient availability and vegetation type and cover. It was also discovered that some humans have high values due to unknown

physiological reasons. They based this theory on the fact that vegans have similar values to herbivores, but published measurements of other human trophic level effects seem larger than for carnivores. Thus, humans are concluded to be systematically different in their nitrogen isotope values, possibly with a physiological cause (van Klinken et al. 2000:56). To conclude, there is a diversity of  $\delta$  <sup>15</sup>N values across Europe, with variation between sites from different countries being about 4‰ (van Klinken et al. 2000:57). This was explained as being possibly due to local differences in plant and animal  $\delta$  <sup>15</sup>N values, and some dietary choices as previously mentioned.

Modern carbon and nitrogen delta values reflect atmospheric pollution (burning of fossil fuels) and modern agricultural practices (application of chemical fertilisers to crops), and there are estimates of likely carbon and nitrogen delta values in marine and terrestrial food chains in antiquity (Table 3.1). These values are approximate and are estimated by taking into account the effects of fossil fuel burning and the application of chemical fertilisers on modern values.

Type of organism	$\delta$ <sup>13</sup> C values (‰)	$\delta$ <sup>15</sup> N values (‰)
C <sub>4 plants</sub>	-11.5	+6.5 (Mays 2010)
plante		+3-9 (Ben-David and
		Flaherty 2012:316)
C <sub>3</sub> leguminous plants	-26.5	0 to +4.0 (DeNiro +
		Hastorf 1985)
C <sub>3</sub> non-leguminous plants	-26.5	+5.0 (DeNiro+Hastorf
		1985)
Meat	-26.0	+8.5
Marine fish	-17.0	+13.0
Marine mammals	- 17.5	+15.0

Table 3.1 Approximate mean  $\delta^{13}$ C and  $\delta^{15}$ N stable isotope values for some major food classes in antiquity (adjusted to take into account fossil fuel burning and use of chemical fertilisers on modern values) (after Mays 2010:268, Ben-David and Flaherty 2012 and DeNiro and Hastorf 1985).

(iii) Carbon and nitrogen stable isotope ratios in human bone collagen

The  $\delta^{13}$ C value of human bone collagen is about 5‰ less negative than diet and the  $\delta^{15}$ N in collagen is about 3–5‰ greater than diet (Ambrose 1993). Carbon in bone collagen comes mainly from dietary proteins although other food groups will contribute when a person has a diet that is low in protein (Craig et al. 2013:346). Almost all of the nitrogen comes from protein and therefore nitrogen stable isotopes directly reflect those in dietary proteins. Bone collagen turnover in adults occurs at the rate of about 1.5% - 4% per year in cortical bone (Hedges et al. 2007:808). Hence stable isotope data from bone collagen yields information about the diet of that individual over the last few decades of their life. However, stable isotopes in collagen from dentine give an indication of diet at the time the dentine was forming, reflecting the childhood diet of an adult (Mays 2010:269). In a young individual who is still growing, bone turnover is more rapid than that of adults, with a rate of about 10%-30% per year for cortical bone in adolescents, this being more rapid in younger children. The general rule is that the younger the child, the faster the rate of bone turnover (White and Folkens 2005:42, Hedges et al. 2007:809).

In analysing bone collagen to generate isotope values, it is assumed that the collagen isotope ratios have not been modified by post mortem processes. However, it has been proved that post mortem alteration of bone collagen isotope ratios does occur, but it is possible to identify prehistoric bone where the collagen has not been significantly altered in this manner (DeNiro 1985:806). They also showed that collagen extracted from prehistoric animal bones with C/N ratios between 2.9 and 3.6 had a range which encompasses the equivalent values for collagen from fresh animal and human bones (Ibid. 1985:807). It was also discovered that some prehistoric collagen samples with C/N ratios outside the 2.9 to 3.6 range have <sup>13</sup>C/<sup>12</sup>C and/or <sup>15</sup>N/<sup>14</sup>N that fall substantially outside the range exhibited by their modern counterparts (Ibid. 1985:807). This observed post mortem alteration of bone collagen isotope ratios is the result of the age of the sample (that is the length of time which it has been buried), but the factors involved in this alteration do not affect all of the bones from a particular burial environment. Although acknowledging the fact that all prehistoric bones have undergone diagenesis as a result of the sum of the physical, chemical and biological processes occurring after burial, the exact diagenetic processes causing the alteration of bone collagen have not been identified (Ibid. 1985:808). De Niro suggests that until these collagen damaging diagenetic processes are better understood, collagen samples with C/N ratios outside of the 2.9 to 3.6 range of collagen C/N ratios in fresh bone should be discarded. This is because their  $\delta^{13}$ C and/or their  $\delta^{15}$ N values may also have shifted as a result of this diagenesis (Ibid. 1985:808). van Klinken (1999:691) tightens this range further by rejecting any samples which fall outside of the 3.1-3.5 range for C/N ratios. However, in 2002, the evidence to date was reviews on the survival of organic matter in bone and three pathways of diagenesis were discussed (Collins et al. 2002). These are:

- 1. Chemical deterioration of the organic phase of bone (primarily collagen),
- 2. Chemical deterioration of the mineral phase of bone, and
- 3. Microbiological attack of the composite.

Whilst pathway 1 is rare in most environments, bones undergo pathway 2, which is then followed by microbial attack (pathway 3.) (Ibid. 2002:383).

However, one problem with analysing collagen through stable isotope values is that collagen can decay during burial. How this can be measured and the cut-off values for collagen preservation are argued. It has been previously suggested that collagen retains its isotopic signature until around 5% of the original protein content remains (Stafford et al 1991:35). Below this, the predominant amino acid compositions differ from that of collagen. It is therefore advisable to analyse the protein extracted from bone to determine whether or not it retains the collagen amino acid signature or not (van Klinken 1999:690). A method of combatting this issue would be to restrict analysis to bones retaining about 5% or more of their original protein. Fortunately, most bones from archaeological contexts are in temperate zones, as are the samples analysed in this thesis, and thus usually satisfy these criteria.

Undertaking an analysis of screening parameters for the identification of dubious quality collagen in archaeological bone samples, van Klinken (1999) investigated existing indicators of collagen quality to discover whether they can reliably detect contamination or degradation (van Klinken 1999:688). They found that modern, fresh bone is composed of approximately 22% collagen by weight. This collagen content drops steadily during burial, the speed of this depending on the climatic conditions with faster decreases taking place in hotter zones (Ibid. 1999:689). Eventually, the collagen content drops below 0.5% and these should be regarded as unsuitable for analysis; van Klinken and his colleagues use a cut off point of 1%. However, Ambrose (1990) found collagen was well-preserved only down to 3.5%, but these bones were mainly from Africa while van Klinken's bone samples were mainly from Europe (van Klinken 1999:689). In further support of the 0.5% by

weight cut off point, amino acid analyses were found to reveal amino acid compositions that do not vary much within the collagen weight yield range of 20% down to 0.5%. Nevertheless, when bone samples yield lower than 0.5% weight of collagen, amino acid profiles sometimes show lowered levels of glycine, alanine, proline and hydroxyproline (Beeley and Lunt 1980:371, van Klinken 1999:689-690). van Klinken concluded that the use of C/N ratios (DeNiro 1985) and % C content to detect low collagen levels in bone are useful indicators, because low collagen bones are a problem in palaeodietary reconstruction due to isotopic signatures changing in these bones (van Klinken 1999:694).

3.4.3 The study of migration using strontium and oxygen isotopes.

Isotope investigations of the mobility histories of people have concentrated on oxygen and strontium isotope ratios in hydroxyapatite, a form of calcium phosphate which makes up the mineral component of bones and teeth. Strontium has similar chemical properties to calcium and is incorporated into hydroxyapatite during mineralisation of bones and teeth in a person's lifetime. The phosphate part of the compound contains oxygen atoms, as do the carbonate ions, the latter of which are present in hydroxyapatite. Oxygen isotope analysis targets oxygen in either phosphate or carbonate. In the laboratory, the strontium and carbonate and/or phosphate components of the tissue are isolated from the tooth sample and the isotopic ratios are measured using a mass spectrometer (Evans et al. 2006:269, Chenery et al. 2010:154).

# (i) Strontium isotopes

Strontium naturally occurs as <sup>84</sup>Sr (0.6% abundant), <sup>86</sup>Sr (9.9% abundant), <sup>87</sup>Sr (7.0% abundant) and <sup>88</sup>Sr (the most common at 82.6% abundant). One isotope, <sup>87</sup>Sr, is radiogenic and is formed from the radioactive decay of rubidium-87. The other isotopes of strontium are all stable (Capo et al. 1998:198). The radioactive

decay of rubidium-87 is very slow with a half-life of 49 billion years, so this means that the proportions of <sup>87</sup>Sr vary in different types of rocks. Older rocks with high <sup>87</sup>Rb have had time to accumulate more <sup>87</sup>Sr than younger rocks or rocks with a low initial <sup>87</sup>Rb level (Capo et al. 1998:198).

As previously mentioned, the amount of <sup>87</sup>Sr in rocks or biological tissues is expressed as a ratio of  ${}^{87}$ Sr/ ${}^{86}$ Sr and is not reported using delta (  $\delta$  ) values (Prowse 2016:206). This can be measured very precisely, with the ratio varving from 0.7030 in volcanic rocks of recent age to around 0.7400 in some continental granites (Åberg 1995:309). The strontium isotope ratio of seawater has varied between 0.7070 and 0.7090 over geological time and so marine sedimentary rocks (such as the chalks and limestones of the burial environments in this study) have values within this range (McArthur et al. 2001:155). Mineral weathering normally predominates in the supply of strontium so the underlying geology, which is reflected in the isotopic ratio of the soil, is the driver for the strontium isotope ratios of the biosphere. However, airborne dust in some areas or sea-spray in coastal locations can significantly contribute to the strontium available (Bentley 2006). Strontium becomes incorporated into plants through their root systems, reflecting the soil in which they grow. Strontium enters the body through the diet (Prowse 2016:207) and isotope ratios are not altered by metabolic processes in living organisms so they remain unfractionated throughout the increasing trophic levels of food webs (Blum et al. 2000:87). This means that if an individual sources his or her food and drink from the area in which they live during development of the teeth, the <sup>87</sup>Sr/<sup>86</sup>Sr in their bones and teeth will reflect this geological area. Minerals that are ingested in the foods can also have an effect on the <sup>87</sup>Sr/<sup>86</sup>Sr values of an individual; for instance, common salt is rich in strontium and, even if consumed in small amounts as it would be if used as a food preservative, it can significantly influence <sup>87</sup>Sr/<sup>86</sup>Sr values (Wright 2005:556).

Pollard suggests the use of caution when interpreting strontium isotope results. He acknowledges that while the solid rock geology of an area plays a large part in determining the strontium isotope ratio of the local biosphere, it must be

remembered this is only one of several factors that contribute to the overall isotopic signal of humans living in the area (Pollard 2011:635). Other factors include drift geology, water supply sources, farming and culinary practices, trade in foodstuffs and dietary taboos (Montgomery 2010:327). Because of a combination of these contributory factors, strontium isotope ratios of people living within short distances of each other may vary markedly.

We need to also be more knowledgeable about the biology of strontium uptake in dental enamel (Montgomery 2011:636). Until weaning, it would appear that most strontium will be taken up by a child from the breast milk of the mother or wet nurse. However, this means a child may acquire a non-local strontium ratio if its mother is an immigrant to the area (Ibid. 2011:636). In order to mitigate against such complications, looking for a discernable dominant value within the teeth of a large number of individuals which could be identified as being a local signal is suggested (Ibid. 2011:636). This can then be compared with data from local fauna, local flora, and the geology. Roberts et al., in their research at Hull, were able to better define local biosphere strontium isotope signatures by sampling cattle bone, snail shell and soil from around the site and using these samples in strontium isotope analysis (Roberts et al. 2013:277). A point that the researchers would have needed to consider is when taking samples from which to estimate biosphere strontium isotope values in the past, is that animal bones and shell must be contemporary with the skeletal samples being analysed. Consideration also needs to be taken of contamination by modern fertilisers and run off from landfill sites. Roberts et al. obtained soil samples from depths of at least 60 cm to minimise modern anthropogenic contamination and to better represent contribution of local geology to the isotope signature. Ideally, the geological evidence would support the other evidence and a local signature can then be confidently identified (Montgomery 2011:636).

#### (ii) Oxygen stable isotopes

Oxygen occurs in three isotopes, all of which are stable. These are <sup>16</sup>O, <sup>17</sup>O and <sup>18</sup>O and they have relative abundances of 99.8%, 0.04% and 0.2%, respectively (Coplen 1994:274). Oxygen isotope ratios are expressed in terms of the amount of <sup>18</sup>O they have with respect to the amount of <sup>16</sup>O, and these variations are very small, and like carbon and nitrogen isotope ratios they are expressed as delta values. The standard used for comparison, as previously discussed, is Vienna Standard Mean Ocean Water (VSMOW), although for carbonate  $\delta$  <sup>18</sup>O, the VPDB standard is also sometimes used. Human skeletal tissues usually have less <sup>18</sup>O than the VPDB standard and so  $\delta$  <sup>18</sup>O measured against this standard is usually a negative value. However, skeletal tissues and most rainwater are enriched in <sup>18</sup>O compared with VSMOW, so values are positive when measured against this standard (lbid. 1994:274).

Humans obtain oxygen from the air during respiration and from ingestion of water, and to a lesser extent from food. Oxygen isotope ratios in the air are fairly constant, and hence it is ingested values that cause variation in the  $\delta$ <sup>18</sup>O values of animals and humans. Oxygen isotopes are fractionated, that is, the ratios are altered by metabolic processes. This means foods differ in their  $\delta$ <sup>18</sup>O values. This is of minimal concern in this study as human tissue  $\delta$ <sup>18</sup>O tends to be incorporated from drinking water (Levison et al. 1987, Kohn 1996, Daux et al. 2008). However, suckling babies obtain all of their water from the breast milk of the mother. This milk is enriched in <sup>18</sup>O compared with the local drinking water and therefore it must be noted that the  $\delta$ <sup>18</sup>O values in the bones of breastfed infants, and in the dentine and enamel of the teeth that form during this suckling time may be elevated by up to around 2‰ (White et al. 2004:234). For the purposes of this study, if possible, teeth that were mineralising during this period of the individual's life were not utilised in this study in order to avoid this confounding issue, although this was unavoidable in some cases.

Oxygen isotope ratios in precipitation vary with climate and thus reflect different geographical areas if their climates are different. Lower temperatures lead to lower  $\delta^{18}$ O, and so this means that the seasonal differences in  $\delta^{18}$ O can be quite considerable. This tends to be evened out if drinking water is sourced from large surface bodies of water, such as lakes, or directly from groundwater (Brettell et al. 2012b:118). However, if drinking water comes from small bodies of surface water such as ponds, seasonal temperature variations need to be considered and taken into account (Mays 2010:279). Another point which needs to be remembered when interpreting human  $\delta^{18}$ O data is that the  $\delta^{18}$ O value of drinking water is increased significantly by the boiling of that water during cooking (Bryant and Froelich 1996, Daux et al. 2008). Brewing may increase the  $\delta^{18}$ O value of ale by 1.3‰ over the original water used. Boiling water raises the oxygen isotope composition by approximately 10.2‰ after three hours of cooking (Brettell et al. 2012a:778).

Another variation in  $\delta^{18}$ O comes about from precipitation falling at high altitudes, because this will generally have a lower  $\delta^{18}$ O value than that falling at lower altitudes. Additionally, mean annual  $\delta^{18}$ O in precipitation generally decreases further inland in the direction of the prevailing wind. This is because clouds progressively lose <sup>18</sup>O as they move inland due to this heavier isotope being preferentially "rained out" first. In Britain, the prevailing winds are south-westerly and  $\delta^{18}$ O is about -5‰ in the south-west. This decreases northwards and eastwards to reach approximately -8‰ in the east of Scotland (Darling et al. 2003). Oxygen isotope ratios of groundwaters and precipitation below -9‰ and above -5‰ can be considered 'non-local' to the majority of the UK (Evans et al. 2006:268, Chenery et al. 2010:152).

It should also be noted that for skeletal hydroxyapatite, both carbonate and phosphate  $\delta^{18}$ O values are directly related to ingested values, but  $\delta^{18}$ O values are not the same in carbonate and phosphate. They are consistently around 8.6‰

higher in carbonate than in phosphate when both are compared on a VSMOW scale (Bryant et al. 1996:477, Chenery et al. 2012:209). Pollard et al. (2011:499) discuss some of the issues of converting  $\delta^{18}$ O values measured in the phosphate fraction of dental enamel in to the corresponding  $\delta^{18}$ O values in ground or meteoric water. There are several regression equations which have been used to convert  $\delta^{18}$ O from phosphate into  $\delta^{18}$ O values in groundwater. They suggest that the differences between measurement protocols for both  $\delta^{18}$ O of phosphate and  $\delta^{18}$ O of groundwater would benefit from increased standardisation (Ibid. 2011b:499). However, the main focus of their work was on the consequences of the mathematical process used to convert  $\delta^{18}$ O of phosphate into  $\delta^{18}$ O of water. They were particularly interested in the error associated with the predicted value because the total variations in the  $\delta^{18}$ O of groundwater across the UK are quoted as 4.5% (-4.5 to -9% Darling et al. 2003). This means the conversion error must be much smaller than this if useful and accurate places for origin are to be pinpointed. It would appear that the uncertainty in  $\delta^{18}$ O for groundwater is greater than the total range for these values which have been measured across the UK and the western parts of continental Europe (Pollard et al. 2011:503). The reason that the equation of Daux et al. (2008:1143) is used in the current study is that this was found to calculate an estimated value of ±0.73‰ for the uncertainty in  $\delta^{18}$ O of groundwater, which is the lowest error value for all of the conversion equations Pollard et al. examined (Pollard et al. 2011:503). Recent research has used a geostatistical model to predict human skeletal oxygen isotopic values ( $\delta^{18}O_p$ ) in Britain (Pellegrini et al. 2016:1). This model can be employed to study characteristics of local human populations and the mobility of individuals and provides a baseline for comparison without the need for any of the affore mentioned controversial conversion equations from skeletal to water  $\delta^{18}$ O values (Ibid. 2016:6).

# Chapter 4: The Roman Empire

This chapter examines the extent of the Roman Empire and methods of mobility in the Roman world to show the ways in which people could move around the world in Roman times. It also reviews mobility studies on the Roman era in some of the sites from which skeletons were used for isotope analysis in the current research project.

# 4.1 Introduction

It is important for the purposes of a study on the relationship between mobility and infectious disease that the extent of the Roman Empire is understood. The results of the isotope analysis can, after all, only provide indications of likely places of origin for individuals. It is only with knowledge of areas of the world which were and were not under Roman control at the time the people who are the subject of this research were alive that we can understand and narrow down their origins to the most historically likely locations for these individuals. With that in mind, a very brief history of the Roman Empire is now considered.

# 4.2 The extent of the Empire

The Roman Empire at its height in the 2<sup>nd</sup> century AD had a population of approximately 60 million people who lived across an area of around five million square kilometers. This huge empire stretched from Hadrian's Wall, which formed a barrier across northern England, to the banks of the Euphrates river in Syria, and from the Rhine-Danube river system in Europe and on to the North African coast and the Nile valley in Egypt (Figure 4.1). The Empire completely surrounded the Mediterranean Sea (Hekster 2008:ix, Kelly 2006:1).

The Roman Empire reached its full extent after a series of fiercely fought campaigns beginning around the 4<sup>th</sup> century BC when Rome was just a city built

through a network of alliances with populations surrounding it. The first series of successful battles meant that the Romans took over territory along the river Tiber valley and around the Bay of Naples (Mitchell 2007:4, Kelly 2006:4). By the middle of the  $3^{rd}$  century BC, most of the Italian peninsula was under the control of the Romans and, over the next hundred years, the Romans came into conflict with the North African city of Carthage, then the dominant power in the western Mediterranean region. The first of the three Punic wars resulting from this conflict took place between 264 - 241 BC and forced Rome to rapidly develop a permanent navy. This dispute was over the escalating Carthaginian military presence in Sicily, which the Romans saw as a threat. By 241 BC the Romans eventually triumphed and forced a Carthaginian withdrawal from the island (Kelly 2006:5).

The resulting peace that followed the First Punic War lasted only 20 years. The Second Punic War started in 218 BC and lasted until 201 BC and was the result of the Carthaginian general, Hannibal, marching his vast army into Italy from Spain, via southern France and the Alps. This bold venture resulted in Hannibal occupying Italy for 15 years (Kelly 2006:5). Although they avoided pitched battles, the Romans undermined Hannibal by burning crops and starving Hannibal's army out and, in 202 BC, Hannibal was recalled to Carthage to defend the city. Sixty years later, a much-revived Rome won the Third Punic War (149 – 146 BC), which ended with complete destruction of Carthage and enslavement of many of its 50,000 inhabitants (Ibid. 2006:6). By 146 BC, war in the east ensured that all major cities in the Balkan peninsula were ruled by Rome. The following century saw a series of difficult campaigns where Asia Minor was finally secured by the Romans. This took place in a number of stages, with Syria being annexed by the general, Pompey 'the great', and Julius Caesar conquering Gaul from the Pyrenees in southern France up to the Rhine in around AD 50. Caesar's adopted son, Octavian, defeated Cleopatra VII in 31 BC. She was the last independent ruler of Egypt, and this kingdom then fell to Rome (lbid. 2006:6).

The Roman State has proven itself to be a ruthless military power capable of inflicting major damage on its enemies. However there were great rewards to peoples who submitted to Roman authority, for example the Roman model of provincial government which allowed devolution of a signifivcant of power to local elite classes who formed the local authorities. With this power came wealth (Mattingly 2006:7).

Britain was a rather late addition to the Empire with Claudius invading Britain in AD 43. There followed periods of unrest with the Boudican revolt (AD 60 - 61), civil war in the Empire at large and specifically with the Brigantes in Britain (AD 68 - 70). In AD 83, Agricola gained major victory in Scotland, which was short-lived. The Romans withdrew from northern Scotland in AD 87. By AD 117, Hadrian was Emperor and Hadrian's Wall was being built. The Roman Empire had probably reached its full extent by this time.

The full extent of the Roman Empire in AD 117 is shown in Figure 4.1.



Fig. 4.1 A map of the Roman Empire in around AD117. (Donn 2016)

In the 2<sup>nd</sup> century BC, the Romans had to maintain a vast army of around 130,000 men to maintain control of this vast empire that they now ruled. They did this by enlisting around 13% of the young adult male citizens; around 60% of all 17 year olds were conscripted over a period of seven years. This means that over half of all Roman male citizens were expected to serve in the Roman army until their mid-20s (Kelly 2006:10). At the beginning of the Roman Empire, military service lasted for 20 years, followed by five years "under the standards" (*sub vexillis*), which meant that these men were attached to the camp but excused from routine duties. However, by the second century, the military rule was for soldiers to enlist for 25 years of full service (Millar 1981:120) during which time soldiers could be expected to move around to new locations. The majority of migrants within the Roman

Empire, apart from elite groups, are portrayed as people working on the land or serving in the Roman army (Whittaker 2004:202). Later in the second century, conditions improved somewhat for soldiers, and legionaries tended to stay for long periods in the same camps, which were usually built of stone and around which civilian settlements would develop (Millar 1981:121). By the third century, the spread of Roman citizenship had made it possible to recruit legions in the provinces in which they served. Italians now occupied only about one fifth of the military posts whereas in the first and second centuries all soldiers tended to come from Italy (Ibid. 1981:127). Military movement around the Empire would therefore have been diminishing by the time of the later Roman Empire, in the third century (Mitchell 2007:47).

### 4.3 Roman Britain

This section provides a brief history of Britain in the Roman period and examines the effects of Rome on different geographical regions of the islands. Its aim is to provide some background history to the provinces at the time the people in this study were alive. This knowledge may help to further constrain the likely origins of these people based upon their mobility histories.

Caesar invaded the south-east of Britain in two campaigns dating to 55-54 BC, because he thought the islands contained mineral resources worth accessing (Frere 1978:42, Mattingly 2007:64, Wacher 2000:84). However, Caesar did not receive the support in Britain that he had been led to expect, and failed to acquire the expected mineral resources to take to Rome. Instead, the only "booty" which returned with Caesar was in the form of many slaves (Mattingly 2007:67). However, the main Roman conquest of Britain did not take place until AD 43 under the rule of Claudius (Frere 1978:55, Mattingly 2007:47), and therefore Britain was a relatively late addition to the Roman Empire. Nevertheless, parts of Britain did not become part of the Roman Empire in AD 43 and some never did. Rome never incorporated regions facing Britain on the east side of the North Sea, nor was

Ireland part of the Roman Empire (Mattingly 2007:47). While campaigns did take place in Ireland, which was known through diplomatic and trading contacts, these appear to have been insufficient to generate much historical literature (Millet 2005:15). In the north of Britain, Rome never fully subdued a large part of the highlands of Scotland, which resulted in the necessary development of complex land frontiers from the second century AD onwards (Frere 1978:74, Mattingly 2007:24). Under the Governor Agricola (AD 77- 83), and again under Emperor Severus (AD 208-211), campaigns did in fact reach far into Scotland, but full conquest was never achieved (Frere 1978:74, Millet 2005:6). That said, the Romans ruled Britain from the Claudian invasion in AD 43 until around AD 410 (Millet 2005:8-14). Figure 4.2 shows a map of Roman Britain with major towns and military sites. The effects of Rome on the different parts of Britain are now discussed following a general overview of Britain's role in the Roman Empire.



Fig. 4.2 A map of Roman Britain showing major towns. (Mattingly 2006:263)

The incorporation of Britain into the Roman Empire was a very gradual process, as it included regular contact with the Roman world, which commenced in the mid 1<sup>st</sup> century AD. (Mattingly 2007:48). Strabo mentions the exports of corn, cattle, gold, silver, iron, hides, slaves and hunting dogs from Britain. Finds of Gallic and Roman pottery in Britain indicate pre-conquest imports were also being made (Ibid. 2007:48). The population of Britain was in the process of evolving towards having

a more centralised form of social organisation prior to the Roman conquest, and it was arranged into "tribal" groupings which differed in both size and sophistication. These groups came under the leadership of a single chief or king, or occasionally a pair of magistrates (Millet 2005:24). After Caesar's attempts to invade Britain, diplomatic links were maintained with selected British tribal rulers from south-east England; this is supported by archaeological evidence, such as the use of and minting of coins, and an increase in trade between Gaul and Britain following the invasion. The trading ships involved with these exchanges were no longer predominantly using the south-western harbours of Mount Batten in Plymouth Sound or Hengistbury Head in Dorset but were moving towards the use of ports in the new client kingdoms on the Solent, the Essex coast and the Thames estuary (Mattingly 2007:68). Client kings of areas in Britain in contact and collaborating with Rome were controlled and manipulated by Rome in a number of ways, for example in producing levies for the Roman army and paying customs taxes on trade with the Empire. When a client king died, Rome intended to have him or her replaced with a candidate favoured by Rome, although this was not always accepted smoothly by Britons, as seen in the case of the revolt against Rome led by the *Iceni* Queen, Boudicca. For example, Rome attempted to annex the territory of the Iceni people in East Anglia after the death of its client king and Boudicca's husband, Prasutagus (Mattingly 2007:75).

The events in south-east Britain during the period between Caesar and Claudius marked a departure from previous Iron Age traditions, but these occurrences were not mirrored over the rest of the province. Several other regions produced and used coins and also showed patterns of settlement and social change similar to those observed in the south-east (Frere 1978:37). However, some coin-producing areas such as Norfolk, the East Midlands, the Severn/Cotswolds area and Dorset were thought not to have embraced Rome so wholeheartedly, with access to imported goods from the Roman world being lower than in the south east (Mattingly 2007:80). Beyond the coin-producing regions, the situation was different again, with the *Brigantes* and *Parisi* of north-eastern Britain having some access to

Roman and Gallic imports, but being unlikely members of the client network in the pre-conquest years (Mattingly 2007:83).

The activities of the Roman army during the conquest phase had effects on the local population (Mattingly 2007:91-92, Millar 1981:163). Newly conquered people were regularly recruited into the Roman army to serve on other frontiers, with two cavalry units and at least 16 infantry/mixed units being formed in Britain from defeated people during the first century. The names of these units reflect two designations of Britons (Britanni and Brittones) but it is unknown if these names suggest any geographical origins of the soldiers (Mattingly 2007:92). The majority of the regular auxiliary units were posted to the Danube provinces of Pannonia, Moesia, Dalmatia and Dacia. Additionally, a number of other units are known on the German frontier from the second century. Hence, the British auxiliary units required a total of approximately 12,000 men, with extra annual recruitment of between 500 and 750 per year to account for men leaving service or dying (Ibid. 2007:92). However, by the time of the reign of Hadrian, it is likely that some Britons were directed to service within Britain in line with similar situations elsewhere in the Empire, although at this time the majority of legionaries and auxillaries stationed in Britain would have still been recruited from elsewhere. The auxiliary garrison in Britain was by then around 30,000 men in size, which would require a "top up" of 1,200- 1,500 men per year. This was still not on the huge scale of military recruitment in Gaul, Spain, Syria/Palestine and Thrace, but British recruitment to the Roman army was above average when compared to other provinces. Recruits were more likely to have been taken from some areas rather than others (Mattingly 2007:92-93).

The role of Rome and the effects of the Empire in different parts of Britain are now examined in more detail. Knowledge of these areas varies because of differential exposure to the influences of Rome, and is derived largely from literary sources; for convenience and the purposes of discussion, Britain and Ireland have been divided into four main zones; south-east England, Wales, western and northern England, southern and central Scotland, and finally northern Scotland and Ireland.

These are considered separately and in more detail below. South-east England, which was under the influence of Rome from an early date and, as previously mentioned, was in close contact with Rome from the middle of the first century BC eventually formed the core of Roman Britain. Wales and the west and the north of England were less directly affected until after the conquest of AD 43 when they gradually came under the influence of Rome. Periodic campaigns ventured into the central and southern areas of Scotland, but northern and western Scotland and Ireland never came under Rome's control (Millet 2005:23).

#### 4.3.1 Southern and eastern England

Prior to the arrival of the Romans, the social organisation of these regions was largely centred on small clans, several of which had merged into larger tribes. These tribes were under the leadership of a chief or a pair of magistrates. Rapid changes were occurring as tribal territories expanded and alliances shifted, with organisation only becoming formalised after AD 43 when Rome recognised the groups with which she had formed an alliance (Millet 2005:25). It is, however, unknown how far the Roman presence in nearby Gaul had led to the destabilising of the tribes after Caesar's conquest in the 50s BC, but the contact with Gaul and the events occurring in these areas can be traced in the archaeological record using coin evidence. By the first century BC, the tribes under Roman influence started to use coins imported from Gaul. Eventually, these tribes minted their own coins and their presence across any given area has led archaeologists to infer the boundaries of that tribe's territory (Millet 2005:26). The archaeological record also documents the presence of imported Roman objects, indicating that trade with nearby Gaul took place, as well as perhaps with regions further afield in the Roman Empire.

Settlements changed format from the basic pattern of farmsteads and small hamlets consisting of a few clusters of houses, with very few larger settlements in the form of hill-forts, such as those in the Dorset territory of the Durotriges tribe

(Millet 2005:27). However, around the beginning of the first century AD, a number of other types of settlement began to appear. These sites were generally established in river valleys and referred to as oppida. (Latin for "towns"), a misleading term since they were different to towns in other parts of the Roman world (Ibid. 2005:28). The largest oppidum in England was at Colchester, which formed the tribal centre of the Catuvellauni at the time of the Roman conquest. These tribal centres attracted traders from abroad, and they brought material goods that enabled tribal leaders to display their status. Local manufacture also became more closely associated with the oppida and the pattern of settlements and display of material wealth began to more and more closely resemble neighbouring Roman Gaul (Ibid. 2005:29). Archaeological evidence confirms the development of British towns at this time. The first colonia was founded in AD 49 at Colchester, which was known as Camulodunum in the Roman period (Cunliffe 1994:240). The forum of Verulamium was dedicated in AD 81 (Tomlin et al. 2009), and the town wall was completed at the beginning of the third century, enclosing an area of around 200 acres; Lincoln and Gloucester were founded as coloniae by the AD 90s (Ottaway et al. 2012:17).

Another form of town was the *civitas centre*. These are generally understood to be the designated chief town of a distinct group of people (Mattingly 2007:261). Whilst these towns may have been of lower status than *coloniae*, they were sites of local government and administration of customary laws (Ibid. 2007:261). Winchester was an example of a *civitas centre* (see Figure 4.2).

# 4.3.2 Wales, western and northern England

As previously mentioned, these areas were eventually incorporated into the Roman Empire, but this took place at a later date than for south-east of England; they remained marginalised with limited evidence for the urban growth (Millet 2005:29). These areas were in peripheral contact with the Roman world, forming a boundary between the zones the Romans saw as barbarian and the areas of

Roman influence (Ibid. 2005:30). That said, the last 30 years of the first century saw an extension of Roman power into both Wales and the north of England. A legion was established at York, which was probably not developed into a *colonia* until the third century AD (Millar 1981:163-164). The physical geography of these areas tends to be upland in nature, including moors, but there is strong evidence for a relatively large population who lived in a wider range of types of enclosure farmsteads than those found in the south and east. These settlement types continued throughout the Roman occupation with very few villas and small towns developing, and very little evidence for the presence of Roman material goods in civilian settlements. This is in direct contrast to the rich Roman material culture found on military sites in the area, leading to the impression that traditional native systems existed alongside and independently of the Roman military, which Northern England and central and north Wales experienced for a long time; the native people often deliberately rejected the material culture associated with Rome (Millet 2005:30-31).

Northern England and central and northern Wales were occupied by the Roman military for a long period. Rome had great difficulty in controlling the native tribes of Wales and so a large contingent of troops were deployed there to peace-keep and to guard the gold mines until the fourth century (Ibid. 2005:31-32). Meanwhile, northern England remained a frontier zone throughout the Roman occupation, and from AD 120 onwards had a substantial garrison billeted in forts throughout the region and along the frontiers, these varying in size throughout the occupation. The army was based on the frontier itself and also at York and Chester, and demands were made upon the whole area for food, pasture and fuel (Ibid. 2005:32).

### 4.3.3 Southern and central Scotland

The central lowlands of southern Scotland had a similar settlement pattern to that of Wales and northern England prior to the conquest, and they were incorporated

into the Roman Empire by Agricola before being mainly evacuated, leaving just a few forts along communication lines in the region. Four native tribes occupied this area but they did not appear to pose a threat to Rome, and vice versa (Millet 2005:33).

# 4.3.4 Northern Scotland and Ireland

These areas were outside direct Roman intervention save for occasional contact with Roman military and traders. The evidence for trade of diplomatic gifts has been substantiated by finds of a modest amount of Roman material culture, and Christianity reached Ireland in the early fifth century from the Roman empire (Millet 2005:34). It is thought that the presence of the Roman military in the south of Scotland and north of England caused changes in the organisation of the tribes of northern Scotland. Written sources from around the end of the second century describe Caledonian tribes breaching a treaty with Rome and causing revolts in the AD180s, and again around AD 200 (Millar 1981:166); these were eventually quelled by bribes (Millet 2005:35). Diplomatic gifts were then regularly used by Rome to bribe the tribes to keep the peace. Over a prolonged period, this could have led to social centralisation and tribal dependence on Rome, but this stimulus probably had a direct effect on barbarian incursions, which played a significant part in the eventual destruction of Roman Britain (Ibid. 2005:35).

# 4.4 The population of Britain during the Roman occupation

The Roman army peaked in size at around 55,000 and may have dropped down to approximately between 10,000 to 20,000 by the fourth century. If dependents are included, the number of military people could have been as high as 125,000. The combined populations of all Roman towns in Britain can be estimated at around 240,000 (Millet 2005:37). Whilst these numbers are only estimates based upon archaeological remains, excavated burials and written sources from the time, it

can be seen that there were a large number of 'non-native people' in Britain during this period. Observations also show that the pattern of settlement across most of Britain during the Roman period had changed dramatically. This included more and more people settling in towns instead of in rural homesteads, with small hamlets dispersed throughout the countryside. It is this coming together and living in close proximity with people from a wide variety of places, and new contacts through increased trading, that potentially allows (and probably allowed) for the introduction and spread of air-borne infectious diseases. It is therefore understandable why there seems to have been an increase in the numbers of people with tuberculosis in the Roman period compared to the Iron Age (Mays and Taylor 2003, Roberts and Buikstra 2003:132, Taylor et al. 2007:1243), although far fewer Iron Age burials have been excavated in Britain than those from the Roman period due to different "disposal" methods being used for the dead in the Iron Age (Cunliffe 2005:114), thus making human remains less invisible to archaeology. Therefore, just because there is an absence of evidence of Iron Age TB, this cannot be blindly interpreted as evidence of absence of the disease during these times.

It is important to now consider how these non-native populations, described above, might have entered Britain. Therefore, the evidence for the different means for mobility in the Roman Empire are now considered, and in particular in Roman Britain, so that possible modes of movement can be explored in relation to the spread of infectious disease. Essentially, if there are no means to travel then infections would have a harder time establishing themselves in new "countries" and regions of countries.

# 4.5 Mobility in the Roman world

### 4.5.1 Introduction

It is easy to travel long distances around the world today, and indeed to also move shorter distances, quickly and reasonably cheaply; many people move on a daily basis, for example, for work or for leisure. This section examines the practicalities of movement in the Roman period and the drivers for movement, particularly in Roman Britain where the individuals in this study were buried.

Mobility was of great importance in the Roman world. The Roman Conquest, which began in AD 43, incorporated Britain into an Empire comprising Europe, North Africa and the Near and Middle East and resulted in extensive voluntary and forced movement of people (Mattingly 2007:7). Roman migrants included the army, traders and slaves. In Britain, as will be seen in the following sections, there is a legacy of extensive Roman roads, imported pottery, tombstone inscriptions, shipwrecks, and evidence of imported foodstuffs and wines, proving goods and therefore people were moving around on a regular basis. There is published isotope evidence to suggest women were migrating in the Roman era, Eckardt et al. (2009:2821) in their study of Lankhills cemetery in Winchester, found that the three individuals most likely to be immigrants on the basis of their oxygen isotope results, were women.

This section begins by examining the evidence for Roman roads and ports in Britain because they were an important part of the infrastructure that supported mobility of people. It then looks at evidence from the Roman Diaspora project (Eckardt et al. 2010), which is a recent piece of research examining movement and migration in this period and where epigraphic and burial evidence was examined in order to identify incomers to England. Evidence of other material culture to support the presence of migration is then discussed, along with a short section on the evidence for the presence of slaves from other regions of the world. Finally, stable isotopic analysis is considered, perhaps arguably the best tool that
is available to aid bioarchaeologists in identifying immigrants/non-locals buried in a particular area. This is of interest and concern within this chapter as the scientific basis for the current project is based upon isotope analysis.

To expand on the key project mentioned above, one of the major pieces of research into Roman migration in Britain is a recent comprehensive study covering many aspects of evidence for mobility (Eckardt et al. 2010). The identification of incomers to Britain in the archaeological record and their interactions with the local population was the focus. The 'Roman Diasporas' project used a variety of methods in order to examine migration, including stable isotope analysis of human remains, material culture, (eg. pottery types), funerary contextual evidence in the form of grave goods, and also written documents. They suggested that there is scope for future work in examining both military and rural cemeteries, in addition to considering the relative importance of short and long distance movements. This is alongside the impact that "return migration" causes (when people return to their homelands after a period living elsewhere), and the effect that high mobility levels had on the original homelands of these individuals (Eckardt et al. 2010:11). The Roman Diasporas project is thus a baseline for this part of the background chapter because it covers such a range of different types of evidence for the movement of people. However, it does not explore the transport infrastructure used to facilitate the movement of people, such as the position and condition of roads. Hence, firstly, and by way of an introduction to the practicalities of mobility, the Roman road system of firstly the wider Empire and then Britain is examined to understand how this system was developed and why. This is followed by a consideration of the evidence for Roman ports, and thus the evidence of transport of goods and people by water. Finally, the Eckardt et al. (2010) project is considered with reference to all other aspects of the evidence of mobility within, and migration to, Roman Britain.

## 4.5.2 The great roads of the Roman Empire

Outside of Italy, the first great roads built by Rome in the conquered territories all began in Rome. When Augustus took on the title of "superintendent of roads" in 20 BC, he had a column erected in the Forum of Rome representing "mile zero", i.e. the starting point of the entire road system of the Empire (Staccioli 2003:83).

The first road which was built outside of Italy began at *Apollonia,* right in front of Brindisi, and then ran straight from *Dyrrhachium* on the coast at Epirus, through Macedonia, to Thessalonis (Salonika) in Greece. It continued to Philippi and along the Thracian coast (Ibid. 2003:83). From there it travelled along the Hebros River Valley, turning inland and ending at Byzantium (which became Constantinople in AD 330) (Ibid. 2003:86).

In the west, the Via Domitia was built around 120 BC. This road reached from the Alps to the Pyrenees (Ibid. 2003:86). Meanwhile, in the Iberian Peninsula, by the time of Augustus's death in AD 14, Rome has built more than 2,000 miles of roads. This total was later extended to reach almost 7,000 miles (Ibid. 2003:88). In Gaul, the road system was linked and integrated with numerous river routes and underwent a major overhaul after Augustus's journey there in 27 BC. The Via Domitia at Monginevro was the start of a road system which extended along the upper Rhône Valley to Lyon from where two important roads started. One of these ran to modern-day Marseilles whilst the other ran to Bordeaux where it was joined by another road from Nabonne via Toulouse (Ibid. 2003:92). Continuing the Rhône route was another important road that headed north and branched out at Reims. The main branch continued on to Boulogne on the English Channel where it crossed and continued into Britain from the port of *Dubrae* (Dover) (Ibid. 2003:92).

That these roads were well built and maintained is attested to by the documentation of exceptional feats of rapid travel. For example, Ovid mentions a private letter arriving in Rome from Brindisi after nine days, averaging around 59 km per day. Cicero travelled from Rome to *Ameria,* in Umbria by fast carriage,

covering 83 km in ten hours, while Caesar is said to have covered a distance of 1,182 km from Rome to Lake Geneva, on horseback in only eight days, averaging 148 km a day (Ibid. 20013:104)! A map of the main roads of the Roman Empire is shown in Figure 4.3:



Fig. 4.3 Some of the main roads in the Roman era. (Staccioli 2003:83)

# 4.5.3 Roads in Roman Britain

There was an extensive network of roads in use during the Roman occupation of Britain. The following section examines the literature to establish who built these roads and where they were located. Figure 4.4 (following) is an overview map of the position of some of the main roads of Roman Britain, although there were many more minor roads in use, as will be discussed.



*Fig. 4.4 A map of the main Roman roads in Britain in relation to Roman towns and forts.* (Mattingly 2006:264)

## (i) Types of Roman roads

It is without doubt that the Romans built great roads, with direct routes (see Figures 4.3, 4.4 and 4.5). Many of our current roads have been developed on top of and following Roman roads, which proves this point, but also means that

archaeological evidence for the structure and size of these roads has now been destroyed. Disappointingly and perhaps surprisingly, there is little written about the Roman roads of Britain, but the available literature, as it stands, is reviewed. Although generations of school children have been taught that the Roman army built the roads of Britain, this was not completely true for all routes. There were three types of roads (Jones and Mattingly 1990:175):

- Type 1 roads those built by the state,
- Type 2 roads- those built by local government bodies (*coloniae* or civitates)
- Untyped roads those built by smaller communities or individuals purely for local convenience

Type 1 roads were constructed by the army, mainly for military purposes. These were useful for the rapid deployment of the army and for receiving supplies, such as food, although river and sea transport was used in preference to the latter wherever possible. Many of the early military roads radiate outwards from London, highlighting the importance of that city's key strategic and administrative roles (Jones and Mattingly 1990:175, Davies 2002:114). Davies (2002:115) suggests that the types of military roads (which were state-built) can be subdivided into a further three categories, namely:

(a) Penetration roads: these roads were built directly into territory that was being invaded. Forts were then built along them to provide security. Dere Street, which runs up the east of Britain from York into Scotland, is an example of this type of road.

