THE APPLICATION OF POST-MORTEM COMPUTED TOMOGRAPHY (PMCT) FOR THE ANTHROPOLOGICAL EXAMINATION OF JUVENILE REMAINS

Comparison of PMCT with traditional anthropology, guidelines for data acquisition, and the development of a standard PMCT anthropological reporting form.

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ABSTRACT

The application of post-mortem computed tomography (PMCT) for the anthropological examination of juvenile remains

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Post-mortem computed tomography (PMCT) is a non-invasive medical imaging technology that could be a valuable adjunct to traditional techniques in forensic practice. However, despite numerous theoretical advantages, integration of PMCT into forensic pathology, anthropology and odontology is currently restricted by the lack of scientific evidence.

This thesis reviews the literature regarding the anthropological investigation of juvenile remains. The experimental chapters use PMCT images of the Scheuer Juvenile Skeletal collection, a unique collection of remains, that span the full age range of the developing human held in Dundee, and cases from the PMCT image archive at the East Midlands Forensic Pathology Unit. Images were acquired using multi-detector CT scanners and analysed using OsiriX three-dimensional imaging software. This thesis considers 1) if anthropological measurements are reproducible using PMCT, 2) if PMCT-derived measurements are accurate, compared with dry bone and orthopantomogram (OPT) examinations 3) what images and data are required to conduct a full anthropological examination to determine an individual's biological profile using PMCT and finally 4) how to format and display these images appropriately to facilitate data sharing, international interpretation and future development of this method. These techniques were also used in the anthropological investigation of Richard III.

Using age as the principle parameter, and assessment of both long bones and dentition, I have shown that 1) measurements used in the most frequently applied forensic anthropology techniques can be extracted from PMCT data, 2) PMCT measurements are accurate, and repeatable by multiple practitioners of various professional backgrounds and experience and 3) the information required to conduct a comprehensive anthropological examination can be condensed into a concise two-page 'minimum data-set' form.

The results of this thesis provide new evidence to support the implementation of PMCT for anthropological examination in events requiring forensic investigation and disaster victim identification.

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LIST OF CONTENTS

INDEX OF FIGURES

CHAPTER 1

CHAPTER 3

CHAPTER 4

CHAPTER 5

CHAPTER 6

CHAPTER 7

CHAPTER 8

INDEX OF TABLES

CHAPTER 3

CHAPTER 4

CHAPTER 5

CHAPTER 6

CHAPTER 7

LIST OF PRESENTATIONS

ORAL PRESENTATIONS

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CHAPTER 1: INTRODUCTION

"The skeletons in my laboratory...have tales to tell us, even though they are dead. It is up to me, the forensic anthropologist to catch their mute cries and whispers, and to interpret them for the living."

- William. R. Maples (1937-1997)

From 'dead men do tell tales' page 280.

1.1 Forensic Anthropology

For many years law enforcement agencies have relied on a range of scientific professions to aid criminal investigations (Isçan, 1988). These experts are often required to testify in court (Isçan, 1981a). They are therefore referred to as 'forensic' sciences, from the Latin 'forensis' meaning 'of or before the forum' (Black, 2003).

Anthropology, from the Greek word 'anthropos' meaning 'human', is the science that deals with the origins, physical and cultural development, biological characteristics and social customs and beliefs of humans (Isçan, 1981b). When a human body or human remains are at the centre of the investigation, anthropologists (or anatomists) are used to provide in-depth knowledge of skeletal biology. Sub-disciplines such as biological anthropology (the study of the biological evolution and development of humans) and physical anthropology (skeletal variation and osteology) can provide valuable information to a forensic case.

Forensic Anthropology has developed as a multidisciplinary field of forensic science that has become more prominent in contemporary society, in both the United Kingdom (UK) and the United States Of America (USA). Although the roots of the discipline lie primarily in the USA, its development in other continents has progressed to compliment the specific requirements of different cultures.

As the complexity of the forensic investigation gradually increased, it ultimately led to the development of forensic anthropology, a profession specializing in the techniques of human identification (Isçan, 1988). The earliest developments occurred in the mid-19th century, when American anatomists and physicians became involved in medico-legal casework (Ubelaker, 2006), and as a result many of the techniques employed by forensic anthropologists require a familiarity of human anatomy (Pickering and Bachman, 1997). Two cases that were particularly important to the development of the discipline are the Parkman and Luetgert murders of the late 19th century (Snow, 1982; Christen and Christen, 2003; Byres, 2008).

The Parkman case (1849) was the first documented investigation in the USA that required a professional body to act in the manner of a forensic anthropologist. In 1849, Jeffries Wyman, a professor of anatomy at Harvard University was asked to investigate the death of his colleague, Dr George Parkman. In order to convict the defendant of murder, the state was required to establish that the 'victim' was actually killed, and in order to do this the remains had to be positively identified. Wyman testified that several fragments of bone recovered from Professor Webster's laboratory furnace were consistent with those missing from an incomplete, fleshed, dismembered body found in Webster's privet. By reassembling the body parts he was able to estimate the stature, sex and age of the body, all of which matched Parkman's biological profile. This case still serves as a model for the identification of unknown remains, as the investigatory protocol carried out by Wyman is still in place in forensic anthropological practice today, that is: 1) establish if the remains are human/non-human; 2) calculate the minimum number of possible individuals (MNI); and 3) and create a biological profile by estimating age, sex, stature and ethnicity. When this protocol is used in combination with other evidence (in this case witness testimonies and dental evidence) it is an extremely reliable tool to identify an individual beyond reasonable doubt.

The Luetgert case (1896) is important for two reasons; first, it was the earliest case documented that had an anthropologically trained professional (Dr Dorsey) acting as the expert witness and secondly, in comparison to the Parkman case previously discussed, no soft tissue structures or complete bones were recovered to act as age, sex or stature indicators - since the evidence comprised only of four extremely small fragments of bone. These bone fragments were found in a large cooking vat, in a sausage factory owned by Adolph Luetgert. The bones were suspected of belonging to his wife Louisa Luetgert who had been reported missing. Using his anthropological knowledge of skeletal variation and osteology, Dorsey was able to deduce that the bones were human and that they belonged to a woman. His testimony coupled with other evidence helped to convict Adolph Luetgert of his wife's murder.

The consolidation of knowledge, gained from the previous years, was encapsulated in the work of Krogman, with the publication of *Guide to the*

Identification of Human Skeletal Material (Krogman, 1939). Krogman's work brought the skills of osteologically trained physical anthropologists to the attention of law enforcement officials. However, it was ultimately the outbreak of warfare that eventually resulted in the establishment of forensic anthropology as a professional body in its own right. World War 2 and the Korean War marked the first time that trained physical anthropologists and anatomists contributed to the recovery, identification and repatriation of the remains of US soldiers. Anthropological techniques were employed to identify the skeletal remains of the young victims, resulting in an extensive quantity of data. McKern and Stewart (1957) and Trotter and Gleser (1952) subsequently used this osteological evidence to produce a number of tested anthropological methods, which could be used to establish age from skeletal markers, and stature from bone length, respectively.

From these beginnings forensic anthropology has slowly evolved. In 1972 Ellis Kerley established the physical anthropology section of the American Association for Forensic Scientists (AAFS) and for the first time forensic anthropologists could congregate to discuss research and casework for the further development of the discipline (Ubelaker, 2006). Shortly after, the American Board of Forensic Anthropology (ABFA) was founded to regulate the standards of practitioners (Isçan, 1981a). The ABFA controls the quality of practicing anthropologists and encourages research in the field. There are also certain requirements the individual must meet, including a doctoral degree, an examination and professional experience (Murad, 2008).

The profession has developed relatively slowly in the UK, probably due to a reasonably low violent crime rate providing limited opportunities for practitioners to gain practical experience (Brickley and Ferllini, 2007).

The earliest input from the UK came from Karl Pearson in the late19th Century, a mathematician (Pearson and Whitely, 1899). Pearson used data collected by French scientist Étienne Rollet, which compared long-bone length to cadaver length in a sample of 50 females and 50 males (Ubelaker, 2006) and presented these figures in a form of regression equation. Interestingly most of Pearson's work focused primarily on evolutionary issues, however his data inadvertently offered a method for stature estimation and greatly influenced the development of forensic anthropology (Ubelaker, 2006).

Involvement in overseas work has, however, recently escalated its development and it is now also a well-established profession in the UK (Black, 2003). The UK Council for the Registration of Forensic Practitioners (CRFP) was set up in 1999. It was hoped that accreditation from the CRFP would eventually become mandatory to provide evidence in a court of law (CRFP, 2008). The CRFP demanded that practitioners had the appropriate qualifications and practical experience before certification (Brickley and Ferllini, 2007). The aim was to restore public confidence in the profession and avoid miscarriages of justice (CRFP, 2008). However, in 2009, the CRPF was subsequently closed down, as it was deemed surplus to requirements.

There is a lack of non-academic forensic anthropology posts in the UK and employment in this field tends to be in the academic environment, where both teaching and research are carried out (Brickley and Ferllini, 2007). Registered anthropologists in these posts do, however, have professional responsibilities outside of academia and those eligible for deployment tend to be part of the Centre for International Forensic Assistance and do valuable work for Disaster Victim Identification (DVI). However, since the closure of the CRFP in 2010 and the Forensic Science Society (FFS) in 2011, independent forensic science providers have also played a significant role in the landscape of forensic anthropology in the UK. These providers offer services across all of the forensic science disciplines commonly encountered in criminal or civil investigations and in the evaluation of evidence (PFS, 2014). As a result, forensic anthropologists and other forensic experts are now much more frequently deployed from these private forensic science providers, in addition to academic departments. The Association of Forensic Science Providers (AFSP) is an independent, representative body, which facilitates the delivery of these forensic science providers in the UK (AFSP, 2014). The association was formed in 2010 and acts as a network for independent providers (i.e. non-police) of forensic science services within the United Kingdom and Ireland, to ensure quality and best practice standards are maintained, in the wake of the previous regulatory bodies.

Throughout history, conflict has been a catalyst for the development of forensic anthropology. These situations generally create large populations of deceased individuals, and present an opportunity for experts to enhance their skills and develop investigative techniques. International work has been undertaken in the Balkan region (Thompson, 2003a) and events such as the Asian Tsunami in 2004, and the London Bombings in 2005, called for the expertise of individuals in human identification, predominately, but not exclusively, from the soft tissue level. Soft tissue pathology provides useful information in identification, as well as being “fundamental in determining cause and manner of death” (Milroy, 2007). Progressively, police forces in the UK are also recognising the expertise of these professionals, in the sense that the forensic anthropologist can be given bodies, at any tissue level, to be analysed for identification. This includes such cases as the “part of human leg found on beach” (BBC, 2007) and “suitcase containing body parts in Arbroath Harbour” (BBC, 2008), where Tayside Police called in forensic experts at the University of Dundee to examine the respective bodies. This has taken the work of forensic anthropologists from the analysis of primarily human skeletal remains to remains with soft tissue at any stage of decomposition.

Facial reconstruction is another branch of forensic anthropology, utilising individuals from artistic or scientific backgrounds with extensive knowledge of the musculature and soft tissue anatomy of the face to produce reconstructions of the individual, which is becoming increasingly well established in routine UK forensic casework.

At present, forensic anthropologists no longer work on the periphery of a crime scene (Isçan, 1988). Their extensive training and experience in human identification provides a valuable contribution to a forensic investigation. The forensic anthropologist has two main functions (Isçan, 1981b):

1. Assign the victim a biological identity by classifying the individual as a member of a certain population with a specific sex, age and stature.

2. Provide a personal identification, using a detailed knowledge of skeletal biology and an understanding of associated anomalies and pathologies.

This data builds a detailed biography of a victim which, when related to living morphology, can lead to a positive identification. Additionally, forensic anthropologists are also becoming involved in establishing the age of living individuals. The age of an individual can affect what legal representation or treatment they are entitled to and therefore, establishing an individual's age is often mandatory for legal reasons (Isçan, 1988).

As public awareness of forensic anthropology has increased, so has the number of board-certified diplomats (currently about seventy-seven), and the number of institutions offering advanced degrees in forensic anthropology or physical anthropology with a forensic emphasis (ABOFA, 2014). In the 1970s most practitioners of forensic anthropology held academic positions and offered only occasional assistance to investigative agencies. Once rare, forensic anthropology service laboratories affiliated with universities are no longer unusual. Some organizations employ a number of full-time forensic anthropologists (e.g. Joint POW/MIA Accounting Command/Central Identification Laboratory [JPAC/CILHI], National Transportation Safety Board [NTSB] and private disaster management corporations), and an increasing number of large medical examiner establishments employ full-time forensic anthropologists. Consequently, the presence of forensic anthropologists providing case reports, depositions and expert testimony in civil and criminal courts and in tribunals around the world has increased considerably in the past two decades (Bernardi, 2008).

1.2 Typical Case Progression

Cases requiring the services of a forensic anthropologist arise in a variety of ways. Excluding mass fatality scenarios, the appearance of unknown human remains may involve skeletal components and scavenged fragments scattered about the landscape, clandestine burials, submerged remains or the occasional bone discovered incidentally during an excavation for an unrelated reason. Arson investigators, for in situ examination and recovery of fragile remains prior to transportation to the mortuary, also increasingly request anthropologists.

When remains come to light, law enforcement agencies may have a theory regarding their identity or manner of death. In such cases, anthropological information can be used to help confirm or refute this theory. When the remains are presented to an anthropologist with no background information a complete examination must be conducted. This includes; 1) deciding if the bones are human vs. non human, 2) determining the minimum number of individuals that could potentially be present (particularly important when dealing with mass graves) and 3) establishing the biological profile of the remains (age, sex, stature and ethnicity).

1.3 Biological Identity

Establishing the biological and personal identity of human remains is of fundamental importance for a forensic anthropologist in an investigation (Budowle *et al.*, 2005). Legally, religiously, and culturally it is recognised that all human beings have the right to retain their identity, even in death (Interpol, 2014). As a result, recovery and preservation of all human remains is of vital importance to law enforcement authorities. Preliminary determination of age-at-death, racial affiliation or ancestry, living stature and biological sex is the first step towards arriving at a positive identification. These four basic parameters are supplemented by more '*individuating*' traits that include: evidence of dental or surgical intervention, body modifications, evidence of trauma, fingerprints and DNA. These features help to narrow the wide boundaries established by the basic biological criteria to arrive at a physical identity that will, with reasonable certainty, confirm the provenance of the human remains.

Identification requires the comparison of two data sets to establish their likelihood of belonging to one and the same individual. Aristotle's law of identity states that for two objects to be identical then the predicate must be equal (not approximate) to the subject (i.e. A=A). Yet the concept of biological change requires greater flexibility in this law, so it is perhaps more appropriate to refer to it as A=A* where the asterisk introduces the possibility of accountable change. This reflects Leibniz's time-indexed law, which states "*X* is the same as *Y* if, and only if, *X* and *Y* have all the same properties and relations" (Black, 2007; pg3); thus, whatever is true of *X* is also true of *Y*, and vice-versa in order to account for identity over time. In the field of biological human identity it is essential that the value attributed to that asterisk is as small as possible. The distance between the two data sets, in terms of statistical probability, must be low if the two are to be linked with confidence and forensic credibility. It is vital that one of these data sets is verified to be that of an individual of known identity. Confirmation of identity therefore should accept biological change, and judicially we interpret that change through the realms of statistics, probability, and rationality (Black, 2007).

The evaluation of biological identity is a requirement in a number of circumstances, which can generally be grouped into three wide categories:

1. ***Criminal investigations*** resulting from unexplained natural deaths, homicides, or suicide.

2. ***Accidents and mass disaster incidents*** whether as a result of forces of nature or human intervention, either accidental or intentional.

3. ***War crimes and Genocide***.

However, it is not always possible to achieve a confirmed identity and as a result Jenson (1999) describe three possible categories of identity:

1. ***Positive or confirmed identity,*** which is possible when two sets of information are compared and enough unique data markers match to conclude that the same individual with all likelihood, created both ante-mortem and post-mortem records. Additionally, no irreconcilable differences are detected.

2. ***Possible or presumptive identity*** (believed to be), which occurs when several individual factors are considered and, although no single marker alone justifies a positive identification, when all factors are considered

together they provide sufficient evidence for a presumptive identification. These factors include biological identification parameters age, sex, stature and racial characteristics.

3. And finally, Jenson proposed a third ***'exclusion' category*** of identification, which occurs when all individuals in a closed event have been accounted for and therefore the only remaining individual, could not be anyone else.

A biological profile is a qualitative and biometric description of the remains. The accuracy with which each category of biological identity can be determined depends on the preservation and availability of skeletal elements for examination. The profile may be complete or partial, tentative or robust, depending on the developmental status (i.e. child or adult), quality and quantity of the remains recovered. Generally the more complete (less fragmented) the remains are, the more likely a positive identification will be made.

1.3.1 Estimation of sex

The majority of age and stature determination methods are sex-specific and therefore sex determination is normally the first step of the biological identification process. Natural selection has exaggerated differences in those aspects of skeletal anatomy most closely related to reproduction. There are therefore two main morphological differences between male and female skeletons: 1) female bones are typically less robust and smaller than male bones, which reflects the difference in muscle mass between the sexes, a feature which is particularly evident in the skull and 2) size and shape differences of the pelvis reflect the biomechanical adaptations of the female pelvis not only to tolerate the compressive loading of locomotion, but also provide the expansibility and protective architecture required by late gestation and the birthing process (Schultz, 1949). Hence, female pelves display flared ilia, a large pelvic outlet, a wide subpubic angle (i.e. the arch formed by the two ischial bones) and a sacrum that extends dorsally, increasing the cross-sectional area of the birth canal (Hoyme and Iscan, 1989). It is important to note however, that a significant number (approximately 5%) of individuals in most populations will be androgynous, i.e. will possess an equal number of male and female skeletal traits (Schwartz, 2007). Not infrequently, the skull might appear to be of one sex while the pelvic bones indicate the opposite conclusion. In this case, the pelvis is the more reliable predictor of sex. The main cause of this discrepancy is unknown however, it is thought that genetics, early infant activity pattern and diet could be contributing factors.

For adults, using the pelvis and skull, which are widely accepted as the most sexually dimorphic skeletal elements, a variety of single and multivariate measurements have been identified for the relatively accurate determination of sex, with reports of 90% accuracy using the pelvis alone (May and Cox, 2000).

However, the majority of sexually dimorphic differences between the male and female skeleton are not discernible until puberty. For example, one of the characteristic traits of a female pelvis is the triad of parity (Kelly, 1979; Putschar, 1976; Houghton, 1975; Houghton, 1974). The three features of this triad are pitting of the dorsal surface(s) of the pubic bones, modification (lipping) of the sacroiliac joint and deepening of the pre-auricular sulcus, formed by the stretching of ligaments transecting the pelvic inlet when a term infant passes through the pubic canal. Therefore, the consensus tends to be that skeletal remains cannot be sexed with any degree of reliability from morphological observations until pubertal modifications have taken place (Scheuer and Black, 2000). This is undoubtedly the largest single problem in the analysis of immature skeletal remains. Growth spurt occurs at different times, both in individuals of the same sex (early and later maturers; Tanner, 1962) and also between girls and boys. As a result, any estimated age category is necessarily wider than it would be had the sex been known.

The relationship between skeletal maturity and sexual maturity is complex. Sexual maturity occurs at puberty, when hormones involved in sexual reproduction act as a catalyst for physical changes that increase the dimorphism between male and female individuals. These physical changes do not necessarily coincide with skeletal changes that differentiate between sexes. Some individuals reach sexual maturity much earlier than others and as a result there might be large discrepancies between that individuals skeletal and sexual maturity. This also results in quantitative differences of size and rate of growth being of little use in sexing juvenile skeletal remains. For this reason, most publications on this topic focus on morphological differences, generally in those regions most sexually dimorphic in adults, such as the pelvis and skull (Fazekas and Kosa, 1978; Schutkowski, 1987; Schutkowski, 1993; Hunt, 1990; Mittler and Sheridan, 1992). However, in the majority of these investigations it would appear that these traits are useful only in attempting to sex juveniles under carefully controlled conditions in a specific population (Molleson *et al*., 1998). When the same traits are scored on large assemblages of unknown sex there is a much greater concordance between these traits and they are therefore much less useful for sex discrimination.

Although there are undoubtedly skeletal morphological differences between the sexes from an intra-uterine stage onwards, it appears that they do not reach a sufficiently high level for reliable determination of sex until after the pubertal modifications take place (Scheuer and Black, 2000).

1.3.2 Estimation of stature

Calculation of the height of an individual from skeletal remains is based on the relative proportions of different body parts, which correlate with the overall stature of the individual and is generally the most straightforward biological criterion to establish. Height and stature are interchangeable terms, with stature referring to the height of an individual in an upright position. One major difficulty in studies examining stature is the mismeasurement of stature in the living. For example, the height of the same individual can be almost an inch greater after a reclined rest (overnight) than those taken after 6 or more hours without lying down.

The most accurate calculations will be obtained from undamaged bones of known sex and ethnicity, as stature is both sex and race dependent (Byres, 2008). Regression equations that use the femur and tibia have the lowest standard error, but when these preferred skeletal elements are not available, estimations can be made from fragmented long bones, metacarpals, metatarsals and clavicles, but with less accuracy.

The estimation of stature in subadults is of limited utility because the rapid growth of a child, before the cessation of growth at puberty, renders stature records quickly out of date. Although parents may periodically record the growth development of their child on a chart at home, height is not as generally used as a defining biological characteristic when describing a child's biological identity.

1.3.3 Estimation of ethnic identity

Presently, there is considerable debate within the anthropological community concerning the attribution of race. Many researchers argue the nonexistence of race due to the increasing complexity of geographical migration and population cohabitation. Although there may be a tendency for natural groupings formed by persons who have similar morphological characteristics, most anthropologists prefer to use the term ethnicity or cultural affiliation, instead of race.

Throughout time, the human population has gone through a "constant process of change" (Baker, 1966) and along with it, so has the socially perceived notion of race. Many attempts have been made to define race, in the form of the 'essentialist' concept based on anatomical differences and the 'population' concept which relies on genetic differences (Long, 2004). Neither theory has been validated and both have faced so much controversy that race classification is such a problem for anthropologists that many scientists have decided to dismiss the theory altogether, in an attempt to maintain a "politically correct" position (Billinger, 2007; Brace, 1995). In contemporary society, it is believed that only 5-10% of total human variance is attributable to race (Brown and Armelagos, 2001), with a much higher proportion due to individuation. This has led scientists to believe that the differences in phenotype interpreted by society are now largely due to environmental and cultural factors, and how we perceive them, and are not due to major biological differences consistent with genetic evidence (Owens and King, 1999). This, and the inability to define race add to the transitory nature of the four primary ancestral groups (Caucasoid, Mongoloid, Negroid and Australoid). Since there is no accepted biological definition, it has become increasingly difficult to accept the concept of race. As the constantly changing ancestral groups throughout evolution make it difficult, at any point in time, to define them.

In today's population, not only is the concept of ancestry "constantly evolving," (Winker, 2004) but so is ethnicity and culture. In practice, racial classifications are based on genetic traits and morphological features to support the preconceived notions of race. These are adapted to fit an individual's phenotype to a certain population or ethnic group, and generally not one of the four main ancestral groups. This is due to individuals defining themselves as belonging to a specific population or ethnic group, for example Hispanic and not to a primitive ancestral group. These factors bring further ambiguity to the concept of race. As the ethnical categories continue to evolve over time this gives an indication of changes in society, and how individuals perceive and define themselves.

Human evolution has been characterised by locally differentiated populations "coexisting at any given time" (Templeton, 1998; pg92). This entails a high level of superficial phenotypic variation across all areas of the world and it has become increasing difficult to assign an individual to a specific region. Unrelated and independent selective and random forces are exerted upon populations all over the world, and in turn, individuals adapt to them. The US Government census also dismissed the idea of the four main ancestral groups, allowing individuals to self-designate themselves into up to 126 racial and ethnic categories. This reflects the considerable amount of human variation that exists in the US, where individuals can assign themselves to more than one social group. This constant evolution, caused by both environmental and cultural factors, has lead to 'race' and 'ancestry' entailing different meanings to different individuals.

This ongoing debate and the continual migration and co-mingling of the entire population make attribution of ancestry extremely difficult. In addition, with the exception of a couple of dental features, the majority of characteristics that help to determine ethnic identity apply exclusively to adults. Even within an adult population, the validity of racial characteristics is questionable; therefore it is unlikely that it would be possible to determine the race of juvenile remains

1.3.4 Estimation of age-at-death

Postnatally, chronological, or true calendar age is calculated from the day of birth. Although this may appear simple, as with all biological criteria it is not always accurate. Dates of birth are often recorded incorrectly or falsified for various reasons. Todd (1920) reported that in the Terry collection, the listed ages at death for adults displayed peaks around 5-year intervals, suggesting that individuals in later life perhaps tend to round to the nearest quinquennium. This was supported by Lovejoy et al (1985) who discovered gross discrepancies between ‘stated’ and ‘observed’ ages at death for cadavers in the Hamann-Todd collection (Cleveland, Ohio). Also, in the prenatal period chronological age *per se* does not technically exist, as it is not possible to establish a starting point (i.e. fertilization) with any certainty. Therefore, biological age is used to state the age-at-death of an individual. Biological age encompasses both skeletal and dental age and is an indicator of how far along the developmental continuum an individual has progressed; in contrast to chronological age, which refers to a person’s time since birth.

Methods used to estimate age can be divided into those based on the growth of the skeleton and those based on skeletal deterioration. The first category focuses on ossification and fusion events in sub-adults, from fetuses to older adolescents. While the second category deals with adults, from early maturation to death (Ubelaker, 1987).

Unlike the other three parameters of biological identity, which are more accurately determined after puberty, age estimation methods are conversely more accurate and easier to apply to sub-adults (Scheuer and Black, 2000).

Once growth is complete, the skeleton slowly begins to deteriorate by a combination of biological processes, throughout adulthood. These degenerative changes are extremely variable between individuals making adult age-at-death assignment a difficult challenge. Despite this, there are at least three features that undergo change over time in a roughly predictable timescale: the pubic symphysis, auricular surface of the ilium and sternal rib ends. These are therefore the anatomical regions most frequently investigated for the purpose of adult age determination.

In the juvenile age range there are a vast number of markers that indicate skeletal age-at-death. These can be roughly grouped into dental markers or skeletal markers and further categorized as eruption or mineralization, and appearance, fusion or size and morphology, respectively (Ubelaker, 1987). As the only biological identification feature that can be accurately determined when dealing with juvenile remains, accurate determination of age-at-death is of primary importance. These methods will be discussed in more detail in the following chapters.

1.4 Forensic Radiology

Radiology has played a part in forensic investigation since 1896, almost immediately after its advent in November 1895, when radiographs were used to investigate projectile wounds in a murder-suicide case. The first reported use of radiology for human identification was in 1921, when it was used to detect unique biological features (Elliott, 1953), and its role in mass fatality examinations was established in 1949, when it was used to assist in the identification of the victims of the disastrous fire on the Great Lakes liner “Noronic” in Toronto, Canada (Singleton, 1951). Since then, its role in human identification and mass fatality examination has developed enormously and forensic radiology is now a key component in any multi-disciplinary team dealing with casualty identification (Baggily *et al.*, 2013). A positional statement from the International Society of Forensic Radiology and Imaging (ISFRI) (Ruder *et al.*, 2013) has recently emphasized the role of radiology in mass fatality incidents (Rutty *et al.*, 2013).

Forensic radiology is the application of medical imaging to forensic investigation and is used to determine and document the cause of injury or death, to detect and locate evidence, hazardous or illegal material concealed by an individual and to identify individuals. In current practice, forensic radiology is widely used in a number of circumstances including identification, unnatural death (including trauma and homicide) and natural death.

For identification purposes, forensic radiology can be used on both living and deceased subjects to positively identify an individual for legal purposes both before and after death. The comparison of ante and post-mortem radiographs is one of the most accurate techniques to establish identity. Culbert and Law (1927) made the first recorded identification of human remains in 1927, by comparing ante-mortem and post-mortem radiographs of the frontal sinus. In the UK, clinical images are normally retained for at least 8 years (or in paediatrics, until the patient turns 25 years old) and for 3 years after death. However, with the advent of electronic image storage (Picture Archiving and Communication Systems, PACS) this is becoming longer. In general, they are fairly accessible for investigative purposes and are therefore an excellent source of data. Forensic radiology is particularly important for the investigation process and identification of victims following a mass fatality incident such as a road traffic accident, terrorist attack or natural disaster. In these situations, the remains are often skeletonised, mutilated, decomposed, fragmented or otherwise disfigured. Subsequently, identification by means of the skeleton and the highly resilient dentition assumes a greater importance, and the true benefit of medical imaging is realised.

1.4.1 Medical Imaging methods for identification of human remains

Wilhelm Conrad Röntgen's discovery of X-ray's in 1895 revolutionized medicine. Using the principle of differential attenuation, this 'new kind of ray' was able to differentiate between structures with different density or atomic number. Minimally invasive, relatively cost-effective, permanent and objective - medical imaging techniques are now habitually used to capture and store physical evidence for forensic investigations, although the original 'plain film' radiograph has developed significantly over time.

There is now a wide range of imaging modalities that are of value for the identification of human remains, including:

Fluoroscopy (real time 'live' screening)**:** provides a quick, useful anatomical summary that can be used to detect foreign artefacts such as clinical sharps or explosives. Often the modality of choice by pathologists prior to an invasive autopsy since the Association of Forensic Radiographers (2004) recommended that there should always be an initial radiography examination of the body before or concomitant with a provisional external assessment.

Plain film and dental radiography (X-ray 'photography'): this has higher spatial resolution than fluoroscopy and captures dental and skeletal details, which can be used to complement traditional anthropological, odontological or pathological examination of the remains (Martel *et al.*, 1977; Liechtenstein *et al.*, 1998; Buchner, 1985).

Computed tomography (CT) and magnetic resonance imaging (MRI): acquires information in thin slices with high 2-dimension resolution (good definition between neighbouring structures because of small pixel size). This increases the 'contrast' resolution of the image (i.e. the ability to recognize difference between different tissues). Accurate registration of these contiguous thin slices allows the creation of a full three-dimensional 'volume' dataset, avoiding the super imposition of other structures that occurs with standard radiography. Some MRI protocols actually acquire the information in a genuine three-dimensional manner.

Cross sectional imaging using computed tomography and magnetic resonance are now the gold standard in many aspects of clinical radiology. Magnetic resonance imaging (MRI) and clinical computed tomography (CT) are both non-invasive modalities that produce easy to interpret, three-dimensional images. These medical imaging techniques provide a permanent data source and the opportunity to extract vast amounts of information with minimal damage to the body. Although both techniques successfully image bone in clinical and forensic practice, MRI is a technique based on hydrogen protons in a tissue, which are most abundant in water, and therefore, although MRI gives good detail in the bone marrow, the bone cortex gives no signal. Furthermore, CT has greater spatial resolution and therefore gives better detail of bone cortex. Although it has been considered that CT can damage DNA due to a high radiation dose, these samples can be taken before the PMCT scan is conducted to ensure this does not have an adverse effect on identification. Therefore, whilst MRI has been considered for post-mortem radiology (Thali *et al*., 2003), the financial impact, inherent problems with metallic foreign bodies and the protracted time taken to scan a whole body have limited its wide scale utility in forensic and routine post-mortem investigations. CT is more practical and more time and cost efficient. This has contributed to CT becoming the 'workhorse' of modern hospital medical imaging. Forensic practice has subsequently followed this trend.

Donchin *et al*. (1994) first suggested that computed tomography might provide a viable alternative to invasive autopsies by addressing religious/ethical concerns and the increasing clinical demands on pathologists, both of which were contributing to an overall decline in post-mortems. At the request of the local Jewish community, in 1997 Manchester Royal Infirmary established a dedicated post-mortem MRI service, as an adjunct to the standard invasive post-mortem procedure. Bisset (1998), in the British Medical Journal, reported that radiologists were able to provide a confident cause of death in 87% of cases (Bisset, 1998). Although highlighting the potential application of a non-invasive image-based post-mortem examination, the results of this pilot scheme were not correlated with autopsy, which has lead to some debate over the validity of these suggestions. Nevertheless, a newspaper article in 2008 communicated the Manchester experience to the general public and the topic was subsequently addressed in the House of Commons. This resulted in the Lord Chancellor and Secretary of State for Justice at the time, Jack Straw, announcing that discussions to roll out this service to the rest of the UK were underway. As the UK government considers launching a nationwide post-mortem imaging service, questions remain as to whether there is sufficient validation of the radiological accuracy for forensic identification, pathological detection of identity and cause of death, and whether post-mortem imaging is able to satisfy the needs of the medico-legal system. These are issues which the scientific community must address before this type of project is able to reach fruition.

The provision of radiographers and equipment for the investigation of mass fatality incidents in the UK has been recognized as a weakness in emergency planning, since the Lockerbie disaster in 1988, with many plans relying on local hospitals to provide staff and equipment when requested. With standards of evidence and forensic procedure under the spotlight in a number of high-profile cases during the last 10-15 years, coupled with a much greater awareness of cultural, religious and ethical issues surrounding the examination of the deceased, the recommendations of a number of public enquiries and government reports have culminated in legislative changes including: the Human Tissue Act, a reform of the coronial system and the creation of the Council for Registration of Forensic Practitioners (CRFP). The CRFP was formed in 1999 to provide a level of accreditation to forensic professionals. However, this professional body was subsequently closed down in 2009, as it was deemed surplus to requirements.

The Association of Forensic Radiographers, in conjunction with the Society and College of Radiographers, has led the profession’s response to these changes, producing professional guidelines and developing post-graduate training programs at several UK universities. The University of Teeside for example, now offer a three year Forensic Radiology degree, and both London City University and Birmingham university offer Forensic Radiology modules in conjunction with Masters courses. Which has led to the creation of a team of trained radiographers who have successfully responded to mass fatality incidents including the Asian tsunami and London suicide bombings.

Medical imaging as a whole has seen enormous development and expansion in the last 20 years. Rapid technological advancement now allows CT scanning with multi-planar imaging and 3-D reconstructions of the skeletal and soft tissue structures. MRI scanning allows the clinician to visualize the soft tissue and especially the muscular structures and nervous system in great detail permitting the diagnosis and treatment of disease at much earlier stages.

Despite the obvious advantages offered by the newer technologies to the forensic pathologist, the forensic community in the UK has been slow to explore the potential of these new techniques for post-mortem examination. This is partly due to limited access to the equipment, which is located in hospitals and clinics and simply not available for post-mortem examination, and partly due to the dislocation of departments of forensic medicine from teaching hospitals over the past 10-15 years.

There are currently some UK research projects investigating the use of CT and MRI scanning in place of autopsy to determine the cause of death in non-suspicious deaths, and as an adjunct to autopsy in the forensic investigation of unnatural death or suspected homicide. The advantages of these techniques are clear; if the cause of a sudden but non-suspicious death can be determined without the need for autopsy, the coronial investigation can be achieved more quickly and cheaply without the need for an invasive procedure. This is both more acceptable to relatives, and less likely to be in conflict with religious or cultural belief. In the case of a forensic investigation, an invasive autopsy is likely still to be required, but an initial CT or MRI scan may reveal evidence that will be compromised or destroyed by the destructive nature of the autopsy examination.

With such developments and the huge increase in public interest in forensic investigation and rapidly changing legislative and social background to these issues within the UK, it is likely that radiography and radiographers will play a key role in forensic medical examination in the future.

1.5 Computed Tomography

Godfrey Hounsfield and Allan Cormack developed computed tomography (CT), or computed axial tomography ‘CAT’ scans, in 1971. Within months, CT scanners were developed on a commercial scale and introduced into clinical facilities worldwide. In quick succession, Wüllenweber and colleagues (1977) published the first report using CT in a medico-legal case, to investigate a cranial bullet wound. CT has undergone considerable development in this time including 4 key developments:

- The x-ray tube is capable of delivering x-rays in greater quantities for longer times without overheating.

- The x-ray tube can now continually rotate delivering high-energy x-ray photons and detecting transmitted photons whilst the patient ‘translates’ through the scanner (spiral CT, Figure 1.1).
- Multiple detectors are now used, both in the axial (x-y) plane and also more recently in the longitudinal (z) plane (multi-detector CT, MDCT). This increases speed, but also increases resolution in the longitudinal plane, allowing better multi-plane reconstructions and three-dimensional analysis.
- Increases in computing power and software developments allow faster and more complex image reconstructions.

These developments have changed scan times from many minutes to seconds.

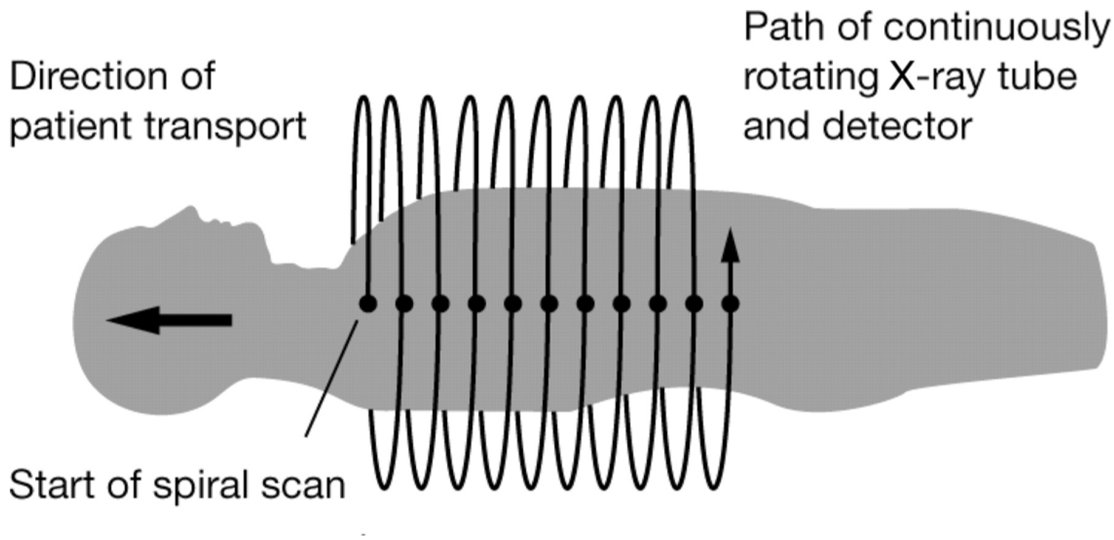


Figure 1.1: Spiral computed tomography (European Heart Journal, 2014).

Three-dimensional data acquisition is achieved through the reconstruction of contiguous two-dimensional cross-sections of the original specimen (Kalender, 2005). Each two-dimensional image is obtained by a large number of equally spaced X-ray projections that pass through the object. Photons are mainly scattered or absorbed by the constituent tissues, such that the emerging X-ray beam has a reduced intensity, a phenomenon known as attenuation (Kalender, 2005). Attenuation in CT is mainly due to electron density, equivalent to density, and therefore CT mainly distinguishes between tissues based on their density, although materials with a high atomic number (such as calcium, contrast agents and metallic foreign bodies) will be comparatively much denser due to the photoelectric effect. The linear attenuation coefficient (μ) of a tissue is displayed as Hounsfield units (HU), which are defined as the attenuation of the tissue relative to the attenuation of water. HU = 1000 x $(\mu_{tissue}-\mu_{water})/\mu_{water}$ (Seeram, 2001) (Figure 1.2).

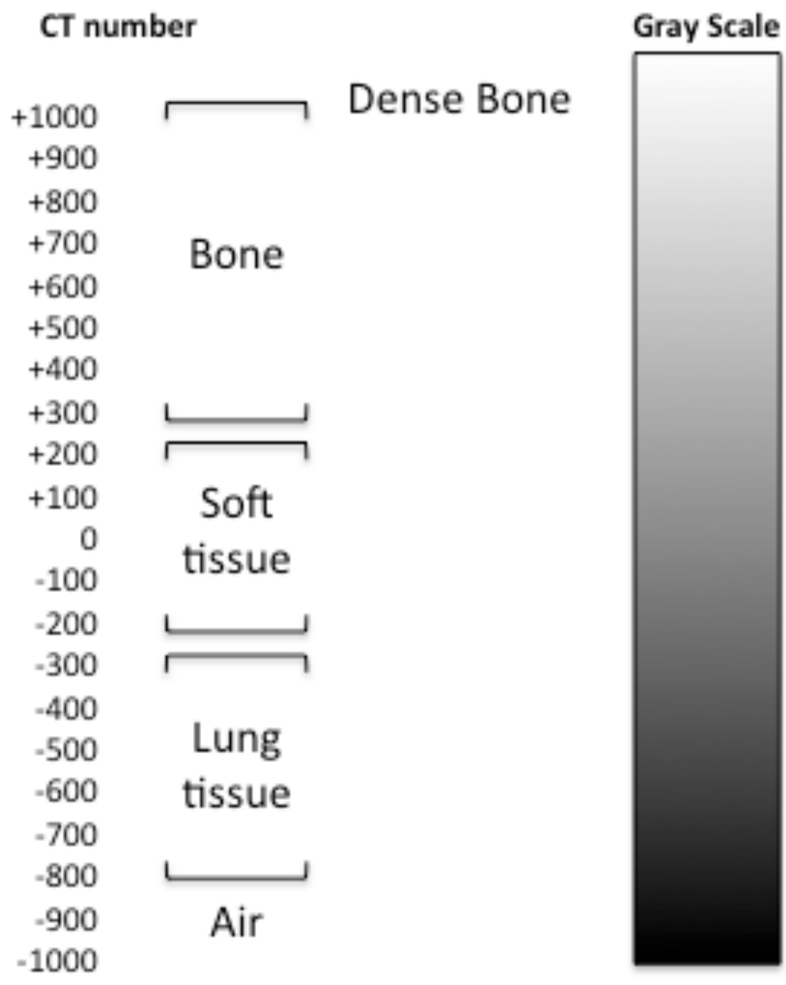


Figure 1.2: Hounsfield scale.

On this scale, water has the value 0 HU and air (with an attenuation coefficient of 0) has a value of -1000 HU. These two values are independent of the energy from X-rays and therefore constitute fixed points on the scale. Other HU values, although correlating with density, cannot be considered a direct measure of density (Kalender, 2005). CT is a non-invasive, non-destructive technique, leading to the extensive utility, of particularly MDCT, for many applications including post-mortem computed tomography (PMCT), angiography, virtual endoscopy and cardiac imaging (Seeram, 2001).

Due to the geometry of the CT scanner images are acquired in the axial plane, or angled axial plane if the tube gantry is tilted. However, for PMCT the images are effectively isotropic (similar spatial resolution in all 3 planes). This means that the images can be reconstructed in any plane. The transverse plane is typically denoted by the x- and y-axes, with the z-axis orientated orthogonal to this (longitudinal plane). The x/y-plane effectively constitutes the image acquisition matrix, while the z-axis is determined by the thickness of the image detector. The image matrix describes multiple (typically 512 x 512) discrete volumes of information (voxels), presented as a 2-dimensional set of pixels. The spatial resolution of the images and therefore overall image quality is an important physical parameter, which is defined by the dimensions of the pixels and their thickness (the voxel thickness) (Seeram, 2001). Spatial resolution is influenced by several factors including, the inherent resolution of the X-ray detector, focal spot size, geometric magnification, stability of the rotation mechanism and the filtering algorithm utilised for PMCT reconstruction (Holdsworth and Thornton, 2002).

As a general rule, high-quality PMCT images can be obtained using a 512 × 512 matrix size. Depending on the field of view, subsequent pixels can be as small as 0.5 mm diameter. Slice thicknesses as narrow as 0.5 mm are also possible, making the voxel isotropic (Petorius, 2006). This size of pixel will naturally generate PMCT images with a high spatial resolution, which is considered fundamental when undertaking measurements on three dimensional image data (Coleman and Colbert, 2007).

1.6 CT Visualization and Analysis

CT scanning produces a large volume of data. This can be processed in a number of ways to extract specific features that the practitioner would most like to focus on. The formation of an image by a CT scanner involves three steps: 1) data acquisition, 2) image reconstruction, and 3) image display, manipulation (post processing), storage and communication (Fishman and Jeffrey, 1998).

1.6.1 Important scanning parameters/potential measurement error

A reconstructed CT image contains quantitative information and the precision (reproducibility) and accuracy (the closeness of the measurement to the true measurement) of the data is dependent on several key performance parameters, which must therefore be considered.

1.6.1.1 Resolution

The spatial resolution of a CT scanner describes the scanner's ability to resolve closely placed objects. Spatial resolution is often measured in two orthogonal directions: axial plane (x-y) and cross plane (z). Historically, there were large discrepancies between the axial plane and cross plane spatial resolutions. However, with the introduction of spiral and multi-slice scanners, the difference between the two has now been reduced and scanners are fully capable of isotropic imaging. The main parameters that affect the spatial resolution are the geometric magnification, slice thickness, reconstruction increment, and the focal spot size. For this investigation, the scan resolution was between 200-300μm.

1.6.1.2 Geometric magnification

The largest factor in determining the resolution achievable in clinical CT is the geometric magnification. For the vast majority of systems, the emitter and detector are held at fixed distance from one another. In clinical CT the sample distance is also fixed, having been chosen at manufacture so that the detector fan can capture the projected image of the part of the body in question. Three factors control geometric magnification; source size, source to object distance, and object to detector distance. Very small focal spot X-ray tubes can be used to create geometric magnification with excellent image sharpness. The source size is obtained by referencing manufacturers specifications for a given X-ray source. The object to detector distance is usually kept as small as possible to help maximize image sharpness.

Geometric magnification can be used to increase spatial resolution of a given detector. This can be very important when trying to detect defects smaller than the pixel pitch of the detector. This is also very important when taking high accuracy measurements. In general, the more pixels that information is spread out across and the smaller the pixel pitch of the detector the higher the spatial resolution of the image and the more likely that measurements recorded from CT images will be accurate.

Geometric unsharpness can occur when the radiation originates over an area instead of from a single point. As the source size decreases, the geometric unsharpness also decreases. When the source size is larger the different paths that the rays of radiation can take from their point of origin in the source causes the edges of the object being scanned to be less defined.

1.6.1.3 Slice thickness and reconstruction increment

On most CT scanners, different slice thicknesses can be selected, ranging from 10 to 0.5mm. The choice of slice thickness will have a fundamental influence on the spatial resolution (Kalender, 2000). As far as image sharpness is concerned, the thickness of the slice is crucial in controlling the partial volume effect, and consequently in enhancing the spatial resolution in the axial plane. Within each voxel, the attenuation of the tissues is averaged. As a result of this volume averaging, the perception of structures that are included only partially in a voxel may be diminished. This is known as the *'partial volume effect'*. This effect is most damaging to the CT image when the scanned object includes many small, high density structures, where only a portion of the structures extends into the slice thickness. Naturally, objects that extend only part of the way into the thickness of the slice will be measured by the detector as less attenuated, and hence less dense than they really are. This could cause information to be lost, which could affect the accuracy of measurements recorded from CT, particularly when measurements rely on the precise determination of the most extreme point of the bone. However, as the slice thickness is generally no greater than 1mm in post mortem scanning, it is unlikely that any loss of information would have a significant impact on the overall outcome of this investigation; as the age determination methods that the measurements are being applied to have an inherent error margin and estimations are therefore given as ranges not definitive figures. Volume averaging can also cause other issues that may be significant in a forensic investigation, for example; soft tissues lesions present in a voxel that contains high-attenuating bones may not be resolved at all. Unfortunately, the partial volume effects can never be eliminated completely. However, by decreasing the slice thickness (depth of the voxel), volume averaging can be reduced. Thus, objects previously blurred by volume averaging will be resolved as discrete structures if scanned and/or reconstructed with narrow slices. Consequently, in this investigation a thin slice setting was applied to reduce partial volume effect and to improve the localization of small structures in the axial slices, such as the precise end point of the long bones.

However, a disadvantage of a smaller slice thickness is an increase in *‘Quantum noise’*. This is related to the number of X-ray photons that are used to generate each image. Decreasing the slice thickness decreases the number of photons contributing to a particular voxel. The quantum noise is therefore inversely proportional to the square root of the number of photons. This means that during image reconstruction there is a trade off between spatial resolution and noise. As a compromise, during post-processing several slices can be combined to reduce the noise. Ford and Decker (2014) illustrated that skulls can be scanned safely at 1-1.25mm so that all crucial detail for 3D modelling is presented. In this investigation, a slice thickness of 0.5mm was used. Therefore, sufficient detail should be present to ensure accurate measurements to be recorded.

In addition to slice thickness, resolution in the z-plane is also strongly dependent on the scan or reconstruction increment between individual slice images. In spiral CT it is possible to freely select the number and the z-position of the images to be reconstructed, this offers the ability to overlap images, greatly improving the spatial resolution in the z-direction. An image reconstruction increment equal to the slice thickness corresponds to sequential CT and does not have the same advantages of spiral CT's overlapping ability, leading to a certain loss of resolution and small detail contrast.

1.6.1.4 Focal spot size

The focal spot size is the origin of X-ray used to produce a radiograph. Fine and broad focal spot sizes are available on general X-ray tubes. It has been established that a fine focal spot size reduces geometric unsharpness however, excessive use of fine focus can impact on tube life and the extent of this benefit on image quality is still unclear. Gorham and Brennan (2010) demonstrated no statistical difference between images produced at each focus. However, they also suggested that a fine foci X-ray source would enhance the visualization of fine details such as trabecular patterns, although there is no evidence to support this practice at present.

1.6.1.5 Additional image artefacts

Beam hardening**:** Beam-hardening artefacts are caused by the polychromatic nature of the X-ray beam. The low energy portion of the spectrum is absorbed more than high-energy portion and consequently the beam exits the object being scanned 'harder' than it entered. This results in the central part of a uniformly dense object being subject to harder X-rays than the outer part. The outcome of beam hardening is that the periphery of the object appears less radio-dense, which produces a 'halo' effect around the centre.

Metal artefacts**:** Effectively the X-ray beams get totally blocked and appear in the reconstructed image in a 'starburst' or 'striped' artefact. This is particularly noticeable in dental reconstructions that contain restorative work but can also effect other areas of the image if the individual is carrying metal objects, have been shot or have jewellery on, for example. Mental artefacts can obscure neighbouring structures and depending on the density of the mental involved can affect a relatively large area of the scan. This could make it difficult to examine these areas and make measurements challenging and less accurate. However, in the juvenile age cohort, metal artefacts are much less common, as children tend to have less restorative dentistry, wear less metal jewellery, and carry fewer metal objects. Therefore, in this investigation, metal artefacts did not significantly affect measurements.

1.6.2 Data acquisition

Data acquisition refers to the collection of X-ray transmission measurements through the patient. Once X-rays have been passed through the patient, they fall onto special detectors that measure the transmission values affected by attenuation (Seeram, 2001).

During scanning, the first image generated is known as a ‘scout’ or ‘localizer’ view or scanogram. This is created by translating the patient through the scanner with a fixed tube, either in the antero-posterior or lateral position. The result is a low-resolution image similar to a plain film radiograph. The scout image is normally up to 150 cm long, although this can occasionally vary slightly from scanner to scanner, which typically produces an image extending from the top of the skull to the proximal femur (patient depending) (Fishman and Jeffrey, 1998). If the entire body bag needs to be imaged then the bag needs to be rotated and the scout image, from the feet upwards, overlapped with the first view. These images are obtained in approximately 8 seconds. The scout images are primarily to plan the field of view and extent of the scan. When a scanning a body bag it is also useful to describe accurately the position of the body being scanned, as if the computer is told the position (e.g. prone, supine, etc.) it will correctly ascribe right-left and front-back on the reconstructed images. The scout views can also help a modern scanner vary beam intensity depending on body thickness through the scan. However, a dose lowering strategy is of little importance to forensic work. An advantage of the scout view in forensic work is that it allows an initial check for abnormalities, foreign objects and pathologies.

CT is an excellent example of digital image processing. The detectors convert the X-ray photons transmitted through the patient and arriving at the receptors into electrical signals (analogue signals). This provides a complex set of raw data with frequencies and amplitude, which then needs to be converted into a digital image (Kalender, 2005).

This array of numbers is sent to the computer for further processing. The benefit of digitisation is that digital images can be processed by specific computerised operations, to produce an output image that is different and more useful than the numerical digital input data. This technique has numerous advantages, such as image enhancement, restoration, analysis, detection, pattern recognition, geometric transformation, and data compression.

The outcome of this reconstruction process is an image with considerably improved contrast between structures, compared with plain X-rays, with no superimposition of structures and the benefit of being able to resolve structures in all planes.

1.6.3 Image reconstruction

After the detectors have collected the transmission measurements, they are sent to the computer for processing. The computer uses complex mathematical techniques to reconstruct the CT image in a finite number of steps called reconstruction algorithms. The application of this iterative algorithm is known as the algebraic reconstruction process (ART). This process acts as an estimate and therefore includes inherent errors (Kalender, 2005).

Mathematical filters can be applied to the CT data before image reconstruction to enhance the edges of the scanned object in a process known as filtered back projection (FBP). Although this ‘de-blurring’ produces a very high resolution image it also creates image noise (graininess). Image noise makes it difficult to differentiate soft tissue structures. Filters are therefore a compromise between resolution and noise creation. Different filter algorithms can be selected to enhance particular features within the image data such as a hard algorithm to enhance bone and lung images or a soft tissue algorithm that provides better soft tissue contrast (Petorius, 2006).

Quantisation is the final step of image reconstruction; in which all pixels in the image are assigned a unique Hounsfield number describing the linear X-ray attenuation coefficient of the 'tissue voxel' as described above.

1.6.4 Image display, manipulation, storage and communication

After the computer has performed the image reconstruction process, the reconstructed image can be displayed and/or recorded and stored for subsequent viewing and further analysis at a later date.

When viewing these images in black and white the brightness value of each sampled pixel is assigned a shade of grey (grey level) based on the Hounsfield number. The total number of gray levels is called the gray scale and can be composed of any number of gray levels. The range of HU values are from -1000 to +1000 allowing 2000 shades of grey to be used. However, the human eye can discern approximately only 16 shades of grey and therefore a reporting practitioner will not differentiate tissues with similar HU values as they will be able to distinguish only between differences of about 125 HU (2000/16) if their 16 shades of grey are spread over the whole spectrum. In this situation, windows can be applied to the image when viewed using an image review workstation, so that tissues of interest can be discerned. For example, a window setting with a 300 HU range would be useful for depicting HU differences of about 20 HU (useful for soft tissue imaging). Window widths of about 90 are used for brain imaging, increasing the contrast of the perceived image. Once windows are applied they will simply return black or white for pixels with HU's outside the window width. Therefore a window level (centre) also needs to be applied that is approximate to the HU of the main structure being examined. Typical clinical viewing window levels and window widths are 500/1500, 40/400 and -500/1300 for bone, soft tissue and lung, respectively.

Image manipulation has become popular in CT, and many computer software packages are now available. Images can be modified through image manipulation to make them more useful to the observer. For example, transverse axial images can be reformatted into coronal, sagittal and paraxial sections. Images can also be subject to other image processing techniques such as image smoothing, edge enhancement, grey-scale manipulation and three-dimensional processing. These techniques ‘enhance’ contrast between structures in an image so that the maximum amount of data can be extracted. Interestingly, these techniques actually involve reducing the data set to minimize ‘noise’ so, although they are generally referred to as enhancing techniques, they typically make specific areas stand out by taking some ‘less useful’ information away.

However, it is this step of the data analysis process and indeed the term ‘image manipulation’ that some professionals are uncomfortable with. As a result, the medico-legal significance of CT findings is often subject to intense scrutiny. It would arguably be more appropriate to call this stage ‘image enhancement’ as it is similar to a radiographer manipulating a patient into the best position for minimal superimposition, a photographer altering the contrast of a picture in order to view fine details more clearly or the enhancement of a pixilated video image. Any digital image could be subject to bias interpretation. However, the secure storage of an entire CT data set allows this process to be monitored by providing an information source that can be revisited indefinitely. Furthermore, the original image provides a Digital Imaging and Communications in Medicine (DICOM) header with each image that has an audit trail of what post processing has occurred within that image. Unlike other methods of ‘image manipulation’ that may alter the primary data, CT image manipulation on image viewing workstations only changes the way in which the scan is viewed.

1.6.5 Micro-CT

The fundamental principles of micro-CT are comparable to clinical computed tomography. However, micro-CT is able to produce images with a much greater spatial resolution and therefore greater image quality and detail. Unlike clinical CT, in micro-CT the sample is revolved between emitter and detector. The extremely small slice thickness and reconstruction increment makes micro-CT voxel spacing inherently isotropic. The isotropy of voxel spacing in micro-CT removes the need for interpolation between slices when rendering samples in 3D, eliminating a source of image artefacts, and allowing for greater versatility during virtual inspection. For micro-CT systems the distance between the emitter and object can be altered, so that smaller objects can be imaged at closer proximities to the emitter. It is this geometric magnification function that is primarily responsible for micro-CT's improved spatial resolution, compared with clinical CT. However, to obtain maximum quality images extremely protracted scan times of up to 6 hours might be required. Although, scan time is proportional to scan quality and resolution so can be reduced slightly, if required.

In a mass fatality incident (MFI), the extremely long scan time and the inability to scan a whole body means that micro-CT is not a feasible option for routine scanning, when their is a high through-put of bodies and time is of an essence. However, micro-CT could be a useful adjunct to a forensic investigation to provide additional information in special cases such as gunshot wounds and stabbings. In these situations the enhanced image quality offered by micro-CT could allow.

1.7 Forensic Radiology-Applications in Anthropology

1.7.1 Mass fatality radiology

Radiology is used for a number of purposes in a mass fatality incident. It can provide vast amounts of additional information to help with the detection of foreign bodies (which may pose a hazard to on-site practitioners), to uncover and pinpoint the exact location of material evidence and, most importantly to this thesis, to aid victim identification. MFI's may generate large numbers of victims, normally suddenly and unexpectedly (Baraybar, 2008). The rapid and accurate identification of individuals is of primary importance for both juridical reasons and for relatives' peace of mind. This is typically achieved by comparing post-mortem and ante-mortem information, on the assumption that each individual exhibits their own unique features and no two individuals are completely identical (Black, 2007).

MFI are often international in scope. For this reason, in 1984 Interpol published the first disaster victim identification (DVI) manual. The aim was to provide information relating to mass disaster handling in general, and the identification process in particular, to increase the efficiency and effectiveness of DVI (Interpol, 1996). At present, 187 member countries utilize a common recording format, and where possible have a structured DVI team in place to react in the event of a mass fatality (Interpol, 1996). Experience acquired during every major disaster event has contributed to the continuous development of this manual, and it now incorporates up-to-date identification techniques (Interpol, 1997).

Although the ultimate aim of Interpol is to create an international standard reporting format, in several countries national laws and regulations influence the selection of victim identification methods. The maceration of bones, for example, to obtain anthropometric measurements, is discouraged or even prohibited in a number of countries (Leclair *et al*., 2007). Therefore, to achieve a unified international approach and improve future disaster victim identification, the introduction of new and innovative techniques that utilise medical imaging would be extremely beneficial.

The current standard approach, as used in previous incidents such as the terrorist bombings in London in July 2005, involves moving fatalities through up to three separate radiology stations: fluoroscopy, to screen remains for potential contaminants or evidence prior to autopsy; standard radiography, principally used for anthropological and pathological examination; and dental radiography. This process requires the procurement and installment of three different imaging modalities in a temporary mortuary environment, sufficient staff to operate them and subsequently a number of health and safety implications.

Mobile fluoroscopy units require the operator to be positioned next to the body throughout the imaging process, to capture images when necessary as they move the machinery systematically along the length of the body. They may also need to open the body bag and manipulate the body to obtain a good imaging plane, exposing them to contamination and often disturbing sights. This surface investigation takes approximately 15 minutes and pathologists who are not specifically trained to report radiological images normally interpret the images.

Plain radiograph stations require a separate team of operators. As with fluoroscopy, the body bags often need to be opened to optimize the imaging procedure. The radiographs are normally examined by either radiographers or forensic pathologists and communicated verbally. A formal radiological report is often not generated.

Dental X-rays are generally undertaken by an odontology team, rather than a radiography team (Schuller-Götzburg and Suchanek, 2007). They face similar issues regarding body manipulation, contamination and staffing. In addition, all of these imaging modalities normally need their own specific electrical power supply and a dry working environment, which presents a number of logistical problems in a chaotic temporary mortuary.

1.7.2 Mobile computed tomography

Using mobile PMCT could reduce disaster management and issues relating to equipment sourcing, operational personnel and health and safety. The use of mobile PMCT scanners in a forensic setting was first suggested in Japan (Hayakawa *et al*., 2006). Rutty and colleagues were the first group to implement this in the UK in 2006 (Rutty *et al*., 2007). This publication reported the authors' experience using mobile PMCT in general autopsy practice to overcome problems with access to clinical scanners. Its potential for DVI has since been considered by the Virtopsy® group in several theoretical papers. They illustrated the benefits of mobile PMCT as a mass fatality-screening tool, and its ability to collect information to complete the Interpol DVI forms (Jackowski *et al*., 2006; Sidler *et al*., 2007). Although mobile PMCT is relatively new to forensic practice, protocols are already well established in clinical medicine and could theoretically simply be adapted to the needs of post-mortem imaging.

Mobile PMCT would also be extremely valuable when dealing with mass fatality events involving chemical, biological, radiological or nuclear materials (CBRN). In this scenario, anthropological examination *in situ* and radiological techniques that involved body handling would not be possible due to the high risk of contamination to trained operators. Mobile medical imaging units would be extremely beneficial, greatly reducing the number of on-site personnel. In theory, an individual could be PMCT scanned on-site using mobile medical imaging modalities and then sent to a suitable identification expert by tele-radiology. The anthropological examination and identification of the individual could therefore be performed at a number of different remote sites, without the need to expose investigators to the contaminated site. PMCT may in the future provide an all-encompassing radiology facility, acquiring all the necessary data in one, more time efficient station (Sidler *et al*., 2007).

1.7.3 Court presentation

During a forensic court case, an expert witness might require visual representations of their evidence in the form of photographs or diagrams to convey their findings. In the UK, over recent years there has been a move away from using autopsy photographs in court, as these images are considered traumatic and could be upsetting for the jury due to the unpleasant nature of forensic casework. Hand drawn black and white, two-dimensional 'body diagrams' can be used to create computer generated anatomical models. These can be printed for paper illustrations or presented to a jury as three-dimensional animations. However, the production of these recreations has considerable cost implications and the image remains the interpretation of the artist, not real findings.

External surface scans could also be used. Although, practitioners rarely use this technique as data acquisition and manipulation of the images to a presentable level is time consuming and therefore has considerable cost implications. Computed tomography is a practical solution to these issues. It can be used by an expert witness to illustrate both internal and external body findings, with scout, multi-planar reconstructions (MPRs) and three-dimensional reconstructions that are less traumatic to the jury than photographs and more easily obtained than surface scanning.

1.8 Aims and Objectives

The overall aim of this thesis is to strengthen and develop the field of forensic radiology and imaging worldwide. This aim is rooted in the direct application of anthropological identification techniques using PMCT in forensic practice. This includes developing a standard protocol for anthropological reporting using PMCT, promoting best practice and developing international quality control and guidelines in the field of imaging. The hypothesis of this research is that:

1. PMCT provides identical measurements to standard osteological techniques and;
2. Protocols can be developed to efficiently use PMCT data to rapidly provide a full anthropological investigation by experts anywhere in the world.

Three key areas of forensic radiology have been identified, representing gaps in current understanding relating to:

1. Data acquisition,
2. Examination methods and,
3. Reading and reporting images.

The research carried out in fulfilment of this thesis will add to current knowledge and literature of these areas. From an operational point of view this translates into the following specific research objectives:

Research Objective 1: Determine the validity of using PMCT measurements for the anthropological and odontological assessment of the developing human skeleton, through the comparison of measurements obtained from CT images, with those attained by traditional anthropological means (the current gold standard).

Research Objective 2: Ascertain the accuracy and precision of age estimations of the developing child by applying anthropological and odontological techniques that are well established in the literature to images generated using PMCT, to assess its prospective contribution to disaster victim identification.

Although many different imaging techniques exist for the visualisation and interpretation of CT information in a clinical setting, there is a discernible lack of research relating to post-mortem imaging. Subsequently, the research conducted in this thesis will help to develop a coherent method that is repeatable and provides standardised measurements that are clearly defined in terms of how they must be taken, statistical reproducibility and with accuracies comparable to traditional anthropological methods - to further future research in this area.

1.9 Delineation of Chapters

To initiate research, it was first necessary to conduct a thorough background review of all age-at-death determination methods frequently used in forensic practice and to consider whether they could be directly applied or suitably adapted to PMCT, replicating the measurements required for the accurate biological profiling of juvenile remains. This preliminary investigation was used as a platform to facilitate subsequent research in this thesis by confirming the vast potential PMCT afforded full body anthropological examination.

Chapters 5 and 6 contribute to part one of this thesis. The investigations conducted in these chapters concentrate on determining the accuracy and repeatability of PMCT measurements, using the clavicle and dentition, respectively. These include comparison studies between anthropological methods and PMCT methods. By using multiple research observers, the inter- and intra observer errors are also examined in both chapters, and results to support the implementation of PMCT in routine forensic practice are presented.

The second part of this thesis includes chapters 7 and 8. Chapter 7 includes a ‘blinded’ test to validate the previous chapters’ work. It was designed to create a radiological reporting form for biological profiling of human remains. This involved identifying a concise collection of PMCT osteological and odontological images from which a full body examination could be conducted for biological profiling by; reviewing the current Interpol form, conducting an extensive literature review and consulting experts in the field of radiology, anthropology, forensic pathology and odontology.

The penultimate chapter, before a detailed discussion and final thesis conclusions, includes an individual case study to illustrate the transferability of the ‘minimum data set’ recording form developed, to adult remains. It also considers the advantages and limitations of a PMCT method of biological profiling from the perspective of the legal, forensic, and radiological communities. Finally, future research projects will be identified and discussed.

CHAPTER 2: LITERATURE REVIEW

"If you cannot - in the long run - tell everyone what you have been doing, your doing has been worthless."

- Erwin Stronger (Nobel Prize winner in Physics).

2.1 Radiology in the Courts

The potential use of radiography as an aid to medico-legal investigations was recognised across the world in quick succession with its discovery in November 1895. The first criminal court case to use X-rays occurred later the same year in Montreal, Canada to help convict George Holder of attempted murder. An X-ray, which took 45 minutes exposure time, was used to illustrate the position of a projectile adjacent to the lower limb bone. Although providing valuable evidence in this case, there was great controversy surrounding the admissibility of X-ray evidence. A number of courts refused to accept them, stating:

> '*It is like offering the photograph of a ghost'* (as cited in Brogdon, 1998; pg. 21).

However, this matter was finally settled in 1896 when Judge LeFevre decided:

> *"We have been presented with a photograph taken by means of a new scientific discovery. It knocks for admission at the temple of learning: What shall we do or say? Close fast the door or open wide the portals? (…..) Modern science has made it possible to look beneath the tissues of the human body, and has aided surgery in telling of the hidden mysteries. We believe it is our duty to be the first...in admitting in evidence a process known and acknowledged as a determinate science. The exhibits will be admitted in evidence."* (As cited in Brogdon, 1998; pg. 21).

Thus, the use of X-rays as evidence in medico-legal cases was established within the court system and is still used to this day. This being said, there is now a similar debate concerning the validity of PMCT as evidence to replace invasive techniques. PMCT images are accepted as supporting evidence however; there is a discernable lack of literature investigating the viability of using PMCT as a replacement to the internal autopsy examination within the legal arena. If the legal profession does not accept PMCT, much of current research in this area will have been in vain. If the prosecuting authorities are unable to prosecute a case with PMCT scan evidence, invasive post-mortem examinations will still be required (Jeffery *et al*., 2011).

2.2 Magnetic Resonance Imaging

In 1977, six years after CT was first introduced into a clinical setting, the first reports of clinical MRI emerged. Unlike the almost immediate application of CT for medico-legal investigations, it was not until 1996 that the first publication relating to MRI being used in this regard was produced. Brooke *et al*. (1996) recorded their experience of MRI in association with perinatal autopsies, which was followed by a number of other studies by numerous researchers relating to paediatric practice.

The first adult MRI studies were produced in 1998, when Bisset (1998) proposed that MRI could potentially be used to investigate adult sudden deaths. Bisset subsequently conducted a four-year research project into MRI as a substitute to an invasive autopsy in cases of adult death, reporting the first operational system of its kind (Bisset, 2002). Roberts *et al*. (2003) presented further evidence in support of Bissets' system. In response to this publication, Rutty and Swift (2004) introduced the concept of a new medical sub-speciality of autopsy imaging which they termed "necroradiology" (Rutty, 2004).

2.3 Post-Mortem Computed Tomography

Despite the early introduction of CT in the early 1970's, the field of PMCT is reasonably new but is developing rapidly, replacing more conventional forms of radiology as novel techniques evolve. The first clinical CT scanner was installed in October 1971 at the Atkinson Morley Hospital in Wimbledon, England. The machine took 160 parallel readings through 180 angles, each 1 degree apart, with each scan taking a little over 5 minutes. Imaging was confined to the head, and images from these scans took approximately 2.5 hours to be processed by a large computer.