(b) Territory-holding roads: once an area had been secured by the advancing military, these roads were built laterally from the Penetration Roads and were perhaps also supplied with forts. An example of this type of road is the Stanegate, running across the territory of the Brigantes in northern Britain.

(c) Frontier-support roads: these were built to provide access to forts when a formal frontier had been built. For example, the Military Way on Hadrian's Wall supplied this function.

It is not thought that the roads themselves would form a frontier due to the difficulties that would be faced in its defence. The three subdivisions of state-built military roads suggested by Davies (2002) are convincing, however, and roads of these types and functions would appear to be likely. Moving on to Type 2 roads built by local governments, these were much less common than military roads because the *civitates* in Britain lacked the resources to undertake a major road-building operation. However, where they were built, they were thoroughly surveyed and constructed with stone foundations and gravel surfaces. It is the Type 3 road that is the most difficult to distinguish and identify in the archaeological record as they were usually not metalled and tended to follow the more sinuous routes of prehistoric roads (Jones and Mattingly 1990:177). However, it is without doubt that numerous roads such as this will have existed despite knowledge of their routes having long since been lost.



*Fig. 4.5 A Roman road in Britain, photographed in the 21<sup>st</sup> century A.D.* (Romans in Britain 2017).

There are some documentary records for Type 1 and Type 2 roads, although much of their routes have been inferred from the analysis of other information. For example, written in about AD 210 the *Itinerary of Antoninus* gives lists of towns and other places with the distances in Roman miles between them. It has taken some time to equate this information with archaeological evidence but ancient Roman names have been attributed to the principal roads with a high level of confidence (Bagshawe 1990:18), and hence the routes of the main Roman roads are now known. Whilst the Romans documented the routes of their roads in this Antonine Itinerary by providing a series of place names, and distances between them, as discussed above, the names of the roads are not known. The names currently used for British Roman roads were coined much later than the Roman period, possibly having Anglo-Saxon origins (Bagshawe 1990:18). For example, the road from London to St Albans, (referred to by Bede in his 7<sup>th</sup> century AD history as *Waclingaceastre*, and later as *Watlingcaceastre*) was known as *Watlingstrete*, later becoming Watling Street (Davies 2002:23).

Starting from these mostly early military origins, the road network of Roman Britain gradually developed. Some roads will have remained primarily military communication links, while others became essentially civilian on which commercial traffic predominated (Davies 2002:113). What is known about the routes of Roman roads in Britain is now examined. Most of the trunk roads in England, Scotland and Wales owe their origin to Roman surveyors and engineers. This is particularly evident for the modern main routes radiating from London. For example, Edgware Road leading north from Marble Arch follows the line of Roman Watling Street, as does the section nearer the City, which follows Ludgate Hill, Fleet Street and most of the Strand. Additionally, Stane Street begins at London Bridge and follows a course towards the east gate of Chichester. Outside of London, there are similarly well-preserved routes, such as Dere Street, the main road north from York, which follows the line of the old Great North Road (the modern A1) as far as Scotch Corner where it diverges into a minor road running via Piercebridge and through Corbridge. Furthermore, another Roman road branches westwards from Scotch Corner that is now the modern road running through Stainmore Pass on to Carlisle (Wacher 2000:121).

For the purpose of identifying the first phase of development of Roman roads in Britain, particularly military ones of early date, the country was arbitrarily divided into three regions that reflected the various stages of the invasion (Davies 2002:116-125). These were Southern England and the Midlands, Wales, finally Northern England and Scotland. The roads in these three regions are now discussed, following a section on dating.

## (ii) Dating Roman roads

Early tracks and routeways across the British landscape will have followed the most convenient route with no requirement for engineering, and no doubt there will have been many such tracks across Britain when the Romans arrived in AD 43. However, apart from in the centre of Iron Age settlements such as Silchester,

where remains of metalled streets have been excavated (Fulford 2000:545), there is no evidence that Iron Age settlements were linked by anything other than unmetalled trackways (Davies 2002:147-8). The Romans quickly implemented a programme of intensive road-building using sophisticated engineering techniques and taking into account planning of routes to provide efficient and direct links between specified points (Ibid. 2002:17). Archaeological evidence shows Roman Britain had a road network comparable in length with our modern motorway system. Many of these roads were so well constructed that they still survive today (see Figure 4.4), but there is little documentary evidence available to inform us of precisely when the roads were built, how they were planned and surveyed, what sort of traffic used them and for what purposes. However, the knowledge that is available is examined below.

Dating Roman roads is notoriously difficult, and hence establishing whether a road is Roman or not is problematic due to difficulties in dating the road building techniques used (Davies 2002:28). Romans used the technique of "metalling" which was uncommon in the preceding Iron Age and also in the subsequent early and later medieval periods, although there is some evidence of metalled tracks at Danebury Hill Fort in Hampshire which were presumed to be Iron Age but are also difficult to date accurately (Ibid. 2002:27). However, 18<sup>th</sup> and 19<sup>th</sup> century roads were often built in a similar manner to Roman roads and it is possible for archaeologists to confuse the two without having sound dating evidence. Of course, the presence of datable artefacts, such as coins or pottery, excavated from sealed layers of deposits above the road surface, would confirm the road to be of Roman origin. These finds are unfortunately a rare occurrence, and even when dating evidence is sufficiently good to confirm a Roman origin for a particular road, there is often a margin of error to be taken into account (Ibid. 2002:28). For example, at Wall on Watling Street, a number of coins were found beneath the metalling of the road, one of which was attributed to Emperor Nero, who ruled between AD 66-68 (Ibid. 2002:28). The coin was in mint condition and this led to the suggestion that the metalling of the road must have taken place about AD 70

(Gould 2001:24 in Davies 2002:28). However, the amount of time between the minting of the coin, its loss, and the metalling of the road is uncertain.

Despite the disappointing lack of direct dating evidence for the roads themselves, other evidence can be used to assist with dating, such as artefacts from roadside cemeteries and settlements, assuming these are contemporary with the road. It is probably reasonable to assume that a road existed from the time of the development of the settlement and was in use for the duration of its habitation. This method has been particularly useful in dating Watling Street, which links around 26 settlements along its length. Due to dating evidence from these towns and villages, it is assumed there was a road along this route prior to the Roman metalled road (Davies 2002:32).

## 4.5.4 Invasion of Britain and the development of the road system

## (i) Southern England and the Midlands

It is generally accepted that the Roman army, under Claudius, started the invasion of Britain from the place where their ships landed in Rutupiae (Richborough in Kent) in AD 43. From here, they crossed the River Thames and captured the Trinovantes' capital at Colchester (Frere and Fulford 2001). A Roman legionary base and later *colonia* at Camulodunum (modern day Colchester, Essex) could have received supplies by boat via the Thames Estuary as it is thought there was a harbour in use there from the Late Iron Age (Crummy 1997). It therefore seems likely that a Penetration Road would have been built to supply the base, and evidence of this has been found both south and north of the Thames (Davies 2002:118). Other routes in this area include at Spital Street, Dartford, Kent, where a lightly-metalled road was found and interpreted as being an early route for Watling Street. North of the Thames at Old Ford, London, a substantial road was built to carry high volumes of traffic, and from Leicester the western part of Watling Street may have started as a penetration road towards Wroxeter and Wales. Akeman Street formed a penetration road from Verulamium (modern St Albans in Hertfordshire) to Cirencester in Gloucestershire, and the southern section of the Fosse Way may have also been a Penetration Road to the south-west of England. From Silchester in Hampshire, it is thought another Penetration Road may have headed towards Maiden Castle and Dorchester, Dorset, with early territory-holding roads linking Towcester in Northamptonshire, Alchester in Oxfordshire, Silchester in Hampshire and perhaps Chichester in West Sussex, with similar roads being built into East Anglia (Davies 2002:118-119).

#### (ii) Wales

Campaigning in Wales may have started a few years after the initial occupation when Legion XIV, which was active in the North, later moved into a fortress in Wroxeter, Shropshire, between AD 52 and 57, and Legion XX arrived at a base in Gloucester at Kingsholm in AD 49 or 50. This first phase of operations was interrupted by the Boudiccan uprising of AD 60-61, with the second phase taking place between AD 74 and 77; by this time Legion XX was at Wroxeter. Of course, both fort and road building reflect these two phases, with Penetration Roads crossing the River Severn at Gloucester and stretching as far down as Cardiff, and one headed from Wroxeter towards Clyro in Mid-Wales. However, perhaps the most important road for the whole of the Roman period was the border road between Gloucester and Wroxeter, which passed through Leintwardine and was later extended to Chester, Cheshire, in the north and Caerleon, Gwent, in the south. This route linked the major fortresses in the area and is named Watling Street West (Davies 2002:120). During the second phase of operations, roads extended both north and south along the coasts towards Caernarfon in the county of Gwynedd and Carmarthen in Carmarthenshire, and beyond. A west-coast route, now known as Sarn Helen, may have linked north and south Wales, although its central section is in doubt. Excavation evidence suggests that the most important and heavily-used road ran from Caerleon to Usk, also in Gwent. It was thickly metalled, which suggests it was used by heavy traffic over a prolonged period.

Beyond Usk, the road forks, with the right fork forming the route to Monmouth and onwards to the link between Wroxeter and Chester via Watling Street West. The left fork passed Brecon in the county of Powys, and proceeded to Llandovery in Carmarthenshire (Davies 2002:120). When Wales was finally subdued at the start of Agricola's governorship, the road network west of Brecon and Clyro, Powys, would have taken shape with the building of three coastal roads running from Caerleon to Camarthen, Carmarthen to Conway, and along the Dee estuary (Ibid. 2002:122). The busiest roads in Wales were in the eastern part of the province and were probably always dominated by military use, with some civilian traffic particularly near towns like Carmarthen (Ibid. 2002:123).

## (iii) Northern England and Scotland

There are two principal roads which dominate this region, namely those running parallel to the east and west coasts and starting at the legionary fortresses of York and Chester, respectively, before both continuing up into Scotland. Both were built as Penetration Roads to support advances by Legion IX in the east and Legion XX in the west. Dere Street is the name now given to the road running up the east side and this appears to have been the most important in Roman times (Davies 2002:123). Two territory-holding roads link the east and west routes, with one crossing the Pennine Hills between Piercebridge, in County Durham, and Brough in Cumbria. The other links the coasts just to the south of the eventual line of Hadrian's Wall, now called the Stanegate (Ibid. 2002:124). Civilian road-building activity is likely to have had an influence on the road system in the eastern part of Northern England where, on the east side of the Pennines, towns and villas grew along Dere Street and also along the complex of roads near Malton in North Yorkshire (Ibid. 2002:124).

#### 4.5.5 Later developments

The network of penetration and territory-holding roads would have been improved over time, with both major and minor links being added (Davies 2002:125). It must be remembered that the Romans ruled in Britain for almost four centuries, and therefore there was plenty of time for refining and adding roads to the network as requirements for their usage altered, new towns developed and forts were constructed or went out of use.

In summary, the layout and direct nature of Roman roads in Britain suggest an overall pre-determined geometric plan. However, no evidence of such a plan exists and neither does a system of road hierarchies, as we see today (eg. major and minor roads). There was also no adherence to a standard width for roads despite there being some similarities in widths (Davies 2002:141). In terms of construction, heavy bottomed wide roads with paving (metalling) are possibly characteristic of military roads, while slightly narrower roads with lighter, gravel metalling are more likely to have been civilian roads (Ibid. 2002:143). The building of a fully-engineered road system across Britain would take many decades to complete, and therefore the army probably made use of pre-existing Iron Age roads in the early years of the occupation (Ibid. 2002:147). There would have been many of these Iron Age roads but, apart from the centre of Iron Age Silchester where the remains of metalled streets have been found, no evidence has come to light that settlements were linked by anything other than basic tracks. Hence it is more likely that the army made road building a priority, and put in place a network of military roads in the early years of the occupation to give them all-weather communication and supply routes to forts. Proof for this comes from early dating evidence of existing roads, such as the mid-first century date given to Watling Street at Richborough in Kent (Ibid. 2002:148). As previously discussed, there is little dating evidence for most roads. Therefore, whether there was an extensive early network or not, a more substantial system began to be put into place in the late 1<sup>st</sup> to early 2<sup>nd</sup> centuries, with repairs and reconstructions taking place in subsequent years. It is suggested that the rise in size and importance of London,

together with the establishment of forts at Exeter, Gloucester, Wroxeter, Lincoln, and later Caerleon, Chester and York, and those on and around Hadrian's Wall, were also major influences on the routes these roads took. The status of *Londinium* as the largest, most important town in Roman Britain necessitated the building of roads for travel in and out of the city. London dominated the transport network of Roman Britain in much the same manner as it does today (Ibid. 2002:149) in that most roads originated there. London also provides us with almost all of the British evidence available on Roman ports and evidence for transport by the sea and along rivers. This part of the transport infrastructure of Roman Britain is examined in the next section.

## 4.6 Transport via the sea and along rivers

Whilst there is little enough evidence for positive dating of Roman roads, there is even less archaeological evidence for Roman ports and shipping in Britain. There was, for instance, thought to have been a major shipping port based in Dover, although this has not yet been confirmed by excavation. However, London once again provides the majority of the evidence such that inferences can be made as to the structure of ports and quays elsewhere in the province, and also the nature of the vessels that used them. This dominance of evidence provided by London is due to the city being so developed over the centuries and new building developments and demolitions bring archaeological evidence to light. There is historic documentary evidence that Romans used ships for moving the military around and to supply resources: Greek and Roman writers such as Strabo (AD14) and Pliny the Elder (AD 77) described trade routes in many parts of the Roman Empire, whilst large numbers of inscribed altars and tombstones provide evidence of individual sailors, merchants or companies involved in transport, and circumstantial accounts of actual sea voyages occur in the writings of Lucian (AD125 – 180) and St Paul (AD 4 – 62) (Greene 1990:18; Noy 2010:14).

Ships are relatively commonly depicted in Roman art but, with a notable exception from Guernsey (Channel Islands, Figure 4.6), and one from Newport, Gwent in Wales (Keys and Hills 1994), wrecks of Roman sea-going ships are virtually unknown outside of the Mediterranean. However, well-preserved river-boats have been discovered in the silts of rivers that were important trading routes, such as on the Rhine (Greene 1990:18) and at Blackfriars and County Hall in London (Marsden 1994). It can be assumed that Britain, being an island with many navigable rivers, was well served by sea shipping as well as boats using inland routes for supplies and trading. Evidence for ports and harbours in Roman Britain, with the noted exception of London, is scarce. Artificial harbours and guays would not be required for smaller ships, which can be beached and unloaded quickly (Greene 1990:29, Perring1999:19), but excavations in the City of London have revealed magnificent Roman timber waterfronts and guayside buildings as well as the remains of boats and artefacts that help with the study of trading routes. The interpretation of these remains can be compared with, and assisted by, historical data from other river ports such as Lyon in France from which a large collection of inscriptions has been preserved giving details of traders, shippers and organisations of river boatmen (Grenier 1937:479-486 in Green 1990:30).



*Fig.4.6 A Roman cargo ship which caught fire and sank in around AD 280 off the island of Guernsey, Channel Islands. It has been excavated and a model is on display in Guernsey Museum (Guernsey Museum 2016)* 

Meanwhile, river transport must also have been seen as an integral part of trade and communication infrastructure because, due to the presence of navigable river routes in Britain, carrying goods by this method must surely have been easier, quicker and more cost effective than taking them by road. River traffic was sometimes organised by the Roman Army; for example, on the river Tyne, northeast England, supplies for Hadrian's Wall must have been unloaded from seagoing ships onto river boats at the mouth of the river. The fort at South Shields, close to the mouth of the river Tyne, was found to contain many granaries thought to serve as storage for supplies during military campaigns into Scotland (Greene 1990:31). An inscription from the site records an army unit of *barcariorum* (bargemen) who were originally recruited from the river Tigris, Italy (Ibid. 1990:31). Knowledge of the size and structure of these vessels has been advanced by finds of Roman river boats elsewhere in the Roman Empire, for example in the silts of the old course of the river Rhine near the fort of Zwammerdam in the Netherlands, and at Mainz in Germany on the middle Rhine (Ibid. 1990:31). It can be inferred from these findings that similar boats were likely used to supply military bases positioned near navigable waterways in Britain. However, it is unknown how many people manned one of these boats and how cramped their living conditions were with regards the possibilities of transmitting diseases such as TB.



*Fig.4.7* Built for river trade in the 1<sup>St</sup> century, this 102 ft long Roman barge was lifted in 2011 from the Rhône River, Aries, France. It is displayed in the local antiquarian museum. (National Geographic 2014a).



*Fig.4.8 An artist's impression of a Roman river barge, similar to that shown in Fig. 4.7 above.* (National Geographic 2014b).

The archaeological evidence for Roman guays and waterfronts in London will now be explored in more detail, although many of the publications used in the section are now dated, more recent authors still mainly rely upon the older works (eq. Sidell 2008, Wallace 2014). The dating of the origins of London as a city is still unknown, but it is thought not to have existed prior to the arrival of the Romans. Causeways were developed through the boggy ground on the south side of the River Thames, and a bridge was built over the river to found Londinium on the northern banks of the river (Miller et al. 1986:1, Perring 1999:1, Wallace 2014:8). Archaeological work in the City and Southwark areas of London suggest that it was not founded until about the late AD 70s and was largely civilian in character (Wallace 2014:4). This new settlement was mostly destroyed in the Boudiccan uprising of AD 60, after which the city was rebuilt. By the end of the first century, London was one of the most important towns in the new province (Miller et al. 1986:1). London was a city of Roman creation (Perring 1999:3), although the Thames was probably in use as a major route for communication and inland trade in the south during the late Iron Age and early Roman period before the creation of the road network (Wallace 2014:5).

However, during the second half of the 2<sup>nd</sup> century, there were great changes to the nature of Roman London that are not clearly understood (Miller et al. 1986:1) or well documented (Perring 1999:76). On many sites that have been excavated, buildings of the 2<sup>nd</sup> century AD are overlain by "dark earth" up to 1.5m thick, which appears to have been a compost-enriched garden soil (Ibid. 1999:78). This signifies large open spaces that could have been used for agriculture. Some buildings appear to have been deliberately dismantled or taken out of use, as seen in the Roman water frontages. The basic cause of these changes is interpreted as being due to a decline in the population of London and the surrounding areas (Ibid. 1999:88). This may have been caused or exacerbated by imported epidemic diseases, of which TB is one possibility. These diseases would have been particularly virulent, due to being newly encountered by people without immunity to them, and they would have been easily transmittable at this busy, major port of entry to the province (Miller et al. 1986:1). However, this quiet period in the history

of the city was not to last: the years AD 190-250 produced public building works on a grand scale. These included the construction of a city wall over two miles long, and what may have been a religious complex of temples in the southwest of the city. Inscriptions from tombstones in the cemeteries outside the city wall show the incumbents to be government officials rather than rich merchants. Therefore, it has been suggested that in the first half of the third century, London had become an administrative centre (Ibid. 1986:2).

With such a large amount of building work being undertaken in a short time, it is reasonable to suggest that the materials required for such a venture should arrive by river. Fortunately, archaeological evidence supports this suggestion, to a large extent. The evidence was excavated in 1974 and 1975 at the former site of New Fresh Wharf, Lower Thames Street, now the site of St Magnus House. Following the excavations, a watching brief on the wider site took place during 1978 (Ibid. 1986:5). The site lay roughly on the Roman waterfront but the line the waterfront took, and the presence of any quay sites along it, were not known prior to this discovery. However, quay sites were inferred by the discovery of a particularly well-preserved Roman quay a short distance to the east at the Custom House site, and immediately to the south of Thames Street. Thames Street was later found to mark the position of the former waterfront during the Roman period (Milne 1985:8). The excavations at the St Magnus House site uncovered structures from three successive periods:

- The fragmentary traces of a 2<sup>nd</sup> century AD river embankment wall. This showed the limit of land reclamation at the end of the 2<sup>nd</sup> century AD.
- A two-part quay installation dated by dendrochronology to AD 225- 245.
- A riverside city wall running along the back of the quay, which was probably built in the years AD 225-270. It limited the useful life of the quay and indicates the approximate time that the quay went out of use.

In addition to these structural remains, many artefacts were recovered from the area, such as imported red samian ware and black Lezoux vessels (400 of which were found) along with a large number of writing tablets with iron styli (Miller et al. 1986:6). This probably reflects the lively commercial activity of the waterfront area in Roman times. Further attempts to interpret the archaeological evidence for the Roman port of London have been made (Milne 1985), and this is still the most recently published work on the port. He agrees with previous authors that the main Roman harbour was established in the 1<sup>st</sup> century AD on the north bank of the Thames near the present site of London Bridge. The port expanded dramatically until the early third century, but seems to then have contracted and became abandoned by the end of the 3<sup>rd</sup> century (Ibid. 1985:9). The line of the Roman waterfront was also considerably different to its present position, and was still affected by tides in the Roman era (Wallace 2014:76). In fact, most of the Roman harbour lies beneath and to the north of the present Thames Street (Milne 1985:18). The principal Roman discoveries in this area include, on the northern side of Thames Street, the natural riverbank and 1<sup>st</sup> century guay structures at St James Garlickhithe (Dyson and Schofield 1981), Miles Lane (Miller 1982) and on both sides of Pudding Lane (Ibid. 1982; Bateman and Milne 1983, Sidell 2008:66). To the south of Thames Street, substantial sections of 2<sup>nd</sup> and 3<sup>rd</sup> century AD quays were also recorded at the Custom House (Tatton-Brown 1975, Sidell 2008:67), St Magnus House (Miller et al. 1986) and Billingsgate Lorry Park sites (Ibid. 1985:19) the presence of these suggests trade continued well into the 3<sup>rd</sup> century.

In further support of the importance of the Roman port of London, as was previously mentioned, a large Roman ship was discovered beneath County Hall on the south side of the Thames in 1910 (Sloane 2008:16). The ship was constructed entirely of oak and was dated to around AD 300. It was thought to have been built locally following the Mediterranean traditions of Roman shipbuilding, and measured some 13m long by 5.5m wide (Ibid. 2008:16). No indications of possible cargo, number of passengers carried or route of travel of the vessel were recorded but its presence here confirms large vessels were using the Port at the time.

Milne (1985:98) and Perring (1999:90) mention the construction of the City wall of London, built in the late 2<sup>nd</sup> to early 3<sup>rd</sup> centuries which enclosed an area on the north bank of the Thames stretching from what is now the Tower of London to Blackfriars, and from the Barbican to Aldgate. An extensive suburb developed on the opposite bank of the river in Southwark at the southern end of the Roman bridge. The position of this bridge, or bridges, has been subject to considerable debate, but understanding their location would provide the marker for the heart of the Roman harbour and, perhaps more interestingly for this particular study, it would mark the point above which large sea-going vessels could not have passed, and thus where cargoes would have been unloaded, unless the bridge construction allowed arches wide enough for sea-going vessels to pass. Since the Roman river Thames was tidal, and large sea-going ships could not have beached at low tide, large vessels would have had to anchor mid-stream in the deep water channel and have their cargoes off-loaded onto lighter boats to bring the goods ashore (Milne 1985:98). This system would have required a lot of human power. Hence, a large number of people would have come into contact with travelling sailors and therefore stood the risk of contracting any infectious diseases these sailors were carrying. As previously mentioned, there has been little archaeological work done on other Roman ports and harbours around England, but it may be assumed a similar situation was present on other large, navigable rivers of Britain. For instance, pottery from Oxfordshire and Surrey is likely to have been moved via river transport (Perring 1999:86). The rivers must have been busy places where lots of people were arriving and leaving on a daily basis. The opportunities for human-to-human contact and the transmission of infectious diseases such as TB would have been enhanced by these conditions.

Discussions on Roman mobility now moves from the transport infrastructure, providing evidence that large numbers of people were moving around into and within Britain, to looking at more specific examples of individuals having been mobile. This starts by examining the evidence for individual and mass migrations into Britain from elsewhere. Of course, the Romans produced good records and some of these still survive today as writing tablets, for example, those found at Vindolanda in Northumberland (Birley 2007:22, Vindolanda Tablets Online 2016) and those found in Roman London (Perring 1999:47, Tomlin 2016, BBC News 2016), and also as inscriptions on stone. Inscriptions on gravestones are one good place to start looking for this evidence, and Noy (2010) is the key source of recent work on this subject, alongside the previously mentioned Roman Diasporas project (Eckhardt et al 2010). A discussion of his findings follows.

# 4.7 Evidence for immigrants in Roman Britain

# 4.7.1 Epigraphic evidence



*Fig. 4.9 A 1<sup>ST</sup> century AD tombstone from Rome, dedicated to Aurelius Hermia and his wife, Aurelia Philmatium, who were both ex-slaves* (Beard 2012)

Noy (2010:13) suggests that the easiest way to identify an immigrant to Rome is via an epitaph (Figure 4.9) on which the deceased's origin is mentioned. However, there are limitations to this type of evidence. Roman burial monuments sometimes mention the place of birth of the buried person, but poor people did not have elaborate stone monuments erected after their death. In the provinces of Rome this was also the case for people who were not very Romanised. Only 20% of

civilian immigrants to Rome known from tombstone inscriptions were female, and therefore there is also a sex imbalance in people represented by tombstones that are preserved in the archaeological record (and made of durable, inorganic substances). Personal names of the deceased could also indicate their immigrant status, although this must be interpreted with care as people with non-Roman names may have been given this for religious reasons, or because of distant non-Roman ancestors (Noy 2010:16). Thus, using epitaphs to identify immigrants will produce biased conclusions because certain sections of society will be missing, but if this is borne in mind when examining "monumental" evidence, it can still be used to help to provide some information about the immigrant status of people who lived in the area in Roman times.

In spite of the biases in the data, Roman inscriptions on tombstones in Britain often give the place of origin of the interred individual (Cool 2010:27), although the proportions of the inscriptions with places of origin to those without was not stated. This is probably because those individuals who came to Britain who wished to have a tombstone with an epigraph tended to bring their burial traditions with them. The majority of British tombstones with inscriptions were erected by the military with the tradition spreading slowly into the civilian population. A large number of people whose inscriptions do not give any indication of a foreign origin may have been the descendants of immigrants, and who would have likely identified themselves as "Roman" and not "British" (Noy 2010:18-19). It was also very rare for the place of origin of a Briton to be given within Britain and, in almost all cases where this does occur, the individual concerned had military connections. However, sometimes people with other occupations, such as merchants, were described as such on their tombstones or on altars that they had perhaps commissioned (Greene 1990:18). One good example of a Romano-British tombstone erected by the wife of the interred and not the military, was that of procurator C. Julius Alpinus Classicianus, who was the financial administrator of the province and was brought in after the Boudiccan revolt which took place between AD 60- 61. He probably came from northern Gaul. The translation of his inscription reads 'To the spirits of the departed and of Gaius Julius Alpinus

Classicianus (son of Gaius)....Procurator of the Province of Britain. Julia Pacata Iniduta, his wife, had this built.' (Millett 2005:45). Another example comes from the military tombstone of a centurion of the Tungrians found at Vindolanda on Hadrian's Wall; it reads as follows 'T. Ann(ius), killed in the war.' (Birley 2007:74). This inscription does not provide the place of origin of the interred, but it is known that he was a centurion of the First Tungrians, who are known to have been from Georgia, north-east Turkey and Iran.

In conclusion, Noy suggests that the use of epigraphic evidence can be informative about how people viewed themselves and how they wanted to be seen, but care must be taken in interpreting this evidence. This reticence is required because inscriptions are heavily biased towards some groups, such as soldiers, those worshipping a local deity, or involved in overseas trade. The poor, who could not afford inscriptions, women, the non-Romanised who did not follow the tombstone tradition, and newly heavily Romanised people who did not want to provide any trace of their non-Roman origins, are not represented. Further, no mention was made by Noy about the representation of children on tombstones. However, in Roman Britain, immigrants are detected on inscriptions more frequently than they are in Rome. This could be due to the habit of recording the place of birth or origin being most common amongst the military of which there was a heavy presence in Britain (Noy 2010:25). Whilst their interpretation requires care, and obviously omits a sizeable section of society, tombstone inscriptions provide evidence of some immigrants and their origins for certain sectors of the population at the time. This is therefore a valuable source of evidence when exploring mobility in Roman Britain, and it would be made even more valuable by the examination of a skeleton associated with one of these tombstones with an inscription suggesting the individual was an immigrant. If strontium and oxygen analysis could be performed on these remains, it would prove a good test of both types of data.

In summary, despite the biases in epigraphic evidence from tombstones, discussed above, the greatest value is their use in studying mobility histories of

people in the Roman period and in particular providing a place of origin. Regarding the current research, as certain portions of the population are invisible epigraphically, despite there being a considerable number of migrants to Britain, identifying migrants isotopically provides some hard and fast evidence.

#### 4.7.2 Burial evidence

In Section 4.7.1, epigraphic evidence for the origins of interred individuals from tombstone inscriptions was explored. However, the actual burial itself – the treatment and positioning of the body and the placement of grave goods - can also provide evidence of the presence of migrants, for example, at Lankhills, Winchester (Evans et al. 2006), in addition to or in the absence of tombstone inscriptions. In examining styles of burial and types of burial goods, however, care must be taken in interpretation. A good illustration of this requirement is in a study by Prowse et al. (2007), who considered stable isotope evidence for migration into the city of *Portus Romae* in Italy. Bruun (2010:120, and 2016:198) suggested they could have also considered epigraphic and burial style evidence along with the isotopic evidence, but Prowse noted this would have been misleading as the skeletons were all excavated from their tombs in the 1920s, and were then returned haphazardly to random tombs at the end of the excavation (Prowse 2016:217). Ucko (1969:273) also discusses the use of ethnography in assisting with funerary contextual interpretation and indicates how much variability exists both between cultures and within a culture. Therefore, the usefulness of this type of data to identify different origin groups for people must be closely scrutinised. Discussing several ethnographic examples to support this argument, Ucko (1969) further indicates that funerary "monuments" are not always good indicators of different religious beliefs or ethnic groups. However, it is argued here that burial context evidence is worth considering, perhaps alongside isotope analysis (Evans et al. 2006:265, Prowse 2016:230), as long as its limitations are considered in the final interpretation, which has been attempted in the examples that follow.

Migration of people has been inferred when a particular burial ritual appears to have been "transported" from its place of origin, or where burials are identified which are atypical in their local setting, but paralleled in another location (Pearce 2010:79). For example, material culture found in grave contexts in cemeteries can be used to explore ethnicity and to identify immigrants to an area. This is the basis of a study that acknowledged that a population's use of material culture is often not straightforward (Cool 2010:27). Indeed, it may be used to create a persona or identity for the deceased that does not reflect their actual "nationality" or ethnicity. For example, burials in the south of Britain at Bartlow, Cambridgeshire, at Grange Road in Winchester in Hampshire, and at Stansted in Essex were all dated to the 1<sup>st</sup> and 2<sup>nd</sup> centuries AD. They are found in rural areas or on the outskirts of small towns and were richly furnished with a range of "Roman" tablewares, strigils (indicating an aspect of Roman hygiene) and styli, indicating literacy (Ibid. 2010:27).

A contrasting group of burials from the 1<sup>st</sup> century AD were found outside the *coloniae* of Colchester in Essex, Gloucester in Gloucestershire and Lincoln, Lincolnshire, but these were not so lavishly furnished with grave goods. It would be easy to suggest that the first group of burials were those of immigrant Romans, due to the nature of the Roman grave goods. However, it is suggested that they are likely to have been the burials of a local British elite who were giving the image of being "properly Roman". It was concluded that a native elite was drawn from the place where these burials were found, that is in rural areas or outside small towns and civitas-capitals (Cool 2010:27). The second group of burials, found outside the *coloniae* of Colchester, Gloucester and Lincoln were far more likely to have been those of incomers, as was indicated epigraphically by a tombstone inscription of Lucius Octavius Martialis who came from Northern Italy. These people did not feel the need to identify themselves as overtly Roman at death, although their "Romanness" would have been very obvious during their lives (Ibid. 2010:27-28).

Further use of grave goods as evidence of migration into Britain can be found amongst male burials at Winchester, Hampshire buried in the Lankhills cemetery,

where some of the men were buried with distinctive crossbow brooches and beltfittings (Cool 2010:28). A total of 17 of these burials were identified and five were subject to isotope analysis. This showed that four people were incomers from southern or central Europe, with the fifth coming from an area not within the immediate vicinity of Winchester, but possibly from southern Britain. It will be interesting to see what further isotope analysis on the remaining individuals reveal, and will establish if burials with these types of personal ornaments indicate that their owners were incomers or natives of the British Isles. However, crossbow brooches tend be durable and there is evidence to suggest they had long lives. Indeed, two of the Lankhills examples had been repaired (Cool 2010:39), so they could have signified something other than the ethnic origin of the person with whom they were buried. For example, they could have been passed down through families to represent an ancestral origin, or they may have been used as a badge of office. It is suggested here that artefacts representing personal adornment, and grave goods, can be useful in deciding which individuals should be further investigated by means of stable isotope analysis. However, control samples would be needed from individuals who do not immediately stand out in terms of their unusual grave goods, and for immigrants not to remain invisible.

Eckardt et al. (2014) have also more recently explored the movement of people in Roman Britain and the impact of these incomers on the local population. They discussed the issues described above, i.e. using unusual objects and burial rites to indicate the presence of foreigners, and some were re-examined, for example;

- inscriptions and epigraphs cannot be used to represent the whole of the population because not all groups are represented,
- people who have become well assimilated into their host society may not be reflected as immigrants in the archaeological record,
- literary sources are strongly biased in favour of the male elite,
- material culture and burial rite are not direct reflections of ethnicity, but instead they are complex expressions of identity which become modified and developed through interaction and over time,

 isotope analysis has been used to establish whether selected individuals are migrants and to attempt to find where these people may have originated, but many of these studies have selected graves for isotope analysis based on the presence of unusual grave goods or burial rites; this must be viewed as not providing a complete picture of mobility in Roman Britain. As previously mentioned, isotope studies need to be expanded to include "control" skeletons from unremarkable graves.

Published isotope work can be used to distinguish four groups of people (Eckardt et al. 2014:537):

- 1. where both burial rite/grave goods and isotope analysis suggests people were of foreign origin,
- 2. where people appear local archaeologically, but are foreign isotopically,
- 3. where people appear foreign archaeologically but are probably local isotopically,
- 4. people are local archaeologically and isotopically.

A good example of Group 1 is the 'Gloucester Goth' (Hills and Hurst 1989, Evans et al. 2012). This individual was buried with an unusual silver belt fitting that has parallels with similar fittings found in south-east Europe and southern Russia. Isotope analysis supported an origin in these locations. Similarly, some males were buried with unusual belt fittings and crossbow brooches at Lankhills, Winchester. Isotope analysis has shown that they did not originate in Britain (for example skeleton numbers 81 and 426; Evans et al. 2006). It must be noted, however, that even when individuals have similar, unusual burial rites, this does not indicate that they originate from the same area, or that they are all "foreign". An illustration is revealed in the study of burials termed the 'Headless Romans' buried in the cemetery at Driffield Terrace in York. As individuals from this cemetery are included in this study, the site is detailed in Chapter 5 but, in brief, this was an exclusively male cemetery where the majority of skeletons had been beheaded and/or had signs of ante- or peri-mortem trauma. A number of these people were found to be isotopically non-local. However, they appear to originate from a number of very different places, which is one of the reasons that this cemetery has attracted so much interest both archaeologically and in the media (Montgomery et al. 2011, Müldner et al. 2011).

An example of the Group 2 of Eckhardt et al. are two individuals from Lankhills, Winchester (0271, a female, and 0281, a male) who, from their burial rites, appear to be local. However, isotopic data showed that they were incomers to the area (Evans et al. 2006). Group 3 is slightly problematic because isotope analysis is not suited to identifying locals. This is because people whose isotopic "signatures" are consistent with an upbringing within a similar climate and geology to the place where they were buried cannot be excluded. Therefore, many parts of England and Wales are isotopically indistinguishable from large areas of western Europe and the Mediterranean, discussed in relation to the interpretation of isotopic results (Chenery et al. 2011, Evans et al. 2012). Most isotope analysts follow Occam's Razor which states that the simplest explanation is usually the best one and that most individuals who appear to be isotopically local probably are local (see Chapter 8; the current author also makes these assumptions). Group 3 people, with mismatches between "foreign" material goods and probably local isotope signatures, could be local people who adopted the use of imported, foreign artefacts, or they could be second generation migrants, or even return migrants, that is people who served or travelled abroad but who returned home in later life and/or to be buried (Eckardt et al. 2014:538). Isotopic studies of Group 4 individuals, as has been previously mentioned, have so far focused mainly on unusual burials, and therefore a shift in sampling strategy is required to ensure isotope data sets are more representative of Roman provincial populations as a whole (Echardt et al. 2014:540).

#### 4.7.3 Forced migration via slavery

The use of slaves is well documented in the Roman era, with enslaved individuals collected from, and sold, all over the Roman Empire (Fitzpatrick 1989; Tomlin 2003; Webster 2010). In this respect, they were migrants, albeit against their will. However, as the current research is concerned with the transmission of TB due to mobility, forced migration also must be considered as a method of transmitting infection and hence this has been included within this chapter.

Some research has focused on identifying the ethnic origin of enslaved individuals and recognising possible "identification strategies" for Roman slaves (Webster 2010), and ancient authors have documented much information on the sources for, and trade in, Roman slaves. However, as slaves were frequently taken as rewards for victory in battle, and ancient historical writers were usually the victors or someone "on their side", the actual numbers of slaves recorded could have been prone to exaggeration. For example, Gracchus reportedly enslaved 80,000 Sardinians in his 177 BC campaign, Caesar reported enslaving one million people in his Gallic Wars, and Trajan is said to have enslaved 500,000 Dacians in AD 105/106 (Webster 2010:48). Whilst these numbers need to be interpreted with care, enslavements undoubtedly did occur and would have been responsible for forced migration of people, but how easy is it to find archaeological evidence for the presence and origins of these slaves? Slaves in the Roman world were given their personal names, either by the seller or the new owner. As is the case today, fashion determined which names were popular, but there appeared to be a general preference for Greek slave names, although in Rome Latin names were preferred. However, these names bore no resemblance to names given to people in their place of origin (Ibid. 2010:48). A good example of this comes from the first Roman deed of the sale of a slave to have been found in Britain. It is in the form of a writing tablet discovered in London at No. 1 Poultry in 1994 and records a slave girl called Fortunata, a typical Latin slave-name. However, in the document it is stated this girl came from Jublains in north-western Gaul (Tomlin 2003).

Nevertheless, in Roman Britain slaves did not have to come from distant shores; some originated in Britain itself. For example, Noy (2010:18) describes a funerary monument from South Shields, north-east England close to Newcastle, on Hadrian's Wall. The monument was commissioned by Barates of Palmyra in memory of his wife, Regina. Barates was Syrian born, but his wife was a former slave of Catuvellaunian origin. She may have been enslaved at the time of the Roman conquest, but orphans, children and individuals sold by families to alleviate poverty and people enslaved as a punishment, could account for slaves being taken from within Britain itself. It has even been suggested that a slave trade existed in Britain before the Roman conquest and that this trade was used to supply the British Roman slave market (Fitzpatrick 1989).

What of material culture being used to identify slaves and possible immigrants? This is more difficult, and there are very few certain examples of slave-made or slave-used material culture in the Roman world (Webster 2010:59). However, Webster suggests that there are many artefacts inscribed with common "slave-names" which could be interpreted as representing the presence of slaves in that particular place, and the use of graffiti attributed to slaves because of its context is mentioned specifically. However, little work has yet been done on graffiti and its use in tracing the slaves of the Roman world (Ibid. 2010:59).

In summary, that there were considerable numbers of slaves moving round the Roman Empire is without doubt, but evidence of where they originated and where they went and were eventually buried is limited in the archaeological record. In order to find this group of migrants, reliance is placed on a scant amount of epigraphy on tombstones of freed individuals (see Figure 4.9, which shows a tombstone of a man and wife who were freed slaves), because there is no material culture deemed specific to slaves that would identify their presence. That said, in the Driffield Terrace, York, cemetery sampled in the current project, one individual (not sampled in the current project) was found buried with heavy iron rings around his lower legs, possibly for the purposes of restricting movement (see Figure 4.10). The excavators stated that the precise nature and function of these rings is still to

be determined (Müldner et al. 2011:282), but they could be interpreted as being slave leg shackles, thus providing evidence of at least one slave buried within a cemetery from which skeletons were examined for the current study. Enslaved individuals were possibly placed under great stress, and were perhaps not well cared for. This may have lowered immunity and resistance to infections such as TB, which tends to occur when the immune system is compromised (Harries and Zachariah 2008:315, Pozniak 2008:343). Therefore, slaves as a category of migrants must also be considered as a reservoir for infection.



Fig. 4.10 Radiographs of the leg rings from Driffield Terrace (Cool 2015)

# 4.7.4 Immigration and material culture in Roman Britain

The extent to which material culture can identify ethnic groups and migrants has long been a subject of debate in archaeology; we have looked at burial evidences and their role in identifying immigrants, and now this section will examine some of the evidence useful to help in the identification of non-locals by means of their associated material culture.

The process of conquest and Romanisation of Britain did not always involve the spread of traditions pertaining to Rome, or even to Italy as a whole. More often it was the native traditions of soldiers from various locations in the Empire that were imported to new areas, for example their cooking traditions (Swan 2009:15). By the 1<sup>st</sup> century AD migration into Britain can be identified due to the influx of the Roman military. These immigrant legionaries predominantly came from around the Mediterranean, and auxiliary regiments came from the more recently conquered provinces; these consisted mainly of areas of Europe stretching from the Atlantic to the Black Sea. In addition, merchants, craftsmen and slaves associated with the military can be added to this influx (James 2001). The total numbers of immigrants to Britain in the early post-AD 43 years may well have exceeded 100,000, with the military accounting for at least 40,000 (Fulford 2010:68). Of course, this increase in immigrants led to a mass of material culture of various types, and food, being imported with them. As an example, the spread and usage of the tripod-vessel, a specialised style of cooking pot has been studied. These vessels first appeared in about the early/mid-1<sup>st</sup> century BC in the Alpine regions of central-southern Noricum (eastern Austria) and central-western Pannonia (western Hungary) (Swan 2009:15). Tripod vessels were synonymous with a particular style of cookery, and Swan suggests that an influx of them into a new area would imply the arrival of a number of people who were familiar with their use (Ibid. 2009:26). Of the tripodvessels recorded in Roman Britain, the majority occur at military sites or at kiln sites supplying the military. They have also been recorded at *coloniae* or former forts, where veterans and their families may have settled and continued their old cooking traditions. Smaller numbers of these vessels have been also found at ports where there were probably more cosmopolitan populations of traders and sailors than in the surrounding districts (Ibid. 2009:31). The past 20 years or so have seen significant advances in knowledge of regional coarse wares in Gaul. As a result it is now possible to determine, from the distinctive character and style of
tripod-vessels found in Britain, whether they are from Gaul and even which part of Gaul this may be (Ibid. 2009:35). Hence, from ceramic evidence here in Britain, it can be concluded that the study of the tripod-vessel is useful for identifying the presence of migrants (mainly soldiers) from different parts of Gaul (Ibid. 2009:51).

Prior to the Roman conquest of Britain in AD 43, there had been some imports of pottery from the Mediterranean provinces and north-west Gaul, but these imports greatly increased after the conquest to meet the needs of the Roman Army, in the first instance (Fulford 2010:68). However, little evidence could be found of research into the types of imported pottery being exclusively linked to a particular ethnic group, the research into tripod-vessels by Swan (2009) being a notable exception. Instead, information on situations such as at La Graufesengue (near Millau, Aveyron in France, located at the confluence of the Tarn and Dourbie rivers) are found, where tablewares were exported to the complete range of army units stationed in Britain, regardless of their place of origin. Military units on the Rhine and upper Danube were also supplied with the same goods, described as a sort of regulation army issue of crockery (Fulford 2010:69). As would be expected due to speed of delivery and reduced expense, a significant amount of Roman military cookware was also sourced from local suppliers. A good example of this can be seen in the fortresses at Kingsholm, Gloucester (Hurst 1985) and Usk in south-east Wales (Manning 1993 and 1995), where the majority of legionaries originated from Italy (Narbonensis and Hispania). However, only a small percentage of the ceramic assemblages originated from the Mediterranean provinces (Greene 1979 and 1993). Indeed, most of the cooking wares came from local native producers, such as the Durotriges in southern England (Fulford 2010:68).