In 1994, a number of technical developments, including the introduction of three dimensional, whole-body, digital image acquisition, lead Donchin *et al*. (1994) to suggest using CT as an alternative to the invasive autopsy, after becoming the first researchers to publish their experience using whole body PMCT imaging for autopsy purposes. Following this suggestion, Professor Gil Brogdon commented that there was still “no general appreciation of the extent of the radiological potential in the forensic sciences,” (Brogdon, 1998; pg15) in the preface to the first edition of his principle textbook *Forensic radiology*. As if motivated by these statements, forensic pathologists and radiologists worldwide began to intensify their research efforts into the potential of advanced imaging technologies in forensic practice. The capabilities of PMCT are now championed within the literature with many researchers agreeing that it has the potential to replace invasive post-mortems in the near future (Rutty, 2007; Leth, 2007; Jeffery *et al*., 2007; Hoey *et al*., 2007; Dirnhofer *et al*., 2006). Analysis of literature between 2000 and 2011 revealed a ten-fold increase in publications relating specifically to forensic and post-mortem radiology during this period. This compares with only a two-fold increase in annual publications relating to general medicine, forensic science and general radiology. This highlights the importance and growing interest in the area of forensic and radiological research.

In 2002, a research group in Switzerland coined the term ‘Virtopsy®’ to describe the notion of a virtual autopsy (Thali *et al*., 2003; Thali, 2009). The Virtopsy group presented the first installment of their research in 2001, at the German Society of Legal Medicine conference in Switzerland. The aim of the Virtopsy project was to implement modern imaging modalities in forensic practice (Thali and Braun, 2000; Thali *et al*., 2003). During this time Bisset *et al*. (2000) also published their experience using magnetic resonance imaging (MRI) in the UK (Bisset *et al*., 2002; Thali *et al*., 2002). From this point, forensic radiology expanded rapidly, with a number of practitioners suggesting it should be recognized as a distinct sub-discipline of radiology and forensic medicine (O’Donnell and Woodford, 2008).

Within the UK, a board of experts appointed collectively by the Ministry of Justice, Ministry of Defense, Department of Health and the Home Office have been assigned the task of pushing forward research into the non-invasive autopsy. In England the three principle research and practitioner groups are based in Leicester, Manchester and Oxford. Since 2002, Leicester has used PMCT routinely in forensic cases. Today they hold possibly the largest validated image dataset in the UK, which is used as a teaching and training aid for both diagnostic and research purposes. They hold a grant form the National Institute for Health Research (NIHR) for the investigation of targeted angiography to investigate natural sudden unexpected deaths and is the largest research group actively producing publications into numerous various aspects relating to a 'near virtual autopsy'. The MRI system described by Bisset is still predominantly in place in Manchester, however together with Oxford the Manchester group have recently published the findings of a Department of Health-funded research project investigating the use of PMCT and MRI as an alternative to the invasive autopsy in natural death (Roberts, 2011). In 2013, the first PMCT-based imaging system was installed into a public mortuary in Bradford, UK.

In May 2005, the Victorian Institute of Forensic Medicine (VIFM), Victoria, Australia, installed a 16-slice PMCT scanner within their mortuary facility (O'Donnell, 2006). The Victorian State Government funded this in an attempt to enhance Victoria's counter-terrorism capabilities (Collett *et al.*, 2006). This is undoubtedly the single best example of service provision using a PMCT scanner within medico-legal premises. Since installation, over 15,000 bodies have been scanned. Australian law related to cadavers has been changed to allow for scanning to occur without relatives' consent or the authority of the coroner, to enable the images to be used as a triage system to avoid invasive autopsy examinations. Early reviews detail a number of difficulties the clinical radiology teams were confronted with during image analysis at VIFM. Publications from this unit are sporadic and generally focus on technical observations from their experiences running a working imaging mortuary. Although mirroring the increasing frequency of clinical cross-sectional imaging, the application of PMCT in the mortuary required different approaches to examination. In the living, CT interpretation involves the administration of an oral or intravenous contrast, which could not previously be done in the deceased. O'Donnell (2006) noted that it was necessary for the reporting radiologist to have a comprehensive understanding of the decomposition process - as the formation of gas can lead to alterations of the internal anatomy that in the living may be considered significant but in death are normal post-mortem features. What has quickly become apparent from the work in VIFM is that PMCT is, quite simply, not the same as clinical CT. For this reason the authors suggest that an individual with training in post-mortem changes is required to report on CT scans, to avoid over-interpretation. However, an investigation by Hogge *et al*. (1994) into the effect of interpreter experience-level on successful identification indicated that individuals with training in radiographic interpretation performed significantly better than other medical groups. This suggests that perhaps specially trained observers specifically for forensic identification are necessary for the successful implementation of PMCT in forensic practice.

Within Victoria, PMCT has proven to be a valuable adjunct particularly in situations where next of kin have objected to post-mortem examinations or when death certificates have come under scrutiny and deemed to require further evidence. It has also facilitated the Australian system of ‘inspection and report’, which is similar to the Scottish ‘view and grant’ system - where the pathologist provides a cause of death without an autopsy, by reviewing the case information together with an external examination - but with the added advantage of radiological evidence (O’Donnell and Woodford, 2008). It is now standard practice, in a large proportion of forensic institutes worldwide, to conduct a post-mortem full body image on every individual before autopsy.

In addition to the scientific community, there is also lay interest in this area of research and several religious groups are championing the proposal for a rapid, non-invasive examination of the dead. There is a perception that the general public and religious faith groups find the thought of an invasive autopsy being undertaken on their relatives as distasteful. However, a number of reviews on this topic suggest that this might actually be the personal perception of the person making the comment, rather than the genuine view of the public. Of the predominant faiths, Islam, Judaism and Zoroastrian faiths only permit autopsy examination when ordered by a coroner (Rutty, 2010). Although there is a general dislike of autopsies in Hinduism they are not strictly forbidden as with the previously mentioned religions (Rutty, 2010).

Despite an ever increasing body of peer reviewed literature concerning the use of PMCT and post-mortem MRI imaging, there is to date only a couple of publications that focus on the perception of the users, professionals and public opinions of such systems. To my knowledge, to date, no large population survey has been conducted to determine the wider consensus on this topic. One study by Rutty and Rutty (2011) distributed a questionnaire to a small audience (n=72) made of both individuals from the general public and pathologist/medico-legal practitioners. They discovered that only 7% of the public cohort indicated that they would actually object to an autopsy authorized by a coroner, much less than the general perception. However, when asked if cross-sectional imaging was available as an alternative to invasive autopsy, whether it would be preferred, there was an overwhelming desire for its use from the general public. Conversely, the pathology group demonstrated a greater number against its use. Rutty and Rutty's investigation suggests that there is not as much resistance to an invasive autopsy as may have been previously suggested but that in general, the public would welcome an non-invasive alternative. Most of the current literature on this subject refers to individual case reports and fails to consider a wider perspective or truly assess the question of whether a virtual autopsy would be fit for medico-legal purposes.

For expert evidence to be presented in court it must be supported by a bank of scientific data, based on a method that has undergone peer review and is generally accepted within the relevant scientific body. Although CT scanning has an established role within clinical science - O'Donnell's observations that PMCT has a much more tenuous position within forensic practice has legal implications. To date there is little peer-reviewed literature to support the presentation of PMCT findings in a court of law. Jeffery *et al*. (2011) conducted an investigation into the criminal justice system's considerations of "so-called" near-virtual autopsies at the East Midlands Forensic Pathology Unit (EMFPU), Leicester. Through a comparative analysis of invasive post-mortem and PMCT findings and a questionnaire based qualitative thematic analysis, the authors considered; 1) if PMCT could provide the same level of information as an invasive Post-mortem (PM) and 2) arguably more importantly, whether it could meet the needs of the end users. They conclude that PMCT is good at providing accurate causes of death and that the interpretation of cases is not significantly altered by the absence of histology. From the questionnaire, which was sent to a panel of experts consisting of a criminal judge, medical coroner, criminal barrister, criminal solicitor and senior police officer, it was evident that PMCT satisfied the needs of these professionals in straightforward trauma deaths, such as road traffic accidents. However, at present, these individuals believed that PMCT did not provide enough information to satisfy the expectations of the criminal justice system in complex forensic cases. A blanket application of the non-invasive approach to all autopsy cases could therefore conceivably lead to missed homicides. Although, a stepwise approach introducing PMCT initially as a pre-screening aid to decide whether further invasive investigation is required is more likely to be accepted into criminal justice system at present.

These publications suggest that caution should be applied by the scientific community, when relaying the benefits of PMCT in a public forum, as the general opinion appears to be in favour of this non-invasive technique. However, many of the preliminary results are yet to be supported by sound scientific research that needs to be conducted before PMCT could be considered within the judicial system as a replacement for an invasive autopsy.

2.4 PMCT Reporting

As the application of PMCT as a pre-autopsy examination tool is becoming increasingly widespread in forensic medicine, it presents the important question of whether the scope and role of reporting should follow an official structured reporting format. This might be dictated by legal requirements as set out by law and landmark court decisions or alternatively whether a free reporting format should be used.

The core task of any post-mortem examination of a body is to provide a comprehensive account of all the relevant findings. With that, the presence of all data gathered, as well as case relevant significant content of any data (including PMCT data with presence as well as absence of relevant findings) have to be explicitly reported. Just capturing and storing data is not legally sufficient in the medico-legal context. Generally, the readers of PMCT can use their judgment in what they want to report, and how they want to formulate their written reports.

In a report by Schweitzer *et al*. (2014) a structured report format was directly compared with a free report format. In this investigation forensically relevant items were missed in critical subject areas in 25-79% of free form PMCT reports. Conversely, for the specific purposes of forensic pathology, structured reporting contained all key features. With PMCT gaining increasing acceptance worldwide it is important to consider how measurements and results can be appropriately recorded to ensure that critical evidence is not missed, whilst not wasting time on irrelevant findings. Structured reporting in PMCT would provide a technically reliable basis for good PMCT reporting. It would force experts that report on PMCT findings to be exhaustive and verbose in their reporting, as a structured form would ensure that all angles of a comprehensive medico-legal investigation were covered sufficiently. A structured form of reporting would also give more weight and credibility to PMCT findings, especially if it was created in conjunction with experts in Law.

2.5 Anthropological Computed Tomography

In line with other research in this field, the majority of previous investigations in the area of anthropological computed tomography have also been case-report oriented, with very few large-scale trials being presented to date. These studies are generally theoretical (Sidler *et al*., 2007; Verhoff *et al*., 2008) or based on qualitative techniques (Lottering *et al*., 2013). The majority of publications use static clinical scanners, but these may be identical to mobile scanners in terms of the actual equipment used. Recent advancement of PMCT and 3D reconstruction techniques has improved the speed of scanning and precision of 3D measurements, which will aid further development in anthropological forensic practice. However, with a reported 22% of forensic radiology publications between 2000 and 2011 relating to identification, and with CT used as the image modality of choice in 53% of these publications, this area of research is slowly changing from an obscure topic to a relevant field in forensic science.

Perhaps the first attempt to determine the repeatability of osteometric measurements using CT was undertaken by Hildebolt *et al*. (1990). The small pilot study used standard spreading calipers to measure five adult skulls, using a number of cranial landmarks already well established in the literature. Comparative measurements using a CT technique were also recorded and compared with the osteometric measurements. The results of the investigation suggested that there was no significant difference between the measurements recorded from surface rendered images (3D-CT images) and those obtained by dry bone analysis. Due to extremely small sample size however, it is important to consider that type II errors (failure to reject the null hypothesis) may have occurred during statistical analysis. A type II error would cause the authors to falsely reject the null hypothesis that is, conclude the means of the two measurement techniques were not different when in reality they were different, leading to biased results. Hildebolt and colleagues also considered the value of measurements obtained from the original CT slices, before they were digitally reformatted to 3D-CT images. The authors established that 3D-CT measurements were superior to those taken directly from CT images, although suggested that this method could not be used to reproduce traditional osteometric measurements with any great deal of accuracy at time of publication. The authors therefore concluded, that until this technique was significantly improved, measurements obtained by this method could not be regarded as precise.

In the last 20 years since Hildebolt and colleagues published their results, technology has clearly improved significantly; for example, multi-slice spiral CT, has now replaced incremental CT used in previous investigations. As a result, although illustrating that the accuracy and precision of 3D-CT osteometric measurements were restricted by technical limitations, the authors also demonstrated for the first time that it was indeed possible to measure human osteological material using 3D-CT techniques. Subsequently, with the vast technological advances in the last two decades, similar measurements can now be taken with much greater precision.

In a more recent study, Cavalcanti *et al.* (2004) attempted to resolve the issues highlighted by Hildebolt *et al*. (1990) by employing a more superior image post processing technique known as 'volume rendering'. Their study produced similar results but concluded that a high degree of precision and accuracy could be achieved with their improved method. Nevertheless, their sample size was also extremely small and negated to consider more regularly applied anthropological measurements generated using an osteometric board; issues that were later considered in a publication by Robinson *et al*. (2008).

Robinson *et al.* (2008) recorded anthropological measurements of the lower limb and foot bones using a virtual osteometric board. The study was undertaken to determine two things; 1) whether PMCT could be used to replicate osteological measurements of fleshed long and small bones, with accuracy comparable to that of traditional anthropological examinations of defleshed limbs; and 2) could this be practically undertaken using medical imaging technology at one site and reported at a remote site via tele-anthro-radiology. The introduction of PMCT significantly improved the three-dimensional image resolution and unlike 2D scout views of CT images and plain radiographs, both of which have previously been used to measure the lengths of long bones with variable accuracies (Helms and McCarthy, 1984; Harris *et al*., 2005; Anderson *et al*., 2006; Aaron *et al*., 1992; Dedouit *et al*., 2007), the 3D images could be viewed on dedicated software in the x, y and z planes. This investigation is of particular importance, as it is the first publication in this area of research to use quantitative techniques and consider the practicalities of using PMCT for disaster victim identification (DVI). Although Robinson and colleagues did not use a mobile PMCT scanner for their investigation, they demonstrated that vast amounts of PMCT data could be sent for remote investigation and therefore, that using a mobile scanner is a viable solution for DVI, to minimize onsite personnel. Although scanning operatives would still be required to visit the scene, forensic experts in odontology, pathology, anthropology and other investigating officers would not need to attend the scene itself.

One of the analysis workstations used a ‘transparent bone‘ algorithm to view the image, while the other created a multi-planar reformation (MPR) from the 3D data set. The transparent bone setting enabled landmarks, even if obscured by neighboring skeletal structures to be observed; a technique that would be of particular value for examining complete human remains, allowing even superimposed personal artefacts such as wrist watches to be excluded from the image. Although MPRs are not strictly 3D images, they still have a potential efficacy for 3D-CT measurements. An MPR is essentially a two-dimensional (2D) slice through the data set that is arbitrarily selected by the observer. It can therefore be used to define the extremities of a bone and even reproduce the planes of the osteometric board. In this study, the base plate of the osteometric board was defined by scrolling through the image with the MPR slice to locate the three points on which the bone would lie. Two additional slices orthogonal to the base plate were then added to mimic the sliding plates and establish the extreme points of the bone. By measuring the distance between each orthogonal MPR slice the authors were able to replicate measurements obtained using an osteometric board.

The results indicated that there was no significant difference between the measurements determined using PMCT and those recorded using an osteometric board. With imaging taking approximately 5 minutes per limb, and Tele-Anthro-Radiology with analysis time taking between 20-30 minutes, depending on internet speed and operator experience, these results suggest that the use of PMCT could accelerate the process of anthropological measurements by a considerable amount (variable depending on condition of the remains and the amount of soft tissue remaining) by removing the necessity to clean bones. In addition, several experts could potentially analyse the same specimen simultaneously in a safe environment, which is of particular importance when dealing with contaminated remains. The mean error between the two techniques was low for all recorded measurements (tibia length, tibia width, calcaneus length, calcaneus height, talus length, talus height), with the largest mean error (1.8mm) resulting from calcaneus length measurements - which agrees with Corner and colleagues’ findings that the overall proportion of error is greater over a smaller distance. Within-subject standard deviation

(WsD) for all the PMCT measurements were extremely low, which in turn amounted to narrow 95% confidence intervals of 5mm (7mm for talar length). Variations of this size are likely to be due to inter-observer variability in identifying bony landmarks and alignment in both techniques. In addition, the inter-observer variability of 1.58% for talar length, calculated by Robinson *et al.* (2008), compares well with the previous study by Adams and Byrd (2002), which produced the same result (0.58%) for observers with 10+ years experience. Robinson *et al*. (2008) also determined that sex assessment using PMCT was close to the reliability of direct anthropological measurements using 'cut-off' points described in previous studies of calcaneal and talar length.

By demonstrating the utility of tele-radiology, Robinson el al. (2008) have opened up the possibility for anthropological assessment of contaminated body parts at remote safe sites, rapid international peer opinion, external quality assurance schemes and the international collection of population data. Although clearly illustrating that it was indeed possible to extract anthropological measurements from PMCT images, Robinson and colleagues (2008) did not establish any standardised protocols for this technique, which is a necessary step before this method can be internationally adopted for DVI. Additionally, while producing promising results it is important to note a number of problems that were encountered in this study. Primarily, the accuracy of the assessment appeared to be dependent upon the operators experience and preference with different techniques, as discrepancies were noted between different imaging software programmes in relation to the image handling and measurements achieved. Standardising image processing and measurements will therefore be extremely important for the progression of this technique and specimen quality may affect the accuracy of future studies.

Although Robinson and colleagues briefly introduce the feasibility of PMCT for age and sex determination, a more comprehensive study focusing on this aim is presented by Grabherr *et al.* (2009). Three anthropologists with different professional experience investigated the data from the bodies of 22 specimens. The study group represented ages between 17 and 92 years at the time of death (all adult). As a basic orientation for the age estimation, the complex method according to Nemeskeri and co-workers (1960) was applied. Additional parameters such as state of dentition and degeneration of the spine were also considered for the final estimation however; each researcher according to their personal experience chose these individually. The results of their study clearly illustrate that the estimation of both age and sex is possible using PMCT. As a result, virtual skeletons as presented in this study offer an ideal collection for anthropological studies, because they are obtained in a non-invasive way and can be investigated *ad infinitum*.

Grabherr *et al.* (2009) pick up from the Virtopsy® project which briefly raised the idea of PMCT enabled dental identification for mass fatality incidents in their first paper, followed by a second, more detailed study, which considered the potential of PMCT as a screening tool for mass fatality investigations using INTERPOL DVI forms and how PMCT could be used to fill out these forms. In comparison to the Virtopsy® papers, Grabherr and colleague's (2009) objective was to investigate different parts of the skeleton opposed to a single parameter, which resulted in them using the complex method according to Nemeskeri *et al.* (1960). This method is widely used by anthropologists in German-speaking regions, especially those who are working with historical skeletal materials. Age estimations with confidence intervals of 3 years or less were interpreted by the authors as correct estimations. Results using only the complex method were poor with over 50% of estimations being incorrect for all observers. When other parameters were added these results improved significantly however, observer 3 still presented a high number (45.5%) of wrong interpretations. Despite these results the authors conclude that PMCT can be used to perform estimations of the sex and age of a deceased persons and attribute some of the initial difficulties with age estimation on a lack of experience in evaluating digital images. In addition they consider the error rate to be similar to that experienced in previous literature utilizing anthropological techniques. In this study a slice thickness of 0.63mm was used which allowed even small structures such as the texture of the symphyseal surface of the pubic bone to be investigated. Currently a slice thickness anywhere between 0.5mm and 5mm can be used and where this may have previously caused discrepancies in image quality, with modern scanners there is very little difference between scans 0.5mm thick and those 1.5mm thick.

While defining a PMCT protocol Grabherr *et al*. (2009) did not establish any standardised measurement protocol, as their approach was purely qualitative. Although morphological approaches are commonly used by forensic anthropologists and are arguably more accurate at producing age estimations, these techniques are not as judicially sound as metric methods and are therefore less useful in a forensic scenario. In addition, a statistical analysis was not performed by the authors due to the small sample size, an issue they look to correct in the future by examining a larger collection of bodies. However, this study, as with future proposed studies by the author, utilize forensic case CT scans and comparative studies with macerated bones of the bodies is not an option as it is beyond the ethical and juridical scope of the research. In addition, Grabherr and colleague's study does not extend to juvenile cases, therefore creating a large void in this area of research that must be considered.

What is highlighted by all of these publications is the necessity for more comprehensive guidelines regarding the correct procedures for collection, processing and presentation of PMCT anthropological measurements. All of the publications discussed in this short review use different scanning procedures, software packages, skeletal elements, morphological features, anatomical landmarks and/or statistical tests. Therefore comparison of the results is particularly challenging and compilation of data is near impossible.

This opinion appears to be supported by Lottering *et al*. (2014), who have attempted to introduce a 'standardized' protocol for anthropological measurements of virtual sub-adult crania using PMCT. The authors used Amira (VSG, FEI Company, united States) and Geomagic Design X (3D Systems, Inc., United States) image software workstations to create 3D reconstructions. The authors subsequently identified three anatomical "reference" planes (mid-sagittal, transverse and coronal), from which "off-set" planes were defined to correspond to the most extreme positions or contours of the surface model. These planes were used as fixed points to make automated plane-to-plane measurements. Maximum cranial length (GOL) was measured between the anterior and posterior extreme position planes parallel to the coronal plane, while the maximum cranial breadth (XCB) was measured between the bilateral extreme position planes parallel to the mid-sagittal plane. The authors also quantified the connective tissue area corresponding to the anterior fontanelle and contiguous sutures (sagittal, coronal and metopic sutures), using a high-quality 3D isosurface mesh model function on Geometric Design X. The measurements were recorded on ten juvenile skulls by five independent observers with various experience and professional backgrounds. The authors concluded that all participating investigators could reliably replicate the measurements when comprehensive instructions were available. Suggesting that this measurement protocol should be used in all further morphometric analysis of skeletal elements for forensic investigation.

The measurement method outlined by Lottering and colleagues utilizes 3D high-quality surface models to perform complex shape analysis. There is undoubtedly a requirement to "standardize" the measurement procedure for anthropological PMCT reporting. An offset plane technique is an interesting suggestion that would certainly help to clarify terminology and provide fixed reference points, which could be easily transferred to other anatomical regions. Although this publication demonstrates the utility of Amira and Geomagic Design X for morphometric analysis, these programmes are expensive resources and therefore may not be readily available to all research institutes. However, the measurements and techniques discussed in this technical note could be achieved on a number of more accessible software packages (i.e. OsiriX, Voxar, etc). Therefore, specific reference to a particular software workstation might be too prescriptive for adoption of a "standardized" protocol.

The reasoning behind using a juvenile sample population for measurements of the skull is also questionable. Although it is appreciated that skull measurements are used for sex and ancestry determination, these discriminate function techniques are generally applied to adult remains, as the majority of sex and ethnicity characteristics do not develop until after puberty. Whilst this is a minor concern, it limits the practical application of the method described in this publication.

The data collected in this investigation seems to be of limited value for inclusion in similar studies because of the minimal utility of the selected measurements in any juvenile anthropological methods used for biological profiling. However, quantification of the connective tissue area is an interesting topic raised by the authors. Calculation of this complex region of interest demonstrates superior measurement opportunities afforded by PMCT data. Using sophisticated reverse engineering capabilities there is the potential to attain areal measurements, as demonstrated in this investigation. This study is one of the first to apply anthropological measurements to 3D isosurface mesh models rather than 2D axial and lateral views or 3D MPRs. It would be interesting to know how long this measurement procedure took, which was unfortunately not discussed by the authors, to assess if it would be practical in a DVI or forensic investigation, where time is often limited.

While the literature regarding post-mortem computed tomography is growing, there is still much to learn. Although, there are undoubtedly many advantages to adding sophisticated imaging to the post-mortem procedure there needs to be more critical assessment of the limitations of PMCT before we can consider it an alternative to the current post-mortem examination. We also need to consider whether radiological examination is robust enough to meet the requirements of the judicial system.

CHAPTER 3: STUDY DESIGN

"Progress, of the best kind, is comparatively slow. Great results cannot be achieved at once; and we must be satisfied to advance in life as we walk, step by step."

- Samuel Smiles (Scottish author).

3.1 Overview

Through CT imaging of the Scheuer Collection (See Section 3.2.1) and comparison with the original collection the first part of this thesis will primarily consider:

- The translation of traditional anthropological techniques to PMCT for the osteological examination of the developing human skeleton.
- The accuracy of PMCT measurements of the clavicle, in comparison to dry bone osteological measurements, for the determination of age-at-death in the developing human skeleton.
- The accuracy of a PMCT dental examination, compared with traditional orthopantomographic (OPT) examination, for the determination of age-at-death of the developing human skeleton.
- The repeatability and reproducibility of PMCT techniques between the same observer and between different observers, of various professional backgrounds and experiences.

The knowledge gained in part one of this thesis will then be validated by ‘blind’ testing against a known population. The methods and protocols developed in chapters 4-6 will be applied to a sample of forensic autopsy cases with known demographics that have undergone PMCT, which the second part of this thesis will consider:

- The actual implementation of PMCT into forensic anthropological casework, including: a system of best practice for image acquisition; the information required to conduct a comprehensive analysis; the best format to present this information so that the resulting ‘PMCT biological profiling form’ is internationally translational for juvenile remains.
- The application of the PMCT biological profiling forms to adult remains.
- Future work and scope for further development of this research.

3.2 Materials

A selection of specimens from the Scheuer Collection, with no known pathologies or damage, aged from *in-utero* to approximately 18 years, will be used. These were scanned using a mobile PMCT scanner and the images were analysed using both Voxar 3D workstation (Toshiba Medical Visualization Systems Europe, Ltd) and OsiriX 3D imaging software (version 3.7.1; distributed freely as open-source software under the GNU licensing scheme at the following Website: http://homepage.mac.com/rossetantoine/osirix. Pixie: Switzerland).

A series of forensic cases undergoing PMCT will also be used. The only restriction placed on the selection of these cases was that they were aged between 0-18 years. These remains had various levels of trauma and were scanned at Leicester Royal Infirmary, in line with the East Midlands Forensic Pathology Unit's examination protocol. As a result, no specific scanning restrictions or protocols were enforced for this particular investigation. The data from these cases was analysed using only OsiriX 3D imaging software.

3.2.1 Scheuer collection

The Scheuer Collection is believed to be the only active repository for juvenile skeletal remains held anywhere in the world (Scheuer and Black, 2000). The collection contains the skeletons of over 100 individuals, from archaeological and historical anatomical sources and is held at Dundee University's – Centre of anatomy and human Identification (CAHID). The material offers significant opportunities to address areas of education and research into skeletal development that have largely been ignored in the past due to a paucity of material. The Scheuer Collection is a very eclectic mix of remains but it is irreplaceable and therefore invaluable as a teaching and learning resource.

Only a selection of the Scheuer collection was scanned for this investigation. The skeletons selected where those that had most elements represented i.e. they were the most intact. The Scheuer collection contains hundreds of individual parts of skeletons that were not scanned for the sake of time and value. The collections curator selected the skeletons that were to be scanned, and there were no other exclusion criteria.

The provenance of the material is mixed. Some of the specimens are archaeological, some forensic and others are anatomical specimens. Many of the demographics for these remains are documented but for those where information is unknown, each skeleton has been examined by trained forensic anthropologists and the biological identity of each has been estimated using standard anthropological techniques (Scheuer and Black, 2000). During the curatorial process of the collection, and as a mechanism of internal audit, this approach was also performed on the material of documented demographics to ensure consistency of approach and was found to be highly accurate.

Tables 3.1 and 3.2 outline the known demographics of the specimens from the Scheuer collection used in this investigation.

3.2.2 Ethical considerations

Ethical approval is not required for this study, as the specimens in the Scheuer Collection are all from archaeological or historical sources and consent was received from the appropriate coroner for each of the forensic cases used. Their use for research purposes is therefore not governed by the regulations of the Anatomy Act 1984.

Table 3.1: The demographics of the clavicles used in chapter 5 of this thesis.

CLAVICLES				
Case Number	**Age (months)**	**Sex**	**Side**	**Provenance**
1	132	M	Left	Archaeological
2	132	M	Right	Archaeological
3	0	M	Left	Anatomical
4	0	M	Right	Anatomical
5	84	F	Left	Forensic
6	84	F	Right	Forensic
7	48	M	Left	Forensic
8	48	M	Right	Forensic
9	204	F	Right	Archaeological
10	72	F	Left	Forensic
11	72	F	Right	Forensic
12	84	F	Right	Anatomical
13	168	M	Right	Anatomical
14	204	M	Left	Forensic
15	204	M	Right	Forensic
16	60	M	Left	Anatomical
17	60	M	Right	Anatomical
18	6	F	Left	Anatomical
19	6	F	Left	Anatomical
20	6	M	Right	Anatomical
21	48	F	Left	Forensic
22	48	F	Right	Forensic
23	168	F	Left	Archaeological
24	168	F	Right	Archaeological
25	0	M	Left	Forensic
26	0	M	Right	Forensic
27	0	F	Left	Anatomical
28	0	F	Right	Anatomical
29	204	F	Right	Archaeological
30	144	F	Left	Forensic
31	144	F	Right	Forensic
32	228	M	Left	Archaeological
33	168	F	Left	Archaeological

Table 3.2: The demographics of the mandibles and maxilla used in chapter 6 of this thesis.

DENTITION			
Case Number	**Age (months)**	**Sex**	**Provenance**
1	0.25	F	Anatomical
2	144	F	Archaeological
3	47	M	Anatomical
4	34	F	Anatomical
5	2	M	Forensic
6	16	F	Anatomical
7	108	M	Forensic
8	10	M	Forensic
9	10	F	Anatomical
10	1.75	M	Forensic
11	216	F	Archaeological
12	1.5	M	Anatomical
13	196	F	Archaeological
14	7	F	Forensic
15	120	F	Anatomical
16	60	M	Anatomical
17	72	M	Anatomical
18	7	F	Anatomical
19	120	M	Archaeological
20	60	M	Anatomical
21	72	F	Forensic
22	1.5	M	Forensic
23	24	F	Archaeological
24	60	F	Archaeological
25	1	M	Forensic
26	0.3	F	Forensic
27	5.75	M	Anatomical
28	1	M	Forensic
29	156	M	Archaeological
30	192	F	Archaeological
31	132	F	Archaeological

3.2.3 Data visualisation and analysis

The Digital Imaging and Communication in Medicine (DICOM) images of each case were processed using Voxar 3DTM (Barco, 2005) and OsiriX [http://www.osirix-viewer.com/Snapshots.html] software. Both 3D workstations are advanced visualisation and analysis tools that offer a variety of different reconstruction techniques for visualising 3D image data, including surface rendering, volume rendering and multi-planar reformations (MPRs). These software packages also provide advanced measurement and quantification tools, which allows anthropological measurements to be simulated quickly and easily.

Voxar 3DTM is a proprietary clinical software system for image analysis. It can be used to manage the large number of images from a PMCT scan, MRI scan, or a PET scan. Voxar 3DTM offers a full suite of advanced visualisation and analysis tools, designed to optimise productivity and produce good-quality multimedia reports. It is an efficient workstation to process large volumetric data sets, at any thickness with a consistently high image quality, speed and proficiency.

Voxar was also integrated with the picture archiving and communication system (PACS) at Leicester Royal Infirmary (LRI) and the concurrent license program gave multiple users access to “floating” licenses within the LRI, where Voxar was already installed. Access to more advanced program options was available, although these were also restricted to a workstation within the institute.

OsiriX is a similar multi-dimensional image navigation and display workstation, with a source code available to be freely downloaded and distributed under the GNU General Public License. OsiriX, represents the transition to a completely new platform with the added ‘X’ indicating the migration to the new Macintosh operating system version 10, also called MacOS X. The X also indicates the compatibility with underlying Unix platform and the adoption of the open-source paradigm. OsiriX is a very useful system with a simple, clever interface and synchronization features, which work bilaterally between the imaging center and its users.

Unlike Voxar 3DTM, OsiriX is an open source software application. Developed by Antoine Rosset, a radiologist and software developer, it has been designed specifically for the needs of advanced imaging modalities. The software program turns an Apple Macintosh into a DICOM PACS workstation for medical imaging and image processing. Other open source DICOM viewers are available for use on PCs and Macintosh computers.

The OsiriX program offers all the basic image manipulation functions of zoom, pan, intensity adjustment and filtering with real-time performance. Additional functions, such as; multi-planar projections, convolution filters, variable slice thickness adjustments, volume rendering, minimum and maximum intensity projections, and surface rendering, are also accessible in quasi-real-time (depending on the hardware used, as well as the number of slices to reconstruct). One of the key features of the OsiriX software is its flexible user interface allowing users to customise the program by adding and removing tools and functions from the toolbar and menus of the program. This means there is the potential to produce “customised” versions of the program for specific applications – so that the software can be adapted in the long term to anthropologists, radiologists, pathologists, odontologists etc., that may not be computer experts.

The integrity and quality assurance of open software developed by a community of users does not follow the traditional conformance and certification required for commercial medical software programs. However, being an open source, OsiriX has been developed by its users, whose innovative solutions to imaging problems have resulted in a program that is better suited for specific tasks. Open development has ultimately resulted in a program that is tailored to specific needs and clinical assignments of its users, which was beneficial to this project due to the unique measurement requirements. OsiriX now has a medically certified version, which is available to purchase but does not add any extra measurement precision or function relevant to this study. However, a 64-

bit upgrade to the OsiriX software was used in this project, to aid the manipulation of large data files that was not a ‘free’ open source upgrade.

For this project OsiriX was an attractive and cost-effective solution, facilitating information sharing between radiologists and forensic professionals on a global scale, including those based within institutes that could not afford high-priced workstations such as Voxar 3D. The availability of OsiriX also enables communication between *in-situ* and remote forensic practitioners by allowing them to share the same convenient platform for image display and navigation. Furthermore, I believe that the wider availability of OsiriX enables quality control and a minimum standard of reporting to be implemented and monitored. However other image analysis systems are available and could be equally as effective.

3.2.4 Computed tomography scanner

The portion of the Scheuer juvenile skeletal collection used in this investigation, was scanned using a truck mounted SOMATOM® Emotion 16-detector CT scanner (Siemens AG Medical Solutions). Scans involved helical acquisition using a 0.75mm slice thickness, 120kVp, and 100mA with bone and soft tissue reconstructions at 1.25mm (Appendix 1). Data were stored on compact disc and transferred to the secure image archive at East Midlands Forensic Pathology Unit.

The PMCT scans of the forensic cases used in this investigation were undertaken using the standard post-mortem protocol used at Leicester Royal Infirmary using a Toshiba Aquilion 64 slice scanner (120 kVp, 300 mA and 64×0.5-mm slice thickness, matrix 512×512) reconstructed to either 1 or 2-mm thick slices.

Table 3.3 gives more detailed information about the exact scanning parameters used, which are the basis of the one used in the Royal Collage of Pathologists and Royal Collage of Radiologist (RCPath/RCR) national guidance. For babies, two head scans would have been taken and then using the CAP (chest/abdo/pelvis) protocol, the rest of the body would have been scanned in one. Older children would have been scanned, as detailed in Table 3.3, depending on individual case requirements.

3.5 Summary

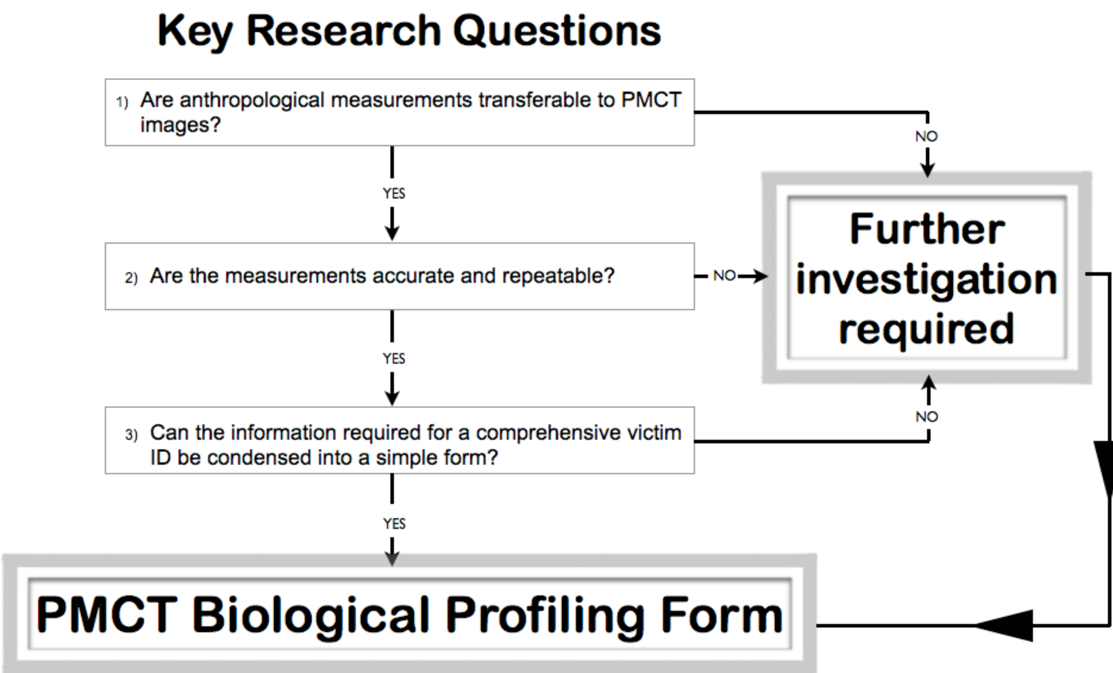


Figure 3.1: Summary of research project.

Table 3.3: Scan and reconstruction parameters.

	1. Brain Angled to base of skull	**2. H & N** Straight tube down to T2	**3. CAP** Above shoulders to below symph	**4. Lower limb** ASIS to below feet	**5. Targeted Angiography** Carina to diaphragm	**6. Whole body Angiography** (fumedica) Top of head to prox femur	**7. Ventilation** Scan Above shoulders to Symph
kV	120	120	120	120	120	120	120
mA	300	300	300	300	R***	300	300
Rotation Time	0.75	0.75	0.5	0.5	0.5	0.5	0.5
Range	250	250	600	850	600	600	600
FOV	220-S	320-M	400-L	400-L	400-L	LL	400-L
Thickness	0.5x32	0.5x32	0.5x64	0.5x64	0.5x64	0.5x64	0.5x64
Recon-thickness Interval	1	1	1	2	0.5		1
Pitch factor	0.8	0.8	0.8	1.6	0.3	See below	0.8
	0.656	0.656	0.828	0.641	0.641	0.825	0.828
Helical pitch	21.0	21.0	53	41	53	53	53
Standard deviation	Sure exp 3D - off	Sure exp 3D - off	12.5	12.5	CTPA-11.5	Sure exp 3D - off	12.5
Recon window	Brain	Body standard Bone sharp	Body standard Bone sharp	Body standard Bone sharp	CTA body	Body standard Axial – 1 on 1 Volume – 1 on 0.8 For 3rd run – Vol 2 – 0.5 on 0.3 CTA	Body standard Bone sharp
Transfer	PACS Volume only	PACS Volume only	PACS Volume only	PACS Axial & Volume	PACS Axial & Volume	PACS Axial & Volume	PACS Volume only

CHAPTER 4: PRELIMINARY WORK

"As those who study them have come to learn, bones make good witnesses - although they speak softly, they never lie and they never forget."

- Dr Clyde Snow, Forensic Anthropologist.

4.1 Introduction

4.1.1 Juvenile age determination

Age at death is one of the four main features of biological identity, along with race, sex and stature (Scheuer, 2002). There is a considerable range of well-established techniques that have been presented and reviewed in the literature for age determination of juvenile remains. Each of these techniques has limitations and may not be applicable to all cases. At present there is no standardised approach to age estimation in forensic cases and there is no consensus concerning which method is the most appropriate and practical (Scheuer and Black, 2000). Juvenile age determination is typically only discussed perfunctorily in general forensic anthropology textbooks (Byres, 2008; Cattaneo, 2009). However, in 1987 Ubelaker presented a literature review and recommendations for estimating juvenile age at death, which has since been updated (Scheming, 2006; Koenigsberg, 2008; Cunha, 2009). Contributions by Cunha and colleagues (Cunha, 2009) provided tables of recommended techniques for specific age groups including foetuses, newborns, infants and children, adolescents and transitory groups. This general overview is a useful summary for both students and practicing anthropologists.

The accurate age determination of juveniles is dependent upon the preservation and availability of skeletal elements (Scheuer, 2002). The most reliable methods for children utilise morphological and developmental changes to the hard tissues of bone and teeth (Byres, 2008). These changes correlate with three distinct phases of an individual’s lifespan; growth and development, equilibrium and senescence (Scheuer, 2002).

In the juvenile age range, before cessation of growth in height; ossification, fusion and eruption events have a somewhat predictable rate of progression and a well-documented pattern of development. As such, age may be estimated relatively accurately, within narrow ranges, from a multitude of skeletal markers (Scheuer, 2002). However, the relationship between skeletal and chronological age is neither constant nor linear, and as a result age estimations are founded on probability (Schmitt *et al*., 2002).

Dental age estimations predominantly use the degree of mineralisation or eruption of the teeth, whereas skeletal age estimations are established from the developmental stage of primary and secondary ossification centers, or the size and diaphyseal length of the long bones. These characteristics have a reasonably predictable rate of progression and a well-documented pattern of development. As such, age may be estimated relatively accurately, from a multiplicity of markers (Scheuer, 2002). It is frequently observed that dental age estimates are closer to actual chronological age than those of skeletal age as they are less affected by environmental insults that can delay maturational processes (Scheuer and Black, 2000). This is probably because the development of all the deciduous dentition and part of the permanent dentition takes place before birth in a relatively protected environment, whereas skeletal growth and development, although having a strong genetic basis, are influenced for a longer time by external factors such as nutrition and disease burden (Mincer, 1993; Maber, 2006).

4.1.2. Traditional anthropological methods

For adults, preliminary determination of age-at-death, racial/ethnic affiliation, living stature and sex are the first steps towards arriving at a positive identification. When dealing with juvenile remains age-at-death is the only factor that can be reliably determined and is therefore the most vital criteria for identification purposes (Scheuer and Black, 2000). Sex, stature, and race/ethnicity cannot be estimated with any degree of reliability from the skeleton until post puberty. Therefore age estimation is of primary importance. Research dating back to 1939 (Maresh, 1939) demonstrates that the relationship between growth and maturity is strong but not linear, displaying periods of both acceleration and stasis. As such, the timing of growth spurts must be considered in any attempt to evaluate the relationship between skeletal (or biological) and chronological age. Children of the same chronological age can have markedly different biological ages and because of this, assessment of age remains something that, while more accurate in juveniles than adults, still has a significant error range. For example, females exhibit advanced skeletal maturation compared to males and therefore it is beneficial to investigations if the sex of the individual is known, but this is not always possible.

Although there are numerous advantages of using dental techniques to identify remains, peri-mortem or post-mortem damage, or complete absence of dentition from the skeletal material can limit the number of applicable methods. When dentition is not present, the age of juvenile remains can be determined from the size and length of the long bones. This method correlates the length of the diaphyseal shaft with a chronological age. This produces a growth curve, or skeletal growth profile, to show the progression of age for a population to complete epiphyseal fusion (Hoppa, 1992). Until birth, the relationship between long bone length and age is relatively linear. Divergence in size, related to race and sex accelerates postnatally and therefore long bone length to determine sub-adult age is only reasonably accurate until approximately 10 years. After this age, particularly if the unidentified remains are from an unknown population, this method becomes unreliable - especially since techniques for determining sub-adult race affiliation are inconclusive. Arguably the most useful data for determining age at death using long bone length was the X-ray standards produced by Hoffman (Hoffman, 1979) and by Maresh *et al*. (1939). However, secular trends (continual, non-periodic change over time) would suggest that this method is perhaps outdated and may lead to discrepancies between Hoffman's standards and juvenile skeletal remains of the twenty-first century.

4.1.3. Virtual anthropology

'Virtual anthropology,' is the investigation of human material, using digital data sets (images), on a computer (Weber *et al*., 2001). This includes the measurement of bones on virtual representations, using traditional anthropological methods. The specific use of imaging methods for identifying human remains is widely recognised and well documented (Hines *et al*., 2006). However, while much literature exists on the application of morphological methods on virtual objects, there is presently a lack of research concerning the accuracy and repeatability of metric analyses using such novel methods. It is therefore important to address this issue, as this new and innovative approach to anthropological examination may provide the key to the challenges faced by mass disaster incidents, particularly if chemical, biological, radiological or nuclear material (CBRN) is involved.

Morphological and metric analyses of so-called ‘virtual skeletons’ relies on the acquisition of image data that can be assessed in three-dimensions, this can be either surface or preferably volume data. Although several different imaging modalities exist for this purpose, the majority of these have a very limited value for analysing human remains. However, as introduced in Chapter 1, CT has demonstrated superiority in this regard, and is generally considered the most practical imaging technique for post-mortem examination (Rutty *et al*., 2007a; Rutty *et al*., 2007b).

The basic principles of PMCT permit the acquisition of ‘volume’ data, allowing both external and internal structures to be visualised in a three-dimensional form (Seeram, 2001). Possibly the most notable and unique quality of CT is its capacity to display information from a wide variety of tissues with differing densities, and with minimal superimposition of structures (Hildebolt *et al.*, 1990). Furthermore, a body can be scanned in a matter of seconds, although other factors such as image reconstruction may prolong this time to approximately 15 minutes per case (Rutty *et al*., 2007a). This makes it particularly useful in a mass fatality incident, as the amount of time available for each case may be limited. Furthermore, with the development of mobile CT, scanning can take place almost anywhere, including the scene of the incident itself. As the scanner and operational suite are mounted within a heavy goods vehicle it can be transported to anywhere with road access, and can even be modified for airlifting to more isolated locations. Most of the current mobile CT scanners are also equipped with telecommunications facilities for the transmission of data to remote locations. However, this process can be time-consuming depending on the size of the data files; therefore a rapid, secure link is ideal (Rutty *et al*., 2007b).

Finally, one of the most noteworthy advantages of PMCT is its capacity to screen whole bodies or multiple fragments within a sealed body bag. This helps prevent any further risk of contamination from CBRN agents, and also reduces the chance of commingling if the remains are fragmented. From the evidence given thus far, there is a good argument to suggest that mobile PMCT and tele-radiology will have a fundamental role in the future of disaster victim identification. However, it is first necessary to validate the accuracy and precision of osteometric measurements acquired using PMCT.

From a medico-legal perspective the development of “virtual skeleton” techniques would have a number of advantages. If for example, a decomposed body requires anthropological age estimation, time-consuming maceration of the bones can be avoided. The virtual skeleton can be repeatedly “handled” without any damage to the original sample. They can also be virtually cut and restored, zoomed and reproduced and the images can be stored as files for further study in the future, if required.

The biggest limitation of the virtual approach is, of course, the limited access to CT machines, at present. In spite of the worldwide use of forensic radiology and the implementation of PMCT in some forensic institutes, the majority of forensic investigation facilities do not have access to MRI or CT equipment.

This chapter considers the possibility of a ‘virtual anthropological examination’ by reviewing the juvenile age determination methods most regularly used by anthropologists at present, and considering how they could be applied or adapted to PMCT data. Using PMCT should prove to be quicker, cheaper and culturally more acceptable in most situations than the more traditional anthropological methods, particularly when processing the remains of deceased children.

4.2 Investigation Summary

Previous studies have provided evidence to support the implementation of PMCT into routine adult forensic investigations. However, none of these studies extend to juvenile age determination, where the techniques used vary from those employed for adult remains. Juvenile age techniques utilise the appearance and fusion of ossification sites and the diaphyseal length of long bones, in addition to dental development, which are not always applicable to adult remains. There is currently no published evidence to validate whether osteological measurements required for juvenile age determination are reproducible on PMCT images, or indeed how accurate this 'virtual approach' might prove to be.

In addition, none of the previous investigations have established a 'standardised' protocol for a virtual anthropological technique, which is a necessary step before this method can be internationally adopted for disaster victim identification or nationally accepted as a forensic standard. Previous reports in the area of anthropological computed tomography have been 'case-report' oriented and typically based on qualitative techniques that compare morphological traits as opposed to quantitative metric anthropological techniques. Although morphological approaches are commonly used (and often preferred) by forensic anthropologists, they rely heavily on subjective opinion and experience and are therefore not as judicially sound as metric methods - that are more objective and therefore facilitate greater inter-operator reliability testing. Standardising image processing and measurements are an extremely important step for the development and acceptance of this technique.

Using the review provided by Cunha *et al*. (2009) as a guide for age determination of juvenile remains, as summarised in Table 4.1, each of the suggested methods were considered as to whether PMCT acquired images can provide equivalent anthropological assessment. To coherently address this question, images acquired from a sample (n=40) of the Scheuer juvenile skeletal collection, housed at the University of Dundee (Appendix 1), which were scanned using a truck mounted SOMATOM® Emotion 16-detector CT scanner (Siemens AG Medical Solutions) were used as an investigation aid. Some of the skeletons in the collection were incomplete or damaged. Therefore, in some instances a method was reviewed on a slightly smaller sample (n~40).

Scans involved helical acquisition using a 0.75mm slice thickness, 120kVp, and 100mA, with bone and soft tissue reconstructions at 1.25mm. Data was stored on compact discs and transferred to the image archive at East Midlands Forensic Pathology Unit. Multi-plane reconstructions (MPR) and 3D volume rendered images were then created for analysis using a comprehensive image software programme (OsiriX version 3.7.1; distributed freely as open-source software under the GNU licensing scheme at the following Website: http://homepage.mac.com/rossetantoine/osirix.

Table 4.1: Recommended age estimation methods (adapted from Cunha *et al*., 2009).

Age Group	Recommended approach	Reproducible by PMCT
Foetuses	Dental Development Presence of ossification nuclei Long bone development	Yes Yes Yes
Newborns	Dental Development Diaphyseal length of long bones Presence of ossification nuclei *Also check mineralisation of cusp of first permanent molar and ossification of femoral distal epiphysis*	Yes Yes Yes
Infants and Children (0-7years)	Dental Development Diaphyseal length of long bones	Yes Yes
Juvenile and Adolescent (8-15years)	Dental Development Long bone development Maturation of hand and wrist	Yes Yes Yes
Transition and adult (16-25years)	Third molar development Fusion of spheno-occipital synchondrosis, iliac crest and vertebral ring. Maturation of hand and wrist Fusion of medial end of clavicle	Yes Yes Yes Yes

4.3 Dental Techniques

4.3.1 Dental development

From the early work of Logan and Kronfeld (1933), Schour and Massler (1937) produced an atlas of dental development that is arguably the most regularly cited in dental and forensic literature because of its regular application in routine forensic practice, and is still widely used today. By matching radiographs of the (preferably entire) maxilla and mandible, to diagrams depicting the expected stage of dental development at each year in a child’s life, an estimated age can be assigned in accordance with the associated diagram.

In order to substitute PMCT into this method, the images produced need to be of comparable quality to the radiographic images (Oral Pan Tomogram or OPT) that are traditionally used. Using PMCT scans of the study sample; curved MPR reconstructions can be created to replicate dental OPTs. This was achieved using a built-in dental platform on the basic OsiriX imaging system, which had been specifically designed to recreate dental images.

By using medical imaging software programs such as OsiriX the mandible and maxilla can be imaged in three planes: axial, panoramic and cross-sectional, with no super-imposition of other osseous structures, unlike standard orthopantomography. The image quality is therefore sharper and clearer, providing better tissue contrast resolution.

The data is displayed in this programme as four views (Figure 4.1, A-C). The first three views show an orthogonal MPR plane in relation to the other views. The curved MPRs are then quick and easy to create by positioning multiple markers on an axial 2D set of images (Figure 4.1, C), which could be adjusted into the correct plane to define the curvature of the jaw and the total area of interest. This plane is defined as 3D Bezier path, which is displayed in lower right view. This curved MPR plane is then automatically processed to produce, by *straightening* or *stretching* this path (Figure 4.1, D), the resulting image - which is a straight representation of the curved jaw. Similar to the panoramic OPT, where the jaws are rendered flat by having the film and beam simultaneously rotating around the head during the exposure, the curved MPR created by OsiriX shows the entire U-shaped maxilla and mandible flattened out on one image.

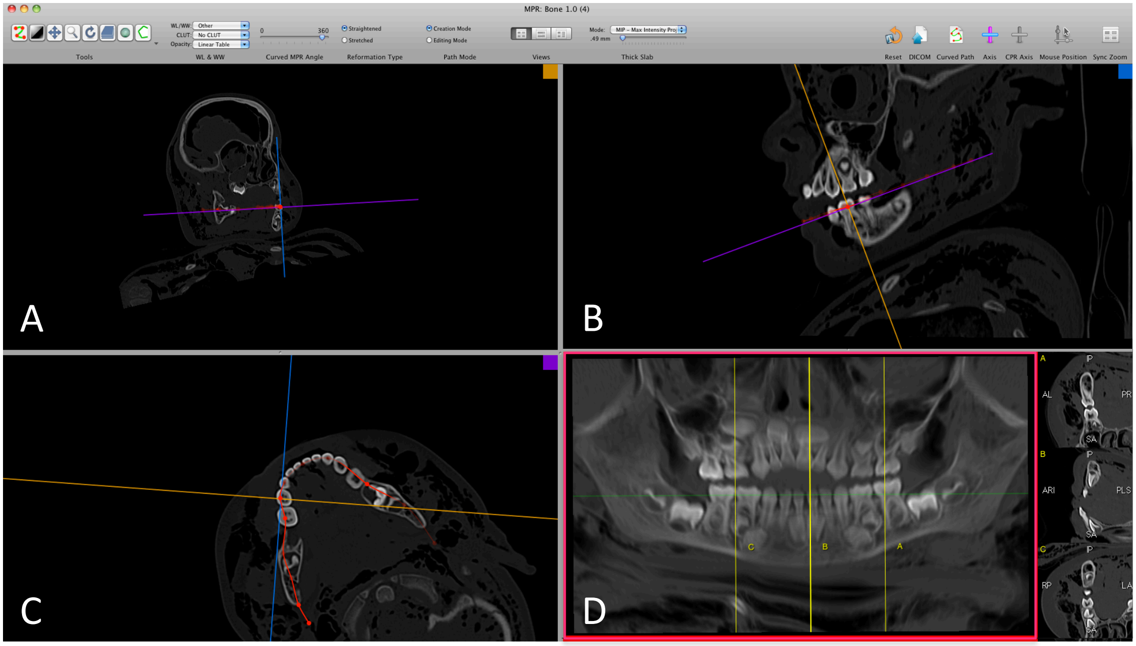


Figure 4.1: 3D Curved MPR viewer window. A-C) Orthogonal MPRs in the x-, y- and z- planes. C) Curved path following the arch of the dentition, using the *curved path* command from the toolbar. D) 3D Bezier path view.

The three views on the right side of the curved MPR (Figure 4.1, D) correspond to three perpendicular views along the 3D Bezier path. These three views are strictly perpendicular to the Bezier path, but are not necessarily parallel to each other's, if the 3D Bezier path is not in a straight line. This function is particularly useful when a more thorough inspection of a particular tooth is required. If there are metal artefacts for example, the surrounding teeth may be obscured-and so the perpendicular views allow the tooth to be viewed individually with less surrounding 'noise'.

The curved MPR views and the corresponding perpendicular views can then be exported as new DICOM images in the database. The outcome of this process is an image that closely resembled traditional OPTs, with the added benefit of containing volume data. The direct comparisons of OPT and curved MPR images in Figure 4.2, clearly illustrates that a sufficient amount of detail is achieved using PMCT to carry out this technique successfully.

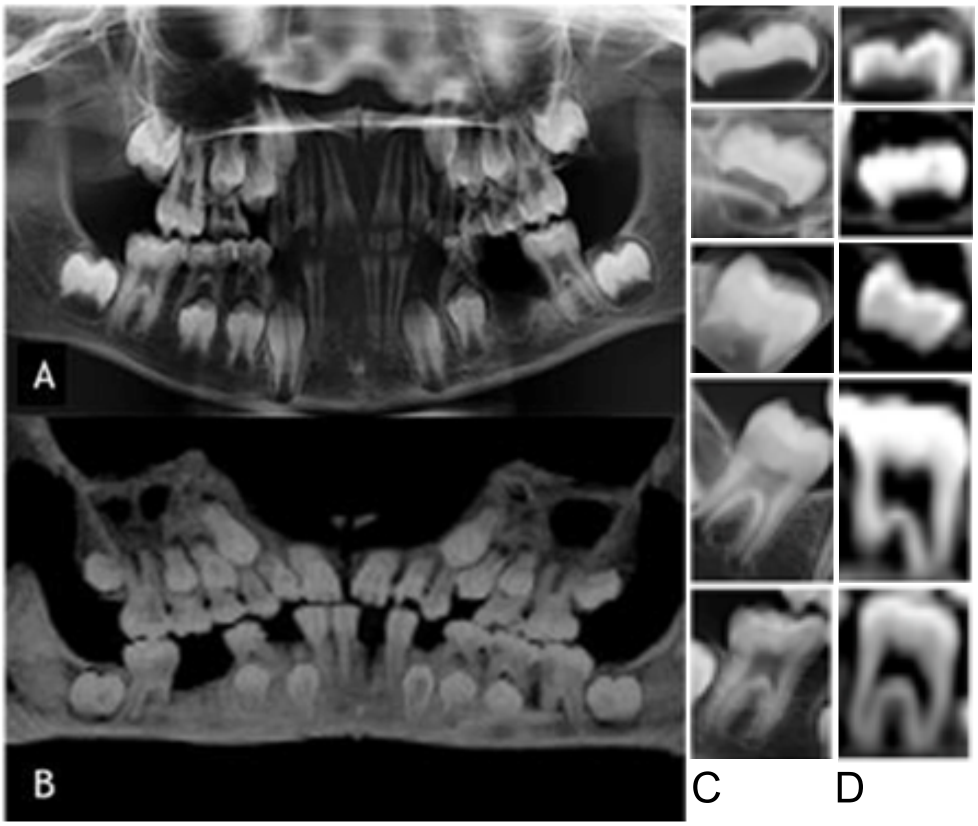


Figure 4.2: Orthopantomograph and PMCT curved MPRs. A) Radiographic images traditionally used in Schour and Massler (1937) dental method. B) OsiriX PMCT comparison image C) Radiographic images traditionally used for Demirjian *et al*. (1976) system for dental age assessment. D) OsiriX PMCT comparison images.

The PMCT dental reconstructions could be used in exactly the same manner as traditional OPTs for age determination, using the atlases of dental development as previously discussed. Furthermore, a number of additional advantages suggest that PMCT could arguably be superior over standard radiographic techniques. These include the ability to: define soft tissues as well as bone, which would theoretically allow stages of clinical emergence to be more accurately assessed (Scheuer and Black, 2000); produce 3D representations of the jaw, so that any distinguishing features can be accurately recorded, which may subsequently help with victim identification; and produce multiple 2D images, which could be rapidly scrolled through to examine every aspect of the dentition, in any plane. By default the curved MPR view is rendered as a thin slice with a thickness identical to the original dataset. However, this thickness can be increased (using the toolbar *Thick Slab* item) so that the virtual OPT can also be viewed as one thick slice, producing an image more similar to a traditional OPT. The image stack could also be exported as a movie file (such as, a QuickTime video) so that they could be viewed as a virtual fly through, completely eliminating issues related to superimposition.

Conversely, OPTs are single 2D images. Care has to be taken to position the jaws in the correct orientation to maximize image quality and minimize superimposition of neighboring structures. Since the dentition varies slightly between individuals, a trained dental practitioner is required to position the head correctly in order to obtain a good quality OPT and ensure the images are taken to the correct standards. In a disaster scenario when time and money are of particular concern this can be a major drawback. Although a trained practitioner is also required by law (IRR, 1999) to operate PMCT equipment, curved MPRs do not require the body to be manually manipulated into a specific position and can subsequently be extracted rapidly from a full body scan, regardless of the position or condition of the remains, in a matter of minutes using a software program such as OsiriX. This being said, it is important to acknowledge that some time does need to be taken by the radiologist producing the PMCT to ensure scanning parameters such as, slice thickness, reconstruction algorithm, and extended CT scale are calibrated correctly to optimize image quality and mitigate for metal artifacts etc. However, a skilled radiographer should be able to do this routinely based on experience.

When processing juvenile remains, it is therefore evident that PMCT cannot only replicate traditional OPTs but also offers numerous additional benefits. However, it important to note that when dealing with adult remains, image artifacts produced from dental amalgam in fillings may limit PMCT's evidentiary value. Although other metal work causes fewer problems and this is clearly less common in younger age groups, it is an important consideration in individuals with extensive dental restoration.

Scoring systems such as those by Demirjian *et al*. (1973) for the permanent dentition and by Mincer *et al*. (1993) for third molar development are also widely used in forensic investigations and are similarly reproducible using PMCT data. Supplementing the 2D "radiograph like" images with 3D representations, which allow each tooth to be individually studied in all planes to accurately score its developmental stage, may enhance these techniques. Adding to the benefits afforded by PMCT. Different reconstruction windows can be used on a case-by-case basis, to maximize the level of detail as illustrated in Figure 4.3.

Figure 4.3: 3D representation of the dentition. Images created using the 3D volume rendering function of OsiriX.

4.4 Long Bone Techniques

4.4.1 Presence of ossification nuclei

Age evaluation of a foetus places a significant emphasis on the location and number of ossification nuclei present (Kosa, 1989). These appear in a relatively well-defined sequence and knowledge of the centres present compared with those yet to form, allows the investigator to assign a possible age range to the individual. The utility of dry bone analysis of ossification centres is variable, depending on the age of the individual and the skeletal element being examined. This is because ossification centres are often initially small, non-specific nodules and are therefore unlikely to be recovered or be of particular significance until they have developed into a recognisable morphology.

Theoretically, it is possible to estimate the age of a juvenile skeleton from three key features of primary or secondary ossification centers; 1) the time at which the centre appears; 2) by its size and morphology; 3) the fusion time of primary to secondary centers (Scheuer and Black, 2000). The deposition of bone at these centers follows a rough sequence, such that their stage of development indicates an approximate age at death of an individual. Unfortunately, these centers are extremely small and fragile, making their recovery unlikely and their utility in forensic investigations minimal.

Clinically, images of these sites are produced by ultrasound, so it does not pose a radiation risk to the unborn foetus. In forensic cases where this is not an issue, radiographic imaging is generally used. However, radiography has significant limitations in this regard as the appearance of ossification centres precedes radiographic visualisation, which depends on calcification (mineralisation). In contrast, ultrasound can visualise ossification sites before ossification, while they are still cartilaginous. PMCT is able to replicate images produced by radiography but with increased contrast resolution of non-ossified structures, albeit at lower spatial resolution (Figure 4.4, A-B) and for this reason could easily be a substitute option in these techniques. In practice the lower spatial resolution is only significant for detecting subtle fracture such as metaphyseal fractures, which are better detected by a plain radiograph (Jarraya *et al*., 2013). Furthermore, using the 3D reconstruction mode of PMCT, the centres can be viewed in the x-, y- and z-planes, offering supplementary information about the structure and morphology of the centres (Figure 4.4, C-F).

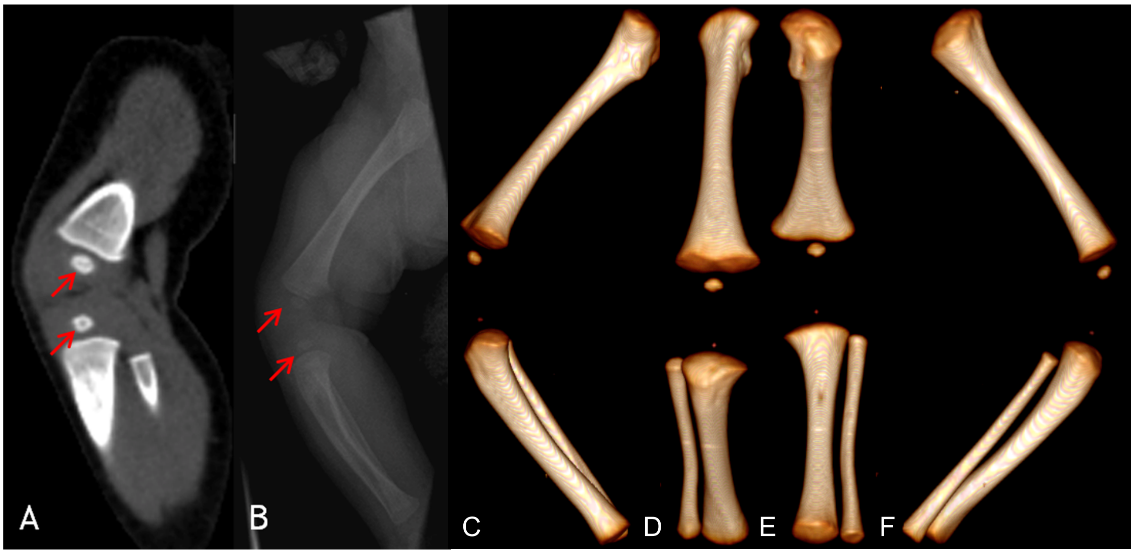


Figure 4.4: Presence of ossification nuclei; PMCT reconstructions of the lower limb. A) PMCT reconstruction; ossification sites can be seen clearly. B) X-ray image; ossification centres are visible, but not as clear as on PMCT. C-F) Lateral, anterior and posterior 3D views; ossification centres are well defined and clearly delineated.

In a forensic investigation, fluoroscopy and surface scanning would generally be conducted on specific regions of interest. It is unlikely that the entire body would undergo a full radiographic examination for the purpose of age estimation. This being said, the appearance and fusion of epiphyseal centres offers a quick and reliable method of age estimations and therefore if a PMCT were being conducted for another primary reason, this secondary information would be extremely useful.

4.4.2 Length and development

As ossification progresses, bones become amenable to recording length, width and morphology, which can be used to assess the age and maturity of an individual. As above, these developing secondary centers of ossification can also be accurately assessed using PMCT.

Common models of longitudinal growth changes (Smith *et al.*, 2005; Smith *et al.*, 2004) of the humerus, radius, femur and tibia, measure all diaphyseal (shaft) long bone lengths parallel to the bone's long axis, from the most proximal to the most distal extremity. Although transverse expansion of the diaphysis also occurs; to maintain proportions within a bone, constant transverse re-modeling occurs, as the bone expands longitudinally. Thus diaphyseal length is used to estimate age, as it is the only dimension that has a linear relationship with growth over time. This measurement can be replicated on PMCT data using built in measurement tools, on software systems such as OsiriX. These measurement tools can be used to generate virtual osteometric boards or calipers. Although many measurement tools have been developed for osteological methods, the most frequently used are sliding calipers and the osteometric board (Figure 4.5). Sliding calipers have a pair of jaws whose variable gape is measured via a scale or dial on the caliper shaft. An osteometric board is used for bones, such as the humerus or femur, which are simply too long to be measured using calipers.

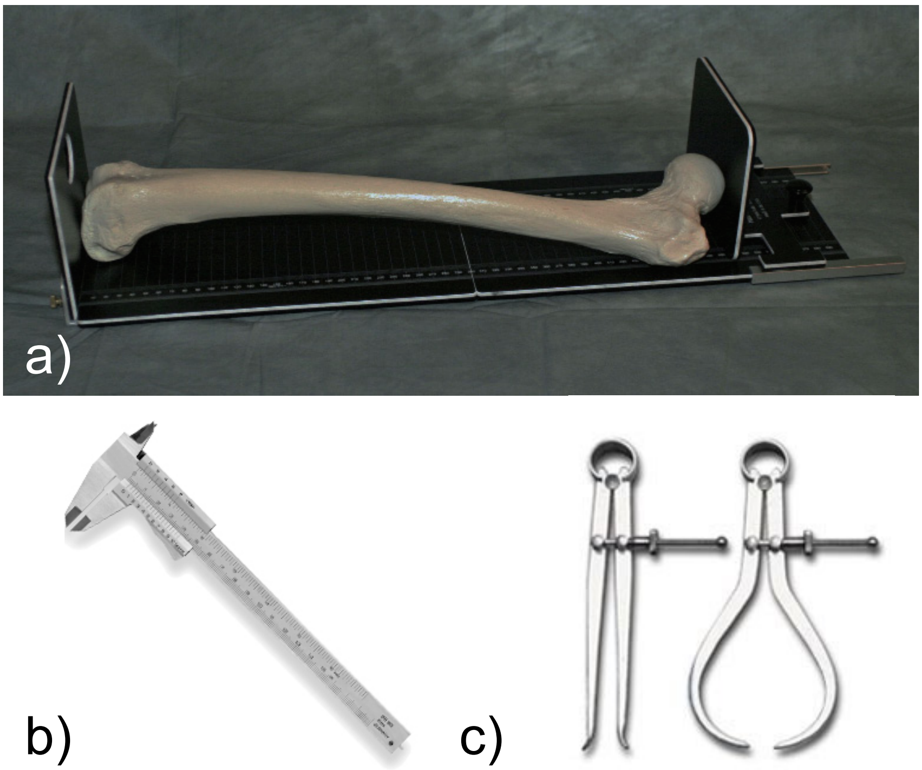


Figure 4.5: Osteometry instruments. (a) Osteometric board (b) sliding calliper (c) spreading calliper (Byres, 2008).

By viewing the PMCT data of a bone in the x-, y-, and z- planes, the most extreme points of each long bone can be determined. The distance between two parallel planes drawn at these points will be the diaphyseal length. This measurement can be attained on both MPR and 3D reconstructions, depending on the specific requirements of the user (Figure 4.6). 3D reconstructions benefit from the ability to "cut away" surrounding bones and soft tissues using different reconstruction windows and bone removal tools. However, measurements using the 3D volume rendering function of OsiriX can be less accurate. Unfortunately, on this setting the 'virtual planes' cannot be fixed to the reconstructed bone by propagating them through the entire series. This can make it difficult to ensure that the true diaphyseal length is being measured, since the measurement planes disappear when the image is rotated to check. However, from experience, measurements generally varied by only a few millimeters compared with those taken using the MPR setting. Therefore, the accuracy of most anthropological techniques would not be significantly affected if the 3D method were chosen.