This native-produced Roman pottery was made at, or near, many towns in Britain, although its manufacture was predominantly a rural industry. However, as previously mentioned, a significant quantity was imported from elsewhere in the Empire. For example, there is evidence for shops in London selling samianware imported from Gaul and which dated from the 1<sup>st</sup> century AD (Hill and Rowsome

2011). A further example can be found in *Albona*, which was was a small town at Sea Mills, a suburb of modern-day Bristol. Pottery found in Albona came from the nearby kilns of Gloucestershire, Gwent, Somerset and Wiltshire and also from the New Forest, Oxfordshire, Northamptonshire and Dorset, along with imported Samian ware from South, Central and Eastern Gaul, and amphorae from Spain. The *amphorae* were imported for their contents (wine, olive oil, fruit or fish sauce) and later recycled as containers for other purposes (Bennett 1988:25). Of course, this does not indicate people living in *Albona* were from Gaul and Spain, although this could have been the case, it does indicate that travelling salespeople, temporary migrants, must have brought the pottery from those areas, but some of these people may have eventually become permanent immigrants by marrying locally and settling in Britain. There is also evidence of these "travelling salesmen" from *Pompeii*, Italy, where a crate of imported samian ware and oil lamps indicated that the pottery was probably exported in "job lots" by intermediaries and not directly from the makers themselves. Furthermore, in terms of evidence of travelling sellers in Roman Britain, the Bloomberg writing tablets record the names of a number of traders who were working in Londinium during the 1<sup>st</sup> century AD (Tomlin 2016).

Closer to Britain, the Roman Puddling Pan Ship Wreck, excavated from the Thames estuary, was seemingly bringing samian ware made by a number of different manufacturers in Gaul into Britain (Bennett 1988:25, de la Bedoyere 2000:16). Again, the crew of this ship was probably not comprised of native Britons and therefore these would have been temporary migrants to the province. That said, little is generally known about how pottery manufacture and its transportation were organised, although it is thought that traders specialised in military or civilian contracts (de la Bedoyere 2000:15). Once pottery reached Britain, it will have been sold on to wholesalers at British ports or by auction to local distributors. Evidence for this comes from Wroxeter in Shropshire where a disastrous fire had destroyed shops and stalls in the late 2<sup>nd</sup> century AD. The remnants of saleable goods were excavated and one trader was found to have been selling Gaulish samian alongside Romano-British mortaria and whetstones.

The samian contained name stamps that showed that the pottery had come from a number of different workshops (de la Bedoyere 2000:16).

In terms of military demand, British-made pottery would have been moved for considerable distances within Britain itself in order to supply the Roman army and associated markets. In the 2<sup>nd</sup> century AD, military positions on the northern frontier of Britain, for example forts along Hadrian's Wall, relied on Black-Burnished kitchen wares from southwest Britain and the Thames estuary. It is likely that these Black-Burnished wares were shipped directly from their place of production to the north, possibly along with other goods for use on the military bases. During the 4<sup>th</sup> century AD, Alice Holt kitchen wares dominated south-east Britain with a scattering found as far west as the Mendips, the Cotswolds and south Wales (de la Bedoyere 2000:51). These wares were more markedly found in areas accessible by river from the kilns and therefore they were probably also transported by boat (Ibid. 2000:17). In addition to the transportation of tableware, both military and civilian Roman populations used olive oil as fuel for lighting ceramic lamps. This oil was supplied from the Mediterranean olive oil industries and shipped to Britain in amphorae. Lamps were imported from Italy, Gaul, Germany and North Africa, but some were also made in Britain, for example in Colchester (Verulamium Region white ware) and in London (Ibid. 2000:54). People trading oil and lamps were obviously moving regularly between provinces and regions.

The movements of individuals involved in the selling and distribution of ceramics and associated goods have now been considered, but it must be remembered that the craftsmen themselves would have moved around the province in order to ply their trades. The movements of some potters into Britain can be traced by looking at potters' marks inscribed into goods they made. This influx of craftsmen was particularly common from the post-conquest period up to the late 1<sup>st</sup> century AD (Fulford 2010:77). However, it is doubtful that the migrant potters can be associated with particular ethnic groups and certain shapes and styles of pottery, because the pottery use could have been adopted by "locals". However, Swan

(2009) did attempt to prove the presence of immigrants with some styles of cooking vessels. She suggested that the tripod-vessel indicated the import of not only a unique shaped vessel, but also the associated cooking style, which would not have happened without the people accompanying it. She viewed the presence of tripod vessels in an area as indicative of the presence of the specific ethnic groups who used those vessels in their native cookery. Nevertheless, it could be argued that the style of cooking was considered somewhat exotic and was thus adopted by native individuals; this is analogous to people in Britain today following fashion and using Moroccan tagines to cook Moroccan-inspired dishes (BBC 2016). Pottery did not enter Britain on its own; it was carried here on ships and transported round the province via waterways and roads. Travelling salespeople will have accompanied it on its journey and these temporary migrants are as important as permanent settlers from abroad in our understanding of population movements during the Roman period. The individuals and groups of people responsible for the movement of pottery into and around Britain likely also brought infectious diseases into the province with them, thus putting at risk individuals with whom they came into contact.

# 4.8 Scientific approaches in the search for immigrants: analysing human remains from archaeological sites.

## 4.8.1 Introduction

The following section considers the direct evidence for mobility of people by analysing their skeletal remains rather than looking at the material culture and structures that they left behind. For example, the more recent use of skeletal remains to establish ethnicity, namely craniometric analysis of the skull, has seen some attention, as has the analysis of both mitochondrial DNA (mtDNA) and Y chromosome DNA to identify the ancestry of individuals. These methods will be examined briefly. Detailed information on the methods and their use is beyond the scope of this study, but it would have been interesting to utilise them to provide a fuller picture of the people with TB than isotope analysis alone can do.

Eckardt et al. (2010) looked at diaspora communities in Roman Britain. As part of this research, they examined 155 skeletons. These were dated to the later Roman period and were from urban cemeteries from different parts of England, namely York, Catterick, Poundbury and Winchester. York was chosen because it was a major military and urban center in the Roman period. Catterick was a northern military site where an unusual "eunuch" burial was located (Cool 2002:41-42), and this individual displayed an unusual gender and religious identity. At Gloucester, a mass burial pit and a surrounding inhumation cemetery were sampled. In previous work at the later Roman cemetery at Poundbury Camp near Dorchester, Molleson et al. (1986) a child migrant from Greece had been identified on the basis of lead isotope ratios, which was supported by stable isotope data (Richards et al. 1998:1251). Eckardt et al. did not attempt isotope analysis on the Poundbury skeletons, but, through the osteological examinations of 364 children, high levels of malnutrition and trauma were found (rickets and scurvy, and rib fractures, respectively). These findings could indicate a poor standard of living and possibly "foreign" food, fasting and weaning practices (Eckardt 2010:111) which could predispose these individuals to infections such as TB. One individual appeared to have suffered from *thalassaemia intermedia* which is a genetic condition characteristic of Mediterranean populations (Musallam et al. 2012:1), hence this child or his/her parents were probably immigrants from the Mediterranean regions. The final site to be sampled for Eckardt et al.'s work was the later Roman cemetery at Lankhills, Winchester. Incomers to this cemetery have been identified previously from interpretation of both grave goods and burial rites (Clarke 1979).

The work to establish if people were likely to be immigrants to these sites utilised a range of techniques, which helped to provide a fuller picture than using one method alone. For example, osteological and forensic techniques were used to assess ancestry in the individuals from York. Just over 200 skulls were examined using craniometry, which quantifies cranial characteristics on an objective scale

and compares them to reference populations from around the world using specialist software, FORDISC 3.0, in order to assess biological ancestry (Leach et al. 2009). Eckardt et al acknowledged that two issues must be addressed when using these methods of ancestry assessment: firstly, the rejection of craniometry by some archaeologists because of potentially racist associations, and secondly, methodological problems associated with such an analysis. However, while "race" is now acknowledged to be a social construct without a basis in biology, they believed that there are definite 'geographically localised aspects of human variability' (Eckardt 2010:111). As we cannot know how ethnicity and "race" were viewed in Roman Britain, and if racism of any form existed or not, if 'geographically localised aspects of human variability' do exist, it will be more helpful to identify these as population affinities rather than definite groupings. Another issue with the use of craniometric software is the comparative reference collections that have been used in the database. These skeletal collections are described as 'early modern reference collections' by Eckardt et al. (2010:112), but do the same geographical variations exist in these collections as was present many years ago, in the late Roman period? Interpretation of the data therefore relies upon understanding that a similarity to a particular reference collection suggests only the closest physical affinity of an individual to that population, rather than a definite direct biological relationship. Misclassifications of ancestry have been reported using FORDISC 3.1, when Hispanic ancestries are found to often classify as Japanese (Dudzik and Jantz 2016:1311) providing recent proof that this method is not without its limitations so results from using the technique must be interpreted with care.

The results from the York study were reported as falling into simplified categories such as "mixed race", "black" and "white" (Leach et al. 2009). This can only suggest, from morphological observations and measurements of the skull, that characteristics could possibly classify an individual into a broad ethnicity category. Used alone, craniometric analysis obviously would be of little value in identifying migrants. However, if access to the whole skeleton is possible and crania are well-

preserved, it is a method which can be used in conjunction with isotope analysis to explore mobility.

Another scientific method which can be used to provide information about migrants is the analysis of mitochondrial DNA (mtDNA) (Giles et al. 1980:6175, Zouros et al. 1994:818). Mitochondria are organelles found within cells and, as each cell contains many mitochondria, multiple copies of mtDNA are present compared to the one copy of nuclear DNA contained in the nucleus of each cell. Because mtDNA occurs in higher copy numbers per cell, it has a better chance than nuclear DNA of being preserved in the archaeological record (Prowse et al. 2010:181).

mtDNA is inherited down the maternal line and therefore its examination allows for tracing the female maternal lineage for any individual, whether they are male or female. Changes in the sequence of DNA (polymorphisms) occurring as a result of random mutations can help to identify the mtDNA signature (haplotype) of an individual. Specific sets of polymorphisms occurring together identify different haplogroups. Due to analysis of many thousands of mtDNA sequences from individuals all over the world, a map has been built up of the origins and migration patterns of different human haplogroups. Hence the analysis of an individual's mtDNA will identify the geographical origins of his or her maternal ancestry (Giles et al. 1980:6715, Zouros et al. 1994:818, Prowse et al. 2010:182).

In addition to mtDNA analysis, male humans have XY as their sex chromosomes, while females have XX. The Y chromosome has a single copy within the nucleus of each male cell and it is a small chromosome. However, it has recently been used alongside mtDNA to try to establish migration routes of the paternal line. Obviously, because only males have Y chromosomes and both females and males have mtDNA, analysing the latter is the most commonly used method of analysis and therefore more comparative data exist (Pinhasi et al. 2012:502). However, the introduction of Next Generation Sequencing (NGS) techniques has led to more accurate studies of Y chromosomal DNA, and several of these studies have taken place over recent years, between Francalacci et al (2013) and Poznik et al. (2013)

and 2016). Although it is generally accepted humans originated around 200,000 years ago in sub-Saharan Africa (Jobling et al. 2014), the dispersal routes towards Asia and Europe are still in debate (Nielson et al. 2017). Current ancient Y chromosome data is limited (Kivisild 2017), but this is expected to increase and so over the next decade, much is to be learned about male-mediated expansion (Batini and Jobling 2017).

Oppenheimer's research into the origins of the British found that detailed Y chromosome data were not available for British males and he tried to address this by analysing the Y chromosomes of over 3000 men from Britain and Continental Europe (Oppenheimer 2007:139). This research provided information about human movements in the past, after the last Ice Age, around 16,500 years ago, and up to around the Bronze Age, between 2,700 and 2,500 years ago. In summary, the problems with the use of mtDNA and Y chromosome DNA analysis are that data can tell us about ancient geographical origins, but not whether the individual in question is a recent migrant to the area. However, if used in conjunction with craniometric and isotope analysis, the technique could shed some light on the ethnic origins of people within an area. This would be of limited use in the current research, and therefore studies using isotope analysis to identify immigrants to Roman Britain are now be examined.

Recent stable isotope analysis of human remains from Roman contexts in Britain has also helped to understand the mobility of these people. There have been several studies undertaken, on skeletons from English sites discussed in the following sections. They include, for example, Leach et al. (2009, 2010) who examined evidence for an immigrant population to Roman York, including the 'Ivory Bangle Lady' who is discussed in more detail in the following section. Evans et al. (2006) researched a possible 4th century immigrant population in Hampshire, Chenery et al. (2011) found a cosmopolitan population in Catterick, North Yorkshire, Eckardt et al. (2009) discovered evidence of mobility in Roman Winchester and Chenery et al. (2011) used isotope analysis to discover origins of the York 'Headless Romans' (discussed in detail following this section), and Montgomery et al. (2010) used lead isotope analysis to identify the origins of a woman from Rome, buried in London. More recent use of isotopes to study mobility in Roman Britain include the work of Redfern et al. (2016), who used analysis of ancestry, mobility and diet to identify immigrants in Roman Southwark, London, and Shaw et al. (2016) who identified migrants in Roman London using lead and strontium stable isotopes.

The following sections review published isotope analysis literature pertaining to the sites used in this study. These have been included for comparative purposes for the skeletons analysed in the current project. (Figure 6.1 is a map of England showing the location of all the sites in the study, for reference).

## 4.8.2 Site 1:York

As previously mentioned, stable isotope analysis was recently utilised in an archaeological mobility study examining an exotic burial of a woman in York known as the 'Ivory Bangle Lady' (due to the nature of some of her grave goods). The information about this individual and her place of origin is now explored in more detail. The Roman conquest of Britain, as previously discussed, resulted in extensive movement of people, both forced and voluntary (Mattingly 2007). In newly conquered Britain, *Eboracum* (York), north-east England (founded in around AD 71) was both a legionary fortress and a civilian settlement. It functioned as one of the provincial capitals for much of the later Roman period (Ottaway 2004). The civilian settlement would have been largely made up of wives and families of soldiers serving in the fortress (Allason-Jones 2005:45). Many soldiers, upon discharge from their military duties, and in their retirement, would have continued to live in the settlement with their families.

Along with this military influence, there were documented visits from the Emperor Septimius Severus who was born in Tripolitania and resided in York between AD 208-211, and later from Constantius I and Constantine the Great, who both visited York in AD 306 (Hartley et al. 2006). These visits would have provided opportunities for probably temporary migration of their slaves, assistants and associates into York and for the city to become a diverse, multicultural society; it appears that this did indeed occur.

In August 1901, a stone coffin was discovered near Sycamore Terrace, Bootham, north of the River Ouse and south west of the legionary fortress. The coffin was aligned approximately north-south with the head to the north. The skeleton inside was that of a woman who lay supine (Boynton 1902:104 in Leach et al. 2010:132). She was accompanied by a number of exotic personal grave goods the exact position of which was never recorded, but which were recently reviewed by Cool (2006). These objects were dated to the second half of the 4<sup>th</sup> century and they included jet and elephant ivory bracelets, earrings, pendants, beads, a blue glass jug and a mirror (Cool 2006). The results of craniometric multivariate analysis of this woman suggests greatest affinity with two reference populations of African-American females from the late 19<sup>th</sup> /early 20<sup>th</sup> century Terry and Hamann-Todd documented skeletal reference collections in the USA. The suggestion of mixed ancestry is also supported by the results of anthroposcopic assessment of morphological traits that showed both "black" and "white" traits (Leach et al. 2010:137). The woman's skull exhibited a low, wide and broad nasal ridge and wide inter-orbital breadth suggestive of "black" ancestry, while the nasal spine and nasal border demonstrated "white" characteristics with the shape of the nasal aperture being inconclusive (Ibid. 2010:135). The data did not show concordance with a particular ethnic group or place of origin, but they did suggest that the York woman's features most closely resembled individuals from these ethnic groups.

Another limitation of comparing the Roman woman's features with individuals who lived around 1800 years after her, is that genetic mutations, normal genetic variation and environmental impacts due to diet and lifestyle, could have lead to changes in the shape of the skull, making these reference features inappropriate for comparison. However, these are the only reference material available and

hence the only comparison that could be made as craniometric data from contemporary Roman Britain and around the Empire are lacking; as such, this limitation needs to be noted. In order to confirm whether this woman was indeed a long-distance migrant to York, oxygen and strontium isotope analysis was performed on her teeth.

The enamel of a second premolar was used for oxygen and strontium analysis because this tooth forms at approximately three to six years of age and therefore after weaning (Smith 1991 in Leach et al. 2010:138); thus, the breast-feeding effect is avoided, which would provide information about the mobility of the mother of the person and not that of the woman herself. The strontium isotope ratio was found to lie within the range defined for the York area, but at 0.7094 it is very close to the <sup>87</sup>Sr/<sup>86</sup>Sr of modern seawater, which is 0.7090. This therefore would also suggest an origin in a number of other places, including most Mesozoic (245 to 65 million years ago) aged terrains as well as coastal areas. However, her  $\delta$  <sup>18</sup>O<sub>dw</sub> (drinking water) ratio was at the very high end of the British drinking water range, making it unlikely that she spent her childhood in the York area. It was concluded that she probably spent her childhood in the west of Britain or, more likely, in the warmer coastal areas of Western Europe and the Mediterranean (Leach et al. 2010:139). In this case, the use of stable isotope analysis showed that the individual was an incomer to York. Craniometric analysis also suggested that she was of mixed ancestral origin, although that tells us more about her ancestry (that is, her line of descent) than her origins and needs to be interpreted with care. This is because differences in the morphology and expression of cranial features (and measurements of the skull) can be very variable for all ethnic groups. However, in the case of this woman, the isotope data support the craniometric data in suggesting that this individual was an immigrant to Britain from warmer climes.

Other related studies have also been undertaken in Roman York. A second, larger scale project looked at burials from the Railway and the Trentholme Drive areas (Leach et al. 2009). This study developed after an earlier examination of the Trentholme Drive burials by Warwick (1968:157), who noted considerable diversity

in the morphology of the male crania excavated. He suggested this variation indicated the presence of "non-locals" and considered the skulls to belong to people who originated in the Middle East or North Africa. The Leach et al. (2009) study reconsidered these data through cranial variation analysis and via isotopic data of the dentitions of 43 individuals from these two burial grounds. The Trentholme Drive "population" represented a small section of the larger cemetery site known as The Mount. Approximately 350 inhumations and 53 cremations were excavated from a densely packed burial ground that was in use from approximately AD 140 until the last decades of the 4<sup>th</sup> century AD. The burials were haphazard in distribution in the cemetery and there were no tombstones and few grave goods; this was therefore interpreted as being a burial ground for individuals of lower social status compared to those buried at The Railway (Ibid. 2009:547).

The Railway site was excavated in the 1870s and the exact number of burials was never recorded. The burial site was in use between the early 2<sup>nd</sup> to 4<sup>th</sup> centuries AD and was more organised than Trentholme Drive, with less crowding and intercutting and many people buried in coffins, with jet jewellery being a popular grave accompaniment (Ibid. 2009:547). A total of 50 individuals were sampled for isotope analysis and, of these, 29 were from The Railway and 14 from Trentholme Drive. In order to avoid the effects of breast-feeding (which raises oxygen isotope ratios – Wright and Schwarcz 1998:1), but to still obtain data representing early childhood, permanent second molars or premolars were used for analysis (Leach et al. 2009:550). These represent an age of crown mineralisation of between three and seven years. In the absence of these teeth, first or third permanent molars were used instead, which represent crown formation from birth to three years, and nine to 13 years, respectively (Hillson 1996:123). These 50 individuals displayed a wide range of oxygen and strontium ratio values, with 46 falling within the expected UK range of oxygen isotope ratios and 54 falling within the <sup>87</sup>Sr/<sup>86</sup>Sr ratios range, consistent with an upbringing in the Vale of York or an area with a similar geology. Of the four that fell outside the York area range for oxygen, one person probably grew up in a cooler or more continental climate, or even at higher

altitude, whilst the remaining three were probably from warmer, more arid or more coastal regions (Leach et al. 2009:552 and 555). The six individuals with strontium isotope values outside of the expected range of the Vale of York had values consistent with older rocks in central and western England or Scotland (Montgomery et al. 2006). Linking the results of the craniometric re-analysis with the isotope analysis, it was concluded that the population of Roman York comprised individuals from a broad ancestral heritage and large geographical origin (Leach et al. 2009:556).

These findings were supported by a study of a burial ground excavated in Driffield Terrace, York (Müldner et al. 2011). Recent excavations unearthed a very unusual cemetery with all male burials, and more than half had been decapitated. The cemetery dates to several phases ranging from the late 1<sup>st</sup>/early 2<sup>nd</sup> century AD to the late 3<sup>rd</sup>/early 4<sup>th</sup> century AD (Ibid. 2011:282). This site is of interest given that some individuals used for analysis in the current research were from this cemetery. Given the highly unusual circumstances of these burials, the study set out to investigate if these individuals were locals or incomers to the York area. A multi-isotope approach (strontium, oxygen, carbon and nitrogen) was used to establish if the individuals buried at Driffield Terrace were different in origin to the wider population of Roman York, based on data discussed in Leach et al. (2009, 2010). Driffield Terrace 3 and 6 excavations revealed a total of 80 inhumations with various burial alignments, and about 16 cremations. Osteological analysis suggested that most, if not all, of the individuals buried there were male and of between approximately 19 and 45 years of age at death. At least 46 of these men had been decapitated, their skulls being placed between the knees or feet, or sometimes by the torso (Müldner et al. 2011:282).

Carbon and nitrogen isotope data were used to examine the diet of the Driffield Terrace individuals to see if it was any different to that of the local population, thus potentially indicating that these people were immigrants to the area. Second premolars of 18 individuals with preserved dentitions buried at 6 Driffield Terrace were analysed fusing oxygen and strontium isotope analysis (Müldner et al.

2011:282), and dietary reconstruction used bone samples (mostly rib) from every individual from both burial sites, namely 52 individuals from 3 Driffield Terrace and 23 from 6 Driffield Terrace (a total of 75 individuals). Dental collagen was also extracted from the 18 teeth from which enamel was previously used for strontium and oxygen isotope analysis (Ibid. 2011:283). The results of the analysis showed that when oxygen and strontium isotope data were considered in combination, it appeared that only five individuals of the 18 from 6 Driffield Terrace had results consistent with a childhood spent in the locale of York. (Ibid. 2011:284).

Overall, the study found that the isotopic signals from the 'Headless Romans' had a different distribution to those of individuals buried in other cemeteries in the city. The oxygen isotope values in particular showed a wider range of values, interpreted as indicating more diverse and "exotic" origins for the Driffield Terrace individuals. The dietary isotope data (carbon and nitrogen) identified a number of additional individuals who would not necessarily have been recognised as nonlocal from their strontium and oxygen isotopes alone (Müldner et al. 2011:287). This is because the isotope ratios of C and N incorporated into their bones from their diets could have been different from the local isotope ratios. This is usually due to eating of C<sub>4</sub> plants such as maize and millet. While only millet (and not maize) was available in Roman times, it was not grown and consumed in Britain during the Romano-British period.

A study by Montomery et al. (2010) has also used lead isotope analysis to further examine some of the individuals from Driffield Terrace. Very few studies have used lead isotopes to track the residential mobility of humans in Britain. However, this study draws together lead isotope and lead concentration data from tooth enamel for over 200 burials at 33 sites in Britain, Ireland and further afield in Rome. This was to establish if immigrants to Britain during the Romano-British period could be identified on the basis of their differential exposure to pollutant lead. Lead is naturally present in small quantities in most of the earth's rocks, soils and waters, from where it can be taken up by plants and then incorporated into animals upon consumption (Montgomery et al. 2010:211). The majority of the lead

content in a human body is found in the skeleton and so it is expected that the isotope ratio of skeletal lead will be indicative of the rock from which it originated; thus, it can provide information about geographical origins. However, natural skeletal lead ratios can be overwhelmed if large amounts of lead from sources of lead ore with different isotope profiles are mined and smelted, thus severing the link between a person and the rocks and soil in which they grew and sourced their food. In southern Britain, this overwhelming effect happened in the first century AD when most people had very similar lead isotope ratios deriving from ambient pollutant lead from ore sources (Ibid. 2010:212).

For the purposes of Montgomery's study, only one lead isotope ratio was used, that of <sup>207</sup>Pb/<sup>206</sup>Pb, but the four lead isotopes (<sup>204</sup>Pb, <sup>206</sup>Pb, <sup>207</sup>Pb and <sup>208</sup>Pb) can be combined in various ways. Analysis of this isotope ratio found that the majority of Romano-British individuals clustered together and within the same region as galena from English and Welsh lead mines (Montgomery 2010:213). However, two individuals fell outside of this cluster. They were an adult female burial from Spitalfields in London and an adult male burial from 6 Driffield Terrace in York. The data for these two individuals suggest they would have been unlikely to have "obtained" their lead isotope signatures whilst in southern Britain, unless they consumed most of their childhood foodstuffs from imported vessels, which seems unlikely (Ibid. 2010:215). Four men from the Driffield Terrace cemetery were found to fall outside of the central cluster of polluted individuals (that is, those with more than 1mg kg<sup>-1</sup> of lead). When compared with skeletons from southern Britain, and also Rome, these individuals were found to have different lead isotope ratios that were consistent with younger ore areas such as the Tertiary deposits of the Mediterranean basin and the Near East (Ibid. 2010:217). No further conclusions could therefore be drawn about their likely geographical origin. However, lead isotope ratios are useful data when other isotope systems such as strontium cannot be used to discriminate between individuals from regions of Mesozoic rocks, which widely occur across southern Britain and Europe. In this respect, it is another tool that can be used to determine if people were raised locally to their burial place.

Finally, a recent study by Martiniano et al. (2016) of seven Roman individuals buried in the Driffield Terrace, York cemetery used genome and isotope analysis to identify the origins of these people. In the genome analysis, mtDNA and Y chromosome DNA analysis was possible, as all of the individuals were male. Strontium and oxygen isotope analysis was also undertaken on all seven individuals (Martiniano et al. 2016:4). The results show that one York Roman (3DRIF-26) has a genome which gave a clear Middle Eastern signal with closest neighbours of Palestinian, Jordanian and Syrian origins. Isotopic analyses supported this genetic result (Ibid. 2016:4). The other six York Romans were genetically and isotopically identified as being of British (most likely Welsh) origin (Ibid. 2016:6).

## 4.8.3 Site 2: Lankhills, Winchester

Forty skeletons from a Roman cemetery at Lankhills School in Winchester, Hampshire, southern England were examined by Eckardt et al. (2009). Previous excavations on this site had discovered a number of individuals thought to have originated from the Danube region, based on grave goods/burial style evidence (Clarke 1979). Roman Winchester (*Venta Belgarum*) was an urban centre of *civitas* capital status (an independent administrative centre), and the late Roman burial site at Lankhills School forms part of Roman Winchester's northern cemetery. This site was excavated by Clarke (1979) between 1967 and 1972, who found 443 inhumations, seven cremation burials, and one empty grave. Oxford Archaeology carried out further excavations on the site between 2000 and 2005 and located another 305 inhumations and 20 cremation burials, and further graves that were not excavated (Eckhart et al. 2009).

Oxygen and strontium analysis was applied to a sample of 18 individuals drawn equally from the perceived immigrant and local populations (Evans et al. 2006). The "locals" followed the predominant pagan burial custom of being buried with grave goods required for the journey to the afterlife, for example footwear, coins to pay passage across the Styx, and food and drink (Ibid. 2006:266). The "nonlocals" did not conform to this burial rite. Instead, they had a large quantity of personal ornaments and equipment with them, which were remarkably consistent in their positioning in relation to the burial (Ibid. 2006:267). This was particularly notable in the manner in which bracelets were worn, ones made of copper alloy being worn on the right wrist, with bracelets of various other materials worn on the left. This fashion is closely paralleled in Hungary, which is further supported by the presence of hexagonal blue cylinder beads found in burials on the Roman frontier on the Upper Danube (Ibid. 2006:268). The stable isotope data showed that seven of the nine "locals" did indeed have strontium and oxygen isotope values consistent with an upbringing in this part of Hampshire. However, two individuals had oxygen isotope values too depleted to indicate they were from southern Britain. These data would be more consistent with an origin on the continent (Ibid. 2006:270). On the other hand, the "non-local" population exhibited a wide range of oxygen and strontium isotope ratios, supporting the suggestion that some are not likely to be from southern Britain. However, with such wide-ranging data, it is also unlikely that all the individuals were from a single origin. Instead, four of the individuals (three men and one woman) had oxygen isotope drinking water values consistent with a childhood in continental Europe, and possibly the Italian/Austrian Alps where populations today have similar values (Ibid. 2006:270). One man had a low <sup>87</sup>Sr/<sup>86</sup>Sr ratio (0.7064), which is consistent with a childhood spent in an area of young, non-radiogenic rocks. These rocks can be found in several areas of central southern Europe, including parts of Hungary. One male had a drinking water oxygen value within the range of UK values, but also typical of parts of Holland, France and Germany. However, his Sr isotope signature is too high for him to have been raised on the local chalk, and therefore it was suggested that he was probably from an area of western Europe not located on chalk or limestone. The other "non-local" individuals provided data in the same range as the "local" group, interpreted as being of local origin, but perhaps a second-generation immigrant (Ibid. 2006:271) for whom burial in the traditional manner of the ancestral homelands was still important.

In a later study, Eckardt et al. (2009) performed strontium and oxygen isotope analysis on teeth from 40 individuals from the same cemetery. These were taken from a broad cross section of the cemetery population in order to encompass the suggested "local" burial rites (Clarke 1979), and also the suspected "non-locals" who were identified by the presence of particular grave goods. The individuals sampled further exhibited a number of different burial positions, with most being supine, but some were also "deviant" (prone and decapitated - Eckardt et al. 2009:2818). It was discovered that 21 individuals had <sup>87</sup>Sr/<sup>86</sup>Sr within the local range of 0.7072 to 0.7092, and an oxygen isotope signature consistent with an upbringing in Britain. Eckardt et al. (2009:2821) suggest that these people quite probably originated in Winchester or the surrounding area. Eight individuals had  $^{87}$ Sr/ $^{86}$ Sr ratios outside the Winchester range, but a  $\delta^{18}$ O value consistent with a childhood in Britain, or elsewhere with a similar climate. Five individuals, four of whom were given "local" burial rites, had strontium isotope values more consistent with western and northern Britain. However, one of these five people had a lower oxygen isotope signature than the others and thus an origin in western Britain was unlikely, these data being more compatible with the Ardennes region of Belgium or western parts of Germany (Ibid. 2009:2821-2). Overall, it was concluded that these 21 individuals may have been born and raised in Winchester and the surrounding areas, with a further eight probably coming from other parts of Britain. The remaining 11 individuals were defined as immigrants, with 10 coming from a warmer area and one coming from a colder place (Eckardt et al. 2009:2823). However, it must be remembered that it is difficult to accurately suggest a place of origin using stable isotope analysis; instead, it is more advisable to conclude only that these people were not "locals". These studies on individuals buried in the Lankhills cemetery do suggest that at least a quarter of them were incomers to the Winchester area after having spent their childhoods somewhere other than Britain (Ibid. 2009:2824). The burial rites and grave goods could not consistently identify these migrant individuals on account of them not differing sufficiently from those of other individuals in the cemetery, but examination of burial rites and customs is still useful in considering how these individuals, and the mourners attending their burial, may have viewed themselves and their ethnic affiliations.

#### 4.8.4 Site 3: Gloucester

Isotope analysis was also used to investigate population and dietary diversity in Roman Gloucester (Chenery et al. 2010). A multi-isotopic approach was again used (oxygen, strontium, carbon and nitrogen) and applied to individuals found in a late 2<sup>nd</sup> century AD mass burial pit at London Road; this was in addition to those found in a nearby cemetery, which contained cremations and inhumations dating from the 1<sup>st</sup> to the 4<sup>th</sup> centuries AD. Roman activity began in the Gloucester area around AD 49 with the construction of a fortress at Kingsholm, located alongside a pre-Roman settlement (Hurst 1985, 1999). In the late AD 60s, a new legionary fortress was built 0.5 km to the south, and this eventually became a *colonia* (a settlement for retired legionaries and their families) at the end of the 1<sup>st</sup> century AD (Hurst 1999, Wacher 1974).

Between 2004 and 2006, excavations at London Road, Gloucester revealed part of a Roman inhumation and cremation cemetery dating from the 1<sup>st</sup> to 4<sup>th</sup> century AD (Simmonds et al. 2008). An unusual 2<sup>nd</sup> century mass burial pit that contained the remains of at least 91 individuals was located. The burials had been placed into the pit in a haphazard manner in what was interpreted as being a single event. While there were no notable osteological differences between those individuals buried in the pit and in discrete graves nearby, there was an excess of young adults in the pit. The pit burials were interpreted by the excavators as the result of a catastrophic event (Simmonds et al. 2008:140).

Chenery et al.'s (2010) study set out to examine whether the isotopic evidence supported these data and also how diverse the population of 2<sup>nd</sup> century Gloucester was in terms of their geographical origins and diet. The teeth of 21 individuals were sampled for Sr and O isotope analysis; 10 were from the main cemetery and 11 from the mass grave. These teeth were all permanent second or third molars, which would represent an age of crown formation of three to seven years for the second molar, and nine to 13 for the third molar (Hillson 1996:123). Bone preservation at the site, particularly in the mass grave, was very poor and

therefore extraction of collagen from bone yielding enough collagen for C and N analysis analyses was not very successful; only 11 human and nine faunal bones, the latter being used to establish a local isotope signature had sufficient collagen (Chenery et al 2010:153). The results suggested that the diet eaten by these people was directly comparable to that from the same period in York, another *colonia*, being a terrestrial (C<sub>3</sub>-based) diet with a large amount of animal protein. No differences were detected in dentinal collagen isotope composition between the individuals in the mass graves and the cemetery (Ibid. 2010:156).

The <sup>87</sup>Sr/<sup>86</sup>Sr ratios from the enamel of individuals analysed showed a broad range of values from 0.7088 to 0.7134, but these are all within the range of bioaccessible strontium for the area of Gloucester and a surrounding 30km radius. There was found to be no statistically significant differences between the discrete burials and the individuals in the mass grave (Chenery et al. 2010:156). However, care must be taken with the interpretation of these data because these people may not have been locals but rather could have moved into the Gloucester area from other places with similar strontium values. Enamel phosphate oxygen ratios place the Gloucester burials into two groups; the individuals in the larger group had phosphate oxygen ratios compatible with a childhood spent in Britain whereas the individuals in the smaller of the two groups probably had origins in a warmer, more coastal or possibly more arid climate (Ibid. 2010:156).

Correlation of data for sex, age at death and isotope values for all the Gloucester burials suggests no statistically significant differences between mass grave and cemetery burials. This would support the hypothesis that the mass grave contained a random sample of the Roman Gloucester population and was excavated after a catastrophic event, such as an epidemic of infectious disease (Chenery et al. 2010:157). However, this study was less successful in identifying which of these people, if any, was an incomer to the area. It was suggested that six or seven of the 21 individuals analysed spent their childhood outside Britain in a warmer/more coastal climate. However, difficulties in interpretation of these data are largely due to the complex geology of the Gloucestershire region, and that it is

known that foods were imported during the Roman period. Consumption of nonlocal foodstuffs would additionally impact on the isotope data generated, thus leading to misleading conclusions about potential immigrants to the area, i.e. was it imported food or was it an immigrant from a place where the food was eaten that produced a particular isotopic value.

#### 4.8.5 Site 4: Poundbury, Dorchester

Stable isotope analysis was also used to look for immigrants to Roman Britain at Poundbury Camp Cemetery, near Dorchester in Dorset. Here, over 1200 inhumation burials were excavated between 1966 and 1982 (Farwell and Molleson 1993). They date from the Iron Age, early and late Roman and post-Roman eras, with 59 burials being dated to the late Iron Age/early Roman periods (1<sup>st</sup> century AD) and the majority dating to the late Roman period (4<sup>th</sup> century AD). Only three burials could be securely dated to the post-Roman period (Farwell and Molleson 1993; Richards et al. 1998:1247-1248). The study set out to undertake carbon and nitrogen stable isotope analysis on the inhumations from the various time periods and burial contexts to see how much variation in diet occurred (Richards et al. 1998). There was found to be little variation in the isotope values of the Iron Age/Early Roman individuals, who had a terrestrial based diet containing both plant and animal protein. The Late Roman individuals had more positive  $\delta^{13}$ C values and higher  $\delta^{15}$ N values than the Iron Age people, indicating that although much of the dietary protein came from terrestrial sources, there was also a contribution from marine protein (Ibid. 1998:1249-50).

The results of the study showed that there might have been immigrants buried in the Poundbury cemetery. Two individuals had more positive  $\delta^{13}$ C values than would be expected for a terrestrial diet but, if this was to be attributed to the consumption of marine foods, then the associated  $\delta^{15}$ N values would have been higher than they actually were. Instead, it was concluded that these individuals

spent most of their life in a warmer climate and then moved to Britain shortly before they died (Ibid. 1998:1251). The evidence of immigrants would match earlier conclusions drawn by Molleson et al. (1986), who looked at lead isotope values of some Poundbury burials and found evidence for one individual (PC1255) having a lead isotope value that most closely matched the Laurion lead source in Greece. Carbon and nitrogen isotopes for this individual were analysed and it was concluded that this person had been born and raised in a warmer area than Britain and had migrated shortly before death (Ibid. 1986:1251).

# 4.8.6 Summary

To summarise the studies of mobility in Roman Britain using stable isotope analysis, the following table, Table 4.1, shows the main techniques used to differentiate incomers from locals at the different sites:

Site	Author	Isotopes (C and N)	Isotopes (Sr and O)	Isotopes (Pb)	Craniometry
Catterick	Chenery et al. (2011)	Yes	Yes	No	No
York	Leach et al. (2009)	No	Yes	No	Yes
	Leach et al. (2010)	No	Yes	No	Yes
	Montgomery et al. (2011)	No	No	Yes	No
	Müldner et al. (2011)	Yes	Yes	No	No
Gloucester	Chenery et al. (2010)	Yes	Yes	No	No
Winchester	Evans et al. (2006)	No	Yes	No	No
	Eckardt et al. (2009)	No	Yes	No	No
Poundbury	Molleson et al. (1986)	No	No	Yes	No
	Richards et al. 1998)	Yes	No	No	No

Table 4.1 A summary of the methods used to identify locals and immigrants.

# 4.9 Isotope analysis and mobility studies in other parts of the Roman world

The published literature on similar studies in other parts of the Roman world is much less, to date, with just a small number on skeletons from Roman Egypt, the Middle East and mainland Europe. As the current research is considering migration from within as well as without the British Isles, isotope studies from other provinces in the Roman Empire only need to be briefly considered.

A study of the variability in oxygen values in Roman period skeletons dating from around AD 250 was undertaken on remains found in the Dakhleh Oasis in Egypt (Dupras and Schwarcz 2001). The researchers found that 32 of 34 individuals had consumed local water during the time of tooth development and were thus concluded to be locals (Ibid. 2001:1199). Geographic origins and mobility were also investigated at sites in southern Jordan dating from the Roman (1<sup>st</sup> to 3<sup>rd</sup> century AD) and Byzantine periods (4<sup>th</sup> to 7<sup>th</sup> century AD). The teeth of 12 adults from the 2<sup>nd</sup> century AD site at Ichirbet edh-Dharih were analysed for strontium isotopes. The results found one male to be not local and likely to have come from further north in Jordan or from southern Syria (Perry et al. 2008:544).

In Europe, a number of similar studies have been completed. These include strontium analysis of bones and teeth from people buried at Neuburg/Donau in Germany. They were dated from AD 330 to 400, and the data suggested that 56% of women and 37% of men from the late Roman fortress of Venaxamordum were non-locals (Schweissing and Grupe 2003). It was concluded that the immigrants originated from areas to the north east of the Danube river (Ibid. 2003:1377). In another study oxygen isotope analysis was applied to the teeth of 61 individuals buried in the necropolis of Isola Sacra, which dates from the 1<sup>st</sup> to 3<sup>rd</sup> centuries AD. It was concluded that one-third of these people originated from outside Rome (Prowse et al. 2007:510). Both strontium and oxygen isotopes were used by Killgrove (2010) in her PhD research, also in Italy. She concluded that, at Casal Bertone, Rome, 37% of the population were immigrants. Meanwhile, at Catellaccio Europarco, Rome, this figure was slightly less with 29% of the population being

immigrants. Further, a combination of mitochondrial aDNA and stable istotope analyses focused on individuals from a 1<sup>st</sup> to 3<sup>rd</sup> century AD Roman site at Vagnari in southern Italy. Oxygen isotope analysis of 23 individuals found that six people were not born locally. Meanwhile, two out of ten individuals analysed for aDNA had biological ancestry consistent with populations from sub-Saharan African and Asia (Prowse et al. 2010:191).

A recent study assessed migration into Rome by using strontium isotope analysis from 105 individuals buried in two cemeteries associated with Imperial Rome (Killgrove and Montgomery 2016:1). These are Casal Bertone and Castellacci Europarco. Oxygen and carbon isotope analysis were also performed on 55 of these individuals. Several outliers were found and these were likely to be immigrants to Rome (Ibid. 2016:1). Demographics of these immigrants show men and children migrated. A comparison of carbon isotopes from teeth and bone samples suggests the immigrants significantly changed their diet after moving to the city (Ibid. 2016:1).

The adoption of stable and other isotope analysis from the disciplines of earth sciences and geology in order to identify burials of incomers to an area is a useful tool to support previous, less scientific, approaches that attempted to find immigrants in the archaeological record (detailed above). However, these isotopic techniques cannot pinpoint the actual geographical origin where these individuals were born and raised. However, instead the data can be used to suggest (if tooth and bone survival condition is good enough) if these people were born and raised in the areas in which they were buried, or if they were likely to have moved from elsewhere. It may even be possible, as shown in some of the studies discussed, to suggest an area or group of areas from which the person had possibly migrated during their lives. As more and more use is made of isotope analysis, and a "bank" of comparative data is accumulated from analysing numerous individuals from different cemeteries, strontium and oxygen isotope maps will become more detailed and so it may become more possible in future to identify with more confidence the actual place of origin of these people. Until such times when a

large enough data sample is available for comparisons, isotope analysis could be used alongside some of the other analytical techniques briefly discussed above, which are at the disposal of the diaspora researcher, such as craniometry and mitochondrial DNA analysis.

The studies discussed in this section show, on the basis of different kinds of evidence, that people were moving around between provinces and within regions of provinces during the Roman period for a wide variety of different reasons. These groups of migrants included the military, slaves and merchants. Evidence for this movement can be seen in imported material goods and foodstuffs, the remains of transport infrastructures, such as roads and ports, and in the isotopic signatures seen in human skeletal remains.

This section has mainly concentrated on movement into and around particular sites in Roman Britain, because the current research focuses on people buried in what is now England. However, there is considerably more documentary and archaeological evidence of migration and travel in the wider Roman Empire at the time. Considering all of this evidence is beyond the scope of the current study. Nevertheless, relative to this study, it is important to consider the extent of mobility in relation to the transmission of infectious diseases during the Roman period. As has been already discussed, migration and travel in general today is frequently responsible for outbreaks of infection, and the same would have undoubtedly been true in the past. In addition to migrants bringing their diseases with them and introducing these to previously uninfected people in new areas, it is known that travelling can place stresses on the body and the immune system making travellers more susceptible to becoming ill, or to the reactivation of a dormant infection, such as primary TB (Galagan 2014:307).

## Chapter 5: Travel and its impact on the transmission of infectious disease

## 5.1 Introduction

In Chapter 4, the methods of and evidence for mobility and the means to move (transport) in Roman Britain were examined. The current research examines the effects of mobility on the transmission of TB in the Roman period. This was an era for which there is no direct documentary evidence for the link between mobility and infectious disease. It is therefore essential that published literature that considers the link between infection and travel today, in its broadest sense, is explored from which some inferences can be made about the past. In the 21<sup>st</sup> century, movement is easier, quicker and probably less expensive, relatively speaking, than it was 2000 years ago, and a larger proportion of the population travel more frequently than in the past. The following section examines the evidence for infectious diseases, including TB, being transmitted as a result of modern transport systems, although these differ markedly from transport methods available during the Roman period. Data from several case studies that document the impact of plane flights, cruises and train and bus journeys on the spread of infections are explored, and then the transmission of TB in relation to modes of transport is considered. While modern transportation methods differ greatly from those available in the Roman era, what they do have in common is the confinement of a large number of people into a small space for prolonged periods of time. These conditions are likely to increase the possibilities of transmission of infectious diseases (including TB).

Migration of humans has been the pathway for disseminating infectious diseases throughout history (Wilson 2004:39; WHO 2016). The advent of modern transportation systems means that these diseases, including those newly emerging, can now be transmitted globally in a very short space of time. In history, when infections emerged in isolated villages, they most likely died out without further spread (Wilson 2004; Miller 2009:1121). It is therefore known that travelling can pose various risks to health depending on the nature of the travel and the characteristics of the traveller. Migrants may encounter sudden, significant changes in altitude, humidity, and temperature, and the range of microbes that survive in different environments to which they are exposed, all of which may result in ill health. Additional health risks may also result in areas where housing, hygiene and sanitation are of poor quality (WHO 2016). According to the World Health Organisation (WHO), more than 900 million international journeys were undertaken in 2010, with Europeans making up the majority of international travellers (Field et al. 2010:330). This increased to 1,087 million worldwide tourist journeys in 2013 (Statista 2016). People travel for a number of reasons, and movement for business and pleasure represents only a small fraction of the total journeys. Migration individually or in groups is recognised in refugees, missionaries, merchant sailors, students, temporary workers, pilgrims or aid workers. The distances travelled can also vary considerably and the length of stay of the person/group in a place may be temporary/seasonal or permanent (Wilson 2004:163). Recently, mathematical modelling has been used to predict the likelihood and extent of the spread of infectious diseases in cities (Dalziel et al. 2013), on public transportation systems (Xu et al. 2013), across the world (Miller 2009), and in predicting how time spent at a destination could impact on the spread of an epidemic (Poletto et al. 2013). This theoretical approach was developed in response to the perceived threat of terrorist-induced biological warfare. However, while this approach is helpful, it can only ever be a tool in predicting possible scenarios in which a complex range of variables must be taken into account when infectious diseases occur.

The type of infectious disease has a large effect on how well it is transmitted during or as a result of travel. Organisms that survive mainly or entirely within the human host are those that are most easily carried to a new geographical area (Chuit et al. 2003:1, Wilson 2004:162). These are the microbes that can be transmitted via droplets and via close physical or sexual contact, for example Human Immunodeficiency Virus (HIV), TB, measles, pertussis (whooping cough), diphtheria and hepatitis B; they can be particularly severe when introduced into a population with no previous immunity (Chuit et al. 2003:1, Wilson 2004:162).

Travelling not only poses a risk to the travellers themselves, but also to the people encountered on the journey and at the final destination. Non-immune native people are subject to diseases brought in by migrants, and both indigenous and migrant groups are exposed to new diseases to which they have no immunity. A good example of this can be seen in the 15th century AD onwards when European explorers travelled to the New World. Until the arrival of Europeans, the New World was free of smallpox, typhus, cholera and measles. When Cortez invaded Tenochtitlan (modern day Mexico City) in 1520, a year after he arrived in the New World, he found many of the inhabitants of the city to be infected with smallpox. Nearly 50% of the population was thought to have died in this first epidemic. Eleven years later, a second smallpox epidemic was introduced from Spanish ships and this also caused widespread infection and death. By 1595, over 18 million people were thought to have died of smallpox, mumps, measles and other infectious diseases brought by the Europeans (Cartwright 1972).

In comparison to the Roman population, in the 21<sup>st</sup> century AD people travel by many modes of public transport that are increasingly available to all budgets. Research has mainly been focused on infectious diseases transmitted while travelling by plane or ship (eg. Nelson et al. 2013, Isakbaeva et al. 2005). Usually, these involve journeys of long distances. However, in the section below on TB, transmission on long or regular bus and train journeys is also discussed. Research has probably concentrated on public transport systems because of the high numbers of people carried on a daily basis, with people being likely to be exposed to an "index" case who could potentially infect large numbers in one journey. However, people probably travel even more frequently by car, especially over short distances, for example when they commute to work while car sharing with a colleague. In such an enclosed space and with no air filtration systems there appears to be great potential for diseases to be passed between fellow travellers, but no research on the likelihood of this occurring has been found. This would appear to be an area where research needs to be done in order to provide a fuller picture of the risks of transmission of infection due to mobility.

# 5.2 Vaccine-preventable diseases

The current research examines the link between mobility in the Roman period and the transmission of TB. The following section sets out to examine diseases transmitted from person to person as a result of travelling today in order to make comparisons and inferences. Whilst vaccination is not directly relevant to the current research, today access to vaccination programmes is available, and therefore their impact on the transmission of infectious diseases as a result of mobility must be considered. One would therefore be forgiven for assuming that travel-related infection is at a lower rate than it would have been prior to wide access to such an extensive vaccination programme, at least in developed countries. If people used these programmes correctly, it is likely that there would be far fewer diseases transmitted as a result of travelling. However, the following section discusses the fact that a large number of migrants are failing to utilise these vaccinations prior to travelling, possibly sometimes due to last minute planning or cost of vaccination, and some are falling ill with diseases that could easily have been prevented (Heywood et al. 2012).

The National Health Service (NHS) in the UK has a free vaccination programme for all children (NHS 2016b, and see Table 5.1 below). This prevents a series of infectious diseases likely to be encountered at home and in many locations abroad.

When to immunise	Protects against		
Two months old	Diptheria, tetanus, whooping cough,		
	polio, <i>Haemophilus influenzae</i> type b		
	(Hib), pneumococcal disease,		
	rotavirus		
Three months old	Diptheria, tetanus, whooping cough,		
	polio, Hib, Meningococcal group C		
	disease (MenC), rotavirus		
Four months old	Diptheria, tetanus, whooping cough,		
	polio, Hib, pneumococcal disease		
12 – 13 months old	Hib, MenC, pneumococcal disease,		
	measles, mumps, rubella		
3 years four months old	Diptheria, tetanus, whooping cough,		
	polio, measles, mumps, rubella		
Girls aged 12 – 13 years old	Human papilloma virus (HPV)		
14 years old	Tetanus, diphtheria, polio, MenC		

Table 5.1: The NHS vaccination programme for all UK children, regardless of mobility status or travel plans (NHS 2016b) NB. BCG vaccination for TB is no longer routine in most areas.

In addition, various vaccines are available in the UK for people of any age who may be travelling to areas of the world where some infectious diseases are endemic/epidemic; vaccinations provided depend on the location of travel (WHO 2016). The currently available travel vaccines are against cholera, hepatitis A and B, Japanese encephalitis, meningococcal meningitis, rabies, tick-borne encephalitis, TB, typhoid and yellow fever.

In order to access these vaccines, individuals consult with their General Practitioner's surgery to discuss their requirements and make arrangements for the vaccinations well in advance of travel. Although these preventative measures are readily available, with busy lifestyles and last minute travel plans, people do not always seek advice and vaccinations before their journeys. Additionally, some vaccinations incur a cost to the traveller, and people may not be prepared or able to meet these additional cash outlays and so take a risk and travel unprotected.

Research done by Heywood et al. (2012) assessed travel-associated health risks and preventative behaviours in a sample of University students in Australia. This consisted of a questionnaire of 1663 students, both Australian and international (University of New South Wales, Sydney, Australia) examining their international travel history, travel intentions, infection control behaviours and self-reported vaccination history. There were 829 students who had travelled internationally in the previous 12 months. However, in 2007 a student returned to Australia with poliomyelitis from a home visit to Pakistan; this was the first case of the disease to occur in Australia since 1977. Additionally, a review of TB in the state of Victoria, Australia found that 37.9% of multi-drug resistant patients with TB were international students. Unlike the USA, Australia does not ask for proof of TB immunisation on entry to an Australian University, and international students only require screening for TB for their visa applications if they are from medium or highrisk TB countries (Heywood et al. 2012:44). Worryingly, although perhaps not unsurprisingly, the study also found that many travellers do not seek or follow appropriate health advice and therefore take their journey unprepared, thus being susceptible to the risks of contracting infectious diseases. In this study only 32.4% had sought preventative health advice before their trip (269 of the 829 who had travelled internationally in the preceding 12 months). Students in the survey also perceived a low personal likelihood of health risks whilst travelling, with the diseases they thought to be of least concern to them (dengue, hepatitis A, hepatitis B and measles) being the ones which are actually the most commonly reported in travellers (lbid. 2012:45).

The spread of vaccine-preventable diseases (VPDs) is a global concern and not restricted to Australia, with the most common VPDs recorded in travellers being typhoid and paratyphoid B, acute viral hepatitis, influenza, varicella (chicken pox), measles, pertussis (whooping cough), poliomyelitis and bacterial meningitis. Apart from the health consequences to the infected individual, VPDs can cause major

public health problems if they are introduced or re-introduced to areas with susceptible populations (Gautret et al. 2012).

One example of serious public health consequences is the spread of poliomyelitis via international travel. Poliomyelitis is close to eradication, with cases in 2002 numbering 2000 in nine countries, following the efforts of the Global Polio Eradication Initiative in 1988 bringing down the number from 350,000 in more than 125 countries. However, between 2003 and 2006, travellers imported poliomyelitis into 24 polio-free countries. To reduce this risk, in 2006 the Advisory Committee on Immunisation Practices proposed that proof of vaccination against polio should be required from all travellers travelling to polio-free countries where polio is endemic (Gautret et al. 2012:78).

A second example of VPDs causing considerable public health issues is that of *Neisseria meningitidis* serogroup W135, which causes bacterial meningitis and has been of particular concern at the Hajj. The Hajj is an annual mass gathering of Muslims. Each year, over two million pilgrims from over 160 countries attend this event in Mecca, Saudi Arabia. The overcrowded conditions mean that infections transmitted via respiratory secretions (droplets) are very common amongst these pilgrims. An outbreak of *Neisseria meningitidis* serogroup W135, which is transmitted via this route, was reported in association with the Hajj in 2000 and 2001. The resulting spread of disease caused outbreaks in nine countries, with isolated cases in a further four; it caused this strain of the disease to become an important public health issue for the first time. As a consequence, since 2002 the quadrivalent meningococcal vaccine, which is active against serotypes A, C, Y and W135, is now a visa requirement for all pilgrims attending the Hajj. Since then, no further outbreaks have occurred, with only sporadic cases of W135 being detected in sub-Saharan countries since 2003 (Gautret et al. 2012:78-80).

There are now extensive health requirements for people on the pilgrimage to Mecca that include rules on vaccinations against yellow fever, meningococcal meningitis, poliomyelitis and seasonal flu. There are also rules on the transport of

food and the reporting of infectious disease outbreaks at the Hajj (Memish and Al Rabeeah 2013), and this legislation appears to have considerably prevented the spread of infection amongst attendees and the wider population when these people return to their home countries.

Another good example of a travel-related infection is Japanese encephalitis (JE) This is a severe disease and constitutes a risk for people travelling to JE-endemic countries, such as Japan and south east Asia (Ekkelenkamp et al. 2002:3091). The virus that causes JE is mosquito-borne and is a leading cause of encephalitis in Asia (Halstead and Jacobson 2003 in Hills et al. 2010:930). Clinical illness develops in less than 1% of people infected with this virus but, when it does occur, infection is usually severe and has a 30% fatality rate. Although there is no treatment for JE, there is a preventative vaccine and the risk of JE in travellers to Asia is considered to be low, with less than one case per million travellers. However, higher rates of infection have been described for travellers from Finland and Sweden to Thailand, with one case per 257,000 and 400,000, respectively. The risk for the individual traveller was therefore interpreted as dependent on a multitude of factors which include duration of stay, season of travel, type of accommodation and activities undertaken (Hills et al. 2010:930). Activities classified as increasing the risk of becoming infected include rural travel, staying on or near a farm, staying in unscreened (i.e. no mosquito deterrent screens) accommodation, and taking part in trekking or other outdoor activities. A JE vaccine licensed for use in the USA was introduced in 1992 but it has a low rate of uptake, with estimations of only 11% of travellers to JE endemic countries receiving the vaccine prior to travelling (Ibid. 2010:931).

In summary, there are readily available, free (or inexpensive), highly effective vaccinations available that protect against most of the infectious diseases likely to be encountered whilst travelling today, although the vaccine against TB is no longer considered effective enough to be routinely given, particularly in adults. However, take up of vaccinations among the general public appears, for several reasons including poor pre-travel organisation (and cost), to be too low. Where

legislation has put in place a requirement for proof of vaccination, such as for pilgrims to the Hajj, vaccines have been highly successful in controlling the spread of infectious diseases contracted whilst travelling and transmitted back to the home countries of travellers. However, in order to reduce the incidence of commonly encountered travel-related diseases, laws should be introduced requiring proof of pre-travel vaccination for all travellers which should be produced, along with passports, as a legally required travel document.

## 5.3 Infectious diseases contracted on planes and ships

## 5.3.1 Air travel

The following section considers modes of travel today as predisposing risk factors leading to infections being transmitted between individuals. Although flights were obviously not an available method of transportation in the Roman period, they are so commonly used today that their role in disease transmission needs to be considered. There has been much research on the transmission of infectious diseases during the process of travelling itself, and a good example detailed here is the transmission of measles on flights (Beard et al. 2011, Nelson et al. 2013).

Measles is a viral infection transmitted via infected droplets that are exhaled when someone coughs, sneezes or even speaks. Due to an extensive vaccination programme, by the year 2000 measles was declared no longer endemic in the United States of America (USA), but cases and outbreaks continue to occur due to importation from other countries (Nelson et al. 2013:82). Studies have shown that measles can be transmitted during flights (Beard et al. 2011) and, to prevent the introduction and spread of the disease within the USA, contact tracing of people identified with measles on flights is conducted by the USA Centres for Disease Control and Prevention (CDC) and the Division of Global Migration and Quarantine (DGMQ). The CDC devised a measles protocol that was revised in 2008 in order to identify contacts on a flight where a person with measles has been reported.

The CDC protocol defines contacts on an aircraft with more than a 30-passenger capacity as all the passengers seated in the same row as the infected person, and the two rows in front of and behind them. All children aged two years and under seated anywhere on the plane, and crew members serving in the same cabin, are also included. Travel companions of the infected person, regardless of where they are seated, are also classified as contacts. On a flight with a capacity of less than 30 people, all people on board are classified as contacts due to differing air flow patterns in these planes compared with the larger planes (Nelson et al. 2013:82).

In Australia in 2010, there was an outbreak of nine people with measles resulting from contact with an infected person on a flight. The "index" case was infectious while travelling on a 12-hour journey from South Africa to Australia for a BMX cycling event, and, as a result, infected four other people who were travelling on the same flight. Of the remaining four people infected, two were health care workers at the hospital where the "index case" was admitted on the day following the flight. The final two were members of the public that were also exposed at the hospital (Beard et al. 2011:31). Only one of the infected people was seated within the two row contact tracing zone, with the others seated three, four and 16 rows behind the index case. However, three of the four individuals could have been infected in South Africa before the flight as they had all been present at an event there. Unfortunately it was not clear where these three individuals were seated in relation to the index case on the return flight, but no other people with measles were found in association with the BMX cycling event (Beard et al. 2011:30). It can be concluded that it is indeed the case that this outbreak resulted as a consequence of travel, and it appears highly likely that the infection occurred whilst on the return flight from South Africa.

## 5.3.2 Cruise ships

In addition to infection being passed from person to person during flights, cruise ships are another modern mode of transport implicated in outbreaks of infectious

disease. On these vessels, a large number of people are, like on an aircraft, in a confined space together for a long period of time. Ships were a common form of transport in Roman times and will have faced the same problems of people being together in an enclosed space for prolonged periods, and hence undoubtedly the same issues of diseases being spread, as occurs today. While there is no documented evidence of TB being transmitted in a modern ship situation, an example of disease transmission today is reflected in an outbreak of Norovirus documented on a cruise ship, which persisted even after a week's sanitation period. This disease can be transmitted to humans in infected food and water and by person-to-person contact, like for transmission of TB, this is particularly possible in close, confined conditions (Isakbaeva et al. 2005). The virus causes acute gastroenteritis and was reported on day two of the first cruise and continued into a second and subsequent cruise on the same ship, with new passengers being implicated. Despite the week of sanitising on board, people on the following four cruises on the same ship also experienced Norovirus outbreaks. After extensive questioning of infected people, the initial source of the outbreak of the virus on the first cruise was found to be one of the ship's restaurants. Subsequent spread of the disease then occurred by person-to-person contact (Isakbaeva et al. 2005:156), and therefore no amount of cleaning and sanitation of the ship would have been successful in controlling this secondary outbreak, especially if cruise ship staff were acting as a reservoir for the virus and were inadvertently passing it on to the subsequent travellers.

In June 2006, despite usually being a winter virus, cases of Norovirus on board cruise ships increased, with 43 outbreaks being reported on 13 vessels, an outbreak being defined as three or more people falling ill within three days of each other (Verhoef et al. 2008:238-239). In these outbreaks, it was again thought that infected food was the primary source, with person-to-person transmission responsible for the rapid spread of this very infectious virus. Crew members were less likely to become sick than passengers, which could have been due to acquired immunity from previous exposure, or to the crew being reluctant to admit to illness (Ibid. 2008:240). Perhaps the ages of the passengers also need to be
taken into account as it is likely that cruises attract older people who may be more susceptible to falling ill with Norovirus than the younger staff members. Influenza ('flu) outbreaks have also been reported on cruise ships (Brotherton et al. 2003; Ward et al. 2010), another disease to which older individuals are particularly susceptible. Like Norovirus, influenza is a viral disease which creates seasonal outbreaks during winter months when infections and outbreaks are more likely but, with both viruses, the outbreaks occurring on the cruise ships were outside these expected times (Verhoef et al. 2008; Ward et al. 2010). This could mean that people who would have normally had an annual 'flu vaccination had not yet been vaccinated and were thus more susceptible to infection whilst on board.

In summary, the large number of people living together in a restricted space on board cruise ships appear to provide optimum conditions for the transmission of infectious diseases. This is true today as it would be in the past when vessels were smaller and living conditions more cramped. Whilst unsanitary conditions could perhaps be blamed for some infections, particularly in the case of Norovirus, diseases that can also spread by person-to-person contact can quickly infect a large proportion of a ship's passengers. As out of season 'flu infections have also been seen to occur, it should be advised that, if available in time for their holiday, potential cruise passengers should receive the latest influenza vaccination regardless of the season in which they take their trip. This would potentially halt outbreaks of vaccine preventable disease on board cruise ships, where close personal contact occurs between a large number of passengers made up of a potentially vulnerable age group who are gathered together for a prolonged period. The knowledge that people confined together in small quarters on a sea vessel and for prolonged periods are more prone to contracting infectious diseases suggests that the same must have applied during the Roman era. Roman ships were smaller than current vessels (see Figures 4.6 and 4.7), and while we cannot be sure how many people travelled on a Roman ship, we do know they would have been confined together for lengthy journeys. This could increase their chances of contracting infectious diseases (such as TB) from their fellow passengers.

#### 5.4 Diseases contracted at travel destination

Of course, it must be considered that in addition to migrants and travellers being exposed to diseases to which they have no immunity, native people are subject to diseases imported by travellers from their "native" countries. In addition, the journey itself is not the only time that travellers will become exposed to new diseases; this can also happen at the destination. Travellers' diarrhoea is one of the most commonly reported diseases encountered in the 21<sup>st</sup> century, and each year between 20% and 50% of international travellers develop it (an estimated 10 million people). Travellers' diarrhoea is mostly due to bacterial pathogens such as enterotoxigenic *Escherichia coli* (ETEC), and these can be encountered when experiencing differences in food preparation methods and levels of hygiene (Wright and Harding 2016).

Another cause of diarrhoea usually resulting from poor food preparation and hygiene is Salmonella. Caused by bacteria called Salmonella enteritidis, this infection is the most common serovar implicated in food-borne salmonellosis in humans, and is responsible for approximately 80% of salmonella cases in Europe (Nygård et al. 2004). For example, data from the Swedish infectious diseases register described a phage type distribution of Salmonella enteritidis bacterial infections in Swedish travellers from 1997 to 2002 (Nygård et al. 2004). In Sweden, about 85% of reported salmonella infections are acquired during travel abroad, with 11,570 cases (87%) of a total of 13,271 notified as being reported as infected abroad. The most commonly visited countries where infection occurred during the study period were Spain, Greece and Turkey, with Spain accounting for the majority of cases (34% - Nygård et al. 2004:2). It was suggested that visitors to foreign countries may be more susceptible to pathogens than local inhabitants circulating in the community being visited, the latter likely having acquired immunity to them. However, tourists often tend to stay in special resorts where food and preparation methods may be different to the rest of the country (and at home). This may lead to a difference in the reservoir of pathogens to which both natives and visitors are exposed (lbid. 2004:5).

Campylobacter infection is another cause of diarrhoea, and estimated to lead to 5 - 14% of worldwide gastroenteritis (Coker et al. 2002 in Ekdahl and Andersson 2004:1). The disease is caused most commonly by the bacterium *Campylobacter jejuni*, with *Campylobacter coli* being less frequently implicated. The epidemiology of the infection is largely unclear but known risk factors for the disease include consumption of undercooked meat, raw milk or contaminated food and water. Direct contact with pets and farm animals, swimming in lakes, and travel abroad, also carry risk of infection, but person-to-person transmission is uncommon between adults. In a study of travellers returning to Sweden, the seasonality of campylobacteriosis in different regions of the world was explored(Ekdahl and Andersson 2004). Of 53,223 people infected with Campylobacter between 1997 and 2003, 28,704 (54%) were travel-associated. Of those with the disease, it was found that travel to the Indian Subcontinent, North and East Africa, and East Asia constituted the highest risk for infection. The study also found that the seasonal pattern of infection was much less distinct in tropical regions compared to temperate places. Evidence of seasonality could only be detected in East Asia (peak incidence in December) and the Caribbean (peak incidence in February-Ibid. 2004).

In summary, travellers' diarrhoea of whatever cause is one of the most commonly encountered infectious diseases today, with up to 50% of international travellers succumbing each year. It can be avoided by not eating raw or undercooked meat, seafood, raw fruits or salad, and avoiding tap water, ice or unpasteurised, raw milk and other dairy products. Of course, this is easier said than done, and therefore the chances for greatly reducing the frequency of this group of diseases are probably minimal. The WHO are working with various agencies (such as the CDC – Centres for Disease Control and Prevention) in order to improve water and food safety around the world, and also to improve insanitary accommodation in places such as tourist resorts. The CDC is also responsible for evaluating sanitation on cruise ships docking in US ports (CDC 2016b).

Education of travellers is also a priority to ensure preventative measures are implemented, with the popular press and women's fashion and interest magazines, such as Women's Health, running articles on how to avoid contracting these diseases. Travellers' diarrhoea is usually self-limiting and requires no specialist treatment in most cases. Of course, no vaccinations are available, due to the multiple causative organisms responsible for the disease, and therefore the traveller must be aware and be wary when eating and drinking abroad. It is questionable that people travelling in the Roman era were aware of these risk factors and whether they were able to take any precautionary measures against contracting diseases at their destination.

# 5.5 Tuberculosis and travel

# 5.5.1 Introduction

As this research examines the relationship between TB and mobility in Roman Britain, the following section examines studies of TB transmission due to mobility today. The account begins by looking at some case studies of TB occurring as a result of using public transport.

# 5.5.2 Air travel and TB

Obviously, travel by aeroplane was not available in the Roman era, but it is a common, fast and efficient method of travelling the world today, and it has been implicated as a method of mobility during which disease transmission occurs. Due to concerns about contracting TB, the WHO have set out guidelines for prevention and control of TB during air travel (WHO 2008). These became necessary after several reports in the early 1990s of TB, and particularly of the most dangerous form, multi-drug resistant TB or MDR-TB, being transmitted from infectious to other passengers and crew during long haul flights. The WHO report this risk to be

quite low but dependent upon several factors including the infectiousness of the person with TB, the susceptibility of those exposed, the duration of exposure, the proximity to the infected person, and the efficiency of cabin ventilation. Susceptibility to infection is increased in immunocompromised people and in children less than five years of age. However, cabin air quality on most commercial aircraft is high but, on long-haul flights of eight hours or longer, increased exposure to people due to close prolonged proximity increases the risk of transmission of *M. tuberculosis*. Nevertheless, this is unlikely to be a higher risk than for any other circumstances where people are together in a confined space (WHO 2008:18). WHO international guidelines for the control of TB require the tracing of flight passengers who sat for longer than eight hours in rows adjacent to someone with pulmonary TB. They also recommend that a person with TB should refrain from all commercial air travel until the patient has had two consecutive negative sputum smears for drug susceptible TB, or two consecutive negative cultures for multidrug resistant TB (WHO 2008; Abubakar 2010).

However, TB is unlikely to be as widely transmitted during flights as a viral infection (such as measles or influenza), due to the larger size of the TB causative microorganism. This is because, whilst there is no agreed standard for air filtration in place, most aircraft used for long-distance flights have high efficiency particulate air filtration that will reduce the risk of TB transmission by filtering out bacteria above  $0.3\mu$ m in size (Abubakar 2010:180). The aircraft ventilation rate is also very efficient, with up to 20 - 30 air exchanges per hour, which is far greater than the rates for offices, homes and other forms of transportation (Byrne 2007:19). Transmission of TB and other infectious diseases within an aircraft cabin is possible, but appears to be due to people being in close proximity for a long duration rather than to the quality of the air (Mangili and Gendreau 2005:991; WHO 2008:18). Whilst flying was not available in the Roman era, this increased risk of disease transmission due to close proximity to fellow travellers would have been true on Roman ships.

Due to concerns of possible transmission of TB during long haul flights, much research has been done to try to establish the risks of this occurring. For example, Byrne's (2007) study calculated TB disease rates among passengers and cabin crew during a five-year period. During this period, a total of 34 TB cases were notified; all were adult passengers on flights exceeding eight hours, with 15 travelling from Africa, six travelling from India, eight from the UK, four from the rest of the world and one from an unknown location, although it was acknowledged that under-reporting could have occurred and true numbers could be higher (Byrne 2007:20). The risk of under-reporting of new cases of infection was also commented on by Abubakar (2010) who reviewed 13 papers on TB transmission during air travel, finding that, only two studies provided convincing evidence of transmission during a flight (Abubakar 2010:179). Until recently it would have been impossible to conclude without unreasonable doubt that the person with TB was infected during the actual flight. However, recent advances in DNA analysis could mean that the strains of TB in travellers can be identified to establish if they are the same. If they are not similar strains, the infection must have been contracted from different sources. Abubakar also pointed out that a large proportion of identified contacts on flights could not be traced, which was frequently due to the lack of information available to the airline companies, and therefore that underreporting of infection could be occurring (Abubakar 2010:177).

#### 5.5.3 Other modes of transport and TB

Although rare, a study of transmission of TB during a combined prolonged train and bus journey revealed that the index case was a 22-year-old man who was smear and culture positive for TB (Moore et al. 1999). The ill passenger travelled on two U.S. passenger trains, the first from Chicago to Pittsburgh over around 12 hours in duration, interspersed with a bus trip from Pittsburgh to Washington D.C. (5.5 hours), and then a second train journey from Washington D.C. to Florida (almost 17 hours). The trains were made up of separate cars, each with its own ventilation system and windows that could not be opened. Air recirculated in each

car via a typical air conditioning filter (not the high-efficiency particulate air (HEPA) filtration method found in aircraft). There were 10 - 15 air exchanges per hour with filters being changed every 15 days (Moore et al. 1999:53). Only four people were identified as having been in close contact with the infected individual. They were exposed to the ill person for approximately one hour in the dining car, where one of the contacts had a conversation with him. The other three were seated at dining tables close by. All four individuals had a TST conversion from initially negative to positive following this contact, although two of them could have been infected elsewhere; this is a Tuberculin Skin Test, administered by the Mantoux Method, using five tuberculin units of purified protein derivative, read at 48 to 72 hours. It was concluded that of 240 people examined after the journeys, only two were likely to have become infected during this trip. This infection resulted from brief proximal contact rather than sharing of airspace for a prolonged period (Moore et al. 1999:52). The study showed that the ill man was highly infectious during this journey but this did not result in TB being passed on to other passengers who shared his airspace during travel. Those that underwent TST conversion did so after brief face-to-face contact, or by being seated near the infected man when he was talking and eating (Ibid. 1999:55). Moore et al. suggest that the risk of infection from someone with active TB when travelling on a long bus or train trip is minimal, although it should be noted that such brief encounters in the dining car resulted in infection being transmitted. It is likely that the initially infected person (the index case), had suffered with active TB previously and was aware from the symptoms he felt that he was possibly suffering a reoccurrence; he was therefore careful to 'bury his face in his hooded sweater' during the journey (lbid.1999:53). This behaviour probably resulted in far less widespread transmission than could have occurred otherwise.

It is generally considered from these case studies, that people need to be in close, prolonged contact in order for TB to be transmitted, but this is not always the situation. A small amount of research has been undertaken on regular, short-term contact during travel. This has mostly focused on school bus journeys; four case studies are now discussed. The first concerns the infected driver of a bus in

Oneida County, New York, USA. A study of the school population showed 32% of the 266 children who took this bus reacted to the tuberculin test, and 19.5% had active primary TB (Rogers 1962:401). Rogers' study successfully established that the exposed and infected children did not have any higher risk of exposure to TB than non-bus users. She also looked at the relationship between infection rates and seating arrangements on the bus. The seats were not allocated; the choice of where to sit was left to chance. However, Rogers noted that the fan heating system within the bus was such that, with the air circulation due to frequent opening of the doors, air would be pushed back from the driver's position at the front towards the rear of the bus (Rogers 1962:404). Another factor of interest concerned the duration of time each of the children spent on the bus. This could be accurately predicted due to the detailed timetable and the use of a limited number of predetermined bus stops. As would probably be expected from previous research, the tuberculin reaction rate tended to increase with longer journey duration times: for children whose journey time was less than ten minutes per day, the tuberculin reaction rate was 21.6%; with increased journey time, by ten-minute intervals, the reaction rate rose to 50% for pupils on the bus for 40 - 49 minutes per day, and to 62.5% for children whose journey lasted 50 minutes or more. The probability of this distribution occurring by chance was calculated as five times in 100 (Rogers 1962:408).

A more recent study concerned the extent of the transmission of *Mycobacterium tuberculosis* among more than 3,300 students in 49 schools in two counties potentially exposed to five school bus drivers with TB in New York, USA (Yusuf et al. (1997). The findings showed that there was no clear evidence of transmission from four of the drivers, but the fifth appeared to have transmitted TB to students and another driver (Ibid.1997:1). The research concluded that the students exposed to TB on a bus were two to 20 times more likely to have a positive TST, compared with students who were not exposed to *Mycobacterium tuberculosis* on a bus (Ibid.1997:3). The driver who infected students appears to have caused 44% of the children he carried on his bus to have a TST conversion. It was noted that this driver was in the habit of arriving at the school 15 to 20 minutes early. The

school would not allow the students to disembark, and therefore they were kept on the bus; as the study time period was during the winter, the windows and doors of the vehicle were kept closed (Ibid 1997:3-4). Of course, this practice would increase the daily duration of contact with the infection for this group of students.

Whilst dated even now, one of the most up-to-date studies of transmission of Mycobacterium tuberculosis among children on a school bus is that of Neira-Munoz et al. (2008). Again, the source of infection was the bus driver, this time working in Hampshire and the Isle of Wight in the UK. Exposed children were assessed using two versions of interferon-gamma release assays. After exposure, 55% (18/34) had positive interferon-gamma release assays and four children developed TB (Ibid. 2008:836). The study found that the bus involved in the incident had a closed ventilation system, combined with a heating system that blew hot air towards the infectious driver and then back towards the children. The incident shows a perhaps surprisingly high transmission rate for the schoolchildren in an enclosed setting after cumulative exposure to an infectious adult on a bus (Neira-Munoz et al. 2008:837). The final, and most recent, study of the possibility of TB transmission on a bus evaluated if using buses and/or minibuses as public transportation was associated with acquiring TB in a high incidence urban district in Lima, Peru (Zamudio et al. 2015). The researchers found that the use of buses/minibuses to commute to work is associated with an increased risk of TB. This has a significant public health implication due to a large proportion of the population relying on this mode of travel for their daily commute (lbid. 2015:2). The highest TB risk was duration of commute time, with TB risk increasing 2.66 fold for individuals with commute times less than or equal to 60 minutes. However, this risk increased by over five fold for people with a commute time of over 60 minutes (Ibid. 2015:5). Regular bus/minibus users were found to be 12 times more likely to have TB than non-bus/minibus travellers (Ibid. 2015:5).

UK guidelines (National Collaborating Centre for Chronic Conditions, 2006) do not recommend the routine investigation of TB contacts on a bus, but the three separate incidents discussed above highlight the need to thoroughly investigate

exposure to TB within these settings. This is especially important since there are strict guidelines pertaining to following up contacts after reports of TB on an aircraft.

Contracting TB during travel is dependent on a number of factors including origin, destination and the duration of travel, with long-term stays at the destination providing the greatest risk (Rieder 2001). If time spent at the destination involves mostly being indoors, then being in a confined space with little air circulation or filtration provides the greatest risk for indoor transmission. However, if a person is spending time outdoors, TB bacteria are rapidly dispersed and killed by exposure to sunlight. Thus the risk of transmission during a single exposure to someone with the infection is very slight (Rieder 2001:1393) although, as discussed above in the case of the man on the train, it is not impossible for this to occur. Of course, people remain at their travel destination for variable amounts of time, and this will have also occurred in the past, with short, fleeting visits for trade and longer visits for other purposes. Today, long-term stays (at least a month in duration) and overcrowded conditions characterise the Hajj pilgrimage, and the likelihood of transmission of TB during this annual mass meeting of Muslims has been explored (Wilder-Smith et al. 2005). The researchers interviewed and tested Singaporean Hajj pilgrims for TB one month before their 2002 trip and again three months following it. A positive result was classed as a conversion of a pre-Hajj negative test result to a post-Hajj positive. The study showed the conversion rate was 10%, which is much higher than that of 1.8% seen after visits of "general" travellers to TB endemic areas. Further cause for concern was that these pilgrims could become a source of TB infection in their own countries when they returned home. As it is a religious requirement that all Muslims who are able to visit the Hajj do so at least once in their lifetimes, this could prove a considerable risk for countries with a high Muslim population (Wilder-Smith et al. 2005:337).

Other research on the risk of TB infection for long-term travellers to areas of high TB endemicity has concluded that the risks are similar to Hepatitis A or malaria for holidaymakers travelling to, in this case, Kenya (Cobelens et al. 2000). However, it

was several times higher than the risks of contracting other vaccine preventable diseases such as cholera, meningitis and typhoid fever (Ibid. 2000:465).

#### 5.5.4 Summary

In summary, there appears to be a conflict between worries and concerns of the potential for TB being transmitted during flights today and between the published data for this occurring, which suggests it is quite a low risk. The data indicate that it is highly unlikely that an individual would contract TB as a result of sharing a flight with an infected person, unless that person was highly infectious and sitting very close, and probably on the flight for a prolonged period (although underreporting is an issue to consider). It is certainly reassuring to learn how aeroplane air filtration systems are set up to work efficiently to remove bacteria and provide regular circulation of clean air to cabins (if they are working correctly and efficiently at all times). However, as discussed, it is concerning that there is no required standard for the filter sizes, and therefore some aeroplanes will be more efficient than others at trapping TB bacteria, with the best infection control being the use of a high efficiency particulate air filtration system. This will remove air-borne particles and micro-organisms above the size of 0.3µm, which would also filter out Mycobacterium tuberculosis bacteria (Abubakar 2010:180). However, all the research papers are careful to point out that contact tracing after a flight is practically impossible, and therefore cases of infection could be under-reported. This under-reporting is even more likely to happen if the onset of illness occurs some time after the flight took place; in this case the link between the two events is less likely to be made.

In conclusion, there are various concerns about infections in travellers themselves and much research has explored the risks of contracting infectious diseases on long haul flights or on cruise ships, some of which have been discussed. However, these studies are only looking at a very specific and small group of susceptible people today. Where research appears to be lacking is in several areas:

- the risks of infectious diseases being passed to native groups in a country by people who have travelled there bringing the disease with them,
- the transmission of infection during car journeys,
- the risks of infection being transmitted during shorter, perhaps regular, journeys such as on the London underground or by bus during the daily commute to work; however, the recent Zamudio et al. (2014) study has examined the risks of TB transmission on buses during the commute to work in Lima, Peru, and appears to be the first study of this type.

Research in these areas would be particularly appropriate for the current project which concentrates on how infectious diseases were transmitted due to travel in the past, when long haul journeys were less frequent, air filtration systems were non-existent and smaller numbers of people were more likely to travel together. In the case of travellers transmitting infectious disease to native populations, it could be difficult to prove where the infection had originated unless the initial source of infection (the "index case") could be identified. However, in the case of TB, strain analysis and next generation sequencing (NGS) of the DNA of ancient TB bacteria could be used to help to trace origins for the infection, as has been previously discussed. Similarly, in the case of the shorter journeys undertaken on public transport, tracing the huge number of people using an underground train in a morning in London, for example, would be very difficult. This may be easier in the case of a bus, which carries a smaller number of passengers on a journey, especially as a few studies of TB transmission on school buses have successfully been undertaken (discussed above). Additionally, it would be easy to research transmission of infection in car sharing passengers because this is another area in which published research appears to be lacking.

A considerable factor to consider with transportation methods and transmission of infection is lack of ventilation. For example, Lygizos et al. (2013) studied natural ventilation in traditional homes in rural KwaZulu-Natal, South Africa as a tool for reducing high rates of TB transmission. They note that there is a paucity of data regarding factors that may affect TB transmission risks in household settings (Lygizos et al. 2013:1). As previously discussed, this paucity of research also exists for the more commonly used day-to-day transportation methods such as buses, trains and cars, but Lygizos et al.'s work is relevant and important to consider in relation to transport methods. This study found that the estimated risk of TB transmission to someone living in the same house, after ten hours of exposure to an infectious TB patient living in a house where windows and doors were closed was 55.4%. This risk dropped significantly upon opening windows (21.5%), and further upon opening windows and doors together (9.6% - Lygizos et al. 2013:4). It is argued that this research could be applied to ventilation in vehicles.

Lygizos et al. go on to note that research into TB transmission has so far tended to focus on health care settings, with airborne infection control strategies having been developed to reduce transmission in hospitals and clinics. However, they indicate that there is increasing recognition of TB transmission occurring in community settings such as households (Ibid. 2013:5). It is argued that transport vehicles, both for public and private transportation, could also be considered. Nevertheless, both scientific and public health communities lack evidence to inform infection control strategies in these settings and this is something which must be addressed, particularly in these times of multiple and total resistance to antibiotics. Also, the time bacteria can survive outside the host needs more investigation as this is a key factor in the extent of transmission within enclosed spaces likely to be encountered whilst travelling. This information would also help to build a clearer picture of how TB was transmitted in Roman transport systems. If survival of the organism outside of the host is long enough, this would perhaps expose more batches of passengers to the disease and one infected individual

could have had far wider ranging effects than just on the people with whom they are closely confined during their own journey.

We have now considered all the background information and research relevant to the transmission of TB due to mobility today, and have, where possible attempted to apply this knowledge to mobility in the Roman period. Now we shall consider the materials and methods used in the current project, starting with a discussion of the skeletal sample chosen for analysis.

#### **Chapter 6: Materials and Methods**

#### 6.1 Introduction

This chapter describes the skeletal sample chosen for study, including the archaeology of the sites from which the skeletons analysed derive, their burial context information where this was available, and biological profiles of each skeleton (sex, age at death, stature and evidence of disease). The author did not examine the skeletons in this study herself due to funding restraints, meaning that travel to the different curation places for the collections would have not been possible, although as the analysis was undertaken by qualified osteologists at each site, this was probably not a major limitation of the study. Nevertheless, the methods used by the bioarchaeologists who originally analysed the skeletons are detailed and discussed. This is important in order to assess how comparable the different methods were for each site. This discussion is followed by the methods used for sample preparation, stable isotopic and statistical analysis.

#### 6.2 The sites and skeletons: introduction

The sites and skeletons in the current project reflect the hypothesis outlined in Chapter 1; that people infected with TB in Roman Britain, namely the part which now forms England, were not brought up locally to where they were buried. As discussed in Chapter 1, the skeletons selected for this study were some of those previously used in the Natural Environmental Research Council funded aDNA study of tuberculosis between Durham and Manchester Universities (*Biomolecular archaeology of ancient tuberculosis in Britain and Europe*, NE/E018564/1, 2007– 2011, Müller et al. 2014a, 2014b, 2014c, 2016). The project at its outset (2007) followed ethical guidelines at both Universities. Samples of bone and tooth had been granted for analysis by curating institutions, and bone and teeth from Roman individuals that were not used in the project ultimately became available for the current research. The individuals studied derive from a variety of Romano-British cemetery sites in England and were selected from the larger Durham-Manchester study samples on account of their burial sites being based on chalk or limestone geology for which the strontium isotope range is known. This was to allow for direct comparisons of resulting data from that isotope analysis. A map of the position of the sites within England, all located on chalk or limestone geology, is shown in Figure 6.1 below:



*Fig. 6.1 Map of England showing the locations of the burial sites used in this project* (Digimap 2016).

Site name	Skeleton number	Lab number	Date range for burial (if known)	Sex	Age at death (vears)
Cirencester,	S	CIRES	, , , , , , , , , , , , , , , , , , ,	Male	18-25
Gloucestershire					
	37	CIRE37		Unknown	9-10
	189	CIRE189		Female	26-35
Poundbury, Dorset	228	POUN228		Unknown	9
	257	POUN257		Unknown	10
	506	POUN506		Unknown	12
	619	POUN619		Unknown	15
	636	POUN636		Unknown	4
	1201	POUN1201		Male	36-45
Driffield Terrace,	4112-13	DRIF13	Late 2 <sup>nd</sup> –	Male?	16-19
York, Yorkshire			early 3 <sup>rd</sup>		
			century AD		
	1124-19	DRIF19		Male	26-35
Easington/Ganstead,	25183	EASN183	2 <sup>nd</sup> century	Male	>45
East Yorkshire			AD		
Baldock,	7230	BALD7230	AD 2-126	Male	26-35
Hertfordshire			cal		
	7498	BALD7498		Female	26-35
Gambier Parry	531	GRPL531		Female?	25-35
Lodge, Gloucester,					
Gloucestershire					
	538	GRPL538		Unknown	8-9
Chester Rd,	512	CHES512		Female	18-25
Winchester,					
Hampshire					
	535	CHES535		Male	26-35
	636	CHES636		Unknown	12-17
Victoria Rd,	96	VICT96		Female	18-25
Winchester,					
Hampshire					
	129	VICT129		Female	18-25

The details of the individuals used for this study are shown in Tables 6.1 and 6.2:

Table 6.1 Sex and age at death of the Romano-British individuals used in this study.