MPRs more closely replicate traditional X-ray techniques, so may be preferred in some instances; if for example, a direct comparison between ante-mortem and post-mortem images was required. Using this method, measurement planes can be fixed to the image and propagated through the entire data series. The accuracy of the measurement can then be confirmed by scrolling through the contiguous MPRs. If PMCT were used as a substitute for traditional dry bone examination by this approach, it would remove the necessity to deflesh bodies. Subsequently, there would be significant practical and aesthetic benefits as well as being considered more socially acceptable. In addition, Robinson *et al*. (2008) have shown that there is no significant difference between longitudinal diaphyseal measurements made by PMCT and those made by traditional osteometry. If this were correct for all long bones, as with the dental techniques discussed previously, it would suggest that traditional techniques could be successfully applied to PMCT data.

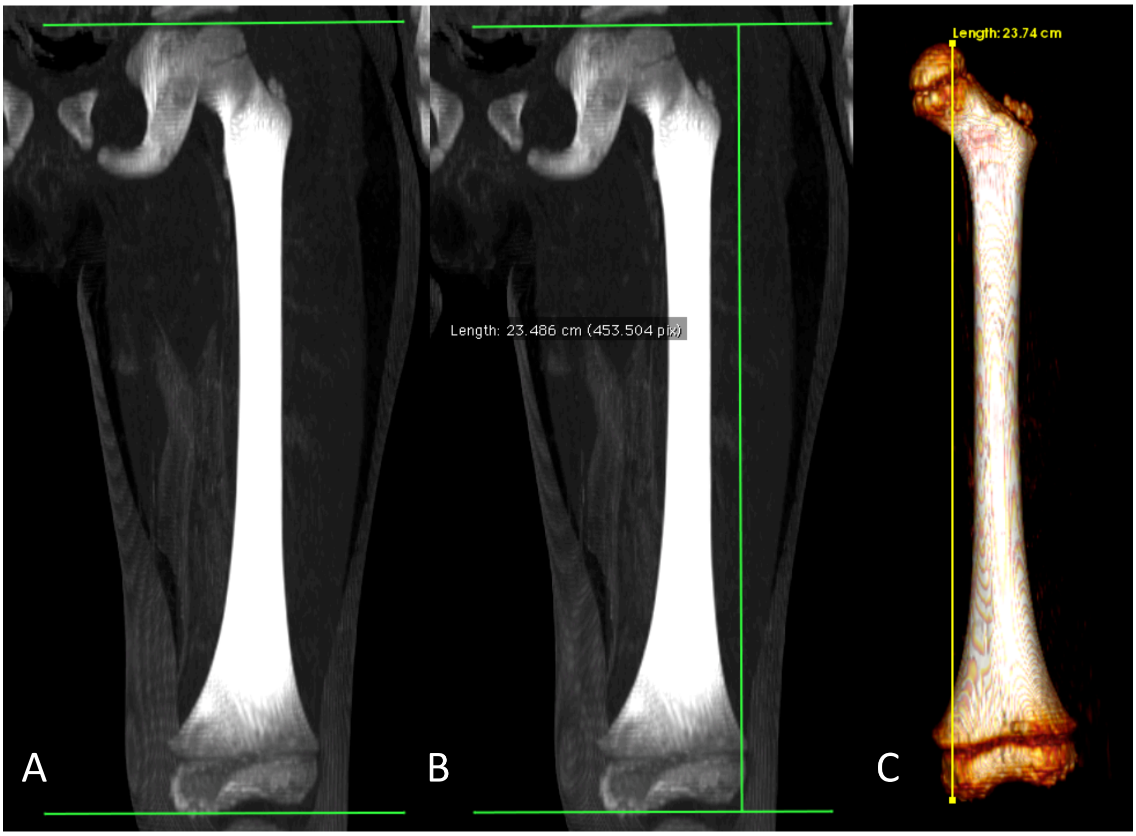


Figure 4.6: Long bone maximum diaphyseal length measurements; MPRs and 3D reconstructions. A, B) MPR long bone measurements. Parallel plates should be drawn at the most extreme point of the bone; the distance between these points gives the maximum diaphyseal length of the bone. C) 3D reconstruction measurement. End plates cannot be used, as only one length measurement can be made at a time.

4.5 Miscellaneous Techniques

4.5.1 Maturation of hand and wrist

The hand-wrist atlas of Greulich and Pyle (1959) and the Tanner-Whitehouse (1975) scoring system both use radiographic images, which can be produced using PMCT data. For these methods it is important that the radiographs are taken in the appropriate plane of view for comparison with the standard image so that all points of interest (ossification centers) can be viewed simultaneously. Although straightforward in clinical practice this can be difficult for post-mortem assessment due to rigor or contractures. PMCT volume rendered images can be sectioned in any plane and rotated in space to better conceptualize the underlying anatomy and therefore could create an appropriate image regardless of the body position during scanning (Figure 4.7). In addition Aaron *et al*. (1992) have shown no significant loss in accuracy when using CT instead of radiographs for the Tanner-Whitehouse system.

Similar issues apply to assessment of fusion of spheno-occipital synchondrosis, iliac crest and vertebral ring and fusion of the medial end of the clavicle. These morphological changes, as with the hand and wrist, should also be accurately assessable by using PMCT.

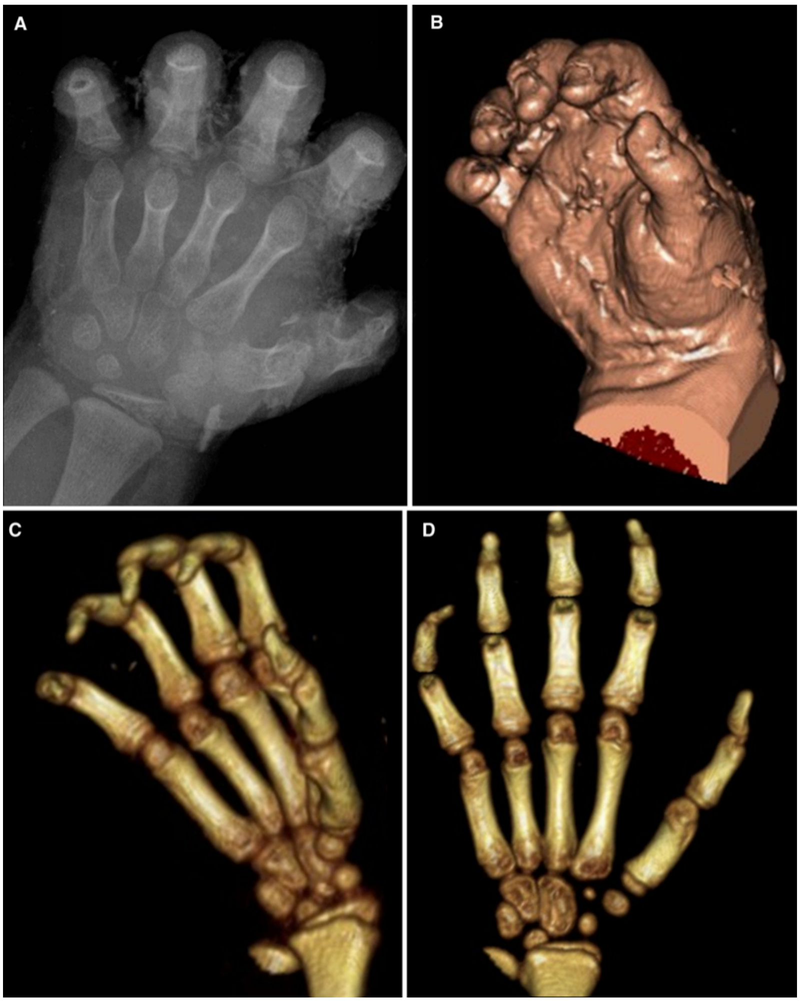


Figure 4.7: Maturation of hand and wrist. A) Radiograph B) Virtual representation of hand from CT C) 3D reconstruction of the hand that can be view in any plane D) Manipulation of 3D reconstruction to open out hand.

4.6 Summary and Discussion

Radiological imaging modalities have been used in forensic practice as an adjunct to traditional anthropological techniques for many years. Recently with the advancement of computer technology and PMCT in particular, there has been an international interest in a near virtual approach to forensic investigation. However, at present there is little anthropological research carried out in this area and currently there are no established protocols outlining which virtual measurements would be the most valuable to obtain a positive identification and the best imaging system to use. This may cause judicial problems were virtual anthropology to be implemented as part of a forensic or DVI protocol.

This review highlights how PMCT is a suitable imaging modality to perform all the major age determination methods for juveniles as stated by Cunha *et al*. (2009). Furthermore, PMCT offers a number of additional advantages, namely: superimposition is reduced, which is particularly beneficial for dental investigations; scans can be taken with the body in any orientation; bones do not need to be defleshed, for long bone diaphyseal measurements; and images can be re-sectioned and viewed in any plane, to make it difficult to distinguish features more visible. However, undoubtedly the greatest advantage that PMCT offers is the ability to perform all of these assessments on the same data set. Unlike traditional anthropological and odontological methods that would require multiple imaging modalities and dry bone examinations to complete all of the age prediction methods discussed in this chapter; each of these features can be assessed simultaneously, by multiple examiners if required, using only PMCT image data. This would be a particular benefit in DVI incidences, when time is limited. Table 4.2 provides a summary of the main advantages and limitations PMCT offers.

This review suggests that it would be a valid possibility to replicate all of the measurements used for juvenile age determination on PMCT data. The next step in this investigation should therefore involve testing the accuracy and reliability of PMCT measurements against anthropological measurements recorded using traditional techniques. This is a necessary step to develop

PMCT anthropological examinations. Considering the added benefits PMCT offers, if measurements were of comparable accuracy to traditional methods this could encourage a shift in anthropological practice towards PMCT examination.

4.7 Key Points

- There has been a gradual acceptance that a ‘near virtual autopsy’ using CT could supplant more traditional imaging and dry-bone investigative methods, driven by its increasing availability and affordability, as well as the development of spiral and multiple detector CT, which has improved scanning speed and resolutions.
- PMCT creates high quality image reconstructions in any plane and 3D modeling of structures; so that bones and teeth can be viewed and measured in any plane without invasive procedures, offering considerable practical and aesthetic benefits.
- This review highlights the suitability of PMCT to perform all the age determination methods for juveniles suggested in a recent review by Cunha *et al*. (2009).
- PMCT and 3D volume rendered images present the opportunity to explore the possibility of using more complex measurements - that can not be easily quantified on physical bone samples (i.e. volume and curvature) - to develop new methods specifically designed for CT analysis, which could improve the efficiency of the age screening process on victims following a mass disaster.

Table 4.2: Advantages and limitations of PMCT anthropology.

ADVANTAGES
• Even if there are no ante-mortem records the amount of soft tissue and bone information may achieve a positive identification of an unknown body • CT is not restricted to a single area of the body • No need for lengthy preparations of bones • Less chance of information or evidence being lost • Fast scan and analysis time • Expert analysis can occur remotely and at any time • CT is non-invasive, therefore virtual anthropology procedures are likely to be culturally more acceptable • A virtual anthropology database could be created improving on traditional anthropological tables
LIMITATIONS
• Access to PMCT scanners may be limited • Equipment is expensive • Metal artefacts might cause viewing obstructions • Not all pathologies can be detected

CHAPTER 5: THE CLAVICLE

"The skin and bones tell a story which the child is either too young or too frightened to tell."

- Johnson, Cameron and Camps.

5.1 Introduction

Data gathering techniques used by forensic anthropologists can generally be classified as; Anthroposcopic, osteometric, chemical or histological. Chemical methods involve analysing the chemical makeup of certain structures of the skeleton and associated matter (e.g. the ground beneath a decomposing body). These methods involve sampling matter and applying special techniques to determine their nature. Histology is the study of the microstructure of bone and teeth. This generally involves cutting off thin slices and staining them for viewing under a microscope to determine demographic characteristics (especially age). However, both of these methods require special instruments that are not always available *in situ* to forensic anthropologists. Anthroposcopic and osteometric methods will therefore be the primary focus of this thesis, as these techniques are the most regularly used in anthropological practice and the two methods that could potentially benefit or be enhanced by medical imaging techniques (such as PMCT).

Anthroposcopy is a qualitative technique that involves recording morphological traits by visual inspection. It is regularly used as it does not require any specialist equipment and is therefore the most accessible anthropological tool. When used by experienced forensic practitioners it is a powerful tool and can lead to a successful identification. However, anthroposcopy relies on judgment of a particular skeleton feature, for example distinguishing wide versus narrow and robust versus gracile bones, and can therefore be highly subjective.

Osteometry provides objective measurements of a human bone. It is less subjective and less ambiguous than anthroposcopy and is therefore preferred by jurisprudence. The results of osteometric methods are normally expressed as a measurement and a range of possible error, giving a more truthful estimation. By condensing the morphological characteristics of a skeletal element into a single quantifiable value, the degree of accuracy and precision of a measurement can be examined. In statistical terms, ‘accuracy’ refers to the degree in which an estimated quantity conforms to its true value (White, 2000). However, it is also often used to describe the abilities of several individuals to reproduce a specific measurement, the so-called ‘inter-observer error’ (Corner *et al*., 1992). In contrast to this, the precision of a measurement describes the closeness of repeated measurements taken by a single individual, or the intra-observer error (Jamison and Zegura, 1974). In osteometry it is vital to limit the sources of inter- and intra-observer error, to obtain results with a high degree of accuracy and precision. A simple way to achieve this is to ensure that the correct measurement is being recorded. It is therefore advised to consult the appropriate guidelines as a matter of course, which outlines the precise definitions of each measurement and how to measure them properly.

The British Association for Biological Anthropology and Osteoarchaeology (BABAO) and the Institute of Field Archaeologists (IFA) are the professional and standard-setting bodies for all those interested in and/or working in all areas of analysis and research in human remains from archaeological and anthropological contexts. Both societies work to promote best practice and implement standards of research to maintain professional credibility.

In 2004 BABAO published 'guidelines to the standards for recording human remains'. Since the formation of BABAO in 1998, the issue of standards for the recording of human skeletal remains in Britain has been a concern to the membership. The need for a guidance document and a specialist framework within which to work was raised at the annual meeting in 2001. A standardised recording format enables human bone assemblages from different sites to be compared with a higher degree of precision. Although this document will undoubtedly have a limited lifespan, due to the inherent evolutionary nature of research into human skeletal remains, it provides a useful current guide for practitioners involved in various fields of research and analysis.

The BABAO guidelines (BABAO, 2014) state that a standard record of any assemblage should include an inventory, which contains; a record of the bones which were available for analysis and the prevalence of pathological lesions and conditions; a record of the data used to determine the age and sex of an individual; metric data and a record of non-metric traits, which assist in sexing and are necessary for the calculation of various indices to further our understanding of biodistance within and between populations; and an accurate record of pathological lesions.

The field of forensic radiology is very much still in its infancy. Although the advantages of radiology in forensic investigations have long been recognized, its implementation to date has generally been conducted in a sporadic, *ad hoc* manner, varying greatly from case to case. In order for the field of forensic radiology to develop and stand-alone as a professional body in its own right, it is important that the standards established by BABAO and IFA which have been developed for the purpose of recording human remains in Britain are maintained. With this in mind, this thesis follows the guidelines for recording human remains established by these professional bodies and will illustrate that the standards outlined by BABAO and IFA are transferable to PMCT.

5.1.1 Landmarks

The definitions for all osteometric measurements refer to standard landmark data. Osteometric landmarks are clearly delineated “points of correspondence on an object that matches between and within populations” (Dryden and Mardia, 2002). It is assumed that a particular point on a bone is the same between different individuals, due to the notion of biological homology within individuals of the same evolutionary origin. However, in reality, the landmarks used as the endpoints of the measurements in osteometry may vary slightly within a population, as the bones on which they are located are not truly homologous. Measurements are therefore more accurately described as the physical distance relating ‘points to points’ rather than ‘parts to parts’ and as a result have an inherent error margin associated (Brookstein, 1991).

Although metric methods are undoubtedly more objective, and therefore generally more statistically robust, than qualitative techniques, landmarks are not always ‘biologically meaningful’ which can introduce a degree of subjectivity and bias into results, as observers have the responsibility of ‘judging’ where the endpoints of the measurement occur (Adams and Byrd, 2002). The inherent biological variation between individuals therefore translates into an inherent intra- and inter-observer error for any osteological measurement.

All biological landmarks can be classified into three main types, depending on their interaction with other anatomical points and location within the body:

Type 1: discrete juxtapositions of tissues - includes point in space where three or more structures meet, and are typically the most straightforward to locate e.g. the junction between the coronal and sagittal sutures (the bregma) on the skull.

Type 2: maxima of curvature or other morphogenetic process - includes points of a structure connected with biomechanics forces e.g. bony processes where muscle attachments are centered.

Type 3: extremal points - include the most extreme points of a structure and are often much more difficult to isolate than type 1 and 2 landmarks.

An investigation of the landmark-specific error associated with type 1, 2 and 3 landmarks by Slice and colleagues (2004) suggested that Type 1 landmarks were the most reproducible, while type 3 were the least reproducible. Therefore, in osteometry it is regarded as best practice to use the average of repeat measurements to achieve the highest possible degree of precision.

Himes (1989) considered the mean of independent replicate measurements to be more reliable than a single determination. This observation was supported by Corner *et al*. (1992). Corner's investigation into the repeatability of landmarks also suggested that error magnitude is indirectly proportional to the linear distance between them. In this investigation we should therefore expect the smaller the measurement, the larger the proportional error.

5.1.2. The clavicle

The clavicle was chosen for this investigation, as the first bone to examine for several reasons, including: It displays an extremely linear growth with age relationship; is particularly resilient to the taphonomic (decay of an organism) stresses involved with inhumation (burial); was present in the large proportion of the skeletons in our collection; was easy to isolate for measurement purposes; had clear anatomical landmarks so that measurement parameters could be accurately described; was anatomically the first long bone in the skeleton (from a superior-inferior perspective) and therefore was considered a logical start point.

The clavicle forms part of the pectoral girdle, along with the scapula, functioning as a strut to steady and brace the upper limb to the thorax (Figure 5.1). Commencing ossification at approximately 39 days, in the 6th week of intra-uterine life, the clavicle is one of the first fetal bones to begin development. The shaft of the clavicle adopts a distinctive 's' shape by around 8-9 prenatal weeks, and achieves virtually adult morphology by week 11. This unusually early attainment of adult morphology indicates that the clavicle is not influenced much by postnatal mechanical stresses and forces. It is therefore a bone of both considerable phylogenetic (evolutionary) and ontogenetic (development from embryo to adult) morphological stability. It displays a high degree of sexual dimorphism in the adult, exhibits some racial characteristics, can be of value in stature estimation and may even assist in the identification of hand dominance. However, the late epiphyseal union means that the clavicle displays the longest period of growth-related activity of all the long bones of the human skeleton, and so is arguably of greatest value in the estimation of age at death.

Figure 5.1: The pectoral girdle; clavicle and scapula. A) Superior view B) Posterior view.

The extremely early attainment of adult morphology subsequently means that changes in length of the clavicle are most useful for age determination of pre-pubertal individuals. Studies on the foetal growth of the clavicle, both by direct examination of the bone and by ultrasound have shown that the clavicle grows at a surprisingly linear rate of approximately 1mm per week. By term, the clavicle generally measures some 40-41mm and then growth appears to slow down, although later growth spurts can be identified between 5-7 years of age and again at puberty. At puberty, endochondral ossification at the medial and lateral extremities tends to slow down as the clavicle reaches maximum bone length. The medial clavicular epiphysis is slow maturing and is generally recognised as the final epiphysis to fuse in the human skeleton and, as a result, it has proved to be of considerable value in estimating age at death in the post-pubertal period. The transitory nature of the lateral epiphysis renders it of limited value in the estimation of age at death.

Despite this there have been few studies investigating the relationship between clavicle length and age. Of the few studies that have been conducted in this area, arguably the most comprehensive was that by Black and Scheuer (1996). In this study the authors determined that although age ranges did show a considerable degree of overlap it was clear that the correlation between clavicle length and age was strongly positive.

Morphological changes of the medial epiphysis have been more extensively studied, particularly for age estimation during criminal proceedings, to determine whether an individual is above or below the criminal liability threshold (normally 21) as this can have an impact on how the individual must be treated and what legal aid they are entitled to. These research projects generally fall into 2 categories, anatomical or radiological. Anatomical studies assess ossification by autopsy or dry bone inspection (Todd and D'Errico, 1928; McKern and Stewart, 1957; Owings *et al*., 1985; MacLaughlin, 1990; Ji *et al*., 1994; Black and Scheuer, 1996), while radiological investigations used conventional plain film X-rays or CT (Flecker, 1933; Galstaun, 1937; Jit and Kullkarni, 1976; Kreitner *et al*., 1997, 1998). Reports of radiological changes during the ossification of the medial clavicular epiphysis go back to the 1930's. However, Kreitner *et al*. (1977) were the first to describe the maturation process using CT.

Investigations of the clavicle have also considered whether age can be reliably estimated from the cortical index of the human clavicle. Kaur and Jit (1990) calculated the cortical index (proportion of cortical thickness in relation to total diameter of bone) in a population of 210 Northwest Indians, aged 15 to 85 years. The clavicles were cut either horizontally or parasagittally and the measurements were taken at the mid-point of the clavicle. From their investigation, Kaur and Jit (1990) found that from 15 to 30 years of age the cortical index increased proportionally with age and between 31-40 years it decreased, in both sexes. After the age of 40 years, this rapid decrease in the index continued in females, but became slow and gradual in the males. Differences between the left and right sides were statistically insignificant in both sexes ($p > 0.05$) below the age of 40 years. However, the sexual differences were significant ($p < 0.01$) in the age groups from 41 years onwards. A number of other authors (Barnett and Nordin, 1960; Meema, 1963; Sedlin *et al*., 1963; Helela, 1969; Anton, 1969) have demonstrated that, similarly to the clavicle, the cortical thickness of the femur, metacarpal, lumbar vertebra, radius and ribs, also declines steadily after a certain age. While these studies suggest that cortical thickness of several bones could be used as an age indicator, Walker and Lovejoy (1985) found - after a visual examination of a number of radiograph series - that trabecular involution in the clavicle exhibited the most constant relationship to age. Although Kaur and Jit (1990) quantified the cortical changes that occur in individuals from 15 years onwards, no investigations have been conducted on younger individuals. It would be interesting to determine if these changes actually begin much earlier in development, as suggested by Walker and Lovejoy (1985). However, Kaur and Jits' method is destructive and due to the paucity of juvenile skeletal collections, ethical permission to conduct this type of investigation today would be extremely unlikely. By using CT and 3D imaging techniques however, this destructive process would not be necessary.

In addition, according to Urist *et al*, (1962), a decline in the thickness of cortical bone cannot be truly appreciated on a radiograph until at least a 30% decrease in the density of bone occurs. However, this problem would be overcome by the greater spatial resolution and advanced measurement tools offered by CT, compared with plain radiographs and dry bone examination, respectively.

Walker and Lovejoy created radiographic standards for age determination using the clavicle from 18-55+ years, consisting of 8 phases of age-related changes. The authors suggest that in general, the clavicle is superior to the femur with respect to both bias and inaccuracy, and that clavicular age estimates are generally equivalent to those of other major skeletal age indicators (i.e. the dentition). It therefore appears as though the diaphyseal length of the clavicle could be a valuable indicator of age at death, predominantly in the pre-pubertal age range, especially as it often survives inhumation (the death and burial process) more successfully than many of the other long bones. During puberty there are too many factors outside reasonable control for this bone to be used as a reliable indicator of age. However, it regains its predictive value when other growth related indicators have become inactive, and goes on to be a useful predictor of age at death until close to 30 years of age, due to the late fusion of the medial epiphysis.

5.2 Aims and Objectives

It is evident, from the previous chapter, that all the measurements required for anthropological techniques frequently used in current practice can be obtained from PMCT images. The aim of this chapter is therefore to consider whether the measurements we are able to gather using PMCT are accurate, and whether they can be replicated with minimum error by practitioners from various professional backgrounds and levels of expertise.

To fulfil this aim, the clavicle was selected for further investigation. Despite the clavicle’s potential for age estimation of juvenile remains, it has not been extensively examined by PMCT throughout the full developmental period of a human, from embryo to adult. Few studies have published substantial data on the clavicle and therefore in addition to addressing the primary issue of PMCT accuracy, this study will also provide valuable data for this field of research.

The aims of this study were:

1. To determine if dry bone measurements of the clavicle, used in age determination methods, can be accurately replicated using PMCT.
2. To directly compare age estimations made using PMCT with those made using dry bone measurements, to determine their accuracy.
3. To explore whether the inclusion of additional PMCT measurements can improve current clavicle-based age determination methods, by narrowing the associated error ranges.

These aims will be realised by completing the following research objectives:

1. Identifying which measurements are required to apply previously published algorithms, and the best way to attain them.
2. Developing a protocol to extract these measurements from PMCT data of a sample of juvenile human clavicles, which would be comparable or supplemental to traditional osteometric methods.

5.3 Methods and Materials

This project uses remains from the Scheuer Juvenile Skeletal Collection (Appendix 1; Section 3.2.1), which is believed to be the only active repository exclusively for juvenile skeletal remains held anywhere in the world. To the best of the authors' knowledge, this is the first series of PMCT studies of this nature to be undertaken exclusively on juvenile skeletal remains.

Image analysis was performed using the "bone" window, with both OsiriX (version 3.7.1; Web site: http://homepage.mac.com/rossetantoine/osirix.) and Voxar 3D (Toshiba Medical Visualization Systems Europe, Ltd) imaging software. Both software packages created multi-planar reconstructions (MPR) from the data set to allow measurements to be taken in any plane.

Thirty-three clavicles from 20 individuals (all of the specimens which were PMCT scanned from the Scheuer collection for this investigation, see Section

3.4.2) were used for further analysis. The demographics of each specimen used in this chapter are outlined in table 5.1. Measurements on the clavicles were performed by myself, a forensic anthropology graduate (AB); Guy Rutty, forensic pathologist (GNR); and John Bennett, medical student (JB) - under the supervision of Bruno Morgan, radiologist (BM). The measurements were taken three times by each rater, as per best practice (see section 5.1). These measurements were then compared with those obtained by traditional osteometric apparatus, which is regarded as the gold standard at present.

5.4 Measurements

Osteological measurements of the clavicle using PMCT have not been previously reported and thus there is no defined protocol available. As the aim of this investigation was to ascertain the viability of PMCT as an alternative to traditional osteometric techniques - a PMCT measurement protocol that exactly simulated the osteometric board was designed, so that a direct comparison could be made between corresponding measurements using both techniques (A detailed protocol and instructions of how to replicate the PMCT measurements used in this investigation can be found in Appendix 5).

MPRs were used in this investigation. Although not strictly speaking a three dimensional imaging technique, *OrthoSlice* and *ObliqueSlice* modules in both Voxar and OsiriX allows contiguous 2D slices to be viewed in any plane which relies on volume data from a 3D data set. Within this 3D MPR function, the *ObliqueSlice* module was preferred for this investigation as it enabled the observer to interactively orientate the position of the slice compared with the *OrthoSlice* module which was more limiting, allowing the MPR slice to be viewed in either the x-, y- and z- plane only.

The 3D MPR function was used in this study predominantly for the reason that, at the time of investigation, an adequate measurement tool was not available in the 3D reconstruction function. The measurement tool in this function could not be fixed or propagated through the entire data series and therefore disappeared when the 3D bone reconstruction was moved. This subsequently meant that it was not possible to ensure that the true diaphyseal length was being recorded, between the two most extreme points of the bone. In contrast, using the 3D MPR function, the measurement tool could be fixed and propagated through the entire series so that, by scrolling through the contiguous slices, the maximum length could be recorded as accurately as possible.

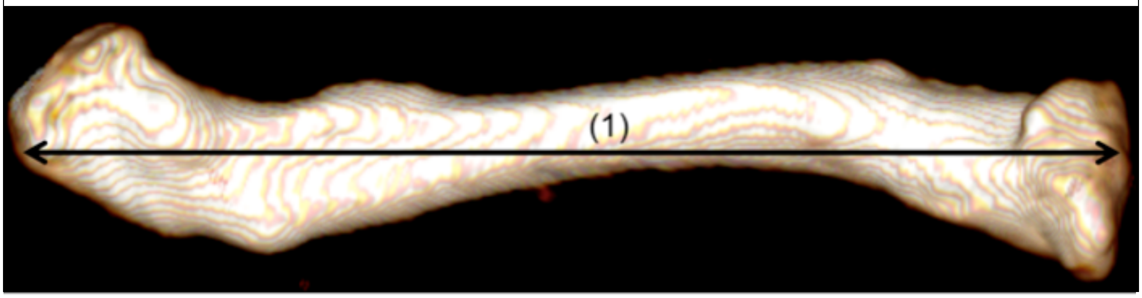
5.4.1 Osteometric measurements

Using an osteometric board, I recorded the ‘osteological’ maximum length of all the dry bone clavicles. Osteometric measurements from each clavicle were recorded to the nearest 0.1mm. Although callipers could have been used for the majority of the cases, some of the older specimens would have been too large to measure with callipers. As a result, an osteometric board was chosen to ensure experimental consistency and minimise the potential to introduce error. An osteometric board consists of a horizontal base plate with two perpendicular sliding planes, between which the bone is measured (see chapter 4, section 4.4.2.). This measurement was recorded 3 times for each bone, as per best practice (see section 5.1), and so that the intra-observer variability could be calculated.

Although several different measurements can be acquired from the clavicle, maximum length is the only measurement known to be of value in age estimation. Maximum length was measured following the standards adapted by Buikstra and Ubelaker (1994) and Moore-Jenson *et al*. (1994), in-keeping with the guidelines issued by BABAO and the IFA. The details of which are outlined in Table 5.1.

Table 5.1: Maximum length of clavicle (adapted from Buikstra and Ubelaker, 1994; and Moore-Jensen *et al*., 1994).

Measurement	Instrument	Description of Measurement
Maximum Length	Osteometric board	Maximum distance between the most extreme ends of the clavicle (1).



5.4.2 PMCT measurements

GNR, JB and AB took radiological measurements. I (AB) had little experience recoding radiological measurements and JB had little experience taking either radiological or anthropological measurements.

The PMCT measurements were taken in planes based on how the bone would lie on a traditional osteometric board. With both Voxar and OsiriX the planes of the board were simulated using the *ObliqueSlice module*, and the linear measurements were recorded using the *measurement too*l.

Measurements were undertaken blind to the knowledge of the actual demographics and blind to the other practitioners' results.

5.4.3 Maximum clavicular length

Maximum clavicle length was recorded by AB and GNR from reconstructed images, using the "bone" window, with both OsiriX and Voxar 3D imaging software. Both software packages created multi-planar reconstructions (MPR) from the data set. In both OsiriX and Voxar the 3D MPR viewer was used so that the bone could be rotated in the x-, y-, and z-planes. This allowed the user to position the bone correctly (i.e. as it would lie on an osteometric board), before taking the required measurements. The most inferior parts of the bone, which would ordinarily be in contact with the surface, if the bone were positioned horizontally on an osteometric board, defined the base plane of the clavicle.

Preliminary results from AB and GNR, using both software systems were compared before JB conducted the measurements-to determine which method, if either, was superior. Although both methods produced similar results, it was decided that JB should conduct measurements using only OsiriX. This decision was made based on ease of use, and software availability. The Voxar workstation is a licensed software program, which is installed on a limited number of computers. For this reason, access to a Voxar workstation was restricted. In contrast, OsiriX can be downloaded free of charge to any Macintosh computer and was therefore a more practical option for this investigation. Furthermore, feedback from both users suggested that OsiriX was easier to use, which is beneficial for the long-term practical application of this project.

Any reference to a measurement protocol from this point onward, will therefore relate to the OsiriX workstation. Although the general “virtual osteometric board” method is transferrable to other software packages, the exact functions and command tools used might be specific to OsiriX.

Orthogonal boundaries (Figure 5.2, A) were drawn at the most extreme points of each clavicle to enable the maximum clavicular length to be measured. The position of these boundaries was determined by scrolling through the multiple reconstructions to find the most extreme point. Once this point was determined the *length* measurement tool was used to create the sliding plain of the osteometric board. This plane was then propagated though the entire series of images so that the rater could confirm that the plane was correctly positioned at the outer-most point, and if not, the sliding plane could be repositioned accordingly. This process was repeated at the other extreme and the distance between these two points was measured using the *length* measurement tool (Figure 5.2, B). This “*virtual osteometric board*” was used for all measurements and each measurement was made to the nearest 1 mm. This was considered the appropriate unit of measurement for this investigation since a 0.5mm slice thickness, and 1mm reconstruction intervals were used as the scanning parameters in this study. Therefore, although both OsiriX and Voxar could produce measurements to the nearest 0.1mm, it is unlikely, with the scanning parameters used, that measurements below 0.5mm would be precise.

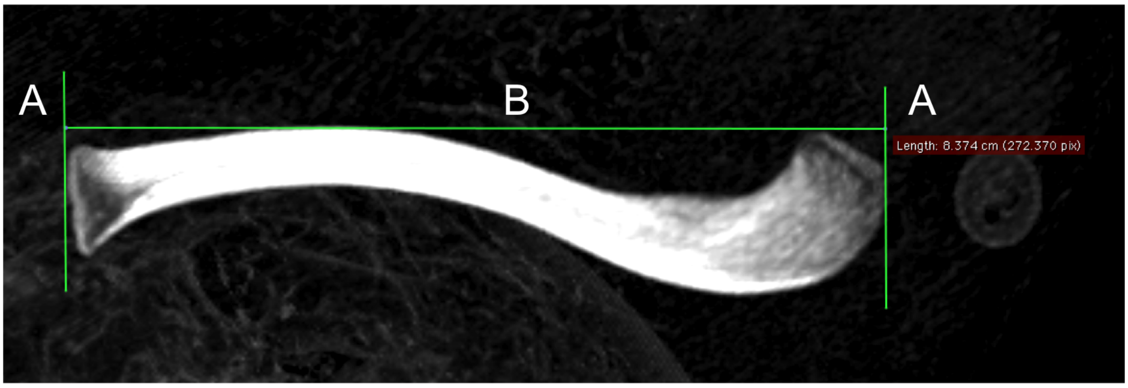


Figure 5.2: Maximum clavicle length, using virtual osteometric board. A) Two parallel planes, marking the most extreme points of the bone, to replicate the sliding planes of an osteometric board. B) Maximum clavicle length, the distance between the two parallel planes (A).

After designing a comprehensive measurement protocol - I provided both GNR and JB with detailed list of experimental instructions, including a step-by-step measurement protocol (Appendix 5), before they gathered their results. This guide included instructions on how to navigate within the OsiriX workstation, tailored explanations of the appropriate toolbar commands, and tips to optimize image quality and measurement output. This was deemed necessary to limit confusion regarding what measurements were required and how to attain them, in order to maintain experimental consistency and minimise inter-observer error. The guide included snapshots of each stage of image acquisition, so that there was no ambiguity created by language choice.