Lab	Skeletal	МТВС	Bone	Tooth
number	elements	aDNA	sampled	sampled
	showing	detection	-	•
	TB/possible	results		
	TB lesions			
BALD7230	Vertebrae	Possible	T11	Mandibular
		positive	vertebra	PM1
BALD7498	Vertebrae	Negative	Lower	Mandibular
			thoracic	canine
			vertebra	
DRIF13	Ribs,	Negative	Rib	Mandibular
	mandible,			PM2
	femora,			
	metatarsals			
DRIF19	Pelvis	Not tested	Rib	Mandibular
				PM2
GRPL531	Ribs	Negative	Rib	Mandibular
				PM1
GRPL538	Ribs	Negative	Rib	Maxillary I2
EASN183	Vertebrae	Negative	Т8	Maxillary
			vertebra	PM1
POUN228	Ribs	Negative	Rib	Maxillary
				M1
POUN257	Ribs	Negative	Rib	Maxillary
				M1
POUN506	Ribs	Not tested	Rib	Mandibular
				M1
POUN619	Ribs	Not tested	Rib	Mandibular
				M2
POUN636	Ribs	Not tested	Rib	Deciduous
				maxillary
				M2
POUN1201	Ribs	Not tested	Rib	Mandibular
				M2
CHES512	Ribs	Not tested	Rib	Mandibular
				canine
CHES535	Ribs	Not tested	Rib	Maxillary
				canine
CHES636	Ribs	Not tested	Rib	Mandibular
_				M1
VICT96	Ribs	Not tested	Rib	Mandibular
				PM2
VICT129	Ribs	Not tested	Rib	Maxillary
				PM1
CIRES	Vertebrae	Negative	Proximal	Maxillary
			left	PM2
			humerus	
CIRE37	Ribs	Not tested	Rib	Maxillary
				M1
CIRE189	Ribs	Not tested	Rib	Maxillary
1	1	1	1	canine

Table 6.2 Details of TB lesions identified in each skeleton in the original report, and skeletal<br/>elements sampled

In the sample used for this study, four of the 21 individuals (19%) demonstrated bone changes of the vertebrae typical of TB. Rib lesions occurred in 17 of the 21 (81%) skeletons. Thus, the majority of skeletons analysed had non-specific new bone formation on their ribs, not pathognomonic for TB. BALD7230 had a "possibly positive" TB aDNA result (Müller et al. 2014a:183), not forgetting that a positive DNA result does not support TB causing the bone lesions, and a lack of a positive DNA result does not mean the individual did not have the disease at some point in their lives.

#### 6.3 The sites

The following section looks at available information from excavations at the sites where the skeletons were found. It also gives any details of the burial positions and contexts of the individuals where these are available. Several sites remain unpublished and more information is available on some places than others. Each site and skeleton will now be considered in turn, but if some information is noted for some skeletons and not for others, this is because the information was unavailable.

#### 6.3.1 Cirencester

A road building development took place to the South West of Cirencester between 1969 and 1981, allowing the Cirencester Excavation Committee to investigate an area lying between the walls of *Corinium Dubunnorum* and the ampitheatre. Excavation before the construction of the western relief road (which was completed in 1974) found over 450 Romano-British burials from Bath Gate cemetery that lies either side of the Fosse Way as it leaves the Roman town travelling in the direction towards Bath. The earliest finds of the Roman period are early military, but these were unstratified and unassociated with any burials. The cemetery appears to have continued in use until the 5<sup>th</sup> century AD, but excavation

evidence suggests that the town was in decline by this time. The geology of the area is oolitic limestone (McWhirr et al. 1982).

## The Bath Gate Cemetery 1969 - 1976

A total of 453 Roman burials were recorded between 1972 and 1976, each of which was given a skeleton record number. A previous 26 burials found in 1969 were given a skeleton record letter, for example, Skeleton S in this study.

## Skeleton 37

This complete burial of an adolescent aged 12 - 13 was not cut or sealed by any other burials. A coin of the House of Constantine (dated AD 330-345) was found in the grave shaft but not associated with the body. The body was oriented south - north with the head towards the south, the skull lying to the right side, and in a supine attitude, with straight legs and hands on the pelvis. The grave was at a depth of 0.22m with stones placed at the head and the feet (Wells 1982).

## Skeleton 189

This grave was not cut or sealed by another burial, so was interpreted as being of the latest chronology in the burial sequence of the area. The skeleton of a female aged 25 – 30 was nearly complete and oriented south-north, with the head towards the south and the skull leaning on the right side. The burial was supine with an indeterminate leg position and arms at right angles to the body. The burial was at a depth of 0.53m and one coffin nail was recovered from the grave. The excavators offered no position for this nail and no interpretation of its presence, although the presence of just one nail does not seem firm enough evidence to suggest the burial was made within a coffin (Wells 1982).

## Skeleton S

This nearly complete skeleton was that of a male aged 20-21 who was oriented south-north with the head at the south. The skull was leaning to the right side and the skeleton was supine with bent legs and an indeterminate arm position. The

grave was at a depth of 0.69m. Inhumation S was identified by the osteologist as being most likely to be affected by TB. This individual's first lumbar vertebra showed a type of cavitation and collapse similar to Pott's disease. (It was acknowledged that this could be caused by some other type of infection as diagnosis was not confirmed at the time) (Wells 1982).

#### 6.3.2. Poundbury

Excavations were carried out on the north-west outskirts of the present town of Dorchester in an area which is now the Grove Trading Estate and has been since 1964. The excavations took place in advance of the industrial development which was due to start in 1968. During the 1914-18 war, an area to the east of Poundbury Camp was used as a Prisoner of War camp and Roman burials were found during its construction. After the First World War, the site then lay unused and uncultivated until 1940 when the camp was extended for the Army and further Roman burials were found. The Army camp was in use until 1964 when redevelopment took place to make the present day trading estate, and further Roman burials were also discovered at this time. Trial excavations took place in 1966 and 1967 with full-scale excavation in 1968, which was extended in 1969 and completed in 1972. Between 1973 and 1976, the main Roman cemetery was excavated with further excavations in 1979, 1980, 1986 and 1987. The late Roman burials were found in this main cemetery, and include all of the five individuals in analysed in the current research. The burials in the main cemetery were single, discrete inhumations in rectangular, vertical sided, flat-bottomed grave cuts aligned west to east. Burial in wooden coffins held together with iron nails was the commonplace rite. The cemetery consisted of 1,114 graves, 1,028 of which were excavated. None of the individuals in the current research project had lead or stone coffins or any grave goods. The geology of the Poundbury area is upper chalk with few superficial periglacial deposits (Farwell and Molleson 1993) which is of relevance to the strontium isotope analysis, and will be discussed in Chapter 8.

## Skeleton 228

This individual was 9 years old and was buried in a grave 102cm deep, 185cm long and 61cm wide. Coffin nails and a coffin stain were present. The skeleton was buried in what the excavators called 'standard attitude' (that is with the head to the west, in supine position, with legs extended and arms by the sides) (Farwell and Molleson 1993).

# Skeleton 257

This was a 10-year-old individual buried in a grave with coffin nails present. The grave cut was 25cm deep, 183cm long and 56cm wide. The skeleton was found with the head to the west, in supine position with legs extended and hands on the pelvis (Farwell and Molleson 1993).

# Skeleton 506

This was a 12-year-old individual buried in a grave 81cm deep, 183 cm long and 61cm wide. There were coffin nails present and the skeleton was found with their head towards the west, in a supine position with the wrists crossed over the lower abdomen (Farwell and Molleson 1993).

# Skeleton 619

This was the burial of a sub-adult aged 15 years old. It was not possible to reliably determine the sex of this individual. The grave cut was 109 cm deep, 185 cm long and 69 cm wide, with coffin nails present. The skeleton position was described as standard, that is with the head towards the west, and the body being laid out supine with legs extended and arms by the sides. The burial had been disturbed, but the nature and extent of this disturbance was not documented (Farwell and Molleson 1993).

# Skeleton 636

This was the burial of a 4 year old in a grave cut 28 cm deep, 124 cm long and 61cm wide. Coffin nails were present and the skeleton was buried with the head towards the west, in a supine position but with disturbance. The grave had been

disturbed, but the nature of this disturbance and how much damage had been caused was not documented (Farwell and Molleson 1993).

#### Skeleton 1201

This was a burial of a 36-45 year old male. The grave cut was 25cm deep, 180cm long and 53cm wide. The skeleton was buried in a supine position, with the head towards the west, and the left forearm flexed slightly inwards. The skeleton was poorly preserved but sufficient remained to see that the upper chest was compacted and this was interpreted as being of pathological origin, although the suspected pathological cause was unspecified in the site report and the nature of the compaction was not described (Farwell and Molleson 1993).

## 6.3.3 Driffield Terrace, York.

Driffield Terrace is located south west of the city walls at the Mount, which is the highest point in the area (Montgomery et al. 2011:141), and was the area through which the road to Tadcaster (Roman Calcaria) ran (Ibid. 2011:145). The site is approximately 0.6km south west of Mickelgate Bar (the south west entrance to the medieval walled city; the medieval walls are thought to correspond to the line of the Roman defensive circuit around the Roman town). Driffield Terrace lies within the boundaries of one of York's most important Roman cemeteries, which was in use for the whole of the Roman period. This cemetery includes examples of the diversity of burial types including cremations and a wide variety of burial styles (Ibid. 2011:141). The geology of the area is Bunter and Kemper sandstones overlain by drift geology that is generally boulder clay over lacustrine clays with deposits of sand and gravel lying within and over the clay in places. Between 20<sup>th</sup> June and 30<sup>th</sup> August 2005, York Archaeological Trust excavated land at 6 Driffield Terrace in advance of alterations to the garden layout. Previous to this work, during 2004, Field Archaeological Services had excavated the site and found Roman burials between post-medieval garden walls. The inhumations at 6 Driffield Terrace numbered 24, all of whom were male and age estimation placed them all

in the young or middle adult age groups (that is 19 – 45 years of age). Eighteen of these individuals had been decapitated (Ottaway 2005).

#### Skeleton 4112-13

This was a 16 to 19 year old presumed male (some epiphyses were still unfused so sex estimation was not confirmed). He was buried in a grave aligned north east to south west, the grave cut of which measured 190cm by 80cm and was 37cm deep. The head of the skeleton was located at the north east end of the grave cut, and had been buried in a wooden coffin, the 62 nails remaining being organised in a rectangle around the skeleton. He had been buried wearing hobnail boots and was accompanied by three small slipped Nene Valley colour coat beakers, all of which were complete. There were also ten sherds of an indented, dimple beaker and a greyware pot. This individual was positioned with his right arm alongside his torso, hand next to right hip and with his left arm gently flexed at the elbow, and his hand over his pubic area. There was visceral surface woven and lamellar new bone formation on the ribs and mandible (Caffell and Holst 2012).

## Skeleton 1124-19 (SK 19)

This individual was the topmost burial of a triple burial with SK 21 and SK 22, all of whom were in a wooden coffin/box with horse bones. SK 19 was buried in a supine, extended position with the right arm away from the body and slightly flexed at the elbow, and the left arm behind the back. SK 19 was a young, middle adult male (aged 26-35). He was 172cm tall and had new bone growth on the ribs and trauma/infection of the left auricular surface. He was decapitated by a single chop through the arch and body of vertebra C3. This blow extended into both ascending rami of the mandible from the posterior side (Caffell and Holst 2012).

## 6.3.4 Easington

Seven inhumations and three cremations and disarticulated bone were recovered from excavations along the route of the Easington to Ganstead gas pipeline,

Holderness, East Riding of Yorkshire. The cremations were dated to the Roman period and the inhumations as Iron Age to late Roman (late Roman being post 3<sup>rd</sup> to 4<sup>th</sup> century AD).

#### Skeleton 25183

This individual was buried on the right side, left arm flexed 90 degrees at the elbow with the legs extended at the hips but flexed at the knees. The body was oriented north to south, with the head to the north, and was buried in a simple grave cut dug into the boundary ditch with deliberately deposited animal bone accompanying it. It was dated as 2<sup>nd</sup> century AD. Although the individual was aged as 46+, he was difficult to age as the multiple ageing techniques gave varied age ranges. Some parts of the skeleton suggested a younger age, but it was decided to place him in the 46+ age group from an "average" of the observations recorded (Caffell, pers comm. 2014). The individual had various pathological changes, including lytic lesions on lumbar vertebrae one, two, three and five and thoracic vertebrae five to 10, probably as a result of TB. There were also signs of *cribra orbitalia* (Caffell, pers comm. 2014).

## 6.3.5 Baldock

The ancient and modern settlements at Baldock lie in a shallow bowl on the northern scarp of the chalk ridge of North Hertfordshire, between London and Cambridge on a north-eastern extension of the Chiltern Hills. Occupation has been continuous here since the third millennium BC with the first nucleated settlement developing on the site during the early first century BC. (Stead and Rigby 1986:84). This occupation was continuous, although no archaeological evidence for settlement from early medieval times has ever been found (Burleigh and Fitzpatrick-Matthews 2010:5). The road south-south-west to *Verulamium* lies along the foot of the hills, climbing the slope away from Baldock. Unpublished excavations which took place in 1988 to 1989 at Jack's Hill, four kilometers to the south near Graveley, showed this road existed as a track by the

early Iron Age. Evidence for continued existence in the Roman period was also confirmed (Matthews and Burleigh 1988). Another road passes between a gap in the hills to the south east of Baldock. This road travels towards Braughing and Camulodunum. There is no existing dating evidence for the origins of this road but it is thought to also have originated in prehistoric times as a trackway (Fox 1923:170). The road is clearly an engineered Roman road and continues from Baldock in a north westerly direction towards the Romano-British settlement at Sandy and the major road junction at Godmanchester (*Duroviguto*). These roads all converge at a point just south of the Romanised road, the Icknield Way (Burleigh and Fitzpatrick-Matthews 2010:5). Baldock lies on the Upper Cretaceous deposits of the Middle Chalk, a rock consisting mainly of white chalk with Melbourn Rock at its base. This chalk bed is approximately 67m thick. Around 60 million years ago, the chalk underwent an uplift, raising it above sea level and resulting in a period of erosion that took place in the early Tertiary period. During the Eocene, the sea flooded the area and levelled out the surface of the chalk. A subsequent Late Eocene marine transgression laid down the London Beds, which are a thick dark grey clay deposit up to 90m thick. These were again eroded, exposing the chalk surface throughout most of Hertfordshire and leaving only isolated pockets of clay remaining on the southern and eastern slopes of the Chilterns (Burleigh and Fitzpatrick-Matthews 2010:5). The chalk is a major aguifer containing hard, generally potable, water with dissolved calcium bicarbonate in it (Hopson et al. 1996:89); this varies seasonally with a maximum occurring around March. It is very susceptible to drought (Burleigh and Fitzpatrick-Matthews 2010:5-6).

The two Roman skeletons used in this study were excavated from an inhumation cemetery which included 191 individuals and approximately 500 cremation burials, which were probably earlier in date than the inhumations. The cemetery was excavated in 1986 beside the Royston Road and had originally been discovered by trial trenching in 1969 (Burleigh and Fitzpatrick-Matthews 2010:19). The excavation report has not yet been published and the skeletal analysis was obtained by pers. comm. from the site osteologist, Jacqueline McKinley (McKinley

1993). Following are her analysis for both of the individuals. Note no burial context report exists for either of these skeletons.

#### Skeleton 7230

Approximately 95% of this skeleton was recovered. The individual was a male aged as an older mature adult (30-45 years). He suffered from ante-mortem tooth loss and cribra orbitalia. Vertebral collapse was observed in thoracic and lumbar vertebrae (McKinley 1993).

## Skeleton 7498

94% of this skeleton was recovered. The individual was a mature adult female, aged between 23-45 years. There was evidence of cribra orbitalia. There were destructive lesions observed in the left first proximal foot phalanx, the lower thoracic and three lumbar vertebral bodies (queried to be tumours). Vertebral body collapse was noted in the 12<sup>th</sup> thoracic vertebra. New bone formation was observed in the thoracic vertebral bodies and osteophytes were noted on the thoracic and lumbar vertebrae. This individual exhibited a dental morphological variation; she retained the mandibular left deciduous second molar and the mandibular left second molar was absent (McKinley 1993).

# 6.3.6 Gambier Parry Lodge, Gloucester.

A large mansion called Gambier Parry Lodge was demolished in 1983 to make way for a housing development. The Lodge itself appears on a 1972 Ordnance Survey map as a large building 100m east of the A38 Tewkesbury Road, and approximately 1.5km north of Gloucester city centre at grid reference SO 8319 NE. Archaeological finds were not expected during demolition and redevelopment of the area, but they were found and they included the remains of a 1<sup>st</sup> century building and a Roman burial ground dating to the 2<sup>nd</sup> to 4<sup>th</sup> centuries containing 125 burials. In 1984, Western Archaeological Trust discovered about 70 more burials; about half of the skeletons were headless and some had unusual coffins.

The report for this site remains unpublished and there is very little information available about the burials. The osteological reports were from C. Roberts (pers. comm. 2014).

## Skeleton 531

This individual was a female aged 25 to 35 who had calculus and periodontal disease. She was reported as having tuberculosis of the cervical vertebra with visceral surface woven new bone formation on the ribs (Cameron and Roberts 1984).

## Skeleton 538

This individual was buried with some accompanying animal bones and was aged between 8 and 9 years, and hence sex could not be determined. Visceral surface woven new bone formation on the ribs was observed in this person (Cameron and Roberts 1984).

## 6.3.7. Winchester

The excavations of the skeletons from Chester and Victoria Roads took place between 1971 and 1986. During this time, excavations were undertaken all around Winchester, in the northern, western and eastern suburbs of the Roman town. Evidence was found of both settlements and cemeteries during this work. At Chester Road in the eastern suburbs, late Roman inhumation cemeteries were discovered, with 117 burials being excavated and divided into 19 phases and grouped into six periods, with the earliest being from the late 3<sup>rd</sup> century and the latest from the late 4<sup>th</sup> century (Ottaway et al. 2012:xxiv). The two skeletons in this project were buried in the Victoria Road West area (a total of 112 inhumation and four cremation burials). These burials were dated from the late 3<sup>rd</sup> century to the late 4<sup>th</sup>/early 5<sup>th</sup> centuries.

#### (i) Chester Road

# Skeleton 512

This skeleton belonged to a female aged between 17 to 25 years old. She formed half of a double burial in a shallow grave cut with skeleton number 535 (see Figure 6.2). She was buried oriented west to east, with her head to the west, in a supine position, and associated with a possible coffin (3 iron nails were found) with no grave goods present. Her skull faced left and was tilted backwards, and her right arm was flexed outwards across the body of skeleton 535. Her left arm was reported as "missing", her spine as having a slight curvature, and her right leg was straight. The position of her left leg was not recorded (Ottaway et al. 2012: 321). She is reported as having one rib affected by an inflammatory reaction and possible cribra orbitalia. (Roberts, pers. comm. 2014).

# Skeleton 535

This individual was a 25 to 35 year old man who formed the other half of the double burial with skeleton 512 above. He was oriented west to east, with the head to the west, and had a possible coffin (3 iron nails found) and no grave goods. The skeleton was reported as having been disturbed and positioned in a supine, extended manner with his skull turned to the right shoulder and his right arm straight by his side. His left leg was bent at the knee and his right leg was straight. His left arm was described as 'apparently absent' and it was suggested that the right arm of skeleton 512 could be the left arm of skeleton 535 (Ottaway et al. 2012:321). The pathology reports shed no further light on this situation. The skeleton also had evidence of periosteal new bone formation of the ribs.



Fig. 6.2 Double burial of Chester Road skeletons 512 and 535 (Ottoway et al. 2012:322)

#### Skeleton 636

This was a sub- adult burial aged between 12 and 17 years of age and thus of indeterminate sex. The burial was oriented south to north, with the head to the south, and the presence of a wooden coffin and one pottery vessel were reported as accompanying the deceased (see Figure 6.3). Ottaway et al. (2012:335) described the burial position as being prone and extended with the skull turned towards the left and arms flexed, with the hands resting on the anterior of the pelvis. The legs were crossed and there was evidence of the presence of a coffin, which was confirmed by the presence of 18 iron nails and charcoal staining on the base of the grave. A miniature greyware jar was situated alongside the right foot. The only pathology recorded was a lung infection causing periosteal new bone formation on the ribs (Roberts, pers. comm. 2014).



Fig. 6.3 Skeleton 636, Chester Road. (Ottaway et al. 2012:336)

## (ii) Victoria Road

## Skeleton 96

This individual was a 17 to 25 year old female who was buried oriented west to east in a prone position with her head to the west and leaning towards the left. There were no signs of a coffin (see Figure 6.4). Ottaway et al. (2012:286) described the grave cut containing skeleton 96 as shallow and irregular. The skeleton was lying slightly twisted on the right side with her skull on the right side and the hands probably meeting the side of the pelvic area. A copper alloy bracelet (now lost) was positioned round the lower arm bones of the right arm, slightly below the elbow (Ottaway et al. 2012). She had a fracture to her right clavicle, and cribra orbitalia. She also had new bone growth on several of her ribs indicating a lung infection which could be TB (Roberts pers. comm. 2014).



Fig. 6.4 Skeleton 96, Victoria Road. (Ottaway et al. 2012:287)

#### Skeleton 129

This individual was a female aged between 17 and 25. She was buried in a south west to north east alignment, with the head to the south west, in a supine position with her legs crossed (see Figure 6.5). A wooden coffin was reported as having been present, the evidence for the presence of a coffin inferred from the presence of 21 iron nails in what were described as 'appropriate positions' and a wood stain on the base of the grave (Ottaway 2012:291). The copper alloy unidentified items and an iron pierced strip were interpreted as possible coffin fittings. A coin, bone pin and copper alloy strips were also discovered in the grave fill. Reported pathology showed this individual was affected by cribra orbitalia. She also had three right ribs with new bone formation indicative of the presence of a lung infection, and possible tuberculosis (Roberts pers. comm. 2014). Ottaway et al. (2012: 291) described the grave cut of this skeleton as being 75 cm deep. The skeleton was well preserved but badly disturbed, mainly in the region of the upper body. The burial position was supine, extended, and with the skull on the left side and tilted slightly backwards. The hands were positioned by the sides and the legs were crossed (right over left) above the knees (Ottaway et al. 2012).



Fig. 6.5 Skeleton 129, Victoria Road (Ottaway et al. 2012:292)

#### 6.4 Osteological methods

The sex and age at death for each skeleton were taken from the original skeletal reports if these were published and available (see Table 4.1) or, if unpublished, directly from the site bioarchaeologist. These reports used standard osteological methods to estimate sex and age at death, which will be discussed below. In the case of non-adult skeletons where sex cannot reliably determined, sex was recorded as 'unknown' (See Table 6.3, following page).

As previously mentioned, it should be noted that the author did not make any attempt to estimate age and sex of the skeletons herself. This was not essential for the current research since the information was available. It is therefore necessary to make a critique of the techniques used by bioarchaeologists at each site in terms of reproducibility and accuracy.

Site and site/osteology report	Age estimation methods	Age estimation references	Sex estimation methods	Sex estimation references
Baldock (McKinley 1993) (unpublished)	Tooth wear, age related changes, skeletal and dental development.	Van Beek (1983), Brothwell (1981), Ubelaker (1989)	Standard sexually dimorphic traits.	Not mentioned
Driffield Terrace, York (Ottaway 2005 and Caffell and Holst 2012)	Skeletal development and fusion, dental development and wear.	Buikstra and Ubelaker (1994), Scheuer and Black (2000a, 2000b), Brothwell (1981). Ubelaker (1989), AlQahtani et al. (2010)	Pelvic and skull morphology (adults).	Buikstra and Ubelaker (1994)
Gambier Parry Lodge (Cameron and Roberts 1984) (unpublished)	Dental development, epiphyseal fusion, pubic symphyseal changes, degenerative change.	Van Beek (1983), Ubelaker (1989)	Pelvic morphology, skull and sacral morphology. Measurements of femoral and humeral heads and ischio-pubic and sacral indices.	Not mentioned
Easington (Caffell 2014) (unpublished)	Pubic symphysis, dental development.	Scheuer and Black (2000a), (2000b) and Cox (2000), Ubelaker (1989)	Skull and pelvis	Mays and Cox (2000)
<b>Poundbury</b> (Farwell, D.E. and Molleson, T.L. 1993)	Pubic symphyses, Dental development.	Schour and Massler (1941)	Sexual dimorphism	Ascádi and Nemeskéri (1970),Phenice (1969),Kelley (1978)
Chester Rd, Winchester (Ottaway, P.J., Qualmann, K.E., Rees, H. and Scobie, G.D. 2012)	Dental development and wear.	Brothwell (1981) Ubelaker (1978)	Sexual dimorphism	Not mentioned
Victoria Rd, Winchester (Ottaway, P.J., Qualmann, K.E., Rees, H. and Scobie, G.D. (2012)	Dental development and wear.	Brothwell (1981) Ubelaker (1978)	Sexual dimorphism	Not mentioned

Site and site/osteology report	Age estimation methods	Age estimation references	Sex estimation methods	Sex estimation references
<b>Cirencester</b> (McWhirr, A., Viner, L. and Wells, C. 1982)	Cranial morphology, long bone measurements, dental development.	Brothwell (1972) Schour and Massler (1941) Gustafson and Koch (1974)	Cranial morphology.	Brothwell (1972)

Table 6.3 Methods of age and sex estimation at each site (as detailed in site reports).

#### 6.4.1 Methods of sex estimation

In an archaeological context, it can be possible to estimate sex through the analysis of skeletal remains, although this cannot be achieved successfully for sub-adults who have yet to pass through puberty. In terms of accuracy of sex estimation, probability states that a pure guess as to the sex of any given individual will be accurate 50% of the time. Some skeletal elements, for example the cranium, if analysed carefully, can be used to estimate sex correctly 80 – 90% of the time (White and Folkens 2005:385). In general, bioarchaeologists note that for all parts of the female skeleton, females are usually smaller in size and are less robust in build. However, normal variation will always produce some small, gracile males and large robust females. For this reason, bioarchaeologists tend, as for the skeletons from the sites in this study, to use the cranium and pelvis to determine sex more accurately (Ibid. 2005:386). All of the following methods are inappropriate for sub-adult individuals who have not reached puberty because their bones have not developed the necessary secondary sex characteristics. In the case of non-adult skeletons, sex was recorded as unknown. In all other skeletons, the methods of sex estimation used were following recognised methods and these are briefly evaluated below.

#### (i) The Skull

Due to variation in cranial shapes and sizes between and within populations, bioarchaeologists are advised to use the pelvis, if preserved, to estimate sex and to use the cranium as corroboration if the pelvic features are unclear or not well preserved. At times there can be intra-population variation and the sex-related features may not be clear, even in the pelvis.

The standards currently used by bioarchaeologists for sex estimation using the cranium are those of Walker (Buikstra and Ubelaker 1994). This method uses a five point scale looking at size and shape of the nuchal crest, the mastoid process, the supraorbital margin, the supraorbital ridge/glabella and the mental eminence, with the more gracile, feminine features falling at one end of the range and the more robust features of males at the other (White and Folkens 2005:387). Previous standards used were those suggested by Giles and Elliot (1963) who used nine standard cranial measurements to sex the skull. In testing these criteria, Meindl et al. (1985) found that older individuals show an increasing tendency towards more masculine features, and so suggested that sex should only be estimated from pelves. It is argued here that the bioarchaeologists in this study considered the sexually dimorphic features of pelves in addition to cranial morphology in order to estimate the sex of the skeletons so sex estimations were probably more accurate than if cranial morphology alone was used.

#### (ii) The pelvis

Female os coxae and sacra are smaller and less robust than those of males, and pelvic inlets are relatively wider. The sciatic notches of female os coxae are relatively wider than those of males, and females have longer pubic bones, including the superior pubic ramus. The sub pubic angle, made up of the inferior edges of the two pubic rami, is larger in females than males. The acetabulum of males tends to be larger (White and Folkens 2005:394). In 1969, Phenice
published a new method of sex estimation using the pelvis which has proved to be the most accurate method of determining sex of an individual from his or her skeletal remains.

Sex estimation using the Phenice method should only be undertaken on fully adult material, (which is presumably why non-adults were recorded by the bioarchaeologists at the sites in the current research as not determined), when accuracy of assigning sex to an individual ranges from 96 to 100%, although Lovell (1989) argued that this becomes less accurate in older adults. White and Folkens (2005:398) suggest that bioarchaeologist's experience still plays a big role in the success of the sexing method which could lead to limitations on accuracy of sex estimation. These considerations also need to be borne in mind when estimating age of skeletons, the methods used by bioarchaeologists in this study shall now be discussed.

# 6.4.2 Methods of age estimation in non-adult skeletons

# (i) Dentition

Teeth tend to survive better than bone in archaeological burial contexts and this is useful for estimating age of archaeological skeletons. Tooth development is more closely associated with chronological age than skeletal bone development because it is mainly affected by genes (White and Folkens 2005:364) and is less likely to be affected by nutrition and disease than bone development (Lewis and Gorn 1960:70, Smith 1991:143). In studies of children, girls reach most tooth development stages earlier than boys (Berkovitz et al. 1986:177, Hillson 1996:211, Hillson 2005:210) with the exception of the third molars (Hillson 1996:211). Some human populations are in advance of others for dental development stages (Hillson 1996:211, White and Folkens 2005:364).

#### How dental development methods were developed

Age may be estimated from the teeth by comparing the unknown individual with a chart or atlas showing the mean stage of development of the dentition (White and Folkens 2005:365). The best-known atlas is probably that of Schour and Massler (1941) which consists of a series of diagrams covering an age range from five months in utero to 35 years (AlQahtani et al. 2010:4). This atlas was produced as a result of radiographic data and anatomical studies of autopsy specimens and this atlas or variations upon it are still used today (Ibid. 2010:4). Since 1941, Gustafson and Koch (1974) used anatomical and radiographical data to construct a schematic representation of tooth formation and eruption for ages ranging from prenatal to 16 years old. Ubelaker (1978) compiled an atlas of dental formation and eruption among Native Americans, which was later updated (Ubelaker 1989) and was also based upon data collected from radiographs and anatomical examinations. AlQahtani et al. (2010) developed an updated atlas to estimate age using tooth development and alveolar eruption for humans aged between 28 weeks in utero and 23 years. This was based upon data from known age-at-death skeletal collections and dental radiographs of living individuals.

# Problems of use of dental development atlases for ageing archaeological specimens.

Dental development can vary with sex (Berkovitz et al. 1986:177, Işcan 1988:203) and population differences (Işcan 1988:203). As has been discussed, it is difficult to estimate sex of a non-adult skeleton, data from archaeological specimens must be compared with dental development ages for both males and females and an average taken (as per the recommendations of Moorrees et al. 1963a:211, Smith 1991:165). Also, the age of development of the dentition of an archaeological individual or population to which that individual belongs may not be directly comparable to the population used for the standard developmental atlases (Moorrees et al. 1963b:1500, Smith 1991:165). Additionally, the experience of the

bioarchaeologists in recognising sequential stages of tooth formation is a known cause of variation of age estimated (Moorrees et al. 1963b:1500). As the seven sites in this study had different bioarchaeologists at different stages of experience, this could indeed be an issue to take into account with comparability of ages in the current study. Finally, as the sites were excavated over a long period of years from the 1970s into the 2000s, different standard atlases were available, and, as can be seen in Table 6.8, different methods were used. As has been discussed, these standards are all based on the same method, but slight differences may occur between them leading to possible differences in age estimation compared to the ideal situation where only one standard developmental atlas would be used.

(ii) Bone growth and development

This method of age estimation is used for sub-adult skeletons alongside tooth development and is now discussed. It utilises the fact that each bone has a main primary ossification centre that appears at an age within an age range, with accompanying epiphyses (secondary ossification centres) that appear and fuse also at an age within certain age ranges (Buikstra and Ubelaker 1994). Epiphyseal fusion starts in the bones of the elbow at age 11 - 12 years and ends with fusion in the bones of the knee around 17 - 19 years of age, with female epiphyses fusing one to two years earlier than those in males (Scheuer and Black 2000a and 2000b). The length of long bones (without their epiphyses) can also be used to estimate age at death in non-adults (Humphrey 2000).

However, there is considerable variation in maturation times within any normal population group. There is then the added complication in that mean differences are present between different groups from around the world, with a great range of variability being observed between different populations, between the sexes and between individuals of the same population (Scheuer and Black 2000a:11). These differences are determined by genetic and nutritional factors (Brothwell 1981:68, Scheuer and Black, 2000a and 2000b). Another limitation is that actual sex and known age at death collections to use as standards for comparison are rare in

archaeology with only two sizeable collections being in the UK. These two collections are from London, one from St Bride's Church, Fleet Street (18<sup>th</sup> to 19<sup>th</sup> century AD) and the other of a similar date from Christ Church, Spitalfields (Scheuer and Black 2000a:12), although these may not make a good direct comparison with the Roman period skeletons in this study.

Although bioarchaeologists at Baldock, Driffield Terrace and Gambier Parry Lodge used skeletal development in their ageing methods, those at Easington, Poundbury and Chester and Victoria Roads, did not mention them at all, so it is assumed they were not used. Although there are genetic and nutritional factors affecting skeletal development rates, it is unfortunate that so few bioarchaeologists made use of this technique, even as a secondary method. As only three out of seven of the sites had skeletal age estimated by this method, this leads to less of a fair comparison of results than for ageing determined by the almost unanimous method examining dental development.

The methods of age estimation of sub-adults using recognised standards of skeletal development shall are now briefly considered and evaluated.

(iii) Skeletal development in sub-adult ageing

As for dental development and eruption, the standards for comparison between known and unknown age skeletons have been mainly established based upon American and European documented samples, and may not directly apply to other parts of the world's population. Climate and diet also have a considerable effect upon maturation rates (Scheuer and Black 2000a:11). Brothwell (1981:64) states that assessments of age at death based on skeletal remains are more likely to be accurate for non-adults or young adults. However, surprisingly, the bioarchaeologist at Poundbury, where many of the dead were sub-adults, did not use this technique, and nor was it used at Easington, Cirencester and Chester and Victoria Roads. The bioarchaeologists who described using skeletal development for non-adult ageing were those associated with the sites of Baldock, Driffield Terrace and Gambier Parry Lodge. It must be noted, however, that there was only one individual who was a sub-adult (GRPL538, who was aged 8 to 9) amongst the skeletal sample. All other individuals from these sites were aged 16 or above.

The bioarchaeologist who examined the Cirencester individuals in this study (Wells 1982) was the only one in this study to look at the skull in terms of ageing of skeletons. Brothwell (1981:65) believes the only accurate feature of the skull to be used in sub-adult ageing is the dentition, as ageing by using the cranial sutures has fallen out of favour, which could explain why none of the other bioarchaeologists used the method at all in their sites. Brothwell does, however, state that in 'very immature' individuals, the ossification sequence of the occipital bone can be useful for age estimation (Brothwell 1981:65), although he gives no indication of the age range that 'very immature' covers.

Sub-adult age has been estimated by the bioarchaeologist at Cirencester by measuring long bone length. This method is not as accurate as other ageing methods due to a range of variability between individuals and populations (White and Folkens (2005:373). White and Folkens suggest the use of data provided by Ubelaker (1989) as standards for comparison. However, this method could be fraught with difficulties; the children in the sample used for this study were sick individuals and could have been sick for a prolonged period, which is likely to have impacted upon their bone growth and development. Long bone length would therefore not be an accurate measure of skeletal age if used on its own, but may be acceptable if used in conjunction with dental development and eruption, which has been proven to be less likely to be affected by illness (Lewis and Gorn 1960:70). This factor is perhaps one that was considered by the bioarchaeologists at the other sites studied before rejecting the method. Or perhaps it was because use of the method for ageing skeletons has questionable accuracy.

Estimating sub-adult age from epiphyseal closure is a better method than long bone measurements because it is subject to less intra- or inter- population variation (White and Folkens 2005:373). The epiphyses fuse at a known rate and

in an orderly fashion. However, the exact chronological ages at which these events occur vary by individual, by sex and also by population. One point to bear in mind is that epiphyseal fusion for several skeletal elements actually overlaps with the conclusion of tooth eruption (White and Folkens 2005: 373) so the use of this technique would complement the use of the dental development and eruption methods previously discussed. Easy to use graphs of the timing of the fusion of different epiphyses for males and females are provided in Buikstra and Ubelaker (1994). However, despite this ease of use, bioarchaeologists must bear in mind that some of the major problems with ageing the skeletal remains of juveniles are the ineffectiveness of most methods for sex estimation for skeletons in this age category (Scheuer and Black 2000a:17).

#### 6.4.3 Adult age estimation methods

It is far more difficult to estimate age of adults than it is for sub-adults, because in the adult, all of the permanent dentition will have erupted, epiphyses will have fused and cranial sutures are very likely to have also fused. Degenerative skeletal changes can be examined but, if present, these will occur at different rates depending on the individual and his or her life experiences (Brothwell 1981:146, Mays 1998:127, Katzenberg and Saunders 2008:358). Dental wear can be examined, but this does not occur at a fixed rate and is highly dependent on the type of diet consumed, with more rough and unrefined foods causing tooth wear to occur much faster than more processed, smoother foods (Berkovitz et al. 1986:176). By far the best and most accurate methods of age estimation in adults would be to use as many methods as is feasible to try to eliminate as far as can be expected any individual variation according to a specific method, and to achieve an holistic picture. Possibly due to time and funding constraints, the use of two different ageing methods for the individuals in this study was the most common finding for the sites of interest, and therefore ages are given as a rather broad range for the adults, and only one individual, the skeleton from Easington, was classed as an older adult, being over 45 years of age. It is difficult to assign a

more accurate age than this to individuals over 45 for the reasons discussed above.

The methods of ageing adults that have been used by the bioarchaeologists working on the sites in this study are now examined. For each one, methods are given and this is followed by a comment on the likely accuracy of the use of the technique.

#### (i) Dental wear

Age at death can be estimated in adult skeletons by assessing the amount of enamel wear on the permanent teeth (Walker et al. 1991:169). Brothwell (1981:72) suggested a tentative method of classification of age in Neolithic to Medieval British skulls based on molar wear. Results were pooled across all of the time eras. Assessment of age is based upon comparisons of tooth wear in a skeleton with a series of graded and age related illustrations. While this would be useful for British skeletons, it must be considered that, although skeletons may have been buried in Britain, they may, like many of the individuals in this study, have migrated from other provinces with different diets which would cause dental wear to occur at a different rate to the standards used for British skeletons (Walker et al. 1991:176). White and Folkens (2005:369) suggest a similar method of assessment of age due to dental wear based upon Lovejoy's (1985) set of illustrated wear patterns in a prehistoric Native American population from Libben, Ohio. This again illustrates the same dietary comparison issues and limitations as Brothwell's method. Dental wear can thus be used as a rough guide to adult age, but cannot really be expected to accurately provide a chronological age for individuals who may have been exposed to differing diets from the standard populations which are used for comparison (Walker et al. 1991:176). Indeed, when the results of carbon and nitrogen isotope analysis are considered later, it can be observed that there was variation in the diet of the individuals sampled, and these dietary differences were particularly pronounced at Driffield Terrace, Chester Road and Victoria Road. Bioarchaeologists at these sites were the only ones in the study who mentioned

the use of dental wear in adult ageing, and therefore the accuracy of this method needs to be called into question.

#### (ii) Pubic symphyseal surface

This is generally described as being one of the most widely used indicators of adult age at death because age-related changes occur on this surface. In young adults, the pubic symphyses have a rugged surface traversed by horizontal ridges. The surface loses relief with age and is bounded by a rim by the age 35 (White and Folkens 2005:374). However, only bioarchaeologists at Easington and Poundbury used the method and, while this is entirely appropriate for the mature male (aged 45 years or above) at Easington, all of the individuals at Poundbury were young sub-adults, and thus the method is not as accurate for age estimation in this demographic group, as will now be discussed. Age-related changes of the pubic symphysis have been formally recognised as a method for determining adult age at death since Todd (1920) based a set of standards on a group of 306 males of known age at death. However, Cox (2000.69) reports that Todd removed individuals from his sample who fell outside of the recognised criteria for their staged age ranges, which would restrict accuracy. Todd noted that the pubic symphysis provided the most accurate age estimations for individuals aged between 20 and 40 compared with after 40 years of age. However, few further independent tests of the method were made until Brooks (1955) found the Todd method tended to overage especially for individuals in their 30s and 40s. McKern and Stewart (1957) attempted to refine the methods of Todd by using the skeletal remains of 349 males killed in the Korean War. However, similar to Todd, McKern and Stewart based their research and standards on a completely male sample with a limited age range, which provides an obvious drawback when trying to estimate the ages of female skeletons.

In order to address any issues of male-only standards for comparison, Gilbert and McKern (1973) used a sample of 103 known-age females to set up some further

comparisons. However, there was found to be a potential for premature changes in the pubic symphysis of females due to the obvious trauma caused during childbearing and birth. It was felt that over-ageing estimates would result for females and, in 1979, Suchey tested the method by asking 23 osteologists to age a number of skeletons. A large amount of inter-observer error was noted. From this work it was deduced that the system is a highly unreliable method for age estimation. Meindl et al. (1985) tested all these ageing methods against the Hamann-Todd collection of known age at death skeletons. They found Todd's original method to be the most accurate. Katz and Suchey (1985) suggested that while Todd's methods were superior they required further development due to being based on an inadequate sample. They worked on the Los Angeles County Coroner multiracial sample of 739 male individuals aged between 14 and 92 at the time of death and recommended the use of a modified Todd approach with six phases defined on the symphyseal face. This approach shows up a large amount of variation in age for any phase, particularly for older individuals (White and Folkens 2005:375). To further develop their research, Katz and Suchey went on, in 1989, to assess "race" differences in the Los Angeles collection. They also extended their work to include the examination of 273 females, and the refined techniques are detailed in Brooks and Suchey (1990). Cox (2000:69) however suggested that Brooks and Suchey's work produced very large age ranges with enormous amounts of overlap between them. This is particularly the case for older age groups.

In conclusion, pubic symphyses rarely survive in large numbers in the archaeological record due to their anterior position and anatomical make-up, meaning they become prone to weathering and mechanical damage (Cox 2000:69). This could explain why the method was not used by all of the bioarchaeologists reporting on the skeletons from the sites in this study. However, it is still a method of value. It would appear that the use of the pubic symphysis is a more useful method of age estimation in adult males than in adult females because we cannot fully know the extent of alteration childbearing would have upon the pubic symphyseal surface and hence on the accuracy of these ageing

methods. That said, based upon testing performed by Saunders et al. (1992), Cox describes the use of the method as performing very poorly in terms of intra- and inter-observer error, with the age ranges being so broad and overlapping as to be almost meaningless (Cox 2000:69), and the current study has different bioarchaeologists working on ageing of the skeletons from the different sites, so this could be a problem. However, as previously discussed, when used in conjunction with other methods of age estimation, its use allows bioarchaeologists to build up a more likely age range for adults than would have been estimated by the use of one method alone.

(iii) Cranial suture closure.

As mentioned in Table 6.3, the bioarchaeologist at Cirencester (Wells 1982) was the only person to use "cranial morphology" in order to estimate ages. Although the precise method was not mentioned, age can be estimated from cranial sutures, and this is assumed to be the "cranial morphology" technique used at Cirencester. This is the oldest method used for estimation of age at death (Cox 2000:68), and possibly had gone out of fashion by the time of the other site excavations which could explain why it was not used by the bioarchaeologists at these sites. Sutures occur at the point where the growing edges of two skull bones come together and are linked by a thin, unossified membrane. This membrane may remain unossified for some years after the individual has reached adulthood. Indeed in some individuals, sutures may remain open indefinitely (Brothwell 1981:43), with later suture closure often being observed in females (Lenhössek 1917). However, these sutures usually begin to close at about 20 years of age, being completely obliterated in later life, although the internal and external surfaces may not show the same degree of fusion (Brothwell 1981:43).

Early researchers (Dwight 1890, Parsons and Boc 1905) showed the possible usefulness of suture closure in age estimation. Todd and Lyon (1924) undertook further research based upon earlier work and as a result of this work, they

concluded that even sections of a suture could produce reliable age estimates (Brothwell 1981:43). However, this was called into question by Singer (1953), Cobb (1955), McKern and Stewart (1957) and Genovés and Messmacher (1959), who suggest that, even if a trend in suture closure exists, it is of little use in age determination. Brothwell (1981:43-44) suggests that suture closure could still be a useful tool in age determination of a skeleton whose facial region, and therefore teeth, is missing, as closed or partially closed sutures do at least show that the individual was an adult. However, care must be taken in interpreting the age of skeletons where the sutures are open because this does not necessarily prove the individual was under 20 years old (Brothwell 1981:45). Cranial suture fusion was re-introduced as an ageing method by Meindl and Lovejoy (1985) who chose a series of 1cm long segments of ten sutures and scored these on a scale of 0 (open) to 3 (complete obliteration). This method proved easy to use and has once again become an accepted method of skeletal ageing and has been further refined by the research of Buikstra and Ubelaker (1994). The Cirencester bioarchaeologists, Wells, did not state which aspects of cranial morphology were used for the ageing of the individuals buried there, but the methods of Meindl and Lovejoy (1985) and Buikstra and Ubelaker (1994) would not have been available by the time of the work on the Cirencester individuals.