With experience, each clavicle could be measured in less than 10 minutes. JB had little experience with either PMCT or anthropological measurements, which was reflected by slightly longer measurement collection times. Although, this initial lag was notably reduced after experience from the first few cases.

Adams and Byrd (2002) provide evidence to suggest that inter-observer variation is a valid concern with osteometrics, and that it must be considered when we interpret the results of quantitative analysis. However, previous studies have demonstrated that inter-observer errors between different professional groups for osteological measurements using PMCT are extremely low (Robinson *et al*., 2008). It was therefore deemed unnecessary, after this initial investigation of maximum length, to perform repeat measurements for every standard test. Alternatively, the appropriate professional (BM) oversaw all further measurements and blind tests of reliability were randomly conducted.

5.4.4 Cortical thickness

As previously discussed, cortical thickness measurements were recorded using only OsiriX. In addition, these measurements were only recorded by AB. Since just one person undertook these measurements, the repeat measurements were taken twice for each bone - with the second set of repeat measurement taken on a different day, a week apart, blind to the first measurement result.

Cortical thickness was measured using the method described by Kaur and Jit (1990) (Figure 5.3). To measure the cortical thickness, the midpoint of the bone shaft was identified, using the previously recorded maximum length measurements (mid-point = maximum length (A)/2). A line, half the length of the maximum length (B) was used to ensure that the midpoint of the diaphysis was accurately marked. Using the *Angle* measurement tool, a line at an approximate tangent to the curvature of the clavicle at this point was then created (c). The maximum diameter of the bone was given by the width of the bone, at a 90° angle to this line (d). Using this plane as a guideline, the cortex of the bone was measured on each side of the medullary cavity, using the *length* measurement tool. The total cortical thickness was calculated by the addition of these two measurements (Kaur and Jit, 1990).

I found that by using the *Manual Contrast* command from the toolbar, the cortex of the bone could be more clearly delineated. This tool works by enhancing the brighter areas (cortical bone) at the expense of the darker regions (air in the medullary cavity) (Figure 5.4). The clarity of the cortical bone edge was also improved using the *Thick Slab* tool, to increase the slice thickness. Increasing the slice thickness ensured that the entire diameter of the cortex was measured by increasing the depth of information included in the principle image.

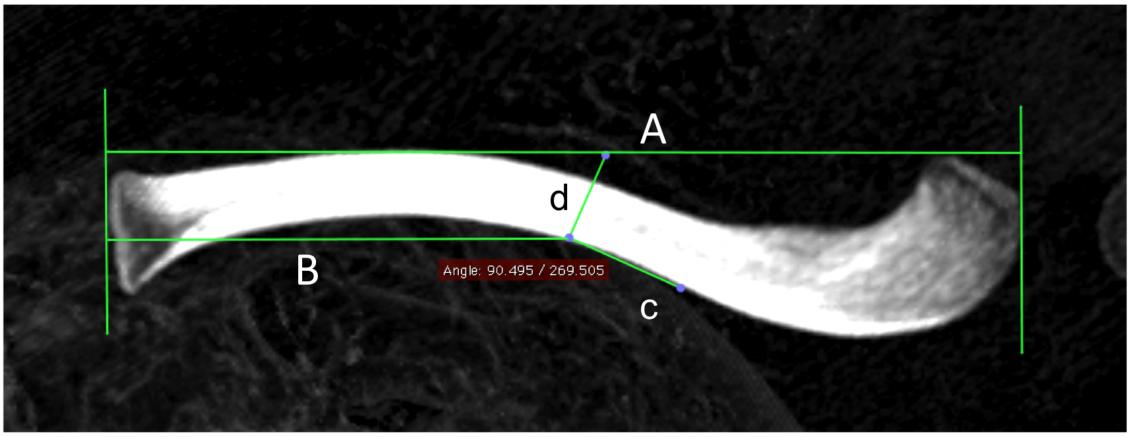


Figure 5.3: Reconstructed MPR illustrating the measurement method for cortical thickness (adapted from Kaur and Jit, 1990). A) Maximum length; B) Mid-point marker; c) Tangent to curvature at the mid-point; d) Mid-point dissection plane, along which the maximum diameter and cortical thickness are measured.

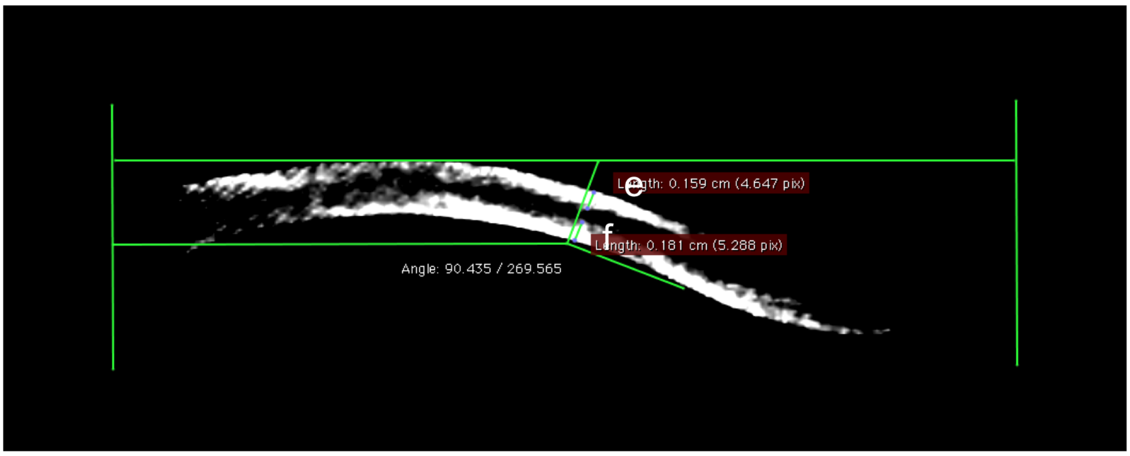


Figure 5.4: Increased contrast and slice thickness MPR. The medullary cavity is more clearly delineated. e, f) Cortical thickness measurements.

5.4.5 Additional measurements

Additional measurements were also recorded from the PMCT images, as a means of future proofing for possible studies in age evaluation from these dimensions. These measurements were taken by JB and AB.

The maximum bone diameter was measured at the medial, central and lateral regions of the clavicle diaphysis (Figure 5.5). The same steps as for maximum length and cortical thickness were used to define the end-plates and the mid-point before the additional measurements were recorded. I also found that using additional length planes, to ensure that the maximum diameter was definitely being recorded at each of these points, extremely helpful. For these additional measurements it was particularly important to ensure that the bone was in the correct anatomical position (i.e. as it would be lying on an osteometric bone) to maintain experimental consistency. The additional measurements were not standard measurements and there were no official guidelines for recording these. Therefore, ensuring that all measurements were recorded in the same plane was extremely important, so that the measurements could be accurately replicated and clearly described to other observers. Measurements were repeated 3 times and the mean was recorded.

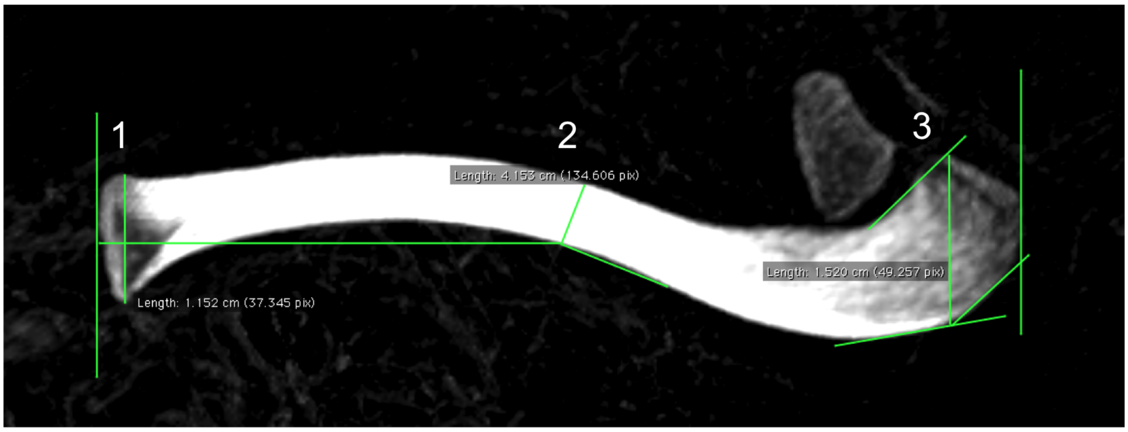


Figure 5.5: Additional measurements recorded. 1) Medial; 2) Central; and 3) Lateral maximum bone diameters.

5.4.6 Age estimation

The relationship between age, maximum clavicle length, and cortical thickness was assessed. Cortical thickness measurements were applied to the formula stated by Kaur and Jit (Kaur and Jit, 1990) to calculate the cortical index of each clavicle. The cortical index was then correlated with specimen age to determine its estimation power.

In the present data set, many of the demographics are estimations and therefore the virtual method has been evaluated by comparing with an established technique rather than the true quantity. For clavicle length, the guide presented by Black and Scheuer (Black and Scheuer, 1996) provides the only meaningful published results comparable to the present study. It was therefore the method selected for this investigation to estimate the age range of specimens from clavicular length.

5.5 Results

5.5.1 Within-subject standard deviation

The within-subject standard deviation (wSD) and 95% repeatability was assessed for PMCT measurements compared to osteological measurements, using AB's results for both methods.

Data for the repeated osteological and PMCT measurements are given in Tables 5.4, 5.5 and 5.6. In this investigation the intra-observer error for repeat osteometric measurements was extremely low. This error was determined by calculating the standard deviations of the repeat measurements on the same specimen; the so-called within-subject standard deviation (wSD) (Bland & Altman, 1996). A low wSD value (0.08mm) suggests that the rater AB did not deviate from the mean, which in turn, indicates minimal measurement error.

Where multiple measurements were compared, analyses of agreement were performed using a method described by Bland and Altman (1996). Firstly the effect on measurement magnitude of intra-observer variability was assessed and then the wSD was calculated. The difference between a subject's measurement and the true value would be expected to be less than 1.96 wSD for 95% of observations. 1.96 x wSD gives the range of difference from the 'gold standard measurement' or mean that 95% of cases will fall within and √2 x 1.96 x wSD gives the range within which 95% paired measurements would be expected to differ from each other.

For both OSIRIX and VOXAR the wSD values were slightly higher than for repeat osteometric measurements. This was expected since osteometric measurements of defleshed bones are the 'gold standard' for anthropological examination in forensic practice, and the standard to which all PMCT measurements are ideally aiming to conform. Nonetheless the wSD values for both OsiriX and Voxar were still consistently low, demonstrating that these virtual techniques also have a high degree of precision and repeatability. The low wSD indicates that each measurement did not deviate far from the mean (0.28 and 0.43mm, respectively).

5.5.2 Osteological-virtual comparison (max length)

Comparison of mean osteological and PMCT measurements is shown in Figure 5.6. There is no evidence that PMCT consistently either over- or under-measures compared with osteometric measurements, with an average difference of only 0.01mm. Also, there is no evidence of error being proportional to the magnitude of the measurement. The wSD for intra-observer variation between osteometric and PMCT measurements was 0.39mm. The inter-observer error is also minimal, indicating there is no statistical significance between measurements taken by individuals with different professional experience. The low error rates signify that the results are consistent.

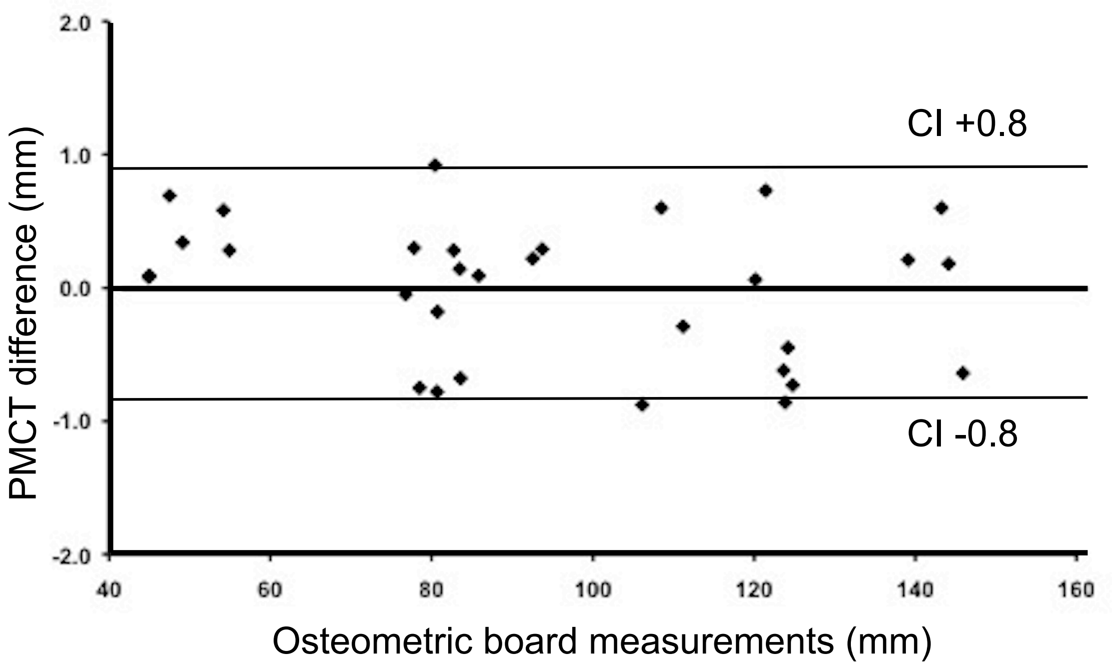


Figure 5.6: Maximum length of the clavicle; Osteological-virtual comparison. PMCT acquired measurements compared with Osteometric Board measurements.

5.5.3 Between-subject standard deviation (OsiriX and Voxar)

The inter-observer error of osteometric measurements has been shown to be extremely low in the majority of influential studies (Black and Scheuer, 1996; Buikstra and Ubelaker, 1994; Maresh and Deming, 1939). This, in conjunction with the fact that the primary focus of this investigation was to determine the accuracy of PMCT measurements subsequently meant that repeat osteometric measurements by the second observer were not deemed necessary in the present study.

However, the inter-observer error for OsiriX and Voxar measurements were calculated, by comparing the measurements obtained by GNR with those obtained by the AB for both workstations (Table 5.7). The consistency of different individuals using PMCT was also analysed by calculating the average "scaled error index" (SEI), which permits the comparison of measurements regardless of scale (Bland and Altman, 1996). Adams and Byrd (2002) devised a Scaled Error Index (SEI) as part of their study on inter-observer error of postcranial skeletal measurements. The SEI is represented by: SEI = [(x–median) /median]*100, where 'x' represents a single measurement and median the midpoint of all measurements. This index is useful because it allows for a comparison of measurements that is unaffected by scale or sample size (Adams and Byrd 2002). The SEI also facilitates comparisons between individuals taking the measurements and the measurements themselves, so it can be used to analyze both inter-observer error and method performance. Differences in mean SEI can be examined using a one- or two- tailed *t*-test, or, for several methods, ANOVA.

In this investigation the SEI therefore allowed the comparison of error between estimated and actual age regardless of scale or sample size. SEI was also used to determine whether measurements were dependent upon experience. It was predicted that those individuals with more experience in skeletal ageing (AB) would have a lower SEI score than those with less experience. However, the results of this investigation suggest that there was in fact no statistical difference between each set of measurements, indicating that there is minimal inter-observer error. The low error rates signify that the results are consistent between observers.

Where 2 different measurement approaches are compared (i.e. Voxar and OsiriX), the mean difference and 95% confidence intervals were calculated to identify any significant measurement bias for each technique. Although wSD values were low for Voxar those for OsiriX were slightly lower (0.43 and 0.28 mm, respectively). In addition the 95% confidence limits for OsiriX were also slightly lower indicating its advanced accuracy and repeatability.

5.5.4 Age estimation

The strong positive correlation between age and clavicle length illustrated by Black and Scheuer was maintained in this investigation, using a virtual approach (Figure 5.7). An r^2 value of 0.923 shows a significant positive correlation, which suggests that PMCT maximum length measurements could be used to successfully predict age. There is therefore supporting evidence to suggest that PMCT measurements closely parallel the dry osteological data available from existing published communications.

Multiple regression calculations show that maximum length is the strongest indicator of age. However, correlation (central=0.912, medial=0.912, lateral=0.863) and regression (r^2=0.832, 0.831, 0.745; respectively) analysis of the additional measurements (medial, lateral and central maximum diameters) (Table 5.2) indicates the potential significance of additional values. Cortical thickness had a negative correlation with age (Table 5.3) and although yielding a result that was highly significant at the level of p=0.01, it must be noted that the correlation coefficient is not necessarily the best indicator of the power.

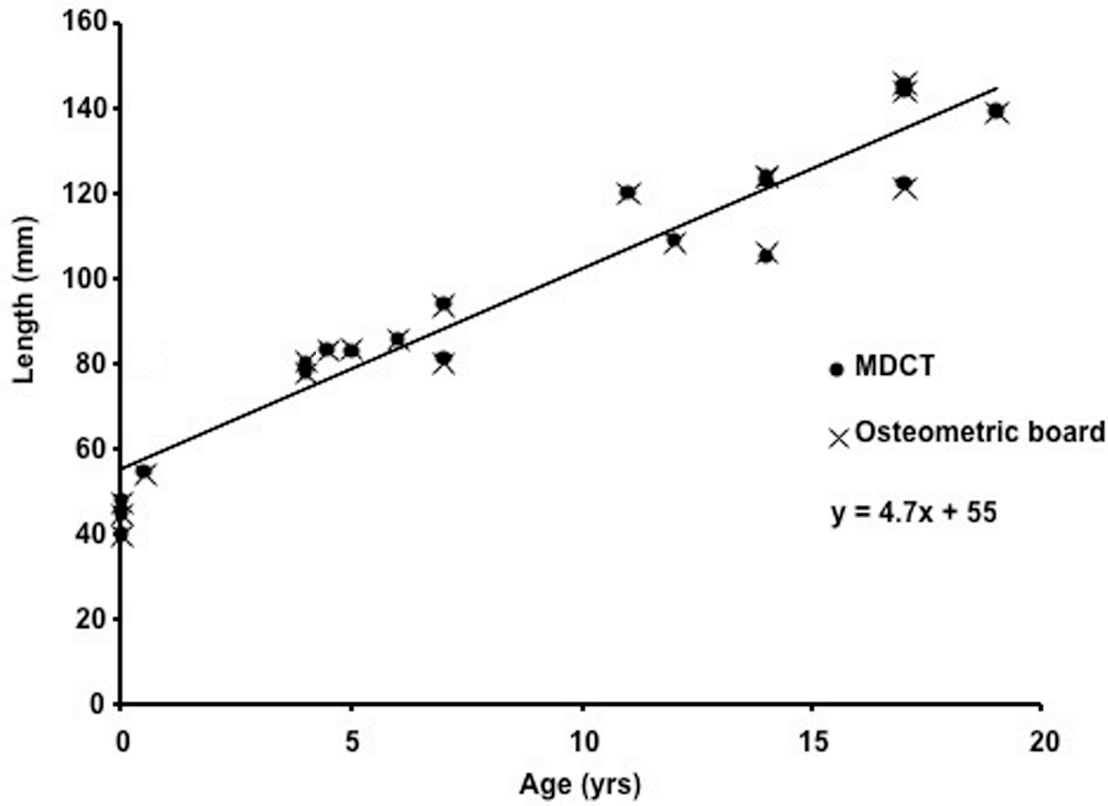


Figure 5.7: Maximum clavicular length measurement using PMCT and osteometric board measurement to estimate age. The trend lines for both measurement overlap.

Table 5. 2: Additional Measurements.

	Measurements (mm)																	
	CENTRAL						MEDIAL						LATERAL					
Case No.	R1	R2	MEAN	WsD	VAR	95% CI	R1	R2	MEAN	WsD	VAR	95% CI	R1	R2	MEAN	WsD	VAR	95% CI
1	9	8	9	0	0	0	18	17	18	0	0	0	20	20	20	0	0	0
2	9	9	9	1	0		18	18	18	0	0		21	20	20	1	0	
3	4	4	4	0	0		6	6	6	0	0		7	7	07	0	0	
4	4	4	4	0	0		6	6	6	0	0		7	7	07	0	0	
5	8	7	7	1	0		15	15	15	0	0		16	16	16	1	0	
6	8	8	8	1	0		16	15	16	1	0		16	15	16	1	0	
7	7	6	6	0	0		12	12	12	0	0		11	10	11	0	0	
8	6	6	6	0	0		13	12	13	0	0		11	11	11	0	0	
9	12	12	12	0	0		22	21	22	1	0		30	27	28	2	1	
10	8	7	8	0	0		14	13	14	0	0		10	9	10	0	0	
11	8	7	7	0	0		14	13	14	1	0		10	8	9	1	0	
12	6	5	5	0	0		11	11	11	0	0		10	10	10	0	0	
13	8	9	9	0	0		17	17	17	0	0		16	16	16	0	0	
14	10	10	10	0	0		22	21	22	1	0		28	25	27	2	0	
15	10	10	10	0	0		23	24	24	0	0		23	22	22	0	0	
16	5	5	5	0	0		12	11	12	1	0		12	11	12	1	0	
17	6	5	6	0	0		11	12	11	0	0		11	10	11	0	0	
18	5	4	4	1	0		10	9	9	1	0		9	8	8	1	0	
19	5	4	5	1	0		8	6	7	1	0		8	8	8	0	0	
20	6	6	6	0	0		13	13	13	0	0		17	17	17	0	0	
21	6	5	5	0	0		11	11	11	0	0		12	11	12	1	0	
22	5	5	5	0	0		10	9	10	0	0		12	10	11	1	0	
23	8	8	8	0	0		19	18	18	1	0		25	24	25	0	0	
24	9	9	9	0	0		16	16	16	0	0		7	7	7	0	0	
25	3	3	3	0	0		4	6	5	2	0		8	8	8	0	0	
26	3	3	3	0	0		7	7	7	0	0		8	8	8	0	0	
27	4	4	4	0	0		8	8	8	0	0		9	9	9	0	0	
28	4	4	4	0	0		9	9	9	0	0		17	17	17	0	0	
29	7	7	7	1	0		16	16	16	0	0		23	23	23	0	0	
30	8	8	8	0	0		15	14	14	0	0		18	17	18	1	0	
31	9	8	9	0	0		12	12	12	0	0		19	18	18	0	0	
32	10	9	10	0	0		23	23	23	0	0		19	19	19	0	0	
33	9	9	9	0	0		18	18	18	0	0		19	18	19	0	0	

Table 5.3: Cortical thickness measurements and calculated cortical index.

		Measurements (mm)				
ID Number	Age (months)	Cortical Thickness		Combined	Total Diameter	Cortical Index
1	132	3	3	6	9	64
2	132	3	3	6	9	68
3	0	2	1	3	4	73
4	0	2	2	4	4	100
5	84	3	3	6	8	77
6	84	3	3	6	8	77
7	48	3	3	6	7	90
8	48	3	3	6	6	90
9	204	4	4	8	3	61
10	72	2	2	4	9	56
11	72	2	2	4	8	58
12	84	2	2	4	5	73
13	168	2	2	4	9	44
14	204	4	4	8	11	65
15	204	4	3	7	13	57
16	60	2	2	4	6	81
17	60	2	3	5	6	82
18	6	2	2	4	5	72
19	6	2	2	4	6	77
20	6	2	2	4	5	74
21	48	2	3	5	6	67
22	48	1	2	3	5	57
23	168	2	3	5	9	54
24	168	2	2	4	9	47
25	0	2	2	4	4	100
26	0	2	1	3	3	100
27	0	2	2	4	5	100
28	0	2	2	4	4	100
29	204	3	3	6	8	75
30	144	3	3	6	8	67
31	144	3	3	6	9	63
32	228	3	4	7	11	62
33	168	2	3	5	9	57

Table 5.4: Osteometric measurements, using osteometric board. Recorded by AB.

OSTEOMETRIC									
ID Number	Measurement Run (mm)					Mean (mm)	VAR	WsD	95% repeatability
	1	2	3	4	5				
1	120	120	120	120	120	120	0.0	0.1	0.2
2	124	124	124	124	124	124	0.0		
3	45	45	45	45	45	45	0.0		
4	45	45	45	45	45	45	0.0		
5	94	94	94	94	94	94	0.0		
6	93	93	93	93	93	93	0.0		
7	81	81	81	81	81	81	0.0		
8	79	79	78	79	79	79	0.0		
9	144	144	144	144	144	144	0.0		
10	86	86	86	86	86	86	0.0		
11	81	81	81	81	81	81	0.0		
12	81	80	80	81	80	80	0.0		
13	106	106	106	106	106	106	0.0		
14	146	146	146	146	146	146	0.0		
15	143	143	143	143	143	143	0.0		
16	84	84	84	84	84	84	0.0		
17	84	83	84	84	84	83	0.0		
18	54	54	54	54	54	54	0.0		
19	55	55	55	55	55	55	0.0		
20	83	83	83	83	83	83	0.0		
21	78	78	78	78	78	78	0.0		
22	77	77	77	77	77	77	0.0		
23	124	124	124	124	124	124	0.0		
24	125	125	125	125	125	125	0.0		
25	40	40	40	40	40	40	0.0		
26	40	40	40	40	40	40	0.0		
27	48	48	48	48	47	48	0.0		
28	49	49	49	49	49	49	0.0		
29	121	122	122	122	121	122	0.0		
30	109	109	108	109	109	109	0.0		
31	111	111	111	111	111	111	0.0		
32	139	139	139	139	139	139	0.0		
33	124	124	124	124	124	124	0.0		
						Mean	0.0		

Table 5.5: Maximum clavicle length, OsiriX measurements. Recorded by AB.

OSIRIX							
ID Number	Measurement Run (mm)			Mean (mm)	VAR	WsD	95% repeatability
	1	2	3				
1	120	120	120	120	0.0	0.3	0.8
2	123	122.8	123	123	0.1		
3	45	44	45	45	0.2		
4	45	45	44.7	45	0.1		
5	94	94	93.5	94	0.2		
6	93	92.9	92.5	92.7	0.1		
7	80	79.9	79.9	79.8	0.0		
8	77	78	78	77.7	0.2		
9	144	144	144	144	0.0		
10	86	86	85.8	85.9	0.0		
11	80	80.7	80.7	80.5	0.1		
12	81	81	81	81	0.0		
13	105	105	105	105	0.0		
14	145	145.5	145	145	0.1		
15	143.8	144	144	143.8	0.0		
16	82.8	82.9	83	82.9	0.0		
17	83.9	83	83.8	83.6	0.1		
18	54.8	54.6	54.8	54.7	0.0		
19	55	55	55	55	0.1		
20	83	83	83	83	0.0		
21	78	78	77.9	78	0.1		
22	76.5	76.8	77	76	0.1		
23	123.5	124	123.7	123	0.1		
24	124	123	124.7	124	0.5		
25	40	39.9	40	40	0.0		
26	40	40	40	40	0.1		
27	48	48	48	48	0.0		
28	49	49	49	50	0.0		
29	122	122	122.7	122	0.2		
30	109	109	109	109	0.0		
31	111	110.5	111	110	0.1		
32	139	139.5	139	139	0.0		
33	122.9	123	123	123	0.0		
				Mean	0.1		

Table 5.6: Maximum clavicle length, Voxar measurements. Recorded by AB.

VOXAR									
ID Number	Measurements (mm)			Mean (mm)	WSD	VAR	WSD	95% repeatability
	1	2	3					
1	120	120	121	120	0.2	0.0	0.4	1.2
2	122	122	122	122	0.4	0.1		
3	45	45	45	45	0.2	0.0		
4	45	46	46	46	0.3	0.1		
5	94	93	93	94	0.3	0.1		
6	92	93	94	93	0.7	0.5		
7	81	81	81	81	0.3	0.1		
8	78	78	79	79	0.3	0.1		
9	146	146	146	146	0.2	0.1		
10	86	87	87	87	0.5	0.2		
11	82	82	83	82	0.5	0.2		
12	82	82	83	82	0.8	0.7		
13	107	107	107	107	0.3	0.1		
14	146	147	147	147	1.0	0.9		
15	145	145	144	145	0.5	0.2		
16	83	84	84	84	0.3	0.1		
17	81	82	82	82	0.2	0.0		
18	54	55	54	55	0.3	0.1		
19	55	56	55	55	0.2	0.2		
20	83	83	83	83	0.2	0.0		
21	80	80	80	80	0.2	0.0		
22	77	77	77	77	0.2	0.0		
23	124	123	123	123	0.3	0.1		
24	126	126	127	126	0.8	0.6		
25	41	41	42	41	0.4	0.1		
26	41	41	41	41	0.1	0.0		
27	48	49	48	48	0.4	0.2		
28	50	49	49	49	0.2	0.0		
29	123	123	122	123	0.3	0.1		
30	109	110	110	110	0.6	0.4		
31	112	112	112	112	0.2	0.0		
32	139	141	139	140	0.8	0.7		
33	124	124	124	124	0.5	0.2		
					Mean	0.19		

Table 5.7: Inter-observer error of maximum length of the clavicle, using OsiriX. Measurements recorded by AB and GNR.

OSIRIX							
ID Number	OBSERVER Measurement (mm)		Average (mm)	Error (mm)	SD	VAR	95% CI of Difference
	GNR	AB					
1	120	120	120	0	0.3	0.1	1.1
2	123	123	123	0	0.1	0.0	
3	45	45	45	0	0.1	0.0	
4	45	45	45	0	0.0	0.0	
5	93	94	94	1	0.8	0.6	
6	92	93	92	1	0.6	0.4	
7	80	80	80	0	0.2	0.0	
8	78	77	78	1	0.4	0.2	
9	143	144	144	1	0.8	0.6	
10	85	86	86	1	0.5	0.3	
11	80	80	80	0	0.2	0.0	
12	81	81	81	1	0.3	0.1	
13	105	105	105	0	0.1	0.0	
14	145	145	145	0	0.2	0.0	
15	144	144	144	0	0.1	0.0	
16	81	83	82	2	1.1	1.3	
17	83	84	83	1	0.7	0.5	
18	53	55	54	2	1.1	1.1	
19	54	55	55	2	1.2	1.3	
20	81	83	82	2	1.1	1.2	
21	78	78	78	0	0.2	0.0	
22	76	77	76	1	0.4	0.2	
23	124	124	124	0	0.2	0.0	
24	125	124	124	0	0.3	0.1	
25	39	40	40	1	0.4	0.2	
26	40	40	40	1	0.3	0.1	
27	48	48	48	0	0.0	0.0	
28	48	50	49	2	1.1	1.2	
29	121	122	122	0	0.2	0.1	
30	109	109	109	1	0.4	0.1	
31	111	111	111	0	0.1	0.0	
32	140	139	140	0	0.2	0.0	
33	123	123	123	0	0.2	0.0	
			Mean	0	0.4	0.3	

5.6 Discussion

The results of this study illustrates that there is no significant difference between the measurements taken by PMCT and those by direct osteometric methods. Any variation is likely to be due to inter-observer variability in identifying bony landmarks or alignment techniques. This variability translates to errors with 95% confidence intervals as low as 1.1mm. These results suggest a high degree of accuracy and reliability, which accordingly satisfies the ultimate aim of this study. Virtual measurements showed similar results to osteometric techniques when used to predict age.

To supplement or replace anthropological techniques (as outlined by the BABAO and IFA) in a forensic investigation, the practicality of a virtual approach was assessed. In general, the processing time was almost immediate, allowing practical visualisation and interaction with the 3D image. The time taken to obtain each measurement, however, varied with experience. Initially, all measurements were completed in approximately 20 minutes. However, as the observers experience developed, each clavicle was measured in less than 10 minutes.

Although producing consistently low error rates, a number of problems were encountered during the investigation, which must be considered. The main problem was the lack of any previous standards relating to either osteometric or virtual measurement of the clavicle. Nevertheless, the PMCT protocol developed and used in this study (Appendix 5) proved repeatable by a variety of practitioners from different professional backgrounds (Table 5.7). The majority of the discrepancies occurred measuring the smallest bones, which agrees with Corner and colleagues findings that the overall proportion of error is greater over a smaller distance. Although being relatively large in terms of percentage error, these discrepancies translated to no greater than 2mm, and therefore did not affect age estimations for any of the specimens.

As an integral component of the identification process it is essential that anthropological measurements used for confirming, or refuting the identity of an individual, exhibit precision and maximum accuracy. In mass fatality events that are contaminated by chemical, biological, radiological, or nuclear waste, this process becomes extremely challenging. As a result, PMCT has been highlighted as a potential solution. However, several questions still need to be addressed before this technique can be internationally implemented for human identification. In particular, it is first necessary to establish a standard protocol that is accurate and precise; and replicable between observers. This study has illustrated that anthropological measurements can be successfully obtained using PMCT with levels of accuracy and precision comparable to osteological techniques. The results present compelling evidence that PMCT measurements have a definite contribution in the future of disaster victim identification. However, to further validate this claim, further research into the repeatability of PMCT measurements on different skeletal elements and fully articulated remains is required.

Using the clavicle, this investigation presented a protocol (Table 5.2, Appendix 5) to allow PMCT measurements, comparable or supplemental to traditional osteometric measurements, to be acquired for application to previously published algorithms. This allows the forensic world to take a step forward in standardising the way PMCT is used for forensic practice. Comparison to dry bone is essential as all accepted current methodologies are based on dry bone evaluation. However, there is a significant paucity of juvenile skeletal remains of documented age and so it is vital that the association between dry bone and PMCT compatibility is initially confirmed, to permit reassessment of existing algorithms and the design and development of new ones.

In light of the National Academies Committee on Science (NAS) report on forensic expert evidence and the most recent Law Commission report for England and Wales (LCR, 2011), it is essential that all existing methodologies be tested on appropriate samples and that protocols be developed for new approaches that are based on robust principles and sound scientific analysis.

This investigation is the first step towards validation of the process of conversion from measurement of dry juvenile bone to PMCT compatibility and offers some suggestions for where further work might progress. The clavicle has been shown to be a valid model for these purposes but it cannot be assumed that this will hold true for all other bones in the juvenile skeleton until these areas are also tested, and so this will also require further detailed investigation.

This investigation presents the results from the assessment of the clavicle only. Further PMCT measurement protocols are being considered for other bones to search for measurements that may prove to be of added value in the estimation of biological indicators such as sex and age. Further investigations will be discussed in following chapters, to form a series of studies that will eventually provide guidance on the collection of metric anthropological data via PMCT for the entire juvenile skeleton.

In conclusion, while this study does not offer an absolute resolution for the anthropological measurement of the developing human using PMCT, it provides a platform for all future studies in this area.

Table 5.8: Summary of measurement protocol.

SUMMARY OF MEASUREMENT PROTOCOL	
Step 1 *Base plane*	Generate base plane of the reconstructed bone by identifying the most inferior part of the bone, which would ordinarily lie in contact with the horizontal surface by scrolling through the contiguous image slices.
Step 2 *Sliding planes*	Create two additional boundaries orthogonal to this base plane to simulate the sliding planes of an osteometric board and define the most extreme points.
Step 3 *Virtual osteometric board*	By scrolling through the contiguous slices, maximum precision can be achieved.
Step 4 *Measure*	Now the bone is correctly aligned, all measurements can be recorded in relation to these fixed planes.

CHAPTER 6: THE DENTITION

“The jaws that bite, the claws that catch!”

- Through the looking glass, by Lewis Carroll.

6.1 The Teeth

The dentition is an important feature of the human skeleton for anthropologists, palaeontologists, forensic scientists and odontologists. They are the only skeletal element of a human body that is visible externally and their composition, development and anatomy differs significantly from the rest of the osseous skeleton. Similarly to the majority of mammals, humans are diphyodont; that is, they have two generations of teeth: the deciduous teeth and the permanent teeth. Dental development follows a strict sequence of events, which varies little between individuals. Formation, growth and development occur within the protective casing of the jaw and the teeth are therefore less affected by environment insults than bone, during this period, before they emerge into the oral cavity. In addition, enamel, which forms the majority of the dentition, is extremely durable - commonly lasting long after the decomposition of bone. As a result, the teeth generally survive inhumation more frequently than skeletal structures (Byres, 2008).

The dentition is one of the four irreversible systems within the human body as well as; bone development, secondary sex characteristics and stature, that can be use to determine biological age. Formation of the deciduous dentition initiates very early *in utero,* at approximately 6 weeks and unlike bone, is not subject to the process of remodeling during life. The development and maturation of both sets of teeth spans the full juvenile life span, from neonate to early adulthood (Scheuer and Black, 2000), although changes in the dentition also occur, through tooth loss, throughout an individual's entire life (Byres, 2008). The extremely early initiation of development, combined with the late completion of permanent tooth formation and then loss, contribute to the dentition being arguably the most accurate and reliable indicator of age-at-death, particularly in immature individuals.

6.1.1 Tooth anatomy and development

The tooth consists of a crown, covered by a very hard translucent layer of enamel, a dentine body, and a root covered by bone-like cementum. The three hard tissues that compose the tooth, dentine, enamel and cementum, are all different in composition to bone. The root and crown meet at a point known as the tooth neck or cervical margin. The dentine surrounds a central pulp cavity which expands at its coronal end into the pulp chamber, and narrows in the root to form the pulp canal which opens near the root tip by one (or occasionally multiple) foramen. At the apical foramen the connective tissue pulp, which contains sensory nerves and vessels to support the dentine, is continuous with the periodontal ligament. The periodontal ligament runs from the root apex to the alveolar bone to anchor the tooth in place (Figure 6.1).

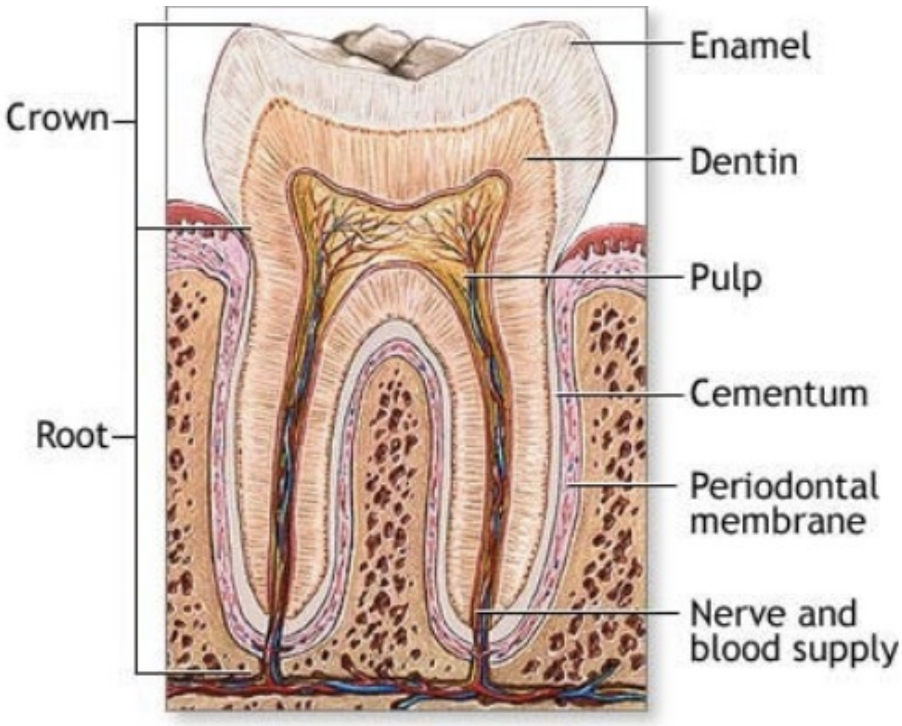


Figure 6.1: Tooth anatomy (A.D.A.M, 2014).

Each tooth develops within the alveolar bone of the jaw from a soft tooth germ that is subsequently mineralised. Early indications of dental development are visible in the embryonic period before the nose and mouth cavities are even completely separated by the formation of the secondary palate.

Thickenings of the ectodermal epithelium form the primary epithelial bands in both the upper and lower jaws. These proliferate to form the dental and vestibular laminae, which gives rise to the teeth and surrounding soft tissue cavity, respectively. Tooth dentine is the first to develop, followed closely by the tooth enamel.

The enamel organs and ameloblast cells, in the inner layer of the primary epithelial bands, induce the outer layer of mesenchymal papilla to differentiate into odontoblasts, which form the dentine. Once the dentine begins to form the enamel organs produce a layer of enamel, which builds a protective layer over the tip of the dentine structure. The rest of the dentine is covered with a layer of epithelial cells, known as Hertwigs epithelial root sheath. In the molars, some of these cells invaginate to produce their distinctive multi-rooted structure (Scheuer and Black, 2000).

Before eruption commences, that is the migration of the tooth from the alveolar crypt to its functional occlusal position in the mouth, the crown and between one-third and one-quarter of the root has already formed. The remainder of root growth, including the closure of the apex foramen, continues after emergence (Figure 6.2).

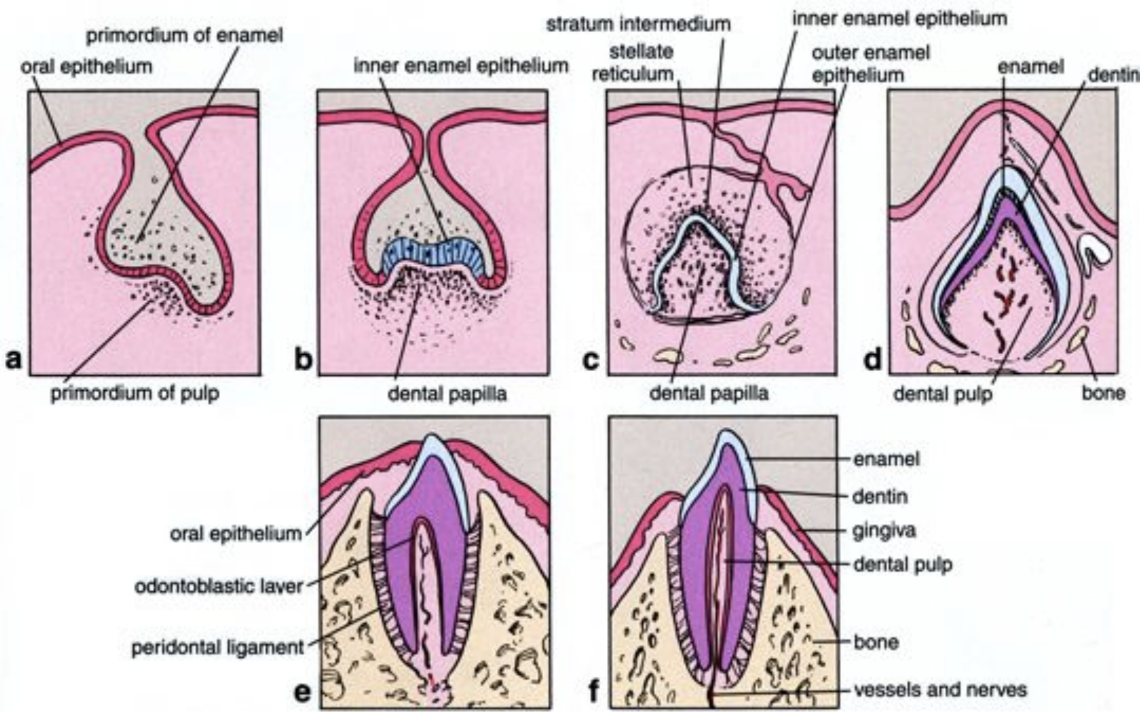


Figure 6.2: Tooth development (A.D.A.M, 2014).

6.1.2 Mineralisation sequence

The sequence of prenatal mineralization of the deciduous dentition begins with the central incisors; followed by the first molar, lateral incisors, canines, and then the second molar. The maxillary central incisors and first molars are usually seen before those in the mandible. The lateral incisors appear first in the maxilla, but subsequent development of the canines and second molars is ahead in the mandible. By birth, all the deciduous teeth and the first permanent molars have initiated mineralization and by approximately three years of age the root formation of the deciduous teeth is complete and they have emerged into the oral cavity. The permanent first molar, incisors and canines begin to mineralise during the first year and the premolars and second molars form between two and four years of age. The third molar starts to mineralise between 6 and 12 years of age (Scheuer and Black, 2000). The emergence of the permanent dentition, except for the third molar, occurs in two stages (between 6-8 years and 10-12 years) (Figure 6.3).

6.1.3 Terminology

In both the upper and lower jaw the teeth are arranged in a dental arch or arcade. Each half of each jaw contains the same number of teeth and is referred to as the left or right, upper or lower quadrant. There are normally 20 teeth in the deciduous series and 32 teeth in the permanent (Figure 6.3). Both the deciduous and permanent dentition contains two incisors and a canine tooth in each quadrant. In the deciduous series, behind these anterior teeth lie two molars (Byres, 2008). These are replaced in the permanent series by two premolars and normally completed posteriorly by an additional three molars. Each tooth has four surfaces: libel, buccal, palatal and lingual, named according to their position relative to the midline of the mouth and to the lips, cheeks, palate and tongue, respectively.

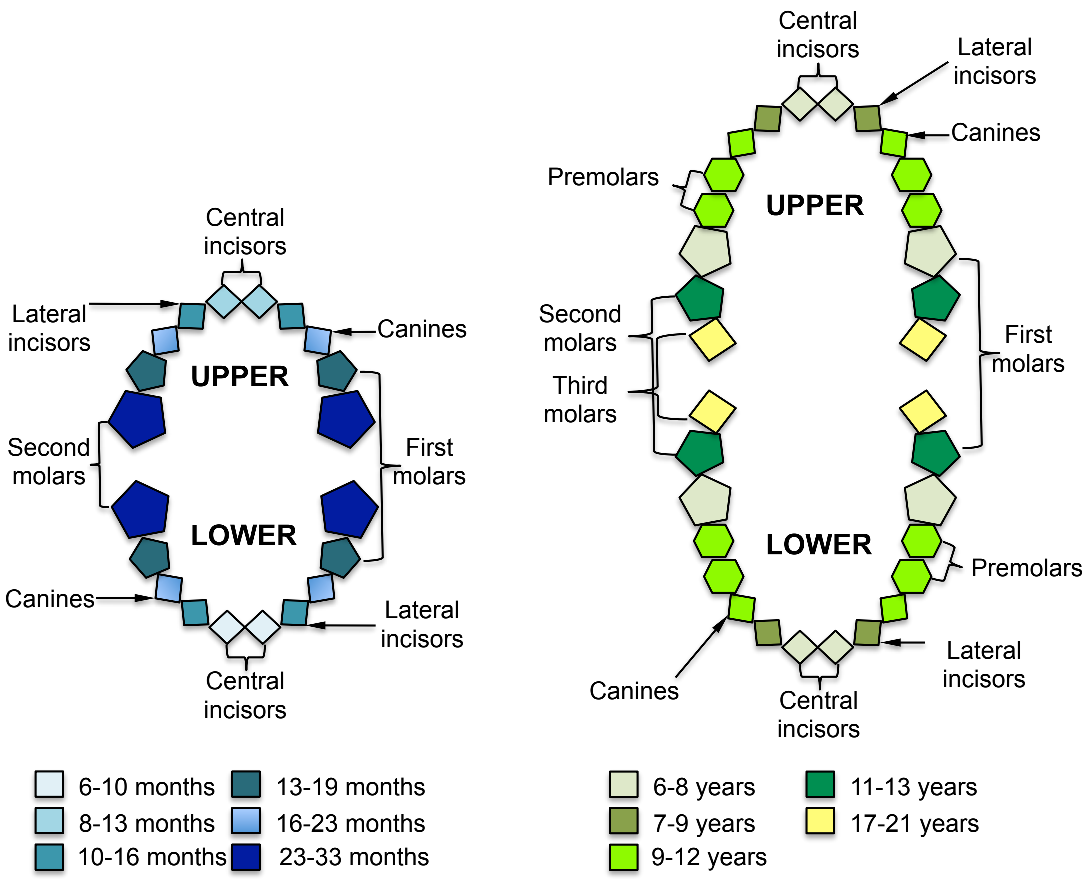


Figure 6.3: Tooth eruption chart. Deciduous (Left) and permanent (Right).

6.1.4 Radiographic appearance

Some of the most frequently used methods for age estimation involve the assessment of dental development as seen on radiographs. It is therefore important to recognise the normal appearance of tooth development stages, as captured in X-rays. Enamel is the most radiopaque tissue of the tooth and therefore the crown, or developing crown can be distinguished from the rest of the tooth as the whitest structure. Cementum and dentine are less radiopaque and so appear grey. The cementum layer is extremely thin and is a similar density to dentine. The two structures can therefore not easily be distinguished from each other (Weems, 2010). The periodontal ligament and pulp cavity are radiolucent so appear dark in radiographs. The lamina dura and alveolar crest of the alveolar bone are also distinguishable. The lamina dura is a thin layer of compact bone, which surrounds the tooth socket, and the alveolar crest is the gingival margin of the alveolus (Figure 6.4).

Figure 6.4: Radiograph of mandible and maxilla.

6.2 Dental Ageing Methods

Under the United Nations Universal Declaration of Human Rights, retaining an identity after death represents a basic human right (UDHR, 2012). When dealing with juvenile remains, age-at-death is an important criterion and odontological examination is arguably the most rapid and practical method available for this purpose. Dental age markers are reported to correlate more strongly with chronological age than skeletal markers, being less affected by the environment (Scheuer and black, 2000). Dental techniques are predominantly based on the degree of mineralization and/or eruption of the dentition and involve matching radiographs to atlases of dental development, or assigning formative stages of mineralization by scoring individual teeth.

Tooth formation is superior to tooth emergence for assessing dental maturation because the majority of teeth can be studied at each examination. In contrast, emergence is only a specific short duration phase in the continuous process of eruption, rarely observed for more than one or two teeth at a time, if at all.

Emergence is also influenced markedly by environmental factors such as loss of deciduous predecessors and the lack of space in the dental arch, explaining some part of the variation in root length at emergence.

Strictly speaking, eruption is the continuous process by which teeth move from the alveolar bone to full occlusion in the mouth. However, this process can be divided into a number of stages that are visible as bouts of emergence separated by more inert periods. Most eruption age determination methods are actually confined to emergence.

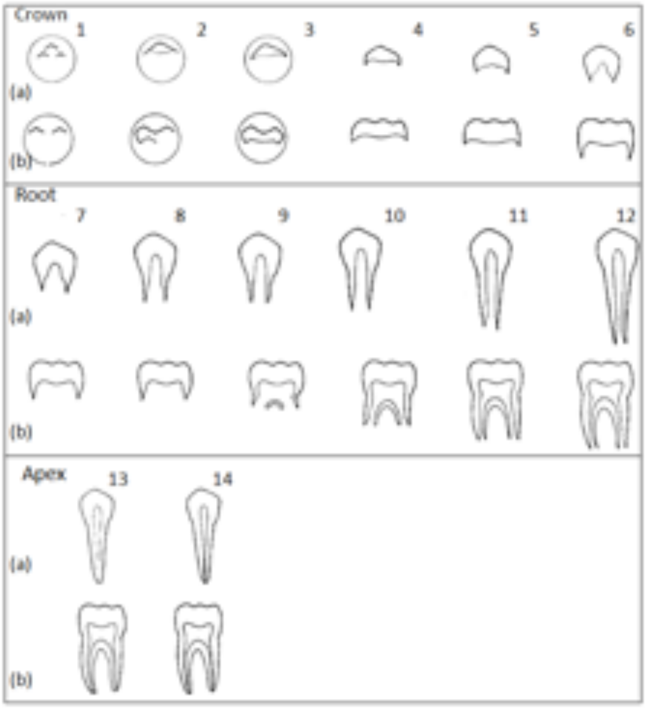
Increased accuracy could be achieved by analysing dental microstructure or calcification. However, these techniques are extremely time consuming, expensive and require expert knowledge to interpret results.

6.2.1 Mineralisation

The process of tooth formation involves the deposition of enamel and osseous material, initiating at the tooth cusp and proceeding towards the roots. Although this process is continuous, the most widely implemented methods developed by Demirjian *et al*., (1973) and Moorrees and colleagues (Moorrees *et al*., 1963), for convenience, divide tooth formation into a number of stages based on the level of calcification (Figure 6.5). Although every tooth passes through each stage of development, the timetable by which this happens varies depending on the type of tooth in question. For example, the deciduous teeth mineralise in advance of their permanent counterparts, the incisors before the molars, and so on. In order to estimate the age of an individual, the level of mineralisation for each tooth is matched with the appropriate stage illustration, which is in turn matched with a time range; developed from samples of known age at death.

Dimirjian *et al.*'s (1973) method, commonly referred to as "Demirjian's Technique" is one of the most widely used methods of forensic age estimation. This technique charts the changes from initial calcium deposition to complete apex formation by dividing this process into eight observable stages. The technique is applied to panoramic radiographs and involves marking specific features of 7 teeth. The individual "score" from each tooth is then added together to provide an estimate of dental maturity on a scale of 0 to 100. This score is then referenced on a corresponding conversion chart with ages ranging from 3 to 17 years, in increments of one-tenth. For this technique to work successfully all seven teeth need to be rated. In many forensic cases, one or more teeth are regularly missing. Although there has been evidence that Demirjian's method has a tendency to over-estimate age (Willems *et al*., 2001), the technique remains a well-regarded forensic tool for determining the age of sub-adults. As with all estimation methods, there is a discernable margin of error, but overall the technique still provides an advantage over other dental methods, especially those based on tooth eruption, which can be highly influenced by health and environmental factors.

The mineralisation status of the deciduous teeth and first permanent molars has been tested against other methods of gestational age. Lemons *et al*. (1972) confirmed that fetal age could be determined with greater accuracy from dental development rather than skeletal development, by correlating mineralisation of the teeth with the appearance of the distal femoral epiphyses.



Standard abbreviations of tooth formation stages

1	C_i	Initial cusp formation
2	C_{co}	Coalescence of cusps
3	C_{oc}	Cusp outline complete
4	$Cr_{1/2}$	Crown half complete
5	$Cr_{3/4}$	Crown three-quarters complete
6	Cr_c	Crown complete
7	R_i	Initial root formation
8	Cl_i	Initial cleft formation
9	$R_{1/4}$	Root length quarter
10	$R_{1/2}$	Root length half
11	$R_{3/4}$	Root length three-quarters
12	R_c	Root length complete
13	$A_{1/2}$	Apex half closed
14	A_c	Apical closure complete

Stage	I1	I2	C	PM1	PM2	M1	M2	M3
Female								
C_i	-	-	0.6	2.0	3.3	0.2	3.6	9.9
C_{co}	-	-	1.0	2.5	3.9	0.5	4.0	10.4
C_{oc}	-	-	1.6	3.2	4.5	0.9	4.5	11.0
$Cr_{1/2}$	-	-	3.5	4.0	5.1	1.3	5.1	11.5
$Cr_{3/4}$	-	-	4.3	4.7	5.8	1.8	5.8	12.0
Cr_c	-	-	4.4	5.4	6.5	2.4	6.6	12.6
R_i	-	-	5.0	6.1	7.2	3.1	7.3	13.2
Cl_i	-	-	-	-	-	4.0	8.4	14.1
$R_{1/4}$	4.8	5.0	6.2	7.4	8.2	4.8	9.5	15.2
$R_{1/2}$	5.4	5.6	7.7	8.7	9.4	5.4	10.3	16.2
$R_{2/3}$	5.9	6.2	-	-	-	-	-	-
$R_{3/4}$	6.4	7.0	8.6	9.6	10.3	5.8	11.0	16.9
R_c	7.0	7.9	9.4	10.5	11.3	6.5	11.8	17.7
$A_{1/2}$	7.5	8.3	10.6	11.6	12.8	7.9	13.5	19.5
A_c	-	-	-	-	-	-	-	-

Stage	I1	I2	C	PM1	PM2	M1	M2	M3
Male								
C_i	-	-	0.6	2.1	3.2	0.1	3.8	9.5
C_{co}	-	-	1.0	2.6	3.9	0.4	4.3	10.0
C_{oc}	-	-	1.7	3.3	4.5	0.8	4.9	10.6
$Cr_{1/2}$	-	-	2.5	4.1	5.0	1.3	5.4	11.3
$Cr_{3/4}$	-	-	3.4	4.9	5.8	1.9	6.1	11.8
Cr_c	-	-	4.4	5.6	6.6	2.5	6.8	12.4
R_i	-	-	5.2	6.4	7.3	3.2	7.6	13.2
Cl_i	-	-	-	-	-	4.1	8.7	14.1
$R_{1/4}$	-	5.8	6.9	7.8	8.6	4.9	9.8	14.8
$R_{1/2}$	5.6	6.6	8.8	9.3	10.1	5.5	10.6	15.6
$R_{2/3}$	6.2	7.2	-	-	-	-	-	-
$R_{3/4}$	6.7	7.7	9.9	10.2	11.2	6.1	11.4	16.4
R_c	7.3	8.3	11.0	11.2	12.2	7.0	12.3	17.5
$A_{1/2}$	7.9	8.9	12.4	12.7	13.5	8.5	13.9	19.1
A_c	-	-	-	-	-	-	-	-

Figure 6.5: Stages of mineralisation; in the development of the crown, root and apex of permanent teeth and charts for age determination as defined by Moorrees *et al.* (1963).

6.2.2 Dental eruption

When the roots of the teeth are approximately one-half to three-quarters of their maximum length, the teeth emerge from their crypts into the mouth. Strictly speaking, eruption is the continuous process by which teeth move from alveolar bone to full occlusion in the mouth. However, as previously discussed, most studies are confined to emergence times, often wrongly referred to as eruption, which can be studied without expert equipment (Figure 6.6). The most commonly used dental atlases are by Schour and Massler (1941), Ubelaker (1978), and more recently AlQahtani *et al*. (2010). The latter, has been developed on a modern London population and is therefore probably the most applicable in current practice. Comparing an individual with a set stage in an atlas is easy and rapid and therefore considered more practical in some fieldwork or forensic situations. However, difficulties can occur with matching, when numbers of teeth, or sequencing between the two, do not match. In addition, due to the specific phase of short duration on which dental emergence techniques relay, tooth mineralisation methods are considered superior, yielding more accurate estimations of age.

6.2.3 Dental microstructure

Dental microstructure is able to provide an even more accurate method of age determination, by counting the perikymata (the incremental lines on the surface of enamel). This is an absolute method of age determination independent of the growth standards of a specific population so could prove to be relevant in individual forensic cases. However, this method is extremely time consuming, expensive and requires specialist expertise.

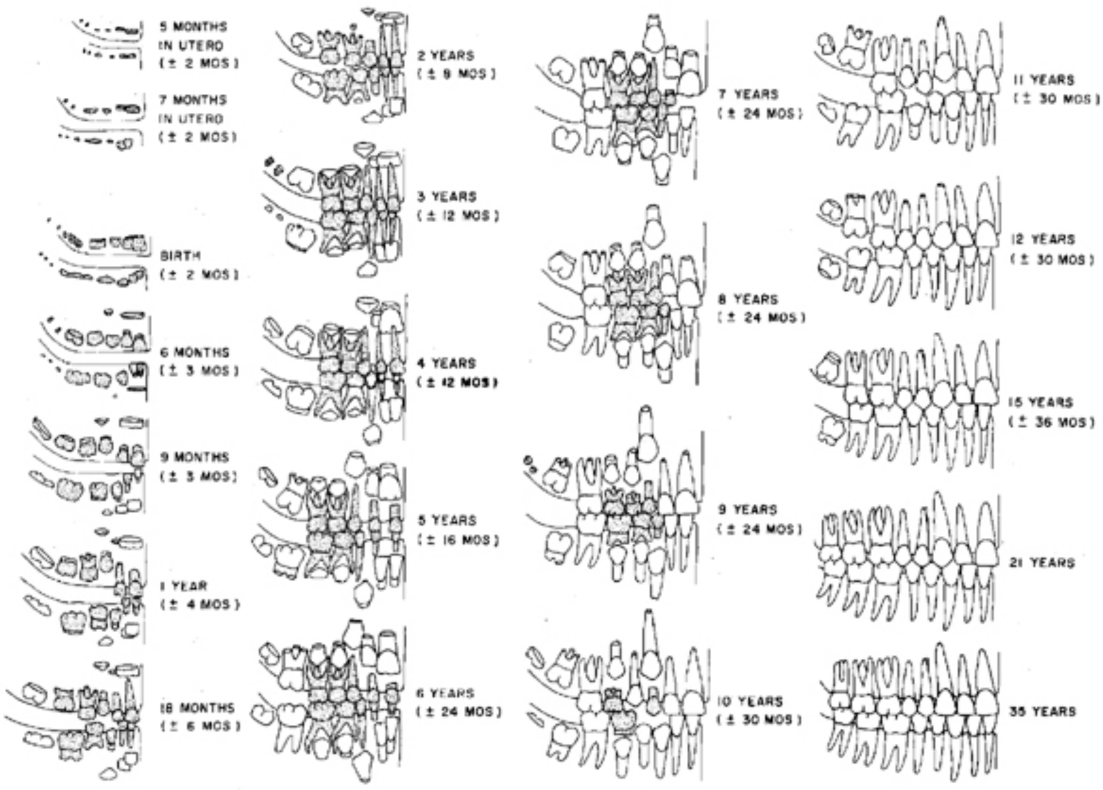


Figure 6.6: Ubelaker (1978) development chart for teeth; Age determination.

6.2.4 Dental markers

There are reportedly at least two billion possibilities in the charting of adult dentition (Hardy, 2007). It is therefore hardly surprising that dental markers are extremely important in human identification. As with fingerprints, even identical twins do not necessarily have identical dentition (Pretty and Sweet, 2001). It is highly improbable that both tooth morphology, and restoration would be the same in both individuals. In contrast to fingerprints however, where there is a deficiency in a comprehensive database, accessibility to reliable ante-mortem dental records is generally much greater, in the UK at least. As well as extensive variation, identifiable dental features and restorations are regularly the only surviving parts of a body, following severe fragmentation or decomposition (Hardy, 2007). For this reason, dental structures have always played, and will continue to play, a key role in both criminal and mass disaster events (Pretty and Sweet, 2001). In addition, as there is no minimum number of concordant points or features that must be establish for a positive identification, a single tooth may be adequate, assuming it contains sufficient features to make it ‘unique’ to a single individual (Pretty and Sweet, 2001). This is a huge advantage over other identification markers, especially in a mass fatality situation where the body can be severely disarticulated. However a single tooth does not necessarily equate to the death and therefore a full or partial dental arcade is preferable.

The primary restriction of dental markers in identification is the quality and availability of ante-mortem data for comparison. This was particularly evident in the Asian tsunami (2004), where a much higher success rate was achieved for foreign victims than for Thai victims (Schuller-Gotzburg and Suchanek, 2006). In this particular event, high temperatures and an aqueous environment resulted in poor preservation of other identification markers, namely DNA and fingerprints, and led to dental evidence playing a vital role and subsequently highlighting the importance of good quality, accurate and comprehensive ante-mortem records (Huckenbeck *et al*., 2008). However, dental records may not exist at all, if the victim has never attended a dental practitioner (Pretty and Sweet, 2001). Additionally, enormous variation in charting systems and dental charts further limits the utility of this marker. Although the majority of forensic odontologists prefer Federation Dentaire International (FDI) notation there is, at present, no worldwide standard ante-mortem charting and subsequently many records lack detail or are illegible.

6.2.5 Dental restorations

Dental restoration markers add to the identification power of a dental comparison. Although, these are less useful for individuating children, adolescents, and edentulous persons, who will have undergone no or very little dental treatment and therefore have less ante-mortem information available for them (Pretty and Sweet, 2001). However, this information can still be used for identification and is becoming an increasingly valuable source of data as the population becomes more educated about dental health (Hardy, 2007). If ante-mortem records are not available and there is no presumptive identity, post-mortem profiling can still be carried out by the forensic odontologist, to narrow possible identities by estimating age, sex and even ethnicity. A number of additional features (Table 6.1) have also led to the development of novel new approaches for identification including; labelled dental prosthesis, orthodontic braces, and comparison of palatal rugae patterns on dental casts to those of the victim (Pretty and Sweet, 2001).

Table 6.1: Dental features used for identification of human remains. (Adapted from; Pretty and Sweet, 2001).

Features examined during the comparative dental identification. This extensive list represents the complexity of these cases, particularly in those instances in which restorative treatment is absent or minimal.

TEETH

Teeth present
a. Erupted
b. Unerupted
c. Impacted

Missing teeth
a. Congenitally
b. Lost antemortem
c. Lost postmortem

Tooth type
a. Permanent
b. Deciduous
c. Mixed
d. Retained primary
e. Supernumerary

Tooth position
a. Malposition

Crown morphology
a. Size and shape
b. Enamel thickness
c. Contact points
d. Racial variations

Crown pathology
a. Caries
b. Attrition, abrasion, erosion
c. Atypical variations, enamel pearls, peg laterals etc
d. Dentigerous cyst

Root morphology
a. Size
b. Shape
c. Number
d. Divergence of roots
Root pathology
a. Dilacerations
b. Root fracture
c. Hypercementosis
d. Root resorption
e. Root hemisections

Pulp chamber/root canal morphology
a. Size, shape and number
b. Secondary dentine

Pulp chamber/root canal pathology
a. Pulp stones, dystrophic calcification
b. Root canal therapy
c. Retrofills
d. Apicectomy

Periapical pathology
a. Abscess granuloma or cysts
b. Cementomas
c. Condensing osteitis

Dental restorations
1. Metallic
a. Non-full coverage
b. Full coverage
2. Non-metallic
a. Non-full coverage
b. Laminates
c. Full coverage
3. Dental implants
4. Bridges
5. Partial and full removable prothesis

PERIODONTAL TISSUE

Gingival morphology and pathology
a. Contour, recession, focal/diffuse, enlargements, interproximal craters
b. Colour – inflammatory changes, physiological (racial) or pathological pigmentations
c. Plaque and calculus deposits

Periodontal ligament morphology and pathology
a. Thickness
b. Widening
c. Lateral periodontal cysts and similar

Alveolar process and lamina dura
a. Height, contour, density of crestal bone
b. Thickness of interadicular bone
c. Exostoses, tori
d. Pattern of lamina dura
e. Bone loss
f. (horizontal/vertical)
g. Trabecular bone pattern and bone islands
h. Residual root fragments

ANATOMICAL FEATURES

Maxillary sinus
a. Size, shape, cysts
b. Foreign bodies, fistula
c. Relationship to teeth

Anterior nasal spine
a. Incisive canal (size, shape, cyst)
b. Medium palatal suture

Mandibular canal
a. Mental foramen
b. Diameter, anomalous
c. Relationship to adjacent structures

Coronoid and condylar processes
a. Size and shape
b. Pathology

Temperomandibular joint
a. Size, shape
b. Hypertrophy/atrophy
c. Ankylosis, fracture
d. Arthritic changes

Other pathologies
a. Developmental cysts
b. Salivary gland pathology
c. Reactive/neoplastic
d. Metabolic bone disease
e. Focal or diffuse radiopacities
f. Evidence of surgery
g. Trauma – wires, surgical pins etc.

6.3. Forensic Dental Radiography

The first non-diagnostic dental radiographs were made within days of Röentgens discovery of the X-ray. However, the first published use of dental radiography to make a successful identification was not until 1943. Today radiography remains the most practical and reliable tool for human identification from dental information. Dental radiographs are even held by most, in higher regard than written notes, if there are any discrepancies between the two, having less potential for human error than charted dental information (Carvalho *et al*., 2009).

6.3.1 Intraoral radiography

Intraoral radiographs including, periapical, bitewing, and occlusal radiographs are all techniques regularly used in general dental practice. Intraoral views are created by placing a radiographic film or sensor inside the mouth. A periapical view is taken of both anterior and posterior teeth and is used to allow the dentist to view the entire tooth (from crown to root) as well as the surrounding bone. In contrast, a bitewing view is taken predominantly to visualize the crowns of the posterior teeth and the height of the alveolar bone in relation to the cementoenamel junctions (the demarcation line which separates the tooth crown from the tooth root). Routine bitewings are generally taken to examine dental caries in the molars; they are not commonly used to view the anterior teeth. An occlusal view is used to obtain information pertaining to the skeletal or pathological anatomy of either the floor or upper palate of the mouth. This technique uses an occlusal film, which is roughly three to four times the size of the film used for a periapical or bitewing image. The film is exposed from either under the chin or angled down the tip of the nose, to produce an image in the axial rather than sagittal plane.

Although dental radiography is moving in a digital direction, intraoral images can be invaluable if a patient's anatomic variations obstruct the possibility of obtaining a clear OPT. In addition, film is inexpensive, easy to place, and produces an exceptional image if proper techniques are used. The draw back is that it requires several minutes of chemical processing, ideally in a dark room setting. In addition, heat, light, pressure, and overactive chemicals can fog film. This can make it an impractical solution for a mass fatality event, when working conditions can be unpredictable.

Direct digital sensors (CCD and CMOS) provide an immediate image with no chemical processing or dark room required. However, digital sensors are delicate, expensive, in some cases difficult to place, and require enormous amounts of digital storage for the resultant image.

6.3.2 Panoramic radiography

An OPT is a radiographic image of the entire mouth, including upper and lower jaw and teeth. The X-ray machine moves around the head while the image is being captured; this allows a plane of interest to stay 'in plane' relative to the film whilst all other structures are 'blurred' out. This provides a two dimensional complete ear-to-ear image of the full dental arcade. This imaging technique is the most frequently used in current forensic practice. It is an extra-oral method in which both revolving X-ray source and radiographic film or sensor are external to the oral cavity. It is also used regularly in general dental practice to look at impacted wisdom teeth, plan orthodontic treatment and help find the cause of dental pain.