#### 6.4.4 Summary

There are other methods of age estimation of adults, for example, examination of the auricular surface of the ilium or the sternal rib end. These methods were not used by any of the bioarchaeologists in this study, but they could have been alternatives had time and money allowed. The author does not go on to critically analyse these additional methods, and it is difficult to assess why the different bioarchaeologists in this study, who appear not to have many preferred age estimating methods in common, did not use any of them. However, it must be noted that time and money are not the only constraints on methods employed in skeletal analysis; bioarchaeologists do tend to work with their own preferred

methods. As has been observed and discussed, none of the methods available are very scientifically precise, so personal preference and experience of use are usually the reasons a particular method, or set of methods, are used by each professional. This does mean that, in the case of this study, there was no consistency of methods used for age estimation in all 21 of the skeletons, which tends to lead to concerns regarding accuracy of age at death and valid comparisons between ages of individuals from the different sites.

In summary, the skeletons in this sample had age and sex estimated by different bioarchaeologists using different methods. These people had varying levels of professional experience and there were different levels of preservation at each of the sites. As discussed above, there is likely to be some variation in the accuracy of these estimations due to these factors. In addition, some of the methods used are reported as having a higher success rate than others, and this may also lead to differences in judgment. However, sex and age is of interest for this research because different groups of people may have moved during their lives, for example young, old, men and women, and it may have implications for the epidemiology of TB. For the purposes of this work, it is therefore accepted that the reports of age and sex fall within the boundaries of acceptable accuracy.

Having now discussed the osteological methods used to estimate age and sex of the skeletal sample, the methods of isotopic analysis are now examined as these make up the analytical laboratory-based aspects of this study on which the conclusions about the place of origin or mobility of the individuals can be established.

# 6.5 Analytical methods.

6.5.1 Bone collagen extraction method for carbon and nitrogen isotope analysis.

Bone sampling was undertaken in the laboratories in the Department of Archaeology at Durham University. Bone demineralising and collagen preparation also took place at Durham University with a sub-sample being repeated at NIGL Keyworth in order to check for method accuracy and continuity. All isotope analysis was undertaken at NIGL Keyworth.

# (i) Bone sampling

Between 90-200mg of bone was sampled by means of a dental diamond cutting disc. The mass of bone was recorded.

# (ii) Bone demineralising

Bone chunks were placed into a 15ml test tube into which approximately 10ml of cold 0.5M HCl was added. The tops of the test tubes were then covered with parafilm into which a hole was pierced to allow gas escapage. The acid was changed every other day, taking care to decant the supernatant acid using an Ezee filter to avoid any sample loss. On intermediate days, the film was removed and the contents of the test tube were shaken. The tube was then re-covered with pierced parafilm. When the samples were soft, demineralisation was achieved and the supernatant liquid was decanted by means of an Ezee filter. The sample was then rinsed three times in deionised water.

#### (iii) Collagen gelatinisation

The liquid surrounding the softened bone was adjusted to pH 3.0. This was achieved by adding deionised water or HCl to the sample and measuring the pH by means of a digital pH meter. The tubes were placed into a hot block and left at 75 °C for 48 hours, until the collagen was dissolved and acid insoluble material was remaining. Ezee filters were then used to transfer the supernatant to pre-weighed plastic tubes. The mass of each tube was recorded in mg. This is initial tube mass. The tubes were covered in parafilm which was pierced. The tubes were then frozen at -50 °C for 24 hours before being placed into the freeze-dryer for 48 hours.

(iv) Calculating collagen yield

The tubes of freeze-dried collagen were then weighed and recorded (in mg). This is final tube mass. Mass of collagen (in mg) was determined by subtracting initial tube mass from final tube mass. Yield of collagen was calculated by dividing the mass of collagen by the initial bone mass. Yields of collagen above 1% were concluded to have sufficient yield for isotope analysis to take place.

6.5.2 Collagen carbon and nitrogen isotope analyses by Continuous-Flow Isotope Ratio Mass Spectrometry

Analyses of  $\delta^{13}$ C and  $\delta^{15}$ N in collagen are analysed using Continuous Flow-Elemental Analysis-Isotope Ratio Mass Spectrometry (CF-EA-IRMS) comprised of an Elemental analyser (Flash/EA) coupled to a Thermo Finnigan Delta<sup>Plus</sup> XL isotope ratio mass spectrometer via a ConFlo III interface. This was undertaken at NIGL Keyworth. Samples were weighed into tin capsules, and loaded into a 'Zero Blank'<sup>®</sup> auto sampler which dropped them onto a quartz combustion column. The temperature of this column was maintained at 900°C and "flash-combustion" was achieved by injecting a pulse of oxygen when the sample enters the tube. The gases produced were then taken by the helium carrier gas through a reduction furnace (680 °C). GC separation of N<sub>2</sub> and CO<sub>2</sub> was achieved before the gas stream passed through a Conflo-III interface before entering the ion source of the mass spectrometer. All reported isotope ratios were expressed using the delta ( $\delta$ ) notation in parts per thousand (per mil: ‰) relative to a standard:

 $\delta(\%) = ((R_{sample}/R_{standard}) - 1) \times 1000$ 

 $\delta$  <sup>13</sup>C and  $\delta$  <sup>15</sup>N results were measured in duplicate and were reported relative to V-PDB and AIR standards, respectively. Internal lab standards were used through the run to correct for instrument drift and to normalise the data to internationally accepted standards. Collagen carbon and nitrogen isotopes ratios were calibrated using an in-house reference material M1360p (powdered gelatin from British Drug Houses - BDH) with expected delta values of –20.32‰ and +8.12‰ (calibrated against USGS40 and USGS41, IAEA) for C and N respectively. The error for analysis is usually calculated based on the reproducibility of M1360P in each run, this is usually better than 0.2 (1 SD) for both elements. In this case the reproducibility was 0.04‰ for nitrogen and 0.03‰ for carbon, which is very good.

6.5.3 Methods of tooth enamel preparation for oxygen and strontium isotope analysis.

The tooth enamel preparation took place in the Archaeology Department laboratory at Durham University. The rest of the preparation and analysis took place at NIGL Keyworth. Dental burrs and drill bits were initially cleaned by placing in ultra-pure deionised water and then by cleaning in an ultra-sonic bath for five minutes, after which they were rinsed three times in ultra-pure water. Tooth enamel was cleaned carefully by means of abrasion to a depth of > 100 microns using a fine tungsten carbide dental burr and dental drill. The removed material was discarded. A small sample of tooth enamel was removed from each tooth by means of a circular flexible diamond edged rotary cutting disc. Removed enamel samples were placed in labelled lidded plastic vials and transported to NIGL Keyworth.

#### (i) Strontium isotope analysis

In the laboratories at NIGL, all enamel surfaces were mechanically cleaned with a tungsten carbide burr to remove adhering dentine. These were then examined under a light microscope to check for full dentine removal. The resulting cleaned enamel samples were transferred to a clean (class 100, laminar flow) working area for further preparation. In the clean laboratory, the sample was cleaned ultrasonically in high purity water, rinsed, then placed in ultra-pure deionised water at 60°C for an hour, rinsed three times in ultra-pure deionised water, dried and then weighed into pre-cleaned Teflon beakers. The samples were mixed with <sup>84</sup>Sr tracer solution and dissolved in Teflon distilled 8M HNO<sub>3</sub>. Strontium was collected using Dowex resin columns. Strontium was loaded onto a single Re Filament with TaF following the method of (Birck 1986), and the isotope composition and concentrations were determined by Thermal Ionisation Mass spectroscopy (TIMS) using a Thermo Triton multi-collector mass spectrometer. The international standard for <sup>87</sup>Sr/<sup>86</sup>Sr, NBS987, gave a value of 0.710256 +/- 0.00003 (2SD, n=21) from May to October 2015 and hence during the analysis of these samples. This value is within uncertainty of the accepted value for this standard and so no corrections are needed. Blank values were in the region of 100pg of strontium.

(ii) The chemical preparation and isotope analysis of oxygen in structural carbonate

For the isotopic analysis of enamel carbonate oxygen approximately 3 mg of prepared enamel was loaded into a glass vial and sealed with a septum. The vials were transferred to a hot block at 90°C on a GV Multiprep system. The vials were evacuated and 4 drops of anhydrous phosphoric acid were added. The resultant

CO<sub>2</sub> was collected cryogenically for 14 minutes and transferred to a GV IsoPrime dual inlet mass spectrometer. The resultant isotope values were treated as a carbonate.  $\delta^{18}$ O and  $\delta^{13}$ C were reported as per mil (‰) normalised to the VPDB scale using a within-run calcite laboratory standard (KCM) calibrated against SRM19, NIST reference material and  $\delta^{18}$ O values were converted to the VSMOW scale using the published conversion equation of Coplen (1988);  $\delta^{18}$ O<sub>VSMOW</sub>=(1.03091 x  $\delta^{18}$ O<sub>VPDB</sub>) +30.91. Analytical reproducibility for this run of laboratory standard calcite (KCM) was 0.09‰ (1  $\sigma$ , n=6) for  $\delta^{18}$ O<sub>SMOW</sub> and ± 0.04‰ (1s, n=6) for  $\delta^{13}$ C<sub>PDB</sub>.

The carbonate oxygen results  $\delta^{18}O_{SMOW(c)}$  were converted to phosphate values  $\delta^{18}O_{SMOW(p)}$  using the equation of Chenery et al 2012: ( $\delta^{18}O_{SMOW}(p) = 1.0322^* \delta^{18}O_{SMOW}(c) - 9.6849$ ) and these are converted to drinking water using the equation of (Daux et al. 2008 – equation number 6) ( $\delta^{18}O_{DW} = 1.54^*(\delta^{18}O_{SMOW(p)} - 33.72)$ ). The calculation of drinking water values involves considerable uncertainties (Pollard et al. 2011) and the values should be used as guidance.

#### 6.5.4 Method of interpretation of Strontium and Oxygen data

It is necessary at this point to provide some background to how the identification of migrants was achieved in this study. For the purposes of plotting local ranges on graphs of istope data (see Chapter 8) and for attempting to identify possible immigrants and their likely places of origin, their isotope results were compared to figures produced by NIGL/BGS for oxygen in Britain (Darling et al. 2003) (See Figure 6.6) and Europe (Chenery 2008) (See Figure 6.7). Local and British ranges for  $\delta$  <sup>18</sup>O<sub>p</sub> were converted back to phosphate values from these figures and have been plotted onto each site graph for ease of identifying locals to the area at a glance. For strontium, the maps of Britain in Evans et al. (2010:2) (See Figure 6.8) and for Europe (Voerkelius et al. (2010:936) (See Figure 6.9), were used for comparative purposes. These maps are useful for giving a broad overview of

comparative isotope data, but the biosphere strontium maps particularly require updating with the addition of new data points in order for tighter spatial predictions to be made for origins of archaeological individuals. Some biosphere data was available for locations closer to some of the sites (Evans et al. 2010 Supplementary data). These locations were identified by comparing their longitude and latitudes with that of the sites in the current study. Mostly, this data fell within the ranges already provided by the maps (Figures 6.9 and 6.10), but where it is available, it has been included in the discussion of the local ranges for each site in Chapter 8.



Fig. 6.6 Oxygen isotope values for modern UK drinking water (Darling et al. 2003:189)



*Fig. 6.7 Oxygen isotope values for modern European drinking water.* (Chenery 2008)



*Fig. 6.8 Spatial variations in biosphere* <sup>87</sup>*Sr*/<sup>86</sup>*Sr in Britain* (Evans et al. 2010:2)



*Fig. 6.9* <sup>87</sup>*Sr*/<sup>86</sup>*Sr values for natural mineral waters from Europe and a spatial prediction (Voerkelius et al. 2010:936).* 

#### 6.6 Statistical Methods

It was decided that, apart from standard deviation and Z scores, there are no appropriate statistical tests which can be applied to the data sets arising from the current study. This is largely due to small sample sizes of suspected TB sufferers available from each of the cemetery sites. Lightfoot and O'Connell (2016) suggest that choosing a statistical test to help with the interpretation of isotope results is difficult in any study, and impossible with a small sample. At least five skeletons would have needed to have been available from each site in order to perform any meaningful statistical analysis. Instead, graphs were produced comparing the isotope results of individuals in this study with other people buried in the same cemeteries and visible differences in these plots were examined and commented upon. Standard deviation (sd) was calculated for each isotope, for all available published data for each site incorporated with the individual site data from the current project. This shows how far the values differ from the mean values for the site. The larger the sd, the larger the range of values for that isotope. Z scores were also calculated for every individual for each of the isotopes analysed. The formula used to compute the Z score for each isotope at each site is (Madrigal 1998:64);

# Z = <u>value for individual – mean value for site</u> standard deviation for site

These values are used when it is required to check whether two samples are likely to have come from the same or different populations (Rowntree 1981:141). If an individual's Z value is below -2 or above 2, that person is considered to be significantly different from the rest of the population for that isotope result. While this is particularly useful for carbon and nitrogen isotopes, Z scores were included for oxygen and strontium results too. This is slightly contentious because strontium results can vary so much within a very short distance, however, as no other statistical analysis is appropriate, these values have been used as an extra tool to aid analysis of these results. This was considered to be acceptable analysis as the

aim of the study is to find out, by isotope analysis, if the individuals sampled were local to the place in which they were buried. This can be observed by visual observation of the graphed versions of results.

For analysis of published data pertaining to the different ages and sexes which were mobile, Chi-square tests were performed on the data to determine if there was a significant difference in the mobility of males compared to females or in the mobility of the different age groups compared with the expected patterns of mobility for these groups.

#### **Chapter 7: Results**

Bone and tooth samples were taken for strontium analysis from a total of 21 individuals. Firstly, all of the bones and teeth were catalogued and assigned a laboratory number for identification purposes, as shown in Table 7.1 These laboratory codes are referred to in order to identify the individuals in Chapter 8: Discussion, and Chapter 9: Conclusion.

The bone elements and types of teeth sampled are also identified in Table 7.1, along with the results of aDNA analysis from the previous Durham and Manchester NERC funded project, which has been discussed in Chapter 1. The lack of presence of MTBC aDNA does not mean TB was not present in the individual concerned at the time of death; osteological examination, detailed in Chapter 6 Materials and Methods, of all the skeletons revealed bone changes which indicated a likely infection with TB in all 21 individuals.

Collagen extraction was performed on the bone samples of all 21 individuals and was successful for 19 of these. However, EASN183 was discounted from further analysis as the C/N ratio was too high indicating a likelihood of diagenetic contamination by humic substances from the burial soil (van Klinken et al. 2000:52). DRIF19 did not yield any collagen despite repeated attempts to extract some. The remaining 19 collagen samples were analysed for carbon and nitrogen isotope composition in duplicate, following the methods detailed in Chapter 6. Mean values of these duplicates were then calculated for each individual in the sample and the results tabulated in Table 7.2. The range for the mean  $\delta^{13}$ C for samples is from -20.6‰ (CHES535) up to -19.1‰ (BALD7230) and GRPL531). The range for the mean  $\delta^{15}$ N is from 8.9‰ (POUN636) to 12.9‰ (CHES512), and all percentages of both C and N were found to be within acceptable ranges (around 35% for C and between 11- 16% for N - van Klinken 1999:691). The full table of carbon and nitrogen results is available in Appenix 1. The significance of these figures in the examination of the dietary habits of the people in this sample will be discussed in Chapter 8.

Site name	Skeletal elements showing TB lesions	MTBC aDNA detection results	Bone sampled	Tooth sampled	Laboratory code
Baldock	Vertebrae	Possible positive	T11 vertebra	Mandibular PM1	BALD7230
	Vertebrae	Negative	Lower thoracic vertebra	Mandibular canine	BALD7498
Driffield Terrace, York	Ribs, mandible, femora, metatarsals	Negative	Rib	Mandibular PM2	DRIF13
	Pelvis	Not tested	Rib	Mandibular PM2	DRIF19
Gambier Parry Lodge	Ribs	Negative	Rib	Mandibular PM1	GRPL531
	Ribs	Negative	Rib	Maxillary I2	GRPL538
Easington	Vertebrae	Negative	T8 vertebra	Maxillary PM1	EASN183
Poundbury	Ribs	Negative	Rib	Maxillary M1	POUN228
	Ribs	Negative	Rib	Maxillary M1	POUN257
	Ribs	Not tested	Rib	Mandibular M1	POUN506
	Ribs	Not tested	Rib	Mandibular M2	POUN619
	Ribs	Not tested	Rib	Deciduous maxillary M2	POUN636
	Ribs	Not tested	Rib	Mandibular M2	POUN1201
Chester Rd, Winchester	Ribs	Not tested	Rib	Mandibular canine	CHES512
	Ribs	Not tested	Rib	Maxillary canine	CHES535
	Ribs	Not tested	Rib	Mandibular M1	CHES636
Victoria Rd, Winchester	Ribs	Not tested	Rib	Mandibular PM2	VICT96
	Ribs	Not tested	Rib	Maxillary PM1	VICT129
Cirencester	Vertebrae	Negative	Proximal left humerus	Maxillary PM2	CIRES
	Ribs	Not tested	Rib	Maxillary M1	CIRE37
	Ribs	Not tested	Rib	Maxillary canine	CIRE189

Table 7.1 Lab numbers and skeletal elements and teeth sampled

Lab number	Age	Sex	Mean δ <sup>13</sup> C	Mean δ <sup>15</sup> N
BALD7230	26-35	М	-19.1	11.6
BALD7498	26-35	F	-19.8	10.1
EASN183	>45	Μ	-20.6	11.4
GRPL531	25-35	F?	-19.1	11.7
GRPL538	8-9	UNKNOWN	-20.3	11.6
VICT129	18-25	F	-20.0	9.3
VICT96	18-25	F	-19.8	9.7
CHES512	18-25	F	-20.3	12.9
CHES535	26-35	Μ	-20.6	11.0
CHES636	12-17	UNKNOWN	-20.4	11.2
CIRE189	26-35	Μ	-19.7	10.4
CIRE37	9-10	UNKNOWN	-20.4	10.9
CIRES	18-25	Μ	-19.9	9.3
POUN1201	36-45	Μ	-20.0	9.5
POUN228	9	UNKNOWN	-19.4	9.5
POUN257	10	UNKNOWN	-20.0	9.3
POUN506	12	UNKNOWN	-19.4	9.6
POUN619	15	UNKNOWN	-19.4	9.8
POUN636	4	UNKNOWN	-20.3	8.9
DRIF13	16-19	M?	-19.6	11.5
DRIF19	26-35	Μ	-	-

Table 7.2 Carbon and nitrogen isotope dataNB DRIF19 and EASN183 omitted

Enamel samples were successfully removed from the teeth of all 21 individuals in the sample. This enamel was analysed for strontium and oxygen isotope composition following the methods detailed in Chapter 6.  $\delta^{18}O_{dw}$  was calculated using the number 6 equation of Daux et al. 2008, (see Appendix 2 for full table of results), as discussed in Chapter 6. The results of isotope analysis are shown in Table 7.3

Lab	_		<sup>87</sup> Sr/ <sup>86</sup> Sr	δ <sup>18</sup> <b>Ο</b> <sub>p</sub>	$\delta^{18}O_{dw}$
number	Age	Sex		(VSMOW)	(VSMOW)
BALD7230	26-35	М	0.70932	17.34	-7.0
BALD7498	26-35	F	0.70904	17.26	-7.1
EASN183	>45	М	0.71031	17.45	-6.8
GRPL531	25-35	F?	0.71008	16.98	-7.6
GRPL538	8-9	UNKNOWN	0.70924	16.99	-7.6
VICT129	18-25	F	0.70943	17.98	-6.0
VICT96	18-25	F	0.70937	17.80	-6.3
CHES512	18-25	F	0.71249	18.18	-5.7
CHES535	26-35	М	0.71252	18.01	-6.0
CHES636	12-17	UNKNOWN	0.70948	19.36	-3.9
CIRE189	26-35	М	0.70884	18.52	-5.2
CIRE37	9-10	UNKNOWN	0.71007	18.50	-5.2
CIRES	18-25	М	0.70992	17.66	-6.5
POUN1201	36-45	М	0.70903	18.37	-5.4
POUN228	9	UNKNOWN	0.70854	18.48	-5.3
POUN257	10	UNKNOWN	0.70886	19.09	-4.3
POUN506	12	UNKNOWN	0.70835	19.35	-3.9
POUN619	15	UNKNOWN	0.70870	21.14	-1.2
POUN636	4	UNKNOWN	0.70875	18.39	-5.4
DRIF13	16-19	M?	0.71327	15.97	-9.1
DRIF19	26-35	Μ	0.70937	17.33	-7.0

Table 7.3 Strontium and oxygen isotope data.  $\delta^{18}O_{dw}$  calculated using Daux et al. (2008) Equation 6

The  ${}^{87}$ Sr/ ${}^{86}$ Sr ranges from 0.70835 (POUN506) to 0.71252 (CHES535) and the  $\delta^{18}O_p$  ranges from 15.97 (DRIF13) to 21.14 (POUN619). The significance of these figures in interpreting mobility of these individuals will be discussed in Chapter 8.

Standard deviation (SD) was then calculated for each isotope system in each site, and tabulated (Table 7.4).

Site/Lab No.	SD δ <sup>13</sup> C	SD δ <sup>15</sup> N	SD δ <sup>18</sup> O <sub>p</sub>	SD Sr
Winchester	0.5	1.1	1.0	0.00112
Driffield Tce	0.7	0.7	1.1	0.00143
Gloucester	0.5	1.0	0.7	0.00125
Poundbury	0.7	1.1	1.1	0.00023
Baldock	0.5	0.8	0.1	0.00020

Table 7.4 SD calculations for all isotope systems

Using the SD for the relevant site, Z values (see Chapter 6 for formula) were then calculated for each sample and each isotope system. The Z values are all tabulated together so that outlying individuals for multiple isotopes could be identified. These values are all tabulated in Table 7.5;

Site/Lab No.	Z score for $\delta^{13}$ C	<b>Ζ score for</b> δ <sup>15</sup> Ν	$\delta^{18}O_p$	Z score for Sr
Winchester				
VICT96	-1.2	0.6	0.1	0.2
VICT129	-1.5	0.3	-0.1	0.2
CHES12	-2.1	3.6	0.3	3.0
CHES535	-2.3	1.8	0.1	3.0
CHES636	-2.2	1.9	1.5	0.3
<b>Driffield Tce</b>				
DRIF13	-0.1	0.4	-1.2	2.1
DRIF19	N/A	N/A	0.0	-0.7
Gloucester				
GRPL531	1.9	1.4	-1.4	0.0
GRPL538	-0.4	1.5	-1.4	0.7
CIRE189	0.8	0.4	0.7	-1.0
CIRE37	-0.6	0.8	0.7	0.0
CIRES	0.5	-0.7	-0.5	-0.2
Poundbury				
POUN1201	-0.8	0.2	-0.7	1.8
POUN228	0.0	0.1	-0.6	-0.7
POUN257	-0.8	0.0	-0.1	-0.1
POUN506	0.0	0.3	0.2	-1.6
POUN619	0.2	0.4	1.9	0.2
POUN636	-1.3	-0.4	-0.7	0.4
Baldock				
BALD7230	0.8	0.8	0.7	0.7
BALD7498	-0.6	-0.9	-0.1	-0.6

Table 7.5 Z scores for all isotope systems.

# **Chapter 8: Discussion**

# 8.1 Introduction

It is pertinent, prior to discussing the results of this study, to return to the hypothesis proposed and tested in this research, namely that people who had TB and who were buried in a number of cemeteries in England and dated to the Roman period were not local to the area in which they were buried. It was suggested that this was because TB had been rare in Britain prior to the Roman Conquest. This was supported by the lack of skeletal evidence, with only one British skeleton documented from the Iron Age who was buried at Tarrant Hinton in Dorset (Mays and Taylor 2003:189). There may be many reasons why this might be the case:

1. This could be because burials in Iron Age Britain are rarely found owing to the methods utilised in the disposal of the dead in that era (Cunliffe 2005:543, Roberts and Buikstra 2014:8),

2. Or because TB was not a common disease at that time,

3. Or because of the impact of the 'osteological paradox' on the skeletal record, where people died in the acute stages of the disease and before bone changes could develop.

What is certainly clear is that, by the time of the Roman Conquest, TB had become relatively much more common, as documented in the bioarchaeological record (Roberts and Buikstra 2003:132). There could be several reasons for this occurrence; the trend for urban living started, with more people living in larger towns than previously (Bennett 1984:7, Cleary 2001:161). It has been discussed that TB is a disease which is spread in more cramped living conditions (Abubakar 2010:76). It is documented that the typical buildings found in pre-Roman Iron Age Britain were thatched roundhouses, and that the Romans introduced rectangular building plans which were more suitable for packing buildings closer together along street frontages in planned cities (Cleary 2001:163). Barnes et al. agreed that there was an increase in TB infection load which was proposed to follow the transition to urban living as a result of increases in population density, pathogen mobility through long- distance trade and also with exposure to the disease through animal husbandry (Barnes et al. 2011:842). However, the researchers investigated the possibility that the process of urbanisation has, through a consequence of increased disease load, shaped the distribution of gene polymporphisms which convey resistance of an individual to infection by TB (Ibid. 2011:843). Their research examined 17 populations for the presence of the gene variant conveying disease resistance and found a strong correlation between the older age of the town and the likelihood of being resistant to TB (Ibid. 2011:843). So this age of towns and urbanisation is indeed a risk factor which could be examined in another future project in order to test the hypothesis that the older a town is, the fewer people living there have TB.

Another risk factor which increased during the Roman period was mobility. It is well documented that the Roman Army in particular were highly mobile, and records were kept of where legionaries and auxiliaries were recruited from, so it is known recruits were made as needed, from wherever the legion was based at the time (Roselaar 2016:138). There are records of the Roman road system within Britain (Davies 2002), and some evidence of ports, for example, in London (Miller et al. 1986). Thus from this evidence, it is clear that people were able to move relatively easily around and within the province, and they could potentially take their diseases with them. As isotope analysis has become a relatively routine method to help to detect mobility in archaeological populations, bioarchaeologists now have the means to investigate potential mobility in the past, if bones and teeth are available for examination.

# 8.2 Discussion of the data

The data resulting from the isotope analysis of the bone and tooth samples are now discussed. The nature of the samples analysed are first considered and this followed by a consideration of the data from each site and a final section provides a summary.

8.2.1 Carbon and nitrogen: the samples analysed, including limitations

It is now necessary to discuss the bone samples used in this project and to explain why some, (namely DRIF19 and EASN 183), were discounted from carbon and nitrogen analysis. As discussed in Chapter 3, the amino acid profile of collagen remains intact until about 5% of the original protein remains present in the bone (Stafford et al. (1991:35). For the same reasons, DeNiro (1985:807) recommends only using samples with C/N ratios between 2.9-3.6, with van Klinken (1999:691) narrowing this further and suggesting rejection of any samples outside of the range 3.1 - 3.5. For the purposes of this project, dietary analyses were therefore restricted to samples of bone which contained at least 1% of the original collagen. and hence the individual from Easington (EASN183) had to be discounted from the carbon and nitrogen isotope results. It was not possible to extract sufficient collagen for any analysis from DRIF19, one of the two individuals from York, despite repeated attempts to do so. The most likely factors to cause this denaturing of the collagen were possibly the burial environment conditions; for example, being buried for a long duration in acidic soils would certainly lead to this sort of damage to the bone collagen (Millard 2001:640). In comparison with another recent isotope study on burials in Roman London, (Redfern et al. 2016), the success rate of collagen extraction in the current project was very good. Redfern et al. found eight of their samples did not yield enough collagen to process, and one sample had a low collagen yield along with a C/N ratio of 4.38, so was also rejected. This left only 10 samples which were successfully analysed (Redfern et al. 2016:17). However, in 2006, Fuller et al. had a 100% success rate at extracting collagen from 81 ribs and six femur. These samples were taken from skeletons buried at Queensford Farm, the largest excavated cemetery of late/sub-Roman period in the Upper Thames Valley, with a total of 164 skeletons interred. The site is near modern Dorchester-on-Thames in Oxfordshire (Fuller et al.

2006:47). This suggests local conditions have a huge effect on preservation of collagen in buried skeletons.

The bone and teeth samples used for this project were, as discussed (see Chapter 1), made available from a previous project and no further samples could be obtained from the original skeletons. The re-use of bones and teeth from the previous research was an advantage because it meant no further destruction of the original skeletons was required and this mitigated against ethical dilemmas. However, it did introduce an element of not being able to compare "like with like" in terms of bone type used for analysis. As discussed in Chapter 3, different bones renew or turn over at different rates, and therefore, by using different bones, isotope analysis is considering different time spans of the lifetime of the individual. However, bone remodelling also varies considerably between individuals, dependent on age, sex, genotype, systemic metabolic conditions of the individual and activity levels (Beaupré et al. 1990:651). That said, bones analysed from 15 out of 19 skeletons were ribs so reasonable comparisons of similar time periods prior to death for most of the individuals are possible. Three of the bones were vertebrae (BALD7230 and BALD7498, and the omitted EASN183), and one was a proximal left humerus (CIRES). CIRES was the only skeleton from whom the bone was used which was not reported as having been directly affected with TB. However, the effect of infection on bone turnover rates is unknown, presumably because different infections and levels of severity of infection (bacterial load) will have differing effects on the bone. The fact that all but one bone sample used displayed pathological legions likely to indicate infection with TB mitigates to some extent against this effect, thus controlling this variable as far as is possible, and allowing comparison to be made between samples. It has been suggested that pathological lesions on bone, namely osteomyelitis, can lead to an elevation in the nitrogen isotope ratios of that bone (Katzenberg and Lovell 1999:316). This was explained as being possibly due to collagen being formed during illness as part of the repair process of pathological bone lesions being derived from catabolism of existing proteins in the body (ibid. 1999:316). However, this research used wholly pathological bone for isotopic analysis, whereas the current project avoided

pathological lesions as far as was possible when taking samples, so minimal effects on the isotope ratios can be expected.

8.2.2 Strontium and oxygen: the samples analysed, including limitations

Due to the restricted sample choices, some of the teeth sampled were formed during the years an individual would have been breastfeeding, and hence it is required to take this "breastfeeding effect" into account when interpreting the oxygen isotope results (Wright and Schwarcz 1998:1). Ideally, second premolars, second molars or third molars are used for isotope analysis, as these teeth are formed after the period where breastfeeding would impact on their composition. However, some of the individuals were juvenile in age and would not have had these teeth by the time they died. Furthermore, for one individual, POUN636, only a deciduous second molar was available for sampling. All of the other teeth used were from the permanent dentition but a range of different teeth were used. Table 8.1 shows which teeth were sampled from each site and the approximate ages at which these teeth develop;

Tooth type	Age at first evidence of crown calcification	Age when enamel of crown completed	Sample numbers of these teeth
Incisor	3 to 12 months	4 to 5 years	GRPL538
Canine	4 to 5 months	6 to 7 years	BALD7498, CHES512, CHES535,
			CIRE189
First premolar	1.50 to 1.75 years	5 to 6 years	BALD7230, GRPL531, EASN183, VICT129
Second premolar	2.25 to 2.50 years	6 to 7 years	DRIF13, DRIF19, VICT96, CIRES
First molar	Birth	2.50 to 3 years	POUN228, POUN257, POUN506, CHES636, CIRE37
Second molar	2.50 to 3 years	7 to 8 years	POUN619, POUN1201
Deciduous second molar	6 months before birth	10 to 12 months	POUN636

*Table 8.1 The types of teeth sampled and ages at which these teeth develop.* (From Berkovitz et al. 1986:176, AlQahtani et al. 2010:485)

Having examined Table 8.1, the following considerations need to be taken into account. The incisor (GRPL538) and canine teeth (BALD7498, CHES512, CHES535 and CIRE189) would have been starting enamel formation when the individual was likely to have been breastfeeding; this needs to be considered when interpreting the oxygen isotope results. Breast milk is isotopically enriched in <sup>18</sup>O, the heavier isotope, compared to water the mother ingests, due to <sup>18</sup>O being less likely to be expired (breathed out) than the lighter isotope, <sup>16</sup>O (Bryant and Froelich 1995, Wright and Schwarcz 1998). Due to breast milk deriving from the mother's <sup>18</sup>O-enriched body water, it is isotopically heavier than her drinking water (Bryant and Froelich 1995, Wright and Schwarcz 1998, Britton et al. 2015:228). Although some babies may have been given water to drink in addition to breast milk, it is expected that breast milk would be the main source of ingested water prior to weaning onto solid foods. Therefore tooth enamel forming during this

period of breastfeeding would be expected have higher in  $\delta^{18}$ O values than teeth whose enamel formed after the cessation of breastfeeding. These early forming teeth would demonstrate enrichment of between 0.5‰ and 1.2‰ in enamel carbonate and/or phosphate compared to later forming teeth (Wright and Schwarcz 1998). The deciduous second molar from POUN636 was completely calcified before the child is likely to have ceased breastfeeding. This tooth would also demonstrate an elevated  $\delta^{18}$ O value compared to later forming teeth. First molars were sampled from POUN228, POUN257, POUN506, CHES636 and CIRE37. Like incisors and canines, these teeth start to form during the time the individual would have been breastfed, and so the breast feeding effect of enriched  $\delta^{18}$ O values needs to also be considered for these samples.

# 8.2.3 The carbon and nitrogen isotope data

#### (i) Introduction

The carbon and nitrogen isotope data from each of the study sites are now discussed individually. Carbon and nitrogen isotope data will be considered first. This will allow dietary comparisons to be made of the individuals in this project with others from published data from the same, or very proximal, cemetery sites. Where published data are not available (eg. for Baldock), a comparison is made with the published isotope data from the rest of the sites in the study. As these are all broadly contemporary, they provide a baseline for diet in Roman Britain and enable detection of major differences in food type consumed by the individuals that are the focus of this research. Following this, strontium and oxygen data will be analysed prior to drawing all the isotope data together to discuss if these people were likely to be migrants to the areas in which they were buried.


Fig. 8.1 Carbon and nitrogen isotope data for Driffield Terrace, York

DRIF13 had visceral surface woven and lamellar new bone formation on his ribs and mandible (Caffell and Holst 2012). These bone changes on the ribs are not pathognomonic for TB, as has been previously discussed in Chapter 2, but they do indicate the presence of a chronic condition caused by a pulmonary disease, likely to be an infection, and possibly TB. The presence of lamellar new bone suggests a long-term infection, and the presence of new, immature, woven bone suggests that the disease was active at the time of death. If this was the case, it could have had some bearing upon the health of the individual and their appetite and type of diet consumed. However, Figure 8.1 and Z scores (Table 7.5) show that DRIF13 does not stand out as having a diet which is different from the majority of other individuals buried at Driffield Terrace, based on other isotope data from the site. This suggests that DRIF13 was eating the same sort of diet as the other people he was buried alongside, without any dietary input from C<sub>4</sub> plants or marine resources. The data and statistics therefore support the conclusion drawn that DRIF13 was not eating a different diet when compared to the other people buried in the Driffield Terrace cemetery, and therefore he cannot be identified as an immigrant from his dietary isotope values.

That said, strontium and oxygen isotope analysis has been done on other skeletons from Driffield Terrace (Müldner et al. 2011, Montgomery et al. 2011), which have proved there were many immigrants to the York area within that cemetery population. Although this conclusion could also be made for other skeletons buried at sites in Roman Britain where there are obviously migrants to the area, it must be considered that just because DRIF13 was not eating a different diet from the majority of the population buried in that cemetery, it does not mean he was not a migrant to York. Consideration of his oxygen and strontium isotope ratios will further clarify this discussion.

(iii) Gambier Parry Lodge and Cirencester



Fig. 8.2 Carbon and nitrogen isotope data for sites in Gloucestershire

There were no available comparative isotope data for the sites of Gambier Parry Lodge and Cirencester, and therefore both of these datasets were compared with the Gloucestershire regional data of Cheung et al. (2012). Figure 8.2 illustrates that the Cirencester individuals fall within the main body of results for Gloucestershire. CIRES had evidence of Pott's disease (Wells 1982) but it is not possible to determine if this meant that individual had active TB at the time of death. However, CIRE37 (Wells 1982), CIRE189, GRPL531 and GRPL538

(Cameron and Roberts 1984), all had visceral surface woven new bone formation on the surface of their ribs suggesting that the disease causing this bone change, presumed to be TB, was active at the time of death. This could have had a bearing upon eating habits of the individuals, for example, leading to reduced appetite. However, this is not indicated by the isotope data; the Z scores for  $\delta$   $^{13}C$  and  $~\delta$   $^{15}N$ (see Table 7.5) show that none of the Gambier Parry Lodge or Cirencester values are outliers. This means that the diets of the sampled individuals were similar to other people buried within the same cemeteries. None of the sampled individuals can therefore be identified as an immigrant based on differences in dietary isotope values. However, Figure 8.3 shows that both of the individuals from Gambier Parry Lodge have  $\delta^{15}$ N results which appear to differ somewhat from most of the other individuals from the area for whom isotope data are available. GRPL531 is observed to have higher  $~\delta~^{13}C$  than GRPL538 and both individuals have  $~\delta~^{15}N$ values that are towards the higher end of the Gloucestershire values. Higher  $\delta^{13}$ C and  $\delta^{15}$ N bone collagen values mean that these individuals were consuming more isotopically enriched foodstuffs, such as seafood or omnivorous pigs (Cheung et al. 2012:68). However, these foods appear to have been consumed in relatively small amounts. Elevated  $\delta^{15}$ N (around 12‰) could also correspond with consumption of freshwater fish, although the corresponding  $\delta$  <sup>13</sup>C values for freshwater fish would be around -21‰, which is not the case for GRPL531 or GRPL538 who have higher  $\delta^{13}$ C values. This may be because they were consuming low amounts of freshwater resources. However, GRPL538 and CIRE37 could have been consuming more freshwater fish than GRPL531 and 538 because they have the lowest  $\delta^{13}$ C values. There is no evidence for fish bones having been excavated from the site, as corroborative evidence for fish consumption, although as these bones are very small and usually only obtained by sieving soil samples, absence of evidence cannot be conclusively stated as being evidence of absence. It is also not stated that any soil sieving took place at this site, so the process is assumed not to have taken place.

Although not identified from Z scores as consuming a different diet to other people buried in the area, Gambier Parry Lodge individuals do plot towards the higher end of the  $\delta^{15}$ N values on Figure 8.2 compared with others in the surrounding cemeteries. This could be due to chance or could mean that these people were immigrants to the area from somewhere with a slightly different diet to many other people who lived in Roman Gloucestershire. Another consideration is that they could have been second generation migrants who continued the dietary choices of their parents' homelands, as suggested by Swan (1992, 2009) who, as discussed in Chapter 4, used the presence of a particular type of cooking vessel to identify immigrants, based upon the assumption these vessels were used for non-local cooking methods. It is also suggested that there were differences between the diets of urban and rural populations buried in Gloucestershire during the Roman period, with people from urban areas having consumed more isotopically enriched foods such as seafood and garum (fish sauce) than their rural counterparts (Cheung et al. 2012:68). Therefore, the Gambier Parry Lodge individuals, and CIRE37 from Cirencester, possibly followed a more "urban" type of diet, perhaps due to more choice of foods being available at town markets than the narrower choice available to the more rural populace of the area. It should be considered that the Gambier Parry Lodge people, along with CIRE37, could perhaps have better afforded to buy seafood and garum than their rural contemporaries, although there is no evidence in their burials to suggest they were of a higher wealth or status than other people buried near by. So perhaps it was more acceptable for people living at Gambier Parry Lodge or Cirencester to eat a more Romanised diet than it would be for people still following more traditional Iron Age diets in more rural areas.

# (iv) Poundbury



Fig. 8.3 Carbon and Nitrogen isotope data for Poundbury, Dorset.

Figure 8.3 shows that the Poundbury isotope ratio values for the six individuals analysed in the current project fall within the range of published results for other individuals buried in the same cemetery. POUN257, POUN506, POUN619, POUN636 and POUN1201 had woven new bone on the visceral surface of their ribs, and one had lytic rib lesions (POUN228). This would suggest that the disease (interpreted as being TB) was active at the time of death, and this again could have impacted on their appetite and dietary choices. However, the Z scores for the

site's  $\delta^{13}$ C and  $\delta^{15}$ N results (Table 7.4) show no outliers in diet for the sampled population compared to the diet of the rest of the cemetery population. This means that they were not eating a different diet when compared to other people buried near them, with no evidence of C<sub>4</sub> plant or marine fish consumption. The carbon and nitrogen isotope data for these individuals cannot therefore identify them as being of a different origin to other members of the Poundbury cemetery burial population. Some of the data from the individuals analysed by Richards et al. (1988) show sufficiently high  $\delta^{15}$ N and  $\delta^{13}$ C bone collagen values to suggest that they were eating some marine food sources. However, this does not appear to be the case for those analysed by Redfern et al. (2012) or for the people in the current project, who perhaps could not afford these possibly more expensive food types.

(v) Victoria and Chester Roads, Winchester.



Fig. 8.4 Carbon and nitrogen isotope data for sites in Winchester

Figure 8.4 shows the current project isotope ratio values from Victoria and Chester Roads in Winchester. While no other isotope results were available from either of these cemeteries, a nearby contemporary cemetery at Lankhills in Winchester was used for comparison (Booth et al. 2010). Two Lankhills individuals with suspected TB (not analysed in the aDNA project), diagnosed from skeletal evidence, have been highlighted in Figure 8.4. The data for those people suggest that they were not eating a very different diet to the rest of the Lankhills cemetery population, although their  $\delta$  <sup>15</sup>N values were towards the lower values for the area. The Z scores for these two people do not show that they are outliers compared to the

rest of the area cemetery populations of the area in either  $\delta$  <sup>13</sup>C or  $\delta$  <sup>15</sup>N values (See Table 7.4).

These people all had TB during their lives, and, due to the presence of woven new bone on the visceral surface of their ribs, the underlying disease was interpreted as being active at the time of their death, and therefore if their illness had made them lose their appetites, their  $\delta^{15}$ N values may have increased. In the classical literature, Hippocrates did not provide any guidance about special diets that people who were ill should be consuming (Hippocrates and Chadwick 1950, Mattock and Lyons 1969), and although no evidence exists from Roman Britain which suggested diets for the sick, it is known that special diets have been recommended in the past. For example, although relatively recent, there is evidence of early 20<sup>th</sup> century specialised diets for hospitalised TB patients in Portugal (Santos 1999:133). This diet consisted of three meals a day, but, unlike the Romans from the current study who had suspected TB, meat or fish featured with every meal. The preferred meats being lamb (eaten once a week) and beef (at least three times a week). Eggs and soup were also eaten, along with fish (at least three times a week). The fish was marine in origin (salted cod) plus other varieties listed as "fresh fish" which could have presumably been of marine or riverine origin (Ibid. 1999:133). This was in direct contrast to the diet of the lower socio-economic groups in Portugal at the time. The local Portugese diet at this time was nutritionally poor and mainly vegetarian with a small but not daily intake of fish (usually sardine and salted codfish) (Ibid. 1999:132). In opposition to early 20<sup>th</sup> century Portugal, the Roman era Winchester individuals could have eaten less meat than their counterparts who did not have signs of the disease, perhaps due to local tradition dictating that ill people should eat certain foods, or because they did not feel well enough to eat the "normal" diet, requiring instead something easier to chew and digest, such as broth or porridge. However, as discussed in the Summary (8.2.3.) for this section, this is difficult to conclude with any degree of certainty due to the isotope results reflecting diet from the last five to ten years of the life of these individuals and therefore presumably not just covering the time when they were ill.

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The results for the two Lankhills individuals with TB were, however, different from those people with TB buried at the nearby contemporary cemeteries of Victoria and Chester Roads. The Chester Road individuals had certainly eaten a different diet to the Lankhills people, and the Victoria Road people had  $\delta^{13}$ C values lower than most of the Lankhills cemetery population. This is confirmed statistically, with all of the Chester Road individuals having Z scores below -2. They are therefore outliers to the rest of the population (see Table 7.5), which means that they were eating a diet which was significantly different to other people buried in the area. Neither of the Victoria Road individuals had an outlying Z score, meaning that their diet was similar to other individuals buried in the same cemetery. For  $\delta^{15}$ N, the SD shows a larger range around the mean at 1.1, but only CHES512 had an outlying Z score of 3.6; this is very different to other people buried in the area.

When compared with the published data from other English sites used in this study (see Figure 8.6), it can be seen that CHES512 (  $\delta$  <sup>13</sup>C -20.3‰ and  $\delta$  <sup>15</sup>N 12.9‰) still stands out as unusual within contemporary populations buried in England, some of whom could have themselves been immigrants. This would suggest that being an immigrant to the area, and perhaps even to the country, from somewhere where the population consumed a different type of diet would be the most feasible explanation for the collagen isotope values of this person. These dietary differences could be due to the consumption of freshwater fish, which the people at Lankhills did not eat. The difference in diet could also be explained by local choice, availability, or what individuals could afford to buy, but because all of the cemeteries in Winchester were situated so close together, it appears that these populations would have had equal physical access to similar foods, even though perhaps not everyone would be able to afford them. However, in terms of excavation evidence, while over 18,000 mammal and 500 bird bones have been identified from this area of Winchester, only a small sample of fish bone was found (Maltby 2015:179) and details of the species of fish was not given, so perhaps not

much fish was available to people in the area, lending more weight to the suggestion the Chester Road individuals were from elsewhere.

It is not possible to begin to completely understand the driving forces behind the foods chosen by individuals or populations for consumption, even when they are available to eat. Presumably, personal tastes and preferences played a part, as they do today. Additionally, perhaps local and family dietary and culinary "taboos" existed, despite all individuals seemingly having access to the same foods (Montgomery 2010:339). However, perhaps access was not equal for all people; some foods could have been expensive and out of the price range of most individuals in an area, and therefore they were only consumed by the more elite classes (Cheung et al. 2012:68), this would be a relevant consideration for individuals from all of the sites in the study, but the Chester Road individuals stand out as being different from other people buried in the area, so perhaps the people buried at Chester Road had arrived in the Winchester area from a location with ready access to riverine resources and where the consumption of these was more common. After all, the bone collagen sampled gives an average of the diet consumed for approximately five to ten years before the death of the individual, and therefore they could have fitted in with local customs and consumed a local diet when they arrived; however, previous food choices would still register in their isotope values. The Chester Road people could also be a population with local origins but with different beliefs about food consumption. If they were indeed in this category, they could be second generation migrants to the area who have continued with the dietary customs from their family homelands (Swan 1992:1, Swan 2009:1).

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I.

Fig. 8.5 All published carbon and nitrogen isotope data for the sites used in this project

### (vi) Baldock

The final two individuals to be considered were buried at Baldock (BALD7230 and BALD7498). There was no indication on the vertebrae of these people to suggest if the TB disease was active or not at the time of death, and there are no comparative isotope data for cemeteries in this area. Therefore, both of these people were compared to all published data for the sites used in this project (Figure 8.5), that is, roughly contemporary burials in England. They both fall well within the range of values for the sites, and thus that these people were eating the same diet as others in England during the Roman period.

# 8.2.4 General discussion and summary

Firstly, whilst it should be considered that local traditions in Roman Britain may have dictated that ill people should eat certain foods, or they may have not felt well enough to eat the "normal" diet, requiring instead something easier to chew and digest, such as broth or porridge. However, this is difficult to conclude with any degree of certainty due to the isotope results reflecting diet from the last five to ten years of life of these individuals and not just the time when they were ill. Although, when the average  $\delta^{13}$ C and  $\delta^{15}$ N values were compared for people with TB with those without TB (see Tables 8.2 and 8.3 below), it can be concluded that on average, males with TB had slightly enriched  $\delta^{13}$ C values compared to males without TB. Apart from this, no obvious differences are observed;

Meanõ <sup>13</sup> C values	Meanõ <sup>13</sup> C values	Mean $\delta^{15}$ N values	Meanõ <sup>15</sup> N values
for Males with TB	for females with TB	for males with TB	for females with TB
(‰)	(‰)	(‰)	(‰)
-17.0	-19.8	9.0	10.7

Table 8.2 Mean  $\delta^{13}C$  and  $\delta^{15}N$  values for males and females with TB

Mean $\delta$ <sup>13</sup> C values	Mean $\delta$ <sup>13</sup> C values	Mean $\delta$ <sup>15</sup> N values	Mean $\delta$ <sup>15</sup> N values
for Males without	for females without	for males without	for females without
TB (‰)	TB (‰)	TB (‰)	TB (‰)
-19.7	-19.9	10.1	9.0

Table 8.3 Mean  $\delta^{13}$ C and  $\delta^{15}$ N values for males and females without TB

Presumably people who were infected with TB were unlikely to be ill and with a resulting reduced appetite for all of the time that their bones were remodelling, or lesions were developing. This is because the isotope data show and average for the whole of the "lifespan" of the bones, which may include varying times before infection occurred. It must also be considered that neither osteological nor aDNA examination can provide information about how long the person had TB prior to their death, although if immature unremodelled bone is present this at least suggests that the disease was active at the time of death.

Overall, the carbon and nitrogen results show that all the individuals in this project appear to be omnivores who ate a diet whose terrestrial food chain was based exclusively on  $C_3$  plants and who were not eating significant quantities of wheat and other grains grown in warmer and more arid climates than Britain (see Chapter 3). As there were no C<sub>4</sub> plants growing in Britain during the Roman period, their consumption is not to be expected in someone not originating from areas of the world where they are cultivated. C<sub>3</sub> plants have  $\delta^{13}$ C values of -20‰ to -35‰ (DeNiro and Hastoff 1985:97), whereas C<sub>4</sub> plants would have  $\delta^{13}$ C values of -9‰ to -14‰ (Ben-David and Flaherty 2012:316). In regions such as Britain were no C<sub>4</sub> plants were grown, people consuming a local terrestrially-based diet would have a  $\delta$  <sup>13</sup>C of -19‰ or lower. Bearing in mind  $\delta$  <sup>13</sup>C values for people with a large intake of marine resources is -12.0% to -14.5% (Schoeninger et al. 1983:1381), it would appear that all of the individuals sampled had a mainly terrestrial diet, with some input from marine resources (such as GRPL531) or freshwater fish, the consumption of which also elevates  $\delta$  <sup>15</sup>N values (van Klinken et al. 2000:56). The three Chester Road, Winchester individuals provide evidence for the consumption

of some freshwater fish. People with lower nitrogen isotope values probably ate a mainly vegetable based diet, although isotopic ratio values cannot distinguish a true meat-eating omnivore from a vegetarian who consumed milk and other dairy products (Makarewicz and Sealy 2015:146). However, it is clear that none of the individuals in the study ate a vegan (purely plant based) diet.

The isotopic evidence for some consumption of fish in Roman Britain is supported in other archaeological research, for example, Locker (2007) and Murphy et al. (2000). Locker (2007:141) examined fish bone evidence at Roman sites in Britain (modern England) and states that there is little Iron Age evidence for consumption of fish, and even by the Roman period, the quantities of fish bones found is generally much smaller than a few centuries later. However, it seems possible that people living close to forts and towns were probably the most likely to try new, available trends in food and culture, such as fish consumption, compared to remote communities, who were perhaps more likely to continue in an Iron Age tradition. This could be due to practical issues such as accessibility factors or to lingering cultural taboos.

Of course, the recovery of fish bones from archaeological contexts is not without its difficulties; fish bones are often relatively small and difficult to find even when sieving is carried out. In addition, because they are small, they may be more subject to diagenesis than larger bones, which would lead to less chance of them being recovered. On sites where all bone collection was done by hand, recovery of fish bones is poor to non-existent and obviously biased to larger species such as cod. In pre-1970 excavations, no sieving was carried out at all (Locker 2000:142). Therefore, it is to be expected that there is little or no physical bone evidence from the cemetery sites in this project for consumption of fish. However, indirect evidence for fish consumption in Roman Britain has been found at some other sites, for example, particular types of amphorae used to transport salted fish and fish sauce across Europe. One good example of such a vessel from the 1<sup>st</sup> century AD was found in Southwark, London, inscribed with a personal name and Antibes (a coastal town in France). It contained six Spanish mackerel heads (Locker

2007:142). There is also literary evidence from the Roman period in England for the consumption of fish products including orders for fish sauce found on writing tablets from the fort at Vindolanda (Alcock 2001:5). It can be assumed that fish and fish products were becoming more widely available to people during the Roman period.

Generally, low levels of fish consumption in Roman Britain were probably influenced by economic issues (perhaps these resources were expensive), and perhaps even a negative cultural perception of fish left over from Iron Age traditions (Locker 2007:141). However, Locker found evidence that there was some development of freshwater fisheries and, to a lesser extent, estuarine and inshore marine fisheries in the Roman period in England, the latter being particularly associated with high status people (Ibid. 2000:154). This usage of some fish resources in the Roman diet (although not necessarily in Roman Britain) is however confirmed by the isotope evidence of the three Chester Road, Winchester, individuals who showed evidence of some freshwater fish consumption, although as previously suggested, these people could have been immigrants to the area and thus ate the fish elsewhere.

In terms of other dietary components, it is pertinent to consider a study of new plant foods in Roman Britain and their dispersal and accessibility (van der Veen 2008). It was suggested that the Roman period brought with it the consumption of many new plant foods from the Mediterranean (eg. olives, figs, grapes, cucumbers, peaches, pine nuts and almonds), as well as a range of other cultivated foods such as apples, plums, cherries, cabbages and turnips (Ibid. 2008:11). These new foods undoubtedly brought a significant diversification of the plant component of the diet as well as being sources of essential nutrients (Ibid. 2008:12). van der Veen does not mention the likelihood of vegetarians in the Roman period in Britain, but if a more varied range of plant based foods was becoming available to townspeople and at markets, survival without eating meat or fish in great quantities, or even at all, would indeed have been possible.

written documents on vegetarianism in Europe dating to the 6<sup>th</sup> century BC (Leitzmann 2014:469S). This diet was part of a religion called the Orphic mysteries, the followers of which were vegan (eating no animal products). Around the same time, the Greek philosopher Pythagoras developed his idea of reincarnation which led to the avoidance of meat consumption (Ibid. 2014:469S). As has been mentioned, several individuals analysed in this research could have been ovo-lacto vegetarians (still consuming eggs and dairy products, unlike vegans who would not eat any form of animal products), as isotope analysis does not distinguish between people eating animal protein in the form of flesh or from the consumption of secondary products of animals.

With regard to the meat aspects of diet, King (1984) researched animal bones in archaeological contexts and their implications in understanding diet in Roman Britain. This study was limited to three main food animals: ox, sheep/goat and pig (Ibid. 1984:2). The conclusions of this study suggest that, during the Iron Age, sheep/goat and pork were the most popular meats, but by the Roman period, the army brought about dietary changes which spread to villas and Romanised settlements. These changes saw the consumption of more ox and less of other meats (Ibid. 1984:3, Maltby 2002:88). It was also found that the more highly Romanised settlements (that is *coloniae* and towns with strong military origins, such as Cirencester) show a further move away from the non-Romanised pattern (Ibid. 1984:4, Maltby 2002:89). King also concluded that trends in the diet would be largely due to the influence of the army, its veterans and associated personnel although less detectable groups such as officials, traders and other foreigners may have also played a part (Ibid. 1984:5). Unfortunately, isotope analysis cannot provide information of which types of meat the people in this study were eating, but it can be assumed that those who ate meat were likely to be partaking in some of these dietary changes, even though their low levels of consumption could be because it could possibly have been too expensive for some people to eat much of it.

Only one other study has attempted to link TB with dietary isotopes. A recent study showed that an individual who had TB was consuming a mixed diet similar to the expected range for other hunter-gatherers in the area he was from (Guichón et al. 2015:92). The remains of a man from Myren in Chile, who was between 18 and 23 years old at the time of his death, were tested for carbon and nitrogen isotope ratios. His resulting data show he was not eating differently from other people from his population group. Despite the tiny sample size, and entirely different archaeological context, this lends some corroboration to the conclusions that most people in Roman Britain who were infected with TB were consuming a diet isotopically similar to others in their locality.

In summary, the purpose of the carbon and nitrogen isotope analysis in this study was to attempt to explore whether the individuals analysed were incomers to the cemetery population with which they were buried. As discussed, the majority of individuals appear to have been consuming foods which were isotopically similar to the foods consumed by other people who were buried in the same cemetery or region, and hence there is no evidence that the majority were migrants based on carbon and nitrogen values. Notable exceptions to this are the three Chester Road, Winchester, individuals, especially CHES512 who still stands out as having eaten a different diet to most people buried in seven sites in Roman Britain for which isotope data exist, namely the area which is now modern England. These people were possibly immigrants, and the strontium and oxygen isotope data will help to establish if this is indeed the case.

# 8.2.5 Strontium and oxygen isotope data



Fig. 8.6 Strontium and oxygen isotope data for all sites

Figure 8.6 shows all the available published data for strontium and oxygen isotopes for the comparative sites, along with the data from the current research. While there is a wide range of data, it must be considered that there will likely be immigrants to both the area and to Britain among the cemetery populations for each of the sites represented on the graph, so not every individual shown will have a British isotope signature, as can be seen by the outliers on Figure 8.6. Although the sites, (with the exception of York), were chosen for their similar underlying chalk geology in order to afford a direct comparison in terms of strontium isotope ratios, there are many other factors, such as climate, proximity to the sea, glacial soils and different water sources (Montgomery 2010:332), which will also be reflected in any underlying differences between the individual strontium and oxygen isotope results from different sites.

As regards statistical analysis of the results in this section, it must be noted that the use of Z scores for strontium isotope ratios is not as useful as for carbon and nitrogen since geology, and thus strontium isotope ratios of tooth enamel, can vary within a very small area; thus a person could be identified as being from a different population when this is in fact not the case. However, these Z score calculations have been used as a tool to further aid the visual interpretation of the graphed results. Estimated local ranges for both <sup>86</sup>Sr/<sup>87</sup>Sr and  $\delta$  <sup>18</sup>O<sub>p</sub> have been plotted as a blue line on each of the graphs to aid with visual interpretation of the results.

8.2.6 The strontium and oxygen isotope data for individual sites





Fig. 8.7 Strontium and oxygen isotope data for Driffield Terrace, York

The Vale of York, in which the city of York lies, was once a large glacial lake but is now a flat floodplain with glacial deposits overlying sedimentary sandstones, mudstones and limestones which increase in age from east to west (British Geological Survey 1977, 2001). The site of Driffield Terrace is located on a large glacial moraine (Clark et al. 2004:47). Surrounding the city of York, the North York Moors, Hambleton Hills and Howardian Hills to the east are formed of Jurassic sandstones, clays and limestones, while also to the east is the Cretaceous Chalk of the Yorkshire Wolds. To the west of the Vale of York are low foothills of Permian Magnesian limestones (Montgomery et al. 2011:144). The strontium isotope biosphere values for the York area range from 0.7090 to 0.7100 (see Figure 6.8).

York has a longitude of -0.5945 and a latitude of 54.0436 which were compared with strontium biosphere data from specific locations close by (Supplementary Data, Evans et al. 2010). These are shown in Table 8.4 below:

Sample number	Nature of sample	Longitude	Latitude	<sup>87</sup> Sr/ <sup>86</sup> Sr value
WH-C	Soil from chalk geology	-0.6057	54.1710	0.7074
WWH-351d	Dentine sample from chalk geology	-0.5762	54.0197	0.7078
E-Wet-1	Plant sample from chalk geology	-0.5762	54.0197	0.7078
WWH-161d and WWH- 173d	Dentine samples from chalk geology	-0.5762	54.0197	0.7079
WWH-121d	Dentine sample from chalk geology	-0.5762	54.0197	0.7082
WWH-275d	Dentine sample from chalk geology	-0.5762	54.0197	0.7087
DH2	Dentine sample from clay geology	-0.6561	54.0905	0.7089

*Table 8.4 Local strontium biosphere values for locations around York* (Supplementary Data, Evans et al. 2010)

These additional values suggest the local <sup>87</sup>Sr/<sup>86</sup>Sr range of the area around York is 0.7074-0.7089.

Although Driffield Terrace is the one site in the current study that is not itself sited upon chalk, it was decided to include it in the research due to the availability of published strontium isotope data from other individuals in the Driffield Terrace cemetery with which to make comparisons. The  $\delta^{18}O_{dw}$  values for York range between -8‰ and -9‰ (see Figure 6.6).

When considering isotope analysis results for DRIF13 and DRIF19, Z scores (Table 7.5) show DRIF13 is an outlier for strontium, although DRIF19 is not. It must however be considered that the previously published isotope data for other individuals buried at Driffield Terrace show evidence of people having origins from outside of York. The fact that no large differences are therefore shown on the figure or in the statistical analysis does not necessarily mean DRIF19 is not an incomer to the York area, although it is likely that DRIF13 is an immigrant. From the strontium (0.71327) and oxygen isotope results for DRIF13 (-9.1‰), and allowing for the ±0.5‰ error margins on the  $\delta$ <sup>18</sup>O<sub>dw</sub>, this person could be from a number of areas including parts of Northern Scotland, Sweden, Finland and the Czech Republic/Austria area of central Europe (see Figures 6.8 and 6.9). Although his precise place of origin is uncertain, it can be concluded that he was certainly not born and raised in York.

For comparison with previous isotope analysis on other individuals buried at Driffield Terrace, Müldner et al. (2011:285) examined a second premolar tooth from skeleton 6DRIF-9 and found <sup>87</sup>Sr/<sup>86</sup>Sr to be 0.7126 and  $\delta$  <sup>18</sup>O<sub>p</sub> to be 16.7‰. These are the closest isotope ratios to those of skeleton, DRIF13, who has a <sup>87</sup>Sr/<sup>86</sup>Sr ratio of 0.71327 and a  $\delta$  <sup>18</sup>O<sub>p</sub> of 16.0‰ ( $\delta$  <sup>18</sup>O<sub>dw</sub> -9.1‰, ±0.5‰), so they could originate from approximately the same location. Müldner et al. concluded that the strontium isotope results for 6DRIF-9 showed that he moved from an area where the rocks are older than those around York, of Palaeozoic or Precambrian date and which are significantly more radiogenic. These terrains are predominantly found in Western England. However, this is not consistent with his relatively low  $\delta$  <sup>18</sup>O<sub>p</sub>, and thus areas of Eastern Scotland or continental Europe are more appropriate origins (Müldner et al. 2011:285). It could be argued that as Scotland was not part of the Roman Empire, this individual and DRIF13 are more likely to be from continental Europe. However, individuals from outside of the Roman

Empire could have moved to provinces within the Empire for a number of reasons, for example as merchants or traders bringing required goods to England from Scotland, or they could have been slaves who were brought to England against their will. Therefore, in conclusion, even if a person has isotope results that indicate a possible origin in a province outside the Roman Empire, this still needs to be considered as possible due to the complex and far-reaching network of contacts across the Empire.

The other Driffield Terrace individual examined was DRIF19, whose strontium value (0.70937) indicates he could possibly have had a local upbringing. However, when considered with his oxygen isotope results, (-7.0‰), other regions with similar strontium and  $\delta$  <sup>18</sup>O<sub>dw</sub> data, allowing for the associated ±0.5‰ error margin, could also indicate an upbringing in western parts of the UK, modern day France, Germany or northern Italy (See Figures 6.6, 6.7, 6.8 and 6.9 and for Italy; Longinelli and Selmo 2013:80).

DRIF19 results are similar to four other individuals from the site, 6DRIF-6, with <sup>87</sup>Sr/<sup>86</sup>Sr of 0.70937 and  $\delta$  <sup>18</sup>O<sub>p</sub> of 17.5‰ (Müldner et al. 2011), DRIF16 with <sup>87</sup>Sr/<sup>86</sup>Sr of 0.709407 and  $\delta$  <sup>18</sup>O<sub>p</sub> of 17.7‰, DRIF33 with <sup>87</sup>Sr/<sup>86</sup>Sr of 0.708920 and  $\delta$  <sup>18</sup>O<sub>p</sub> of 17.4‰ and DRIF35 with <sup>87</sup>Sr/<sup>86</sup>Sr of 0.708924 and  $\delta$  <sup>18</sup>O<sub>p</sub> of 17.4‰ (Montgomery et al. 2011:167). It was concluded that 6DRIF-6 had oxygen isotope ratios which fell within the core British range for oxygen, and his strontium isotope results could be local (Müldner et al. 2011:285). Montgomery et al. (2011:167) concluded that DRIF16, 33 and 35 also have oxygen and strontium isotope results compatible with an origin in north /east Yorkshire, but these results are also compatible with an origin in much of present day France, Germany, Holland, Denmark and Southern Norway. Lead isotope values have also been studied in individuals at this site, finding that DRIF33 and 35 had values unusual for English burials, and which suggest exposure during childhood to non-English lead ores from Mesozoic ores such as those found in the Mediterranean region (Ibid. 2011:167).

It was not possible to analyse DRIF19 for lead isotopes as this was beyond the budget and scope of this study, but when two of four individuals with otherwise similar strontium and oxygen isotope results are found to have a very high likelihood of being of Mediterranean origin, it has to be considered that DRIF19 could also have originated from this region. DRIF19 has an oxygen isotope ratio within the British range and his strontium value could indicate he originated from York. However, equally, he could be from several other places, as discussed. It would be interesting to examine his lead isotope results in future research to better nuance his origin.

To support the isotope findings that a number of Driffield terrace individuals are not of local origin, and has been mentioned in section 4.8.2 but is also relevant here, a very recent study by Martiniano et al. (2016) of seven Roman individuals buried in the Driffield Terrace, York cemetery used genome and isotope analysis to identify the origins of these people. In the genome analysis, mtDNA and Y chromosome DNA analysis was possible, as all of the individuals were male. Strontium and oxygen isotope analysis was also undertaken on all seven individuals (Martiniano et al. 2016:4). The results show that one York Roman (3DRIF-26) has a genome that gave a clear Middle Eastern signal with closest neighbours of Palestinian, Jordanian and Syrian origins. Isotopic analyses supported this genetic result (Ibid. 2016:4). The other six York Romans were genetically and isotopically identified as being of British (most likely Welsh) origin (Ibid. 2016:6).

To place the Driffield Terrace burials in a Roman context to see if any light can be shed on why people moved to York in the Roman period, it is important to know that York was founded by the Romans in AD 71 and became the site of a legionary fortress and *coloniae*. The city was an imperial residence on two occasions, firstly during the reign of Septimus Severus (AD 193-211) and later during that of Constantine the Great (around AD 274-337) (Montomery et al. 2011:145). As such it will have attracted people in from all over the Roman Empire. The town was also well served by roads (Figure 8.8), which means it would have been very accessible.



Fig. 8.8 Roman roads around York. (Patrick Ottaway in Montgomery et al. 2011:144)

As was discussed in Chapter 6, the individuals were buried in the cemetery at Driffield Terrace, to the south-west of the city walls in an elevated area called the Mount (Ibid. 2011:145). The cemetery population from Driffield Terrace comprised mostly young to middle adult males of tall stature. Average height of Driffield

Terrace individuals was 3.5 cm taller than that of average males at the time (Roberts and Cox 2003:142). Of the total of 80 burials in the whole cemetery, only six were younger than the age of 18 (namely two fetuses/neonates aged up to one month, a child aged one to six years, a child aged seven to 12 years and two adolescents aged 13 to 19 years, of which DRIF13 was one) (Montgomery 2011:152). All of the individuals old enough to have their sex estimated (that is, adults/late adolescents), were male or probable male (lbid. 2011:153). Due to the unusual sex, age and height characteristics of this cemetery, and the fact that the Roman army had height standards for recruits (Friedl 1992:35), it is perhaps likely that DRIF13, 19 and the rest of the cemetery population were military men. Since the cemetery is located on the only piece of high ground alongside the main road from the south, these people were likely to have been of high status, as high ground and approach roads to towns were favoured places for burial of the Roman elite (Montgomery et al. 2011:168). Approximately 63% of the 26 individuals from the same cemetery who were analysed for strontium isotopes were certainly immigrants to the York area (Ibid. 2011:166), which would be expected if they were members of the military. While DRIF13 was definitely not born and raised in the York area, DRIF19 could have been. However, this does not rule out a military occupation for him, or for others identified as local/possible locals, because the Roman army recruited locally into the legions as required (Roselaar 2016:140). The inhumations at this site have been dated from the late 2<sup>nd</sup> to early 3<sup>rd</sup> centuries AD, which was a tumultuous period within the Roman Empire. As York was an important city, being the capital of northern Britain in this period, it would be expected that the army would need a presence here at the regional legionary fortress, particularly in turbulent times, and that they may supplement their numbers by using recruits who met the military age and height requirements.

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### (ii) Victoria and Chester Roads, Winchester

Fig. 8.9 Strontium and oxygen data for the Winchester sites

The geology of the area around Winchester is dominated by chalk with Oligocene and Eocene sands, clays and limestone. Direct measurements of the composition of the overlying biosphere in this area are not available, however, an approximation estimates that the values will range between the composition of chalk (0.7072) and rainwater (0.7092) (Evans et al. 2006:266).

Winchester has a longitude of -1.1022 and latitude 54.1114. This location was compared with biosphere <sup>87</sup>Sr/<sup>86</sup>Sr values from Evans et al. 2010 Supplementary Data. The values are shown in Table 8.5 following:

Sample number	Nature of sample	Longitude	Latitude	<sup>87</sup> Sr/ <sup>86</sup> Sr value
JMW17	Water sample from limestone	-1.3128	53.7653	0.7090
	geology			

*Table 8.5 Local strontium biosphere values for locations around Winchester* (Supplementary Data, Evans et al. 2010)

Table 8.3 shows the only available biosphere data for the Winchester area which falls within Evans et al.'s (2006) range already discussed above.

The oxygen composition of modern drinking water in Winchester is approximately -6‰ to -7‰, with modern British freshwaters ranging from around -9‰ to -4.5‰ (Evans et al. 2006:266, Figure 6.6).

The Z scores for the  $\delta^{18}O_{p}$  for individuals in the study (Table 7.5) showed no differences to the rest of the population. However for the <sup>87</sup>Sr/<sup>86</sup>Sr, CHES535 and CHES512 had Z scores above 2 (3.0 respectively), which show they are very different from the main population data. This supports what can be seen on Figure 8.10, as their <sup>87</sup>Sr/<sup>86</sup>Sr ratios are higher than for any other person in the area's extant cemeteries (CHES535 is 0.71252 and CHES512 is 0.71249), and are thus very likely to be incomers to the area. From comparison with the strontium biosphere map of Britain (Figure 6.8), they could have had an upbringing elsewhere in the UK, possibly western (eg. in Wales, Devon or Cornwall) or northern Britain (eg. Cumbria or Scotland). When considering their strontium and  $\delta$  <sup>18</sup>O<sub>dw</sub> results, allowing for the ± 0.5‰ error margin from calculations, and from looking at European maps of known strontium and oxygen isotope ratios, it was concluded that neither CHES535 (  $\delta$  <sup>18</sup>O<sub>dw</sub> -6.0‰, <sup>87</sup>Sr/<sup>86</sup>Sr 0.71252) or CHES512 (  $\delta$  <sup>18</sup>O<sub>dw</sub> -5.7‰, <sup>87</sup>Sr/<sup>86</sup>Sr 0.71249) had an origin in the Winchester area. Instead they are more likely to have originated in the far west of the UK, for example, Western Ireland, parts of Cumbria, Cornwall or Devon or even from Portugal. However, the oxygen isotope results could be slightly misleading due to the possible enrichment by up to 2‰ (Evans et al. 2012:757) because of the

breastfeeding effect, which would push their origins further west than they really were. Nevertheless, the strontium isotope results would not be affected in this manner, and still suggest a more western origin than Winchester.

CHES636 has a strontium result (0.70948) consistent with a local upbringing, but when considered with their  $\delta^{18}O_{dw}$  results (-3.9%), allowing for the ± 0.5% error margin, an upbringing in the far west of Ireland is suggested, with no corresponding values in large parts of Europe. Recent research (Late Iron Age and 'Roman' Ireland – LIARI- project of 2014) suggests although Ireland was outside of the Roman Empire, there were strong links with Roman Britain and Europe. Evidence to support this is in the form of multiple Roman finds along the western coastline of Ireland suggesting trade took place (Cahill Wilson 2014:24) and evidence of the participation of Irish men in the Roman Army (Ibid. 2014:35) proves this origin, whilst outside of the Roman provinces, could have been possible. That said, a first molar was sampled from CHES636, and the enamel of this would have been likely to have entirely formed when he/she was breastfeeding. This would cause enrichment of the oxygen isotope results by up to 2‰ (Evans et al. 2012:757), pushing the possible origin of the person further to the west than it really would have been, so therefore more emphasis is placed on the strontium isotope data, with the conclusion being this individual could have been brought up locally.

Comparing these individuals with the published data for other contemporary individuals buried in the area, CHES535 with a <sup>87</sup>Sr/<sup>86</sup>Sr of 0.71252 and  $\delta$  <sup>18</sup>O<sub>p</sub> of 18.0‰, and CHES512 with <sup>87</sup>Sr/<sup>86</sup>Sr of 0.71249 and  $\delta$  <sup>18</sup>O<sub>p</sub> of 18.2‰ are similar to Ay21-1277 from the Lankhills cemetery with <sup>87</sup>Sr/<sup>86</sup>Sr of 0.7115 and  $\delta$  <sup>18</sup>O<sub>p</sub> of 18.2‰ (Chenery et al. 2010). These results were interpreted as showing the individual could be from the UK but not from the Winchester area. This is because the <sup>87</sup>Sr/<sup>86</sup>Sr ratio is more typical of bottled waters from Palaeozoic (especially Carboniferous and Devonian) sandstones and mudstones which are found in

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western (eg. Wales, the Malverns and parts of Devon or Cornwall) and northern Britain (eg. Cumbria and parts of Scotland) Chenery et al. (2010:427).

VICT96 and VICT192 have strontium isotope ratios (0.70937 and 0.70943 respectively), which fall within the local range for Winchester. Their oxygen isotope results ( $\delta^{18}O_{dw}$  of -6.3% and -6.0% respectively), including allowing for the error margin of ± 0.5%, support this. However, with these strontium and oxygen isotope results, they could equally have originated from north western areas of Britain, for example the Cumbrian coast, or even from southern France. A first premolar was sampled from VICT129, which could still have been calcifying when she was breastfeeding as a child, thus accounting for her slightly enriched oxygen isotope results compared with VICT96, who had a second premolar sampled. This tooth would have calcified after breastfeeding was likely to have finished. In conclusion, VICT96 and VICT192 were likely to have been of local origin.

In comparison with published isotope data for Winchester burials, Chenery et al. (2010) analysed skeleton number Ay21-0776 and found that this individual had <sup>87</sup>Sr/<sup>86</sup>Sr of 0.7096 (local values ranging from 0.7096 to 0.7098) and  $\delta$  <sup>18</sup>O<sub>p</sub> of 17.8‰, which is also similar to VICT96 and VICT192. Chenery et al. suggested the strontium isotope ratios are comparable with biosphere data from vegetation growing on Mesozoic, non-limestone terrains, bottled waters from carboniferous limestone aquifers and Mesozoic mudstones, and sand and clay geologies. Areas compatible with these data are Mesozoic terrains around the areas of Bath, Bristol and Gloucester, and they are also compatible with origins on older rocks in coastal areas where marine strontium has a significant impact on local biosphere values (Chenery et al. 2010:427). This individual was therefore concluded to be non-local to the Winchester area (Ibid. 2010:427), although it could be argued that the strontium result fell just within the local range, and so this person could have been of Winchester origin.

To put the results into context with the Roman period, at the time of the Roman conquest Winchester existed as *Venta Belgarum* – the market place of the Belgae.

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It was the fifth largest town in Roman Britain in terms of the area it covered. However, it does not appear to have had a large population or long distance trade at the time of the conquest (Ottaway et al. 2012:16). Archaeological evidence suggests it may have been a tribal and religious center with the potential to control important communications routes (Ibid. 2012:16). Evidence for a fort is tenuous. The only possible archaeological evidence was in the form of ditches dated to around AD 50. However, the position of these ditches on a valley bottom suggests it is unlikely to have been a fort (Ibid. 2012:17). Around AD 70, the first Roman defences were constructed (Ibid. 2012:17) and a programme of drainage control was implemented until around AD100 when a grid of streets was established in the newly drained area with a forum and basilica at its centre. This was thought to make the establishment of Winchester as a civitas capital (Ibid. 2012:18).

In terms of access to Winchester, there were certainly five and possibly six longdistance roads that converged here (See Figure 8.10). These linked with Old Sarum (west), Cirencester (north-west) and Silchester (north-east). Other roads led to Chichester (south-east) and to the south. The sixth road possibly led to Neatham (Ibid. 2012:19).



Fig. 8.10 Roman roads around Winchester (Ottaway et al. 2012:10)

Whilst a range of crafts was practiced in the town, excavated evidence does not show this to be on a scale beyond that of meeting local needs. However, metalworking (particularly iron) was increasing in the later Roman town (Ibid. 2012:19). It is thought that cloth making took place in Winchester but no buildings (weaving or fulling mills) associated with the textile industry have been excavated (Wacher 1995:299, Ottaway et al. 2012:19). However, the area was certainly a centre of textile manufacture by Medieval times (Wacher 1995:299). Therefore there is little archaeological evidence to suggest what was so attractive about the town to attract migrants such as the individuals from the cemeteries at Chester Road.

### (iii) Gambier Parry Lodge and Cirencester



Fig. 8.11 Strontium and oxygen isotope data for sites in Gloucestershire

These results show quite a range of values for both strontium and oxygen (see Figure 8.11). The geology of the Gloucester area is complex in terms of geological age in addition to outcrop patterns (Chenery et al. 2010:152). The Vale of Gloucester has early Jurassic, Lower Lias mudstones partly overlain by Quaternary alluvia, undifferentiated river terrace deposits and Holocene to recent tidal flat deposits. The Vale is then bordered by three geologically different areas: to the south and southeast lie the Cotswold Hills which are dominated by early Jurassic limestones and subordinate mudstones. To the west lies the Forest of Dean which is comprised of Palaeozoic - Silurian to Carboniferous-sandstones and mudstones. Finally, the Malvern Hills are situated to the north and are comprised of Precambrian and Cambrian igneous and meta-igneous rocks and Palaeozoic sediments. In addition, the Malvern Hills are separated from the Vale of Gloucester by an expanse of Triassic mudstones partly covered by post-glacial rubble drift. Hence within a 30km radius of Gloucester, the geology ranges from Precambrian to Jurassic age rocks and deposits. Due to this complicated geological situation and the lack of published isotope data for the area, Chenery et al. (2010:152) included a study of strontium isotope values from a 30km area around Gloucester to provide a local range of biosphere strontium for comparison with the human data they were analysing. This biosphere study included analysing samples of water, soil, rocks, faunal remains and vegetation. The distance of 30km was chosen because it was assumed a Gloucester population would have sourced most of its food from this area in the past; this is estimated as one day's travel from the town (Chenery et al. 2010:154), and the <sup>87</sup>Sr/<sup>86</sup>Sr ratios were found to range from 0.7077 to 0.7162, which is a wide range for a small area (Ibid. 2010:156).

Cirencester (CIRE) has longitude -1.5935, latitude 51.5501. Nearby locations with strontium biosphere data were recorded by Evans et al. (2010) and are shown in Table 8.6 below:

Sample number	Nature of sample	Longitude	Latitude	<sup>87</sup> Sr/ <sup>86</sup> Sr values
Dwplant-06	Plant sample from chalk geology	-1.7868	51.1924	0.7079
Dwplant-02	Plant sample from chalk geology	-1.7720	51.1994	0.7077
Dwplant-03 and Dwplant- 08	Plant samples from chalk geology	-1.7720	51.1994	0.7087

*Table 8.6 Local strontium biosphere values for locations around Cirencester* (Supplementary Data, Evans et al. 2010)

The biosphere strontium values in Table 8.6 provide a local strontium biosphere range of 0.7079-0.7087 (Evans et al. 2010 Supplementary data) which falls within the range extrapolated above the table.

Gambier Parry Lodge (GRPL) has longitude -0.4929, latitude 52.4640. Nearby locations with strontium biosphere data (Evans et al. 2010) are shown in Table 8.7 below;

Sample number	Nature of sample	Longitude	Latitude	<sup>87</sup> Sr/ <sup>86</sup> Sr value
KET-21	Plant sample from limestone geology	-0.4129	52.6366	0.7088
KET-20	Plant sample from limestone geology	-0.4272	52.6494	0.7088
KET-17	Plant sample from limestone geology	-0.5277	52.6507	0.7089
Amer4	Land snail shell from chalk geology	-0.5963	51.6847	0.7094
KET-12	Plant sample from chalk geology	-0.5885	52.6451	0.7099
KET-13	Plant sample from chalk geology	-0.5102	52.6442	0.7104

Table 8.7 Local strontium biosphere values for locations around Gambier Parry Lodge(Supplementary Data, Evans et al. 2010)

When combined with the data from Chenery et al. (2010), the values from Table 8.5 give an extended local  ${}^{87}$ Sr/ ${}^{86}$ Sr range of 0.7088-0.7104.

For the oxygen isotope range, the composition of UK freshwater has a known range of  $\delta$  <sup>18</sup>O between -9.0‰ and -4.5‰, with Gloucester falling between -7.0‰ and -7.5‰ (Darling et al. 2003).
Figure 8.12 shows that the Gambier Parry Lodge individuals were both found to plot within the strontium isotope data range for modern vegetation taken from a 30km radius of Gloucester (0.7077-0.7162); they were also constrained to within the range of bio-accessible strontium (0.7077-0.7109) for vegetation growing on the Jurassic terrains of the Vale of Gloucester and the Cotswolds (Chenery et al. 2010:156), with <sup>87</sup>Sr/<sup>86</sup>Sr for GRPL531 being 0.71008 and GRPL538 being 0.70924). The Cirencester individuals had strontium ratios which also fell within the Vale of Gloucester and Cotswold local biosphere values, with CIRE189 being 0.70884, CIRE37 with a value of 0.71007 and CIRES being 0.70992. These observations were confirmed by Z scores, which showed no outlying differences from other members of the cemetery population.

When  $\delta^{18}O_{dw}$  is considered, and allowing for the error range of ± 0.5‰, the results suggest CIRES (-6.5‰) could be local. A second premolar was analysed from this individual and therefore the breastfeeding enrichment effect need not be taken into account. CIRE189 ( $\delta^{18}O_{dw}$  of -5.2‰), CIRE37 (-5.2‰), GRPL531 (-7.6‰) and GRPL538 (-7.6‰), had teeth sampled which could have been affected by breastfeeding, so their oxygen isotope results may be more enriched than expected for teeth formed after the cessation of breastfeeding. Thus their  $\delta^{18}O_{dw}$  results as they stand would suggest a more westerly upbringing. It was decided to place more emphasis on the strontium isotope results for these people, and to conclude from these that they were possibly local.

While these individuals could all be local to the Gloucester area, they could also originate from areas on the continent of Europe, and by taking both strontium and oxygen isotope results into account and making comparisons with available strontium and oxygen isotope data from Europe (Figures 6.9 and 6.7 respectively), the Gambier Parry Lodge individuals, GRPL531 and GRPL538, could originate from areas now in modern Denmark, north western Germany, Belgium and southern France. The Cirencester individual CIRE189 could originate from the northern Spanish coast, the coast of southern France or the coast of southern

Ireland, while CIRE37 may have been born and raised on the coast of western Ireland or the northern Spanish coast, and CIRES in the region of modern Cumbria, south Wales, southern Ireland or the south of France.

To put these results into Roman period context to suggest why people may have been attracted to the town, a brief examination of the history of urbanisation in the area reveals that the Roman town of *Corinium* (modern Cirencester) was not built on the site of Late Iron Age activity, but was located some 3km to the south of the *oppidum* of Bagendon (Moore 2014:26). Survey and excavation have indicated that the Bagendon complex probably began as a group of small banjo-like enclosures in the Middle Iron Age, which probably had a communal agricultural role (Ibid. 2014:27). Bagendon's heyday appears to have been in the mid 1<sup>st</sup> century AD with most of the occupation dating from around AD 40-60, although some activity may have started as early as AD 20 (Ibid. 2014:28). The abandonment of the complex at Bagendon seems to have taken place around AD 60, contemporary with the development of *Corinium*, after which *Corinium* replaced Bagendon's role as centre of production and exchange (Ibid. 2014:30).

It is thought a fort was established at Cirencester within a year of the Roman invasion of Britain. By the early AD 60s, a new fort for a cavalry regiment was established and a civilian settlement started to grow (Wacher 1995:304). Wacher suggested that the fort and its occupants would have provided an incentive for a gradual migration of people to live in the surrounding vicus. The Gambier Parry Lodge and Cirencester individuals were all possibly locals, but they could have been short-distance migrants who took advantage of the military presence and developing town and better opportunities for work, to move from more rural areas. Urban living and close contact with military people who could be immigrants could put them at increased exposure to and risk of contracting infectious diseases such as TB. In addition to the military presence, four schools of mosaicists were known to exist in Roman Britain and these were based in Cirencester and Dorchester with two others in Water Newton and Brough–on-Humber (Bennett 1988:26). The Cirencester school was particularly successful with over 50 mosaics credited to its

workshops. It had very close links with a workshop in Trier, Germany. Trier is on the Moselle river not far from Luxemburg, and was the key city of the Roman northern territories, with people converging here from all over the Roman Empire and elsewhere in the world. Presumably there was movement of tradesmen, craftsmen and women between the two cities, and therefore "locals" could have come into either direct or indirect contact with these people and their diseases, which could be one explanation of how they contracted TB.

Meanwhile, the town of Gloucester started as a *coloniae* in the 1<sup>st</sup> century AD. These were settlements of retired legionaries who were granted an area of land when they left the legions. The functions of the *coloniae* were to serve as a model of Roman urban life towards which the natives could aspire. Early in the Roman era, they also formed a convenient reserve of trained soldiers (Bennett 1988:7). The town would therefore have had a large proportion of immigrants to the area in the form of retired soldiers. Italy was the most important source of legionary recruits into the army in the first half of the 1<sup>st</sup> century AD, but after this Italians only made up 50% of the recruits unless a new legion was created (Roselaar 2016:140). The supplementary 50% was made up of recruits drawn from descendants of Italians settled in colonies in Narbobensis, Spain and Africa and also from the area in which the legion was situated (Bennett 1988:7). While GRPL531 (aged 25 – 35 and probably female) and GRPL538 (aged 8 – 9) could not have been military personnel directly, they could have been associated with ex-military people, perhaps working in service and therefore coming into contact with people from all over the world. GRPL531 and 538 were themselves likely to be locally born, but having been in Gloucester during the Roman period would mean exposure to many migrants and their accompanying diseases.

### (iv) Easington and Baldock



*Fig. 8.12 Strontium and oxygen isotope data for Easington and Baldock* 

Whilst Baldock and Easington are two different sites both geographically and historically, there is no comparative isotope data from other skeletons excavated from the same sites, or published reports on either location, both sites have similar local strontium ranges and so for the purposes of identifying possible non locals to this strontium data range, the results have been plotted on the same axis, but are discussed separately below.

Baldock is built upon chalk geology which is overlain with free-draining calcareous to neutral loamy brown soils along with thin drift deposits of boulder clay and glacial gravel (Babtie Group: 2011:147). Based upon Figure 6.8, the local range for <sup>87</sup>Sr/<sup>86</sup>Sr was estimated to lie between 0.7080 and 0.7090.

Baldock has longitude -0.0327, latitude 52.0356. The closest strontium biosphere data from Evans et al. (2010) is shown in Table 8.9 below;

Sample number	Nature of sample	Longitude	Latitude	<sup>87</sup> Sr/ <sup>86</sup> Sr values
GIR-1.1	Plant sample from greensand geology	-0.0813	52.2411	0.7084

*Table 8.9 Local strontium biosphere values for locations around Baldock* (Supplementary Data, Evans et al. 2010)

This additional biosphere data falls within the range already indicated on the map, Figure 6.8.

The  $\delta$  <sup>18</sup>O<sub>dw</sub> for Baldock is between -8‰ and -9‰ (see Figure 6.6).

The burial site of Baldock is now examined (see Figure 8.13). Both BALD7230 (0.70932) and BALD7498 (0.70904) fall just outside the expected strontium isotope range to be locals, although BALD7498 is very close to the local range, as can be observed on Figure 8.12. Their  $\delta^{18}O_p$  values fall within the expected range (16.8 -18.6‰) for the UK. When  $\delta^{18}O_{dw}$  results for these two individuals are considered, in order to try to nuance an origin by comparing the data with the map of values for UK spring and well waters, they are -7.0‰ (± 0.5‰) for BALD7230 and -7.1% (± 0.5%) for BALD7498. In comparison with the map of oxygen isotope values for UK spring and well waters (Figure 6.6), the Baldock area has a range of -7‰ to -8‰, between which these two individuals may fall, and hence they could both be local to the modern Baldock area. However, allowing for possible errors of  $\pm 0.5\%$ , an origin further to the south and the west is possible, and this could corroborate the strontium isotope results. It must be borne in mind that both teeth sampled started to become calcified when the individuals were still breastfeeding, so this could lead to slight enrichment of the oxygen isotope results. However, comparisons with British and European strontium isotope results (Figures 6.8 and

6.9) and oxygen drinking water values (Figures 6.6 and 6.7) show that these two people could have also originated in most of England, some areas of Scotland, modern Denmark, western Germany and Austria. Similar strontium and oxygen isotope results are also found in coastal regions of Italy (Longinelli and Selmo 2013:80).

To put these results into perspective with activity in the Roman period, is known that Baldock started out as a Romano-British "small town" having first developed in the early 1<sup>st</sup> century BC as an *oppidum* of regional importance in the Iron Age. Later in the 1<sup>st</sup> century BC, its role was overtaken by nearby Braughing and *Verulamium* (see Figure 8.13). However, Baldock continued to flourish and had reached its greatest extent during the 2<sup>nd</sup> century AD. It began to shrink during the 3<sup>rd</sup> into the 4<sup>th</sup> centuries AD and by the late 4<sup>th</sup> century the town was in decline (Burleigh and Fitzpatrick-Matthews 2010:viii). Unpublished excavations revealed the presence of a road running south-south-west to *Verulamium* which was present as a track in the early Iron Age but continued to be used in the Roman period. Further prehistoric trackways which continued to be used as Roman roads were located running to Braughing and *Camulodunum*, and to the Romano-British settlement at Sandy. This later road joined the major road crossing at *Duroviguto* (Godmanchester) (Ibid. 2010:5) (see Figure 8.14), thus the town was easily accessible.



Fig. 8.13 Roman roads around Baldock (Burleigh and Fitzpatrick-Matthews 2010:1)

Some 22 cemeteries have been discovered around Baldock (Ibid. 2010:viii), suggesting there was a considerably sized population in the town over its duration. Therefore there was opportunity for the movement of goods for trade to enter and leave Baldock, along with the people who accompanied them, which could have included BALD7230. However, BALD7498 has isotopic signatures that suggest they are local. Of course, they could be regionally local and have moved a few miles from the countryside into the town itself. As has been discussed in Chapter 2, it is well documented that urban living contributed to the spread of TB, and that migrants to the more cramped living conditions in towns were more at risk of contracting the disease, which could possibly explain how BALD7230 and 7498 contracted TB.

The site of Easington shall now be considered. Easington (longitude -0.0931, latitude 53.4105) lies upon Upper Cretaceous chalk geology which is overlain with glacial deposits of till and boulder clays (UK Fossil Collecting: 2010). There are no

nearby strontium biosphere samples from Evans et al. 2010 Supplementary data, therefore in order to define the local biosphere strontium isotope signature, samples of cattle bone and a snail shell contemporary with some late medieval (14<sup>th</sup> to 16<sup>th</sup> century AD) burials found in nearby Hull were used (Roberts et al. 2013:276). A series of soils from across the region were also sampled (Ibid. 2013:276). These were taken from areas free from irrigation, fertilisers and contamination from landfill sites in order to provide a more accurate representation of strontium isotope ratios in the past (Ibid. 2013:276). The soil samples were also taken from depths of at least 60cm to minimise modern contamination (Ibid. 2013:277). The  $\delta$  <sup>18</sup>O<sub>dw</sub> for Easington is between -7‰ and -8‰ (see Figure 6.6).

EASN183 has <sup>87</sup>Sr/<sup>86</sup>Sr of 0.71031, which is not compatible with an upbringing in Easington; this area has a strontium biosphere range of 0.7080 – 0.7090 (Evans et al. 2010:2). This is further supported by the research of Roberts et al. (2013:282) who found the Hull area, within a 50km range of Easington, has a <sup>87</sup>Sr/<sup>86</sup>Sr range of 0.7074 – 0.7086. Within 100km to the west of Easington the range is 0.7083 – 0.7106, and to the north west, it is 0.7075 – 0.7105, with which EASN183's results are compatible. In Britain, strontium ratios of 0.71031 are found elsewhere, for example, in the south-west of England (the southern parts of modern Cornwall) and parts of the north west (Cumbria and north west Wales), and in southern Ireland or southern France. When the strontium values are considered with a  $\delta$ <sup>18</sup>O<sub>dw</sub> of -6.8‰ (± 0.5‰), the oxygen isotope results for EASN183 certainly support the suggestion that this individual came from an area within 100km of Easington or parts of the north west (Cumbria and north west Wales), however they are also compatible with parts of southern France. A first premolar tooth was sampled from EASN183 which could have started calcification whilst he was still being breastfed. This could have led to slight enrichment of oxygen isotope values, and therefore placing more emphasis on the strontium isotope results, it was concluded that this individual was not local to Easington itself.

Placing these results into Roman perspective, there is very little documented evidence of Easington in the Roman period. Prior to around 2003, evidence for

Roman occupation of the area around Easington was limited to the recovery of isolated pottery finds eroding from the cliff edge (Richardson 2003:4). However, the presence of Iron Age and/or Roman settlements is suggested by evidence from aerial photographs which show at least seven enclosures to the north west and west of Easington (Ibid. 2003:4). Evidence of late Iron Age/ Roman settlements and industrial activity was found in 2003 near the town. Dating evidence was in the form of Roman greyware, a sherd of Dressel 20 amphora, mortaria and possible Daleswares from the 2<sup>nd</sup> century AD (Ibid.2003:15). A fragment of a glass bangle dated to the later part of the 1<sup>st</sup> century AD (Ibid. 2003:22). Industrial activity was in the form of a kiln structure with nearby pottery sherds dating from the 1<sup>st</sup> to the 3<sup>rd</sup> centuries AD (Ibid. 2003:25). However, it seems likely that other evidence of activity from the Roman period is likely to be undiscovered at present, because the site is near the mouth of a major river leading to York, and this may have provided work and thus attracted people to the area.

### (v) Poundbury



Fig. 8.14 Strontium and oxygen isotope data for Poundbury

Poundbury lies on Cretaceous chalk bedrock which is overlain with superficial river terrace deposits of sand and gravel (BGS Geology of Britain viewer). The local <sup>87</sup>Sr/<sup>86</sup>Sr biosphere ratios range between 0.7080 and 0.7090 (see Figure 6.8).

Poundbury has a longitude of -2.2322 and latitude 50.5625. Sites near to this location for which strontium biosphere data from Evans et al. (2010) is available are shown in Table 8.10, following:

Sample	Nature of	Longitude	Latitude	<sup>87</sup> Sr/ <sup>86</sup> Sr
number	sample			values
MW-C	Soil sample from chalk geology	-1.9753	50.9209	0.7076
WIN-C	Soil sample from chalk geology	-1.3165	51.0603	0.7076
MW-S1	Soil sample from chalk geology	-1.9753	50.9209	0.7077
MW-S2	Soil sample from chalk geology	-1.9753	50.9209	0.7077
F23E	Dentine sample from chalk geology	-1.9753	50.9209	0.7078
F23A	Dentine sample from chalk geology	-1.9753	50.9209	0.7078
F23D	Dentine sample from chalk geology	-1.9753	50.9209	0.7079

*Table 8.10 Local strontium biosphere values for locations around Poundbury* (Supplementary Data, Evans et al. 2010)

The additional data in Table 8.10 gives a  ${}^{87}$ Sr/ ${}^{86}$ Sr range of 0.7076 – 0.7079, which more or less corresponds to the values from the map, Figure 6.8.

The  $\delta$  <sup>18</sup>O<sub>dw</sub> of the local area ranges between -6‰ to -7‰ (see Figure 6.6).

The Z scores (see Table 7.5) do not show any outliers when compared to the rest of the Poundbury cemetery population; no individuals stand out as being obviously different to anyone else. As can be seen in Figure 8.23, POUN228 has a <sup>87</sup>Sr/<sup>86</sup>Sr of 0.70854, which is certainly compatible with an origin in the area around Poundbury, as are those for POUN257 (0.70886) and POUN506 (0.70835). However, even allowing for the ± 0.5‰ error margins, the  $\delta$  <sup>18</sup>O<sub>dw</sub> of these individuals are all too high to indicate this as being possible, all indicating warmer and/or wetter origins than the -6‰ to -7‰ expected for Dorset (see Figure 6.6).

Unfortunately, the only teeth available to sample for POUN 228 ( $\delta^{18}O_{dw}$  of -5.3‰, ± 0.5‰), POUN257 (-4.3‰, ± 0.5‰) and POUN506 (-3.9‰, ± 0.5‰) were first molars (M1). The oxygen isotope data will have been affected by breastfeeding during the start of the period of enamel formation of these teeth. This is known to increase  $\delta^{18}$ O values by up to 2‰ (Wright and Schwarcz 1998, Evans et al. 2012:757). While oxygen isotope data may fractionate as a result of breastfeeding, strontium isotope results are unaffected therefore the strontium isotope results carry more weight when attempting to nuance the origin for these individuals. In comparisons with strontium isotope maps of Britain and Europe (Figures 6.8 and 6.9), further suggestions of origins could be made. POUN228 could be from areas on the coasts of southern Ireland, northern Spain, southwest France, western Germany and Italy.

A second molar tooth was analysed from POUN1201, and thus avoided confounding by breastfeeding effects. This individual had <sup>87</sup>Sr/<sup>86</sup>Sr of 0.70903, which is definitely compatible with the Poundbury area. However, when considered along with the  $\delta^{18}O_{dw}$  (-5.4‰, ± 0.5‰) this is more fitting with an origin further to the west, such as modern day Cornwall, south west Wales, south west Ireland, and the coast of northern Spain or Portugal. These results suggest that POUN1201 was possibly not local to Poundbury. POUN619 also has <sup>87</sup>Sr/<sup>86</sup>Sr result (0.70870) compatible with an origin in Poundbury. The tooth analysed was an M2 and this also avoids an elevation due to the breastfeeding effect. This  $\delta$  $^{18}O_{dw}$  is very high (-1.2‰, ± 0.5‰) meaning the individual had an origin somewhere much warmer and/or wetter than Dorset. The strontium ratio could indicate an upbringing in many parts of Europe, including modern Spain, France, Germany and Italy, but could also cover parts of central and eastern England including areas that are not far from modern Dorset. However, the very high oxygen result could suggest this individual was not local. Redfern et al. (2016:19) found an individual (sample number BL44) excavated from a 2<sup>nd</sup> to 4<sup>th</sup> century cemetery at Lant Street in the southern burial area of Roman London had a  $\delta$  $^{18}O_{dw}$  of -1.4‰ (± 0.5‰). This was reported as being higher than any other  $\delta$   $^{18}O$ 

values reported for Roman Britain and was interpreted as being from the southern reaches of the Mediterranean (Ibid. 2016:20). POUN619  $\delta^{18}O_{dw}$  were even higher than this and so a southern Mediterranean or African origin could also be possible for this individual.

POUN636 has <sup>87</sup>Sr/<sup>86</sup>Sr ratios compatible with a Poundbury upbringing (0.70875), but could also indicate an upbringing in many parts of Europe, including modern Spain, France, Germany and Italy, parts of central and eastern England including areas that are not far from modern Dorset. The tooth sampled was a deciduous M2, and so was formed before birth and while the individual was breastfeeding, and thus the oxygen isotope values (-5.4‰,  $\pm$  0.5‰) can be expected to be enriched. However, when considered incorporation with the strontium isotope results, POUN636 could possibly originate from Portugal, western Spain or southern Ireland.

To consider the Roman context for these results, Poundbury is located on the outskirts of Dorchester and is on the site of a former Iron Age hillfort (Wacher 1974:323). An early Roman fort was developed at or near Dorchester and people probably drifted towards living in the vicus (Ibid. 1974:324). Dorchester emerged as a town with an amphitheatre (Ibid. 1974:326) and a large bath house (Ibid. 1974:325) and also an aqueduct which extended from the river Frome 17.6 km south of Dorchester, its flow estimated as 59 million litres per day (Bennett 1984:15). In terms of industry, a lead working hearth was excavated in the forum, along with six metal working furnaces (Wacher 1974:331). A local brickworks produced bricks with decorative bearded faces (Ibid. 1974:332) and there were marble guarries at nearby Purbeck, the stone from which was used all over England (Ibid. 1974:332). This quarry could have attracted migrants to the town for work or trade. POUN1201 was a male of 36 to 45 years who was within an age bracket that could have been involved in this work. Meanwhile, the other Poundbury probable non-local was POUN616 (aged 15). If this person was indeed an immigrant to the town, they may have come as part of a family unit who had moved to the area. Poundbury was close to the river Frome and also served by a

Roman road (see Figure 8.15) and Dorchester is positioned within easy access to the English Channel and thus to shipping routes to and from continental Europe so travel to the town would be relatively straightforward.



Fig. 8.15 Roman roads near Poundbury, Dorchester. (Farwell and Molleson 1993:3)

### 8.2.7 Summary

There have only been three recent studies linking transmission of infectious disease with isotope analysis (Roberts et al. 2013, who studied treponemal disease, and then Inskip et al. 2015 and Roffey et al. 2017 who examined the link between leprosy and mobility in just one individual each). The leprosy studies had the benefit of strain data to support the isotopic suggestions of origin for the infected individuals. Inskip et al. concluded the person probably brought the 3I strain of *Mycobacterium leprae* to Great Chesterford, Essex from a possible Scandinavian origin, probably introducing this strain to England. However, Roffey et al. concluded the individual from Winchester that they analysed was not local to

the Winchester area, and carried the 2F strain (currently associated with south central and west Asia), but they could not conclude the infected man brought the disease to the area with him.

So the current study is breaking new ground and has discovered some problems. However, some of the issues encountered during this work were also found by Roberts et al. in their study of skeletons from the 14<sup>th</sup> to 16<sup>th</sup> century AD cemetery of the Augustinian friary at Hull Magistrates Court, England. This attempted to link evidence for treponematosis with data from stable isotope analysis to test the hypothesis that people with treponemal disease buried at this site were not locally born and raised (Roberts et al. 2013:273). This research found that four individuals out of 12 may have originated from outside England. One of these four had bone changes consistent with treponematosis and it was therefore concluded that most individuals with bone changes studied who were buried at this site contracted their infection within Britain (Ibid. 2013:280). However, those without bone changes could not be proved to not have had the infection; they may have died before bone changes occurred (Wood et al. 1992). Thus, it was not possible to support the hypothesis that people with treponemal disease came into England via trade and contact during the late medieval period. The current research identified a similar problem with drawing conclusions about the origin of the TB infections of the people in the study.

Diagnosis of disease is very challenging in paleopathology. Diagnosis of TB in the current study was made by different bioarcharchaeologists and some of the skeletal changes observed may have causes other than TB. aDNA analysis has not been able to provide the definitive answer to diagnosis and strain typing that was hoped when this study was started in 2013. In theory, the aim was to use the TB strain data in conjunction with isotope analysis in order to decide where the individuals analysed had contracted their infection (comparing with the modern genomic data on strains and their geographic location today), and so the lack of this information has limited the conclusions possible from the current study. Roberts et al. stated that whether people with treponemal bone changes had the

infection before moving to Hull, or whether they developed the infection after moving will never be known, but it is still a possibility that treponematosis was brought to Hull through movement of people (Ibid. 2013:281). Exactly the same conclusions can be drawn about the transmission of TB in the current study.

While speculation on a place of origin for individuals using isotope analysis can be made, the method is limited in its ability to pinpoint very specific origins, such as particular places, rather a regional origin, for example a region or a country, is currently more realistic. It is necessary for further research into establishing biogenic strontium isotopic signatures from local plants and animals in order to refine the strontium ranges for more specific geographic areas in order for places of origin to be more tightly defined and constrained in future studies. As can be seen in the discussion of the results from the sites in the current study, more detailed data are currently available for some areas than others, and limiting the sampling of skeletons to those buried in areas of chalk geology was of no advantage in overcoming this paucity of biosphere strontium data. This is because glacial drifts and deposits overlying the geology along with other factors, such as proximity to the sea will also affect skeletal strontium isotope values (Bentley 2006). However, the hypothesis proposed in this study only required identifying people who were local or non-local to their place of burial; as such, this was largely possible to achieve from the methods used and the results described.

In summary, Table 8.11 shows the possible locals and Table 8.12 shows nonlocals at the sites studied:

Possibly local	Sex	Age
DRIF19	М	26-35
POUN228	Unknown	9
POUN257	Unknown	10
POUN506	Unknown	12
POUN636	Unknown	4
BALD7498	F	26-35
GRPL531	F?	25-35
GRPL538	Unknown	8-9
CIRE37	Unknown	9-10
CIRE189	F	26-35
CIRES	М	18-25
CHES636	Unknown	12-17
VICT96	F	18-25
VICT129	F	18-25

Table 8.11 Individuals who are likely to be local to their place of burial.

Non-local	Sex	Age
CHES512	F	18-25
CHES535	М	26-35
DRIF13	M?	16-19
EASN183	М	>45
POUN619	Unknown	15
POUN1201	М	36-45
BALD7230	М	26-35

Table 8.12 Individuals likely to be non-local to their place of burial.

The ages and sexes of locals and non-locals were included in these tables to see if there is an obvious pattern visible, although in-depth analysis of this would require the examination of age and sex of locals and non-locals in all published isotope data for Roman Britain and thus is beyond the scope of the study. The current research had a sample which was restricted to individuals with TB who were buried on chalk geology, with no heed being taken of sex or age when the sample was chosen. For the possible locals, seven of the 14 individuals (50%) were non-adults, five out of 14 were female or possible female (36%) and three out of 14 were male (21%), whereas for non-locals, one of the seven individuals (14%) were non-adults, two (29%) were female and three out of seven (43%) were male or probable male. Although this is a very small sample, it would appear that more locals were non-adults, which may be expected; young children may not have had time to be mobile before they became ill and died. Two of the non-locals were female. We do know that females were mobile during the Roman period (Bruun 2016:176, Holleran 2016:117, Prowse 2016:208) and these findings support this evidence. In addition a study used strontium isotope analysis to examine 70 human skeletons from Neuberg, Donau in Germany (Schweissing and Grupe 2003:1377). This site was associated with a late Roman fortress. Of the skeletons examined, they found 56% of females and 37% of males to be nonlocals. The researchers explained this high level of mobility of females being due to exogamy, that is, females moving to marry, or moving as a result of their marriage, which could possibly be the case in this study. However, it is impossible to know with certainly if women were mobile within their own rights in Roman times. Very young children presumably moved with their mothers.

As osteological ages are provided as a range, and many of these age ranges are similar, it is more difficult to draw conclusions based upon age of individuals relating to their mobility. However, in the published literature, Prowse et al. (2007) state that they were the first to research mobility linked with age in the Roman period. They examined oxygen stable isotope ratios of 61 individuals who had lived in the town of *Portus Romae* and who were buried in the Isola Sacra necropolis near Rome, dated to the 1<sup>st</sup> to 3<sup>rd</sup> centuries AD (Prowse et al. 2007:515). They found that 14 individuals (23%) had migrated to the area before the third molar crown had completely formed (that is between 10 and 17.5 years of age) (Ibid. 2007:515). Clearly, children were moving around in Roman Italy during times contemporary with the current study, and it is assumed this pattern could be

repeated in other parts of the Roman Empire. Therefore it is reasonable to conclude that the non-adult who was found to be non-local in this study was probably not unusual in the Roman period. Two females were also found to be non-locals and again it might be assumed that families travelled in Roman Britain as a matter of normality.

Compared with transmission of TB today, the data from this study appear to show some differences. However, it is difficult to draw conclusions from this because, in addition to the small sample size, as has been discussed above, the data was not collected to demonstrate a link between age and sex and infection with TB. This is therefore beyond the scope of the current research but could be examined in future studies into the disease in the past. In Chapter 2 it was discussed that, clinically, older people are at an increased risk of contracting TB (Cohen and Dye 2014:23, Ponnuswamy 2014:131) and males are at a higher risk of catching TB compared with females (WHO 2015). It has been suggested that lifestyle factors may play a role in contracting TB (see Chapter 2), and if that is indeed the case, age-related roles may have changed since the Roman period, meaning more nonadults caught the disease then compared with now. Another hypothesis is that fewer people reached old age during the Roman era, having already died of TB or other causes, and therefore they are not represented amongst the sample for the current research. However, it is almost impossible to compare modern mortality rates for a disease with those for archaeological populations. It is unlikely that a representative sample of Roman period individuals of different age groups who had TB have been excavated, so it is difficult to comment with accuracy which demographic groups were most affected by the disease in the Roman period. It must also be considered that the presence of pathological conditions in skeletal remains, such as those indicative of possible TB, indicate the presence of chronic health problems. Therefore, those individuals in the current study with apparent poor health and TB, as indicated by skeletal lesions, may have in fact been the healthier people who did not die in the acute stages of a disease (Wood et al. 1992, Groves et al. 2013:471).

Another modern day risk factor for TB, (discussed in Chapter 2), is poverty. It is difficult to infer much about the wealth of the skeletal sample. However the nature of their burials was examined in detail in Chapter 6, and it was observed that none of the burials were obviously "rich" or "poor". That is, the burial styles and grave goods present did not mark them out as being of a different wealth bracket or social status when compared to other people buried alongside them in the same cemetery. Except for a few notable exceptions, the carbon and nitrogen isotope analysis did not reveal anyone as having eaten a diet that showed them to be different from, or perhaps poorer than, other individuals buried in the same cemetery. Unfortunately, therefore, this skeletal sample cannot be concluded to have shown any indications of social status that was obviously different from their contemporaries. This was in some part expected due to them having no obvious differences in burial goods and styles when compared to the rest of the burials in their respective cemeteries. However, this does not mean the individuals were not affected by poverty as they perhaps lived in a poor community and hence everyone was buried in the same manner. It does mean that there are no obvious reasons attributable to social status that would demonstrate why the sampled individuals had TB and their contemporaries, buried in a similar fashion and having eaten a similar diet, did not have the disease.

Underlying weakness of the immune system, such as is caused by infection with HIV is another modern day risk factor for TB (see Chapter 2). While the sampled individuals could have indeed suffered from other infectious diseases in addition to TB, it can be seen from the descriptions of the skeletons in Chapter 6 that very little pathology other than that pertaining to their possible infection with TB was visible on their bones, although a number were reported as having *cribra orbitalia*, which is an indicator of poor general health of several proposed causes, including the presence of an infectious disease (White and Folkens 2005:321). Of course, the general lack of skeletal pathology does not necessarily mean that they did not suffer from other infections, just that there is no skeletal evidence for them having done so. So unfortunately, no definite link can be made between the likelihood of infection with another microbial disease and infection with TB for these individuals.

To summarise, seven of 21 (33%) individuals studied from a total of seven sites were definitely not local to where they were buried, and 14 of 21 (67%) could possibly be of local origin to their burial cemetery. To put this into perspective with published data from some of the sites (see Figure 8.11), at Driffield Terrace, three out of six individuals (50%) were not local (Montgomery et al. 2011), 11 out of 18 (61%) were from York or an area with similar geology and seven (39%) were not local (Müldner et al. 2011). At Lankhills in Winchester, 14 out of 25 (56%) were not local (Evans et al. 2006), and 11 out of 40 (28%) were not local (Eckardt et al. 2009). Meanwhile, in Gloucester, seven out of 21 (33%) were thought to be not local (Chenery et al. 2010). Thus 42 of 110 (38%) individuals in these studies were identified as non-local. From these comparisons, it is impossible to conclude that people with TB were more mobile than those without, or that TB was contracted mainly as a result of mobility.

However, the "possible locals" from the seven sites studies could still originate from other locations and some of these suggested places have been listed with the individual sites in the discussion. Isotope analysis is not sensitive enough to be able to conclude if these people are definitely local or if they could be immigrants, because other geographic areas having similar geology and rainfall patterns could lead to similar strontium and oxygen ranges for the individuals concerned. In future, when further work has been done in isotope research, it may be possible to pinpoint their origins more accurately as it will provide more detailed comparative geographical data than is currently available. However, despite this current limitation, for the purposes of this study, the data available were enough to establish if the sampled individuals are likely to be of local origin or were more likely to have migrated from elsewhere, as the original research hypothesis suggested.

In order to extend this study further, it was decided to test if there was a significant difference in people who were local and who had TB and people who were non-local with the disease compared with non-locals and locals without the disease. Data from the current thesis was compared with the published data and chi

squared was calculated (see Appendices). The value calculated was 3.14, which is lower than the critical value at the 95% confidence level, of 3.84. Consequently the conclusion was drawn that there is no significant difference in the mobility of people with TB compared to those without the disease. However, are there other factors apart from TB which could influence mobility of these individuals, such as age or sex?

It was decided to do two further calculations based upon these findings, namely to find out if there is a difference in the mobility of the sexes and also of the age groups. For the sexes, the data in this thesis and published data were used to calculate a chi squared of males and females who were local or non local. The chi squared had a calculated value of 4.37. This was higher than the critical value at the 95% confidence level of 3.84. Hence there is a significant difference in mobility between the sexes, with males being the most likely people to be non-local to where they were finally buried (see Appendices for full results).

Finally, thesis sample data and published data for all individuals were divided into four age groups; juvenile (under 18 years of age), young adult (19- 25 years), mid adult (26 – 45 years) and old adult (46+ years). A chi squared value of 10.78 was above the critical value of 7.8 at the 95% confidence level. The conclusion here is that there is a significant difference in the mobility of the different age groups. This is not unexpected because there is a higher number of local juveniles than the expected figures would suggest. However, these young individuals have had less time to move since their tooth enamel formed, compared to people a little older, who have had a longer time span in which to become mobile.

To sum up these findings, the presence or absence of TB had no significant effect upon mobility, hence the hypothesis that people with TB in Roman Britain were not local to the place in which they were finally buried has not been proven. However, being male or female and age category all have a significant effect on whether an individual was local to where they were buried. As has been already discussed above, it is not possible to link the transmission of an infectious disease such as mobility with TB. The chi squared calculations show that the mobility of individuals was not possible to prove as being linked to them having TB; however the factors of age and sex were seen as significant characteristics for someone to be mobile or not, with age group appearing to have the biggest effect on mobility.

### **Chapter 9: Conclusion**

This chapter summarises the overall findings from the data generated from this study and their support, or otherwise, of the research hypothesis. It then goes on to discuss limitations of the study and makes suggestions for further research to support the findings of this small-scale pilot project.

#### 9.1 Summary of the data

The hypothesis that people with TB who were buried in Roman Britain were not born and raised locally to the place in which they were interred has been tested (See Tables 8.11 and 8.12). It was discovered that the hypothesis could be supported for two of the Winchester, Chester Road individuals, the Easington individual, two of the Poundbury individuals and one from Driffield Terrace, York, which means seven of the 21 individuals analysed appear to have been non-local to their place of burial. The hypothesis was not necessarily supported for 14 of 21 people, who could have experienced childhoods local to their burial place. These were all of the Baldock, Gambier Parry Lodge, Winchester Victoria Road and Cirencester individuals, four of the Poundbury individuals and one from Driffield Terrace. However, as discussed in Chapter 8, other places of origin are also possible for these people. In summary, the hypothesis was supported for some of the individuals, and could be supported for more; many people with possible TB who were buried in Roman Britain were not originally raised in the area in which they were buried. By use of chi squared tests, it was discovered that there is no significant difference in the number of cases of TB in migrants and non-migrants, so the hypothesis of this research that migrants were more likely to have had TB than non-migrants could not be proven. However, it was discovered that age group and sex of individuals had a significant effect on their mobility, with males and older individuals being the most likely groups to have originated in a location other than that of where they were finally buried.

### 9.2 Limitations of the study

This research project was a unique pilot study looking at the mobility of people who died after contracting TB in an attempt to link mobility to transmission of the disease. It was hoped to have TB strain data from a sister project in order to assist in the pinpointing of origins and mobility patterns of these people, but, as was discussed in section 8.2.6, it was disappointingly not possible to determine the bacterial strains in order to synthesise this with the isotope data. In addition, the techniques of isotope analysis are expensive and therefore only a small sample of individuals were analysed for only four isotopes. This small sample size means statistical analysis was not viable for the study, and, as discussed in Chapter 6, (section 6.6), choosing a statistical test to help with the interpretation of isotope data is difficult, and impossible with a small sample (Lightfoot and O'Connell 2016). Standard deviation and Z scores were therefore used as tools to assist with visual analysis of the graphed results. These statistical methods are not without their disadvantages (see Chapter 8 Discussion), but it was felt it was better to include some statistics to assist with interpretation than not to use any at all.

As discussed in Chapter 8, the teeth and bones analysed in this study were made available from a previous research project. The osteological diagnoses of TB and age and sex estimations were therefore made by a number of different bioarchaeologists, some of whom will have had more experience in this than others. In an ideal situation, one person should have examined all of the skeletons and performed sex and age estimation and diagnosis of TB related bone lesions. Checking those data could also have been done, but the resources to do this were unavailable. Another limitation of having been provided with previously selected bone and tooth samples is that the enamel of some of the teeth which were analysed was being formed during breastfeeding, which meant that the oxygen isotope data generated from these teeth needed to be interpreted with care, because of the breastfeeding enrichment of  $\delta^{18}$ O. In addition, the use of different bones meant that there would be some difference in how many years of dietary history could be produced from the collagen analysed, due to different bone

element renewal and replacement rates. However, this would be variable even if all of the same bone element for each individual had been used for carbon and nitrogen isotope analysis; as previously discussed in Chapter 8, bones remodel at different rates for different reasons and the presence of TB in the bones could also affect this remodelling rate. The reasons for different remodelling rates could depend not only on the bone element sampled but upon the overall age, health and physical activities of the individual.

With regards to the TB infection itself, it is not possible either by means of osteological examination of the bones, or DNA analysis, to establish when in the life of the individual that they contracted or developed the disease. Hence it is not known for how long before their death TB impacted on their life and their behaviour. It is possible that they had a subclinical infection at the time of their death, with their own immune systems controlling the disease, although the presence of woven new bone on the ribs would indicate that the disease causing the lesions was still likely to have been active at the time of death. Hence it is rather difficult to directly link active TB disease with mobility. People may have moved long before they got the infection, so possibly in this case, mobility had no direct effect on the presence of the disease. In addition, migrants could have brought the disease with them and infected other people with whom they had contact but who were themselves not mobile, but who contracted TB as a direct result of mobility. This will not be possible to resolve as there is no means of knowing when in their lives people contracted the disease or became ill with it.

While it is possible to learn much about mobility in the past through various forms of evidence, what we cannot learn from isotope analysis are the specific reasons people moved (Prowse 2016:205). Migration could have led to these individuals contracting TB due to the depression of their immune systems caused by stress, poor diet and coming into contact with new infections during the journey, or could have been the result of them having the disease and perhaps being stigmatised and forced to move away from their home areas; of course, they may have had subclinical/latent TB and not have been ill, and they could have moved as a result

of the Roman invasion and settlement of Britain. However, they could also have brought TB from another place to the region where they died. Nevertheless, humans do not necessarily only move once in their lifetimes, and these people may have lived in multiple locations prior to their death. Isotope studies are limited in their ability to detect this "mobility history", or to account for short-term residency. In this study, strontium and oxygen isotopes were used to examine the place of residence at birth and during childhood. The carbon and nitrogen isotopes provided information about diet during a period of approximately five to twenty years prior to death depending on age of the person at death and bone element analysed. Hence the methods used assume a one-time migration event took place, thus do not detect short-term residency or repeated movement.

### 9.3 Suggestions for future research directions

This study has shown there is potential in using isotope analysis data for exploring mobility and diet, and evidence of TB in the skeletons analysed, to explore how the disease, and potentially other infectious diseases too, impacted upon communities in the past. It has supported findings from previous studies (see Chapter 3) and further demonstrated that people of all ages and both sexes were mobile during the Roman period and that this mobility meant there was potential for the transmission of TB, and indeed other infections, both during and after the journeys they made. This mode of transmission, linked with travel, is seen with clinical infections today.

There follows some suggestions of where to take this important research to in future;

Firstly, in order to be able to pinpoint geographical locations from which the individuals studied may have originated with more accuracy, it is necessary for further research to establish biogenic strontium isotopic signatures from local plants and animals to refine the strontium ranges for specific geographic areas. This has been done for only some of the sites studied in the current project, but

not yet for all of them. These new data would enable more robust conclusions to be drawn about the original locality of the individuals in the project. It is also necessary for further research into establishing oxygen values for local drinking water, and to take into consideration the range of potential sources of drinking water for each of the sites, (eg. water from other areas being carried in via aqueducts) when considering oxygen isotope results.

Secondly, to establish if there were multiple migrations in the life of an individual, further research could analyse multiple tooth and bone samples from each skeleton. Teeth and bones would be chosen to represent different times of formation during the life of the person. Isotopic analysis of incremental dentine samples from the teeth that are already available for analysis in the study would also provide the ability to indirectly infer mobility information for a widened range of years of the life of the individuals (Beaumont et al. 2013:2), and would not result in further destruction of the skeletons.

Thirdly, it would be useful to perform lead isotope analysis on all of the individuals in the study in order to attempt to constrain their places of origin further (Montgomery et al. 2010:213). This in itself would be interesting enough but, if tied in with strain data for Mycobacterium tuberculosis, information about the origins of people with different strains of TB could be strengthened. Unfortunately, this current research was started (in 2013) before the Durham/Manchester aDNA project was completed (in 2016). It was expected that osteological diagnosis of TB in the current project skeletons would be confirmed by aDNA analysis and that strain data for the bacteria could then give some indications of where the disease may have originated which could have been linked with the isotope data studies of mobility from the current project. However, this did not prove possible (Müller et al. 2014a, 2014b, 2014c and 2016). As the Durham/Manchester project progressed, time became a major constraint, and many of the samples could not be analysed within the time frame. As aDNA analysis techniques develop, future aDNA and TB strain data research may address the issues encountered in the Durham/Manchester projects. This may then improve nuancing of information

about the spread of TB strains with the movement of people today, and may be of use to current medicine when predicting the likelihood of spread of resistant strains of *M. tuberculosis* around the world today. However, this was not possible for the current research.

Fourthly, further research is necessary to extend the current project to investigate the link between TB and mobility. The current project was a pilot study and used a small sample of 21 individuals mostly buried in cemeteries on chalk or limestone regions and with TB/TB related bone lesions. However, there are a further 23 individuals from the Durham/Manchester project, buried in other geology in Roman Britain who also had evidence of possible TB, and from whom teeth and bone samples are available for further analysis. These people were not included in the original project because they were not buried on chalk or limestone (chosen in this pilot research because of available baseline strontium isotope values for chalk areas), and due to funding constraints which limited the number of individuals it was possible to analyse. However, limiting the geological background in this way, whilst providing a convenient sample size, did not confer any methodological advantages. In hindsight, it would have been preferable to have chosen individuals from sites which had more available biosphere strontium data due to biosphere values of strontium being affected by more than just the underlying geology of the region. More detailed biosphere values for strontium would have enabled better comparison with skeletal isotope values and therefore more accurate ability to decide if the individuals were local to their burial place or not.

However, following on from the current work, analysing these individuals in future would strengthen the conclusion made in this study that some people who had TB who were buried in Roman Britain were not local to the place in which they were eventually buried. Other archaeological periods were covered in the Durham/Manchester aDNA project too (1<sup>st</sup> to 19<sup>th</sup> centuries AD. Müller et al. 2014a:178), and therefore it would be interesting to conduct isotope analysis on the bones and teeth of the remaining individuals from the original sample of 77 from different time periods, who were buried both in other parts of Britain and also

in continental Europe (Müller et al. 2014a:178) in order to establish if people infected with TB in time periods other than the Roman era were born and raised in areas different to where they were eventually buried.

Finally, in choosing individuals who had possible TB but who were buried on chalk, no considerations were made of the availability of site reports from the excavations of the cemeteries, or for the availability of comparative carbon, nitrogen, strontium and oxygen isotope data from nearby burials. With hindsight, based upon difficulties of accessing unpublished site and skeletal reports, and upon the lack of detailed comparative isotope data for some sites selected (eg. Baldock and Easington), it would have been advisable to have chosen for this research individuals with bone changes and/or positive aDNA data for TB, who were buried on any geology from sites with detailed, published, accessible reports and with other isotopic data with which to compare that of the TB sufferers. This would have made it easier to highlight any differences in burial and isotope data, and would have strengthened conclusions about possible differences in people with the disease compared with those in the same community who remained uninfected.

# Appendices

# Appendix 1

Lab	Collagen	%C	%N	C/N	δ <sup>13</sup> C	δ <sup>15</sup> N	Mean	Mean
number	yield (%)			RATIO	PDB	AIR	δ <sup>13</sup> C	δ <sup>15</sup> N
BALD7230	3.69	43.6	15.0	3.4	-19.1	11.5		
BALD7230		43.6	15.1	3.4	-19.2	11.7	-19.1	11.6
BALD7498	6.44	45.0	14.6	3.6	-20.1	10.1		
BALD7498		43.9	15.3	3.4	-19.4	10.2	-19.8	10.1
EASN183	1.14	13.9	4.3	3.8	-20.6	11.2		
EASN183		16.0	5.1	3.7	-20.6	11.7	-20.6	11.4
GRPL531	19.15	44.3	15.6	3.3	-19.1	11.5		
GRPL531		44.8	15.8	3.3	-19.2	11.5	-19.1	11.7
GRPL538	4.38	43.1	14.9	3.4	-20.2	11.5		
GRPL538		44.4	15.4	3.4	-20.4	11.7	-20.3	11.6
VICT129	6.64	43.7	15.2	3.4	-20.0	9.3		
VICT129		44.2	15.4	3.4	-20.0	9.4	-20.0	9.3
VICT96	7.85	44.3	15.2	3.4	-19.9	9.6		
VICT96		45.3	15.7	3.4	-19.8	9.7	-19.8	9.7
CHES512	4.00	44.0	14.4	3.6	-20.3	12.9		
CHES512		43.6	14.5	3.5	-20.3	13.0	-20.3	12.9
CHES535	7.80	44.7	14.9	3.5	-20.4	10.9		
CHES535		45.7	14.8	3.6	-20.8	11.0	-20.6	11.0
CHES636	7.71	44.3	15.6	3.3	-20.4	11.2		
CHES636		42.9	14.9	3.3	-20.4	11.1	-20.4	11.2
CIRE189	9.60	43.6	15.2	3.4	-19.7	10.4		
CIRE189		43.5	15.2	3.4	-19.7	10.4	-19.7	10.4
CIRE37	5.06	44.2	15.0	3.5	-20.5	10.8		
CIRE37		44.2	15.0	3.4	-20.3	10.9	-20.4	10.9
CIRES	6.20	43.7	14.9	3.4	-19.8	9.2		
CIRES		44.4	15.4	3.4	-19.9	9.3	-19.9	9.3
POUN1201	6.68	43.3	15.3	3.3	-19.9	9.4		
POUN1201		43.9	15.5	3.3	-20.1	9.6	-20.0	9.5
POUN228	8.32	44.5	15.6	3.3	-19.4	9.4		
POUN228		44.5	15.7	3.3	-19.4	9.5	-19.4	9.5
POUN257	7.78	43.5	15.2	3.3	-20.0	9.3		
POUN257		43.8	15.5	3.3	-20.0	9.4	-20.0	9.3
POUN506	9.11	44.5	15.5	3.4	-19.4	9.6		
POUN506		44.4	15.4	3.4	-19.4	9.6	-19.4	9.6
POUN619	6.86	37.9	13.3	3.3	-19.3	9.8		
POUN619		42.9	14.9	3.3	-19.4	9.7	-19.4	9.8
POUN636	9.66	44.4	15.2	3.4	-20.3	8.9		
POUN636		44.5	15.2	3.4	-20.4	8.9	-20.3	8.9
DRIF13	7.12	41.0	13.8	3.5	-19.6	11.6		
DRIF13		49.0	16.2	3.5	-19.7	11.4	-19.6	11.5
DRIF19	0.65	-	-	-	-	-	-	-
DRIF19	0.44	-	-	-	-	-	-	-

Table of carbon and nitrogen isotope data NB DRIF19 and EASN183 omitted

Lab	ppm	<sup>87</sup> Sr/ <sup>86</sup> Sr	δ <sup>18</sup> <b>O</b> <sub>p</sub>	$\delta^{18}O_{dw}$	δ <sup>13</sup> C
number			(VSMOW)	(VSMOW)	VPDB
BALD7230	59.33	0.70932	17.34	-7.0	-13.32
BALD7498	52.44	0.70904	17.26	-7.1	-15.08
EASN183	70.19	0.71031	17.45	-6.8	-14.31
GRPL531	92.12	0.71008	16.98	-7.6	-13.34
GRPL538	80.89	0.70924	16.99	-7.6	-13.68
VICT129	73.50	0.70943	17.98	-6.0	-13.53
VICT96	115.50	0.70937	17.80	-6.3	-15.44
CHES512	34.44	0.71249	18.18	-5.7	-14.40
CHES535	67.82	0.71252	18.01	-6.0	-14.54
CHES636	46.42	0.70948	19.36	-3.9	-14.14
CIRE189	95.70	0.70884	18.52	-5.2	-15.40
CIRE37	74.19	0.71007	18.50	-5.2	-15.65
CIRES	52.19	0.70992	17.66	-6.5	-13.76
POUN1201	58.12	0.70903	18.37	-5.4	-14.43
POUN228	59.04	0.70854	18.48	-5.3	-15.36
POUN257	65.45	0.70886	19.09	-4.3	-14.46
POUN506	92.19	0.70835	19.35	-3.9	-14.19
POUN619	100.90	0.70870	21.14	-1.2	-13.41
POUN636	100.70	0.70875	18.39	-5.4	-14.44
DRIF13	68.59	0.71327	15.97	-9.1	-9.49
DRIF19	82.01	0.70937	17.33	-7.0	-13.78

Table of strontium and oxygen isotope data.  $\delta^{18}O_{dw}$  calculated using Daux et al. (2008) Equation 6

Chi squared data for locals and non-locals infected with TB compared to uninfected individuals.

Class	Observed results	Expected results
Local with TB	14	10.4
Local without TB	35	39.4
Non-local with TB	7	10.6
Non-local without TB	43	38.6
Total individuals	99	99

Chi squared value = 3.14 Critical value at 95% confidence level for 1 degree of freedom = 3.84

Chi squared data for male and female locals and non-locals.

Class	Observed results	Expected results
Local male	18	22.4
Local female	17	12.6
Non-local male	32	27.6
Non-local female	11	15.4
Total individuals	78	78

Chi squared value = 4.37

Critical value at 95% confidence level for 1 degree of freedom = 3.84

### Appendix 5

Percentages of males and females who were local and non-local by site;

## a) DRIFFIELD TERRACE

(NB all individuals were males)

Locals (%)	Non-locals (%)
35	65

### b) WINCHESTER

Sex	Locals (%)	Non-locals (%)
Both sexes	49	51
Female	74	35
Male	26	65

### c) GLOUCESTER

Sex	Locals (%)	Non locals (%)
Both sexes	59	41
Female	54	46
Male	56	44

Chi squared data for locals and non-locals dependent on age category

Class	Observed results	Expected results
Juvenile local	14	8.8
Juvenile non-local	3	8.2
YA local	8	10.9
YA non-local	13	10.1
MA local	28	27.0
MA non-local	24	25.0
OA local	5	8.3
OA non-local	11	7.7
Total individuals	106	106

Chi squared value = 10.78 Critical value at 95% confidence level for 3 degrees of freedom = 7.8

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