Dental panoramic radiography equipment consists of a horizontal rotating arm, which hold an X-ray source and a moving film mechanism (carrying film) arranged at opposed extremities. The patient's skull sits between the X-ray generator and the film. The X-ray source is collimated to produce a beam shaped as a vertical blade, with a width of 4-7mm when arriving on the film, after crossing the patient's skull. The height of the beam is set to cover the complete mandible and the maxilla regions. The arm moves in a motion that can be described as a rotation around an instant centre, which shifts on a dictated trajectory.

Although this technique produces images that are considered high quality, the dental panoramic image suffers from important distortions due to differential variation of the vertical and horizontal zoom along the image. The vertical and horizontal zooms are determined by the relative position of the recorded elements versus the film and generator. Features closer to the generator receive more vertical zoom. The horizontal zoom is also dependent on the relative position of the element to the focal path. Features inside the focal path arch receive more horizontal zoom and are blurred; features outside receive less horizontal zoom and are blurred. The result is a sharp image along the central portion that contains the mandible arch, and blurred image elsewhere. This can potentially cause an issue if a patient's anatomical variation differentiates far from the norm. However, since modern OPT is a digital technology, the image benefits from post-processing (contrast alterations, zooming, positive-negative inversions) that can mimimise extensive motion, or zoom blurring.

Particular preparation of the patient is not necessarily required to carry out a correct OPT; however, it is absolutely necessary to remove all that could distort the final image (i.e. earrings, dentures, hairpins, etc.). The correct technique also entails the insertion of a removable clamp between the central incisors margin to allow the elements of the inferior and superior arcades to be placed in the same line in a vertical position. This is necessary to ensure that the complete jaw falls into the tomographic plane, to produce an optimal radiological representation. For the same reason, it is also important to place the head on a provided base or submental support plane, to accurately regulate the head position. Although this is simple in general practice, where the patient is able to follow instructions to manipulate their own position, in a forensic situation, this process can be labourious and might not be possible in cases of extreme rigor or anatomical disruption. However, handheld X-ray-generating devices may address many of these issues and have come into common use in forensic dentistry, particularly in a multiple fatality incident morgue. For example, a handheld X-ray system was used to produce all of the radiographic images in the Disaster Mortuary Operational Response Teams (DMORT) response to hurricane Katrina.

6.3.3 Dental CT

OPTs may be difficult to obtain in a forensic context, due to a lack of access to equipment or difficulties with the revolving nature of the image acquisition due to severe trauma or rigor mortis. Computed tomography (CT) could therefore aid, or potentially replace, this more traditional imaging method by both reproducing and augmenting the information available from an OPT. The movement towards CT has been driven by increasing availability and affordability, and the development of spiral and multiple detectors CT (MDCT), which has improved scanning speeds and resolution, allowing high quality image reconstructions in multiple planes and 3D modelling of slices (Verhoff *et al*., 2008). A single image acquisition using PMCT allows teeth and bones to be assessed in any plane without invasive procedures, offering considerable practical and aesthetic benefits. Used in conjunction with a tele-radiogy system, such as the FiMAG system (Rutty *et al*., 2009), it would also allow secure global distribution and evaluation of images used for identification purposes. In addition, computer algorithms correct for the geometric distortion that are present within all plane film images. Thus, there is no magnification and the final images are a 1:1 representation of the maxillofacial structures.

Although the availability of PMCT for forensic examinations is not wide spread, if a ‘one stop’ protocol could be produced for the examination and identification of the entire human skeleton using PMCT, this could potentially replace all existing image techniques currently used. A particular advantage of MDCT dental evaluation is that it does not need the placement of image receptors into the mouth, required for intra-oral radiography, nor manipulation of the head to align the X-ray beam as required for OPT. It is also possible to recreate any intraoral or extraoral film image view from one scan, with the operator having the ability to select any desired slice location and orientation. However, there are a number of disadvantages to using PMCT, due to increased sensitivity to metal artefact. CT reconstruction algorithms can now accommodate for metal implants such as Titanium but high-density metals such as gold or mercury in dental restorations are still a problem.

Cone beam CT (CBCT) is also being increasingly used in dental practice, primarily for treatment panning and diagnosis in implant dentistry. It gives similar (but not superior) 3D dental assessment to PMCT. This advanced digital imaging system was first introduced into dental practice in 2001 and is termed cone beam because the beam is not thin or fan shaped such as that used in typical medical CT units. While the enhanced imaging opportunities offered by CBCT fosters its increasing application in dental radiography, technical limitations somewhat hinder its wide scale practical integration into the field. Compared with PMCT, the wider collimation (spread of the beam) in CBCT leads to increased scatter radiation and degradation of image quality demonstrated by artefacts and decreased contrast-to-noise ratio. The temporal resolution of caesium iodide detectors in CBCT slows data acquisition time to approximately 5 to 20 seconds, which increases motion artefact (blurriness caused by even slight motion of the object being scanned). The time required for image reconstruction therefore takes longer for CBCT compared to PMCT due to the computationally demanding cone beam reconstruction algorithm. In addition CBCT cannot be practically used to image the whole skeleton and so could not provide a 'one stop' protocol for forensic practice. Unlike PMCT, that could potentially replace all other imaging modalities currently used, CBCT would need to be used in conjunction with other modalities.

Most practitioners agree that direct digital imaging systems such as OPT and PMCT, are the best image modalities to use in a mass fatality incident - due to providing an immediate image and dealing effectively with the exaggerated flow of victims in the morgue setting.

6.4 Aims and Objectives

For PMCT to become routinely implemented in forensic practice, evidence is required that age estimations using this virtual approach have comparable accuracy and repeatability to methods using conventional OPTs. Although PMCT offers the potential to develop CT-specific techniques that could ultimately enhance forensic practice, at this stage it is only necessary to provide evidence that PMCT is equal to, not necessarily superior to OPT, and therefore could be used instead of, or interchangeably with conventional OPT. The primary aim of this investigation was therefore:

- To determine if age estimations using PMCT curved MPR images, were in agreement with those made on traditional OPT images.

In addition, observers with different professional backgrounds and levels of expertise of using both PMCT and traditional OPT were chosen for this investigation to consider whether this had any effect on the results produced by each observer.

6.5 Methods and Materials

6.5.1 Selection of cases

Nineteen mandibles and maxillae from individuals of known age (between 0-18years) were randomly selected from the Scheuer Juvenile skeletal collection (see Section 3.2.1) for this study by an independent observer. Although the demographics of some of the specimens in the Scheuer juvenile skeletal collection have been estimated, the cases used in this investigation were all selected from the skeletons in the collection that had a know, not estimated, biological profile associated. OPTs were taken of each specimen at the university of Dundee dental school, by a trained dental radiographer. The images were then saved to an external hard drive and transferred to the image archive at the East Midlands pathology unit, before being distributed to the participating assessors.

Each skull was subsequently PMCT scanned, using a mobile truck mounted SOMATOM® Emotion 16-detector CT scanner (Siemens AG Medical Solutions). Scans involved helical acquisition using a 0.75mm slice thickness, 120kVp, and 100mA with bone and soft tissue reconstructions at 1.25mm. Data were stored on compact disc and transferred to the same secure image archive as the OPT images, before being distributed to the group.

6.5.2 Curved MPR protocol

Image analysis for the PMCT data was undertaken using OsiriX imaging software system (OsiriX version 3.7.1; distributed freely as open-source software under the GNU licensing scheme at the following Web site: http://homepage.mac.com/rossetantoine/osirix).

Curved multi-plane reconstructions (curved MPRs) were created from the PMCT data set using the *3D MPR function* on the OsiriX workstation, so that dental analysis could be undertaken in the x-, y-, and z- planes. Although the resulting images were MPRs and therefore not strictly speaking 3D, they were created using the entire volume data set, by drawing a region of interest (ROI) path, which is stretched to produce a straightened view of the curved jaw. This is similar to the process used to create OPT images, but has the added advantage of being able to view the third dimension of volume, by scrolling through multiple slices of the same object on a specific plane, in real time. This software platform also allows the user to apply pre-set contrast and colour intensity windows or manual contrast tools so that specific areas of interest can be enhanced, for optimum viewing. A step-by-step guide of how replicate the image acquisition technique used in this investigation is presented in (Appendix 6).

In this study I processed all the PMCT data. A series of curved MPR images, which replicated the counterpart OPT, were produced for each specimen then distributed to the other examiners. This approach was undertaken for a number of reasons, principally:

1) To avoid any complications associated with transferring large quantities of PMCT data between different research facilities.

2) To minimise post-processing times, by having only one investigator using the raw PMCT data to create the curved MPRs.

3) To minimise introduced error, which could have caused a negative bias against PMCT. Since all the investigators were using the same OPT images, it was considered appropriate for all researchers to also consider the same curved MPR images. This was deemed a more equal comparison of the two approaches.

Although the PMCT image protocol used in this investigation was specifically designed to replicate conventional OPTs, to allow a direct like-for-like comparison of imaging modalities, it was still considered important to reflect the benefit of having 3D PMCT data compared to 2D OPT data. This was achieved by creating and distributing a series of curved MPRs, to accompany the single OPT image, for each case. All observers were therefore given a single OPT, and a series of 3 or 4 sequential curved MPR images for each case. Examples of a typical OPT and OsiriX curved MPRs, are shown in Figure 6.7.

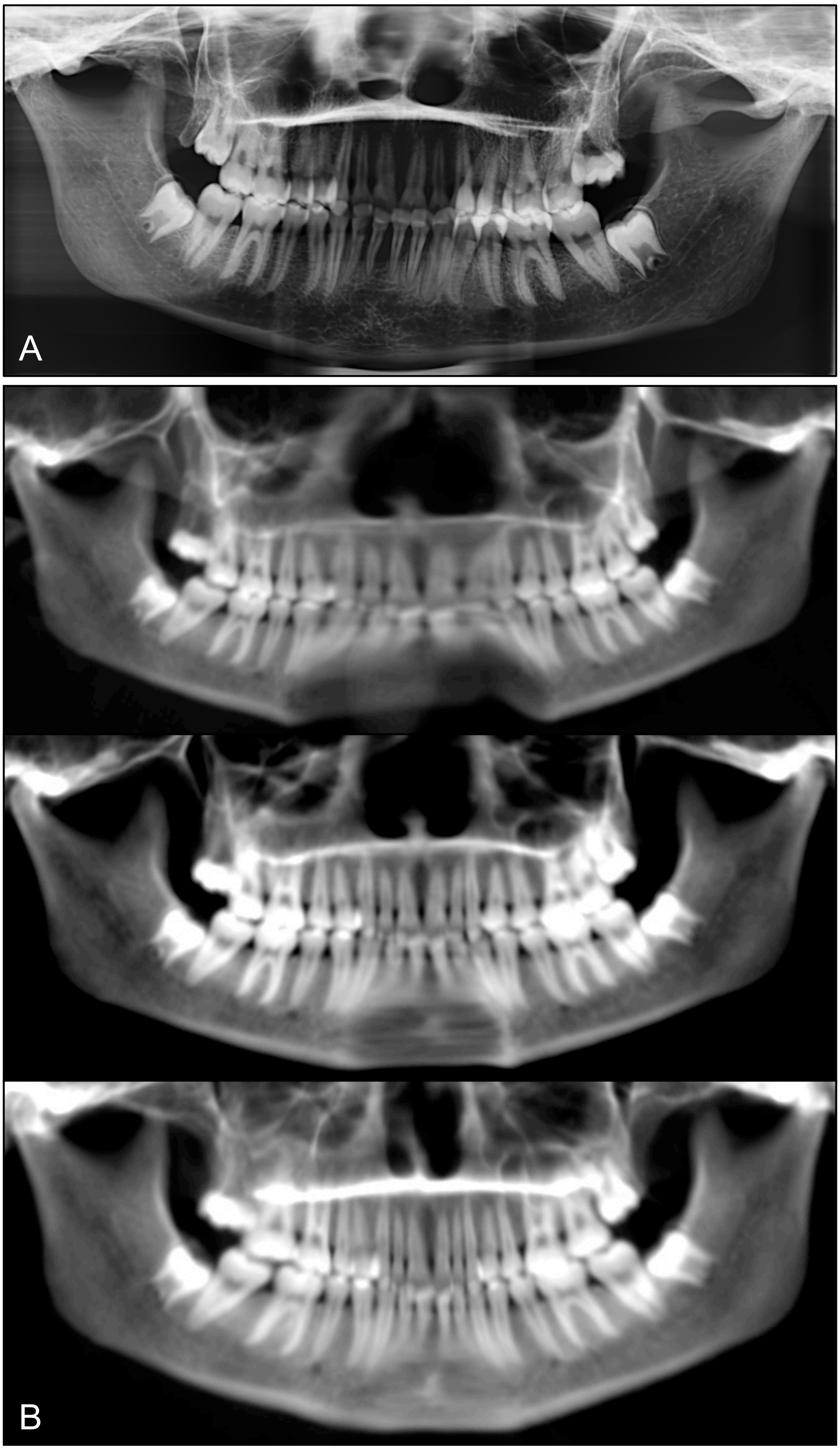


Figure 6.7: Traditional dental radiography Vs PMCT. A) Conventional Orthopantomograph (OPT) image B) Series of PMCT curved MPRs (Case 16, 18 years).

6.5.3 Dental techniques

This investigation will use the dental age techniques most frequently used in general forensic odontological practice in the UK at present; the London Atlas of Human Tooth Development and Eruption (QMUL, Figure 6.9) and Demirjian's developmental stages of mineralisation (Figure 6.8) (AlQahtani *et al.*, 2010; Anonymous, 2012; Demirjian and Goldstein, 1976; Demirjian *et al.*, 1973). The QMUL method is an adaptation of Ubelaker's (1978) atlas (Ubelaker, 1978), using data from modern UK population. It is therefore more suitable for our investigation, as it has been constructed on a demographic that is more closely comparable to our reference collection. In addition, the QMUL method includes more comprehensive illustrations of dental structures such as tooth roots, contains a larger range of age stages, and eruption times that more accurately refer to emergence from alveolar bone instead of from the gingiva; than comparable atlas techniques such as Ubelaker's (1987) (Figure 6.6).

Although Demirjian's technique has been developed on a French-Canadian population from the early 1950's, it is still regularly used in forensic practice today. It was primarily included in this investigation in order to confirm whether it was possible to replicate both a dental eruption, and mineralisation method on PMCT data. However, this method can only be applied to individuals aged 3-17 year old. Since all observers were blind to the documented demographics of the experiment sample, it was appreciated that this could potentially limit the application of this method.

6.5.4 Image analysis

Three 'raters' reviewed the curved MPRs and OPTs independently: two dental practitioners with experience in dental age estimation and one forensic anthropologist with PMCT data analysis experience. Observers were unaware of the age of each individual and were blind to the knowledge of the other raters' results. The raters were provided with the appropriate literature aids (Figure 6.8 and 6.9) and were instructed to carry out their examination of the data as they would for a real forensic case. The raters were asked to apply the QMUL method to each case, followed one-week later by the Demirjian method (where possible – see below). The PMCT curved MPRs were assessed two months after the OPTs, to avoid any study bias that might be introduced from previous recollection of the counter part OPT for each case (a full copy of the OPT data and curved MPR data can be found in Appendix 2). The estimated age of each case, determined by both methods and both media platforms, were then compared to the documented age of each case. The documented age of each case was only revealed to the raters after all age estimations were completed. If there was any uncertainty over an age estimation, the rater was asked to supply further comments and provide a detailed reason for this uncertainty. The results of both assessments are detailed in Table 6.2.

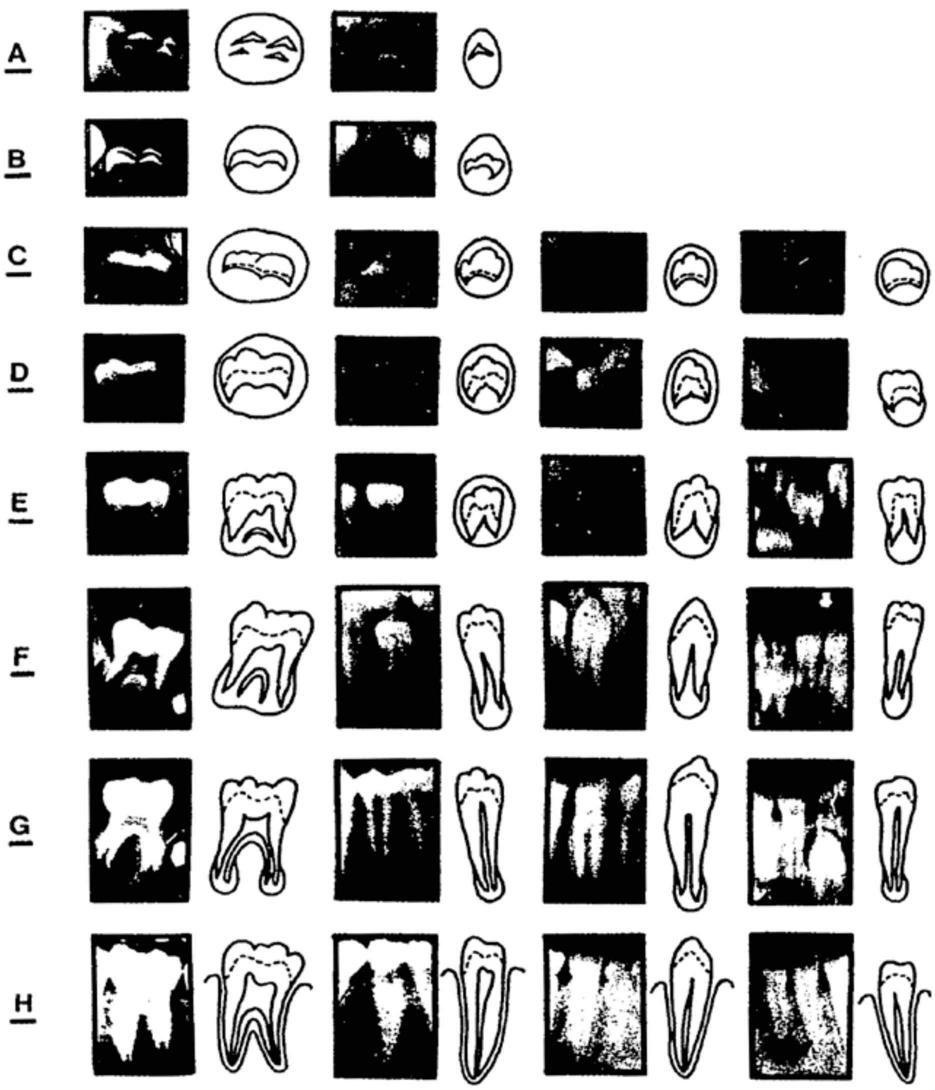


Figure 6.8: Developmental stages of permanent dentition (Dimirjian *et al.*, 1973).

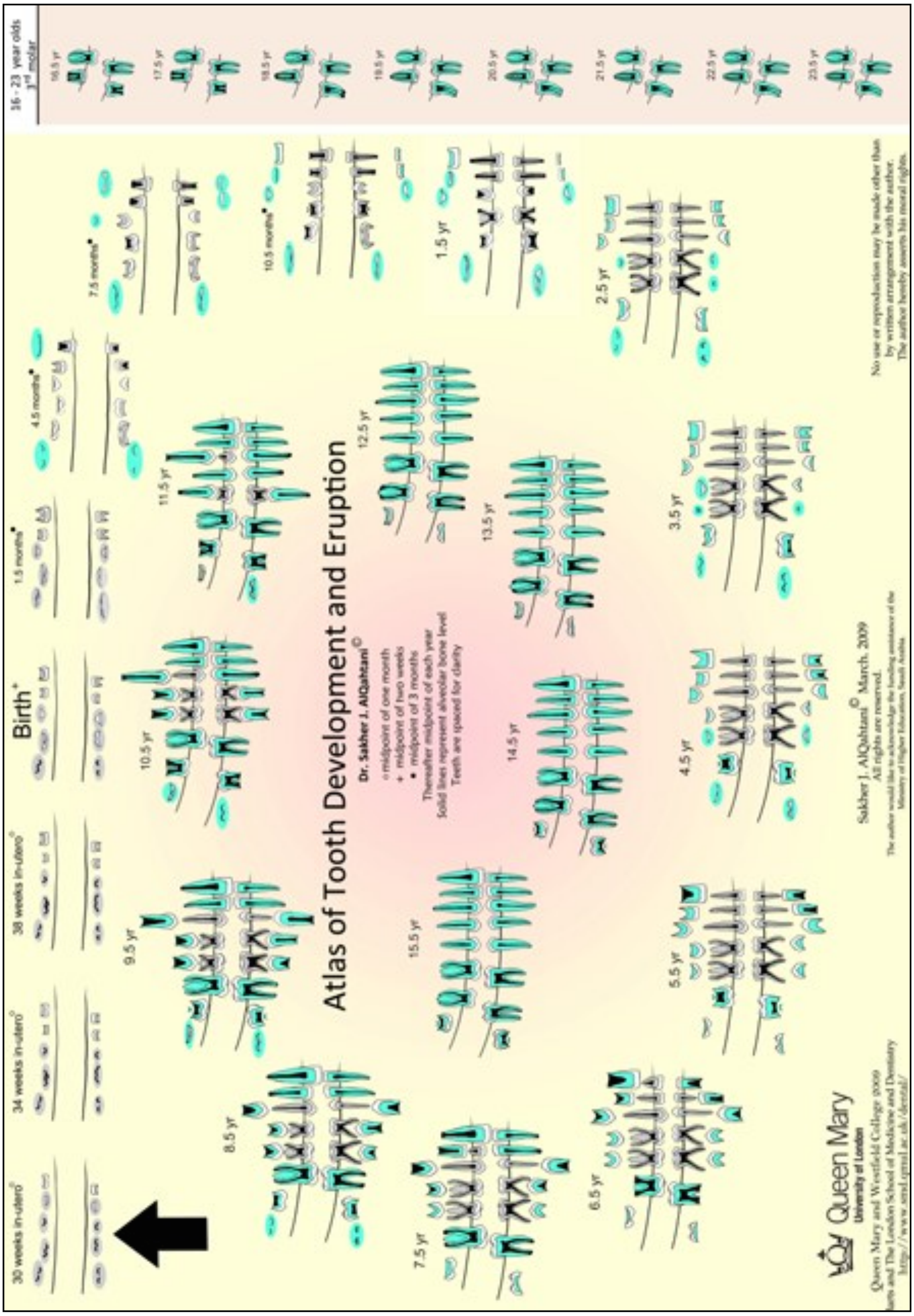


Figure 6.9: QMUL dental ageing charts (AlQahtani *et al.*, 2010).

6.6 Statistical Analysis

Statistical analysis was performed using interclass correlation (ICC) to measure the level of agreement between observers, methods and data sets. Initially, a two-way analysis of variance was performed, and if the column mean sum of squares (between methods of measurements or raters) was not significantly greater than the residual error, concordance was assumed. The ICC for the relationship provided a scalar measurement of agreement, where a value of 1 represented perfect agreement and 0 was interpreted as a lack of any agreement. ICC has advantages over correlation coefficient analysis, as it adjusts for the effects of scale magnitude and can represent the agreements for more than two observers.

6.7 Results

Comparison of the predicted ages using OPT and PMCT are given in Table 6.2 and Figure 6.10. Using the students paired T-test method no significant difference between the age estimations produced using PMCT curved MPRs and those using conventional OPT's, for all raters (as illustrated by 'p' values of 0.45, 0.56 and 0.83 for raters 1, 2 and 3 respectively). No significant inter- or intra-rater variation was found. Almost perfect agreement was illustrated, both between observers, and between repeat estimations by the same raters, for OPT age estimations and PMCT age estimations, using both the QMUL and Demirjian methods with an interclass correlation coefficient of 0.99 calculated for both (Figure 6.11 and 6.12). In Figure 6.11 and Figure 6.12 rater 1 and rater 2 are the two dental practitioner raters. In Figure 6.11, the data point that correspond to these rater's are extremely close for almost every case, compared with rater 3 (the inexperienced rater) which are clearly more scattered, with two points lying outside the confidence intervals. For PMCT estimations however (Figure 6.12) the results for all raters are much less scattered. In addition, fewer estimations lie outside the confidence intervals and Figure 6.13 suggests that the PMCT results have a stronger correlation to documented age. These results suggest that a PMCT technique is more reproducible by individuals with no previous dental experience and produces accurate age estimations. This supports the implementation of PMCT into routine forensic odontological practice. ICC analysis of the results provides further support for this claim, which showed that applying the QMUL method; their was an almost perfect agreement between mean estimated age using PMCT, mean estimated age using OPT's and documented age-at-death (Figure 6.13). We also found age estimations using OPT's and age estimations using PMCT were also in almost perfect agreement (Figure 6.10).

Unfortunately, since the Dimirjian technique is only valid for individuals age 3-16 years, and a large proportion of our sample lay out with this age cohort, sample size was too greatly reduced in this investigation for a similar analysis of data to be carried out for this technique with any degree of confidence. This outcome was not foreseen since the rater's were blinded to the identity of the cases until the results had been compiled after analysis. However, as the agreement between raters for this technique were almost perfect (ICC 0.98) the results do suggest that in cases where this technique is able to be applied, dental age estimations could potentially be improved.

Table 6.2: Comparison of predicted age (months) obtained using the QMUL method, OPT and PMCT.

		QMUL Method						
Case	Documented age (months)	Rater 1		Rater 2		Rater 3		Average
		OPT	PMCT	OPT	PMCT	OPT	PMCT	
1	228	282	270	288	276	282	270	278.0
2	96	114	115.5	114	120	114	102	113.3
3	144	144	150	144	150	150	150	148.0
4	168	174	160	180	160	150	162	164.3
5	228	282	234	288	240	282	234	260.0
6	228	222	246	222	252	234	234	235.0
7	228	186	222	180	186	198	216	198.0
8	PERINATE	1.5	2.25	1.5	2.25	1.5	2	1.8
9	2	1.5	3	1.5	3	1.5	2	2.1
10	18	18	18	18	18	18	18	18.0
11	18	24	30	30	30	30	24	28.0
12	96	90	97.2	90	98.4	90	102	94.6
13	144	150	162	150	162	150	162	156.0
14	60	78	78	78	81	78	78	78.5
15	96	168	174	168	174	168	162	169.0
16	216	198	198	204	205.5	210	210	204.3
17	144	96	92.4	90	88.8	114	102	97.2
18	132	66	42	66	42	66	42	54.0
19	216	72	168	72	168	78	168	121.0

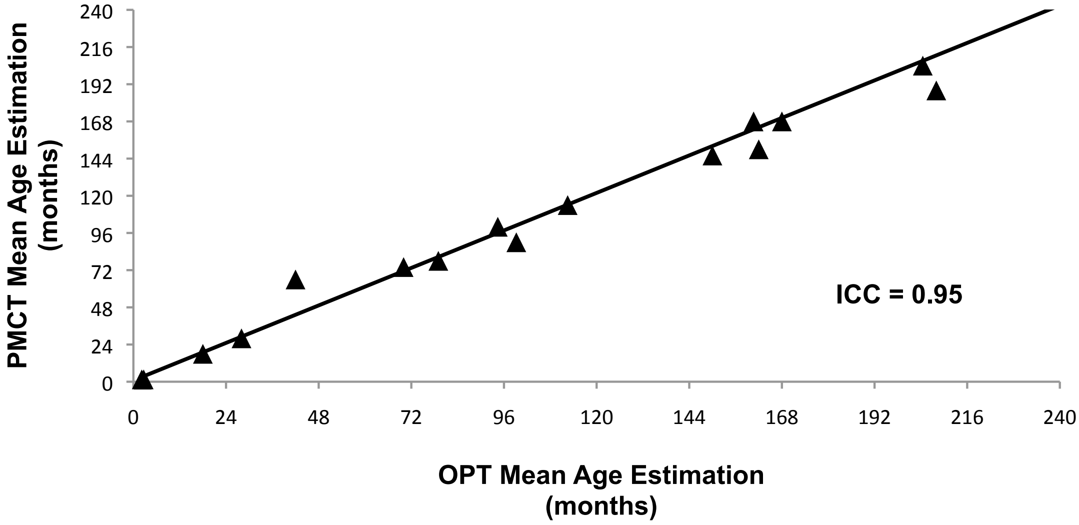


Figure 6.10: Mean age estimations using PMCT images Vs mean age estimations using conventional OPTs.

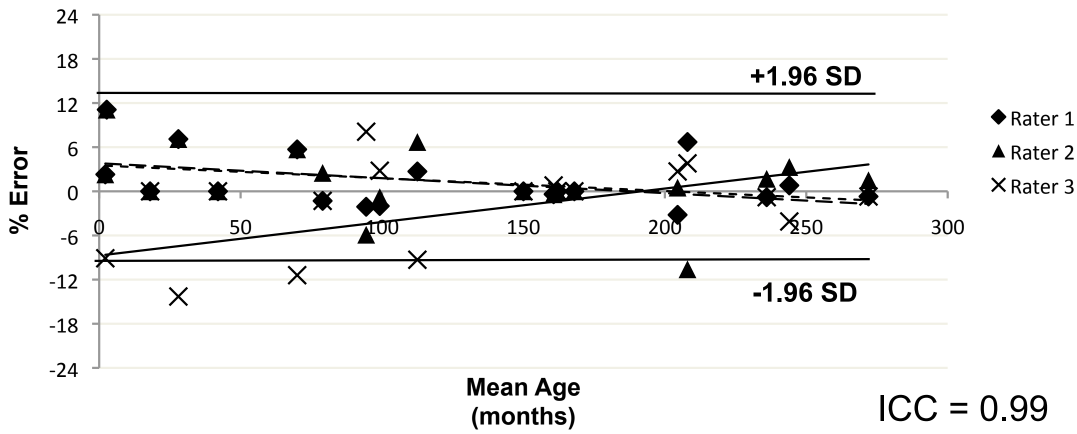


Figure 6.11: Mean age estimation (months) against error for OPT.

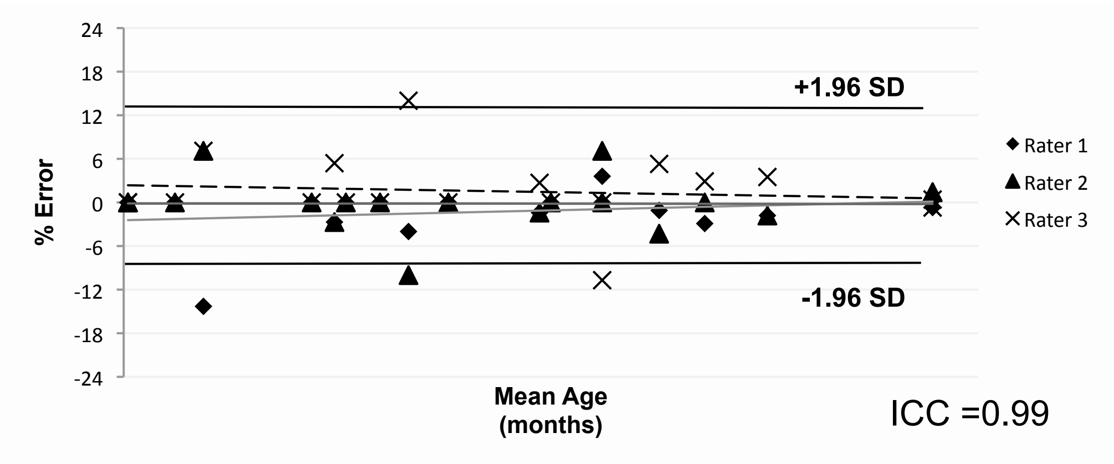


Figure 6.12: Mean age (months) against error for PMCT.

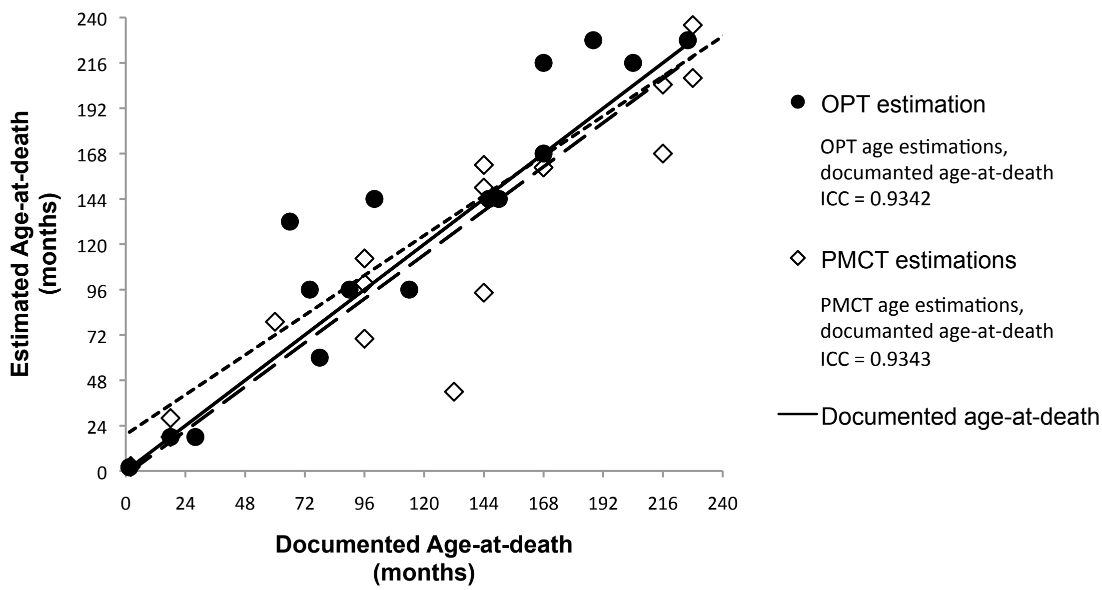


Figure 6.13: OPT and PMCT age estimation Vs documented age. PMCT is in almost perfect agreement with both OPT and documented age-at-death.

6.8 Discussion and Conclusion

Orthopantomography is the standard imaging method for dental evaluation in forensic investigations. Current standards for dental identification are therefore based on this method and there are no recognised standards for PMCT dental evaluation. This study has followed the protocols implemented for OPTs as closely as possible with slight modification where PMCT has offered improved measurement opportunities (i.e. the ability to produce a series of contiguous curved MPR images for improved viewing with minimal superimposition). This image interpretation protocol proved repeatable when used by a variety of practitioners from different professional backgrounds and levels of experience. Figure 6.11 and Figure 6.12 show that the least experienced rater had greater errors in age prediction, although using PMCT, compared with OPT, reduced this error slightly.

This investigation was unfortunately unable to demonstrate the increased measurement opportunities afforded by PMCT improved age estimation. Difficulties readjusting to the multiple 2D data set and issues relating to resolution and slice thickness highlighted additional training requirements. Feedback from the rater's suggested dental practitioners found it difficult to readjust to the multiple 2D data sets - numerous still shots of the curved MPRs were provided to each practitioner to ensure all dental features were clearly represented. However one rater, despite clear instructions, still viewed the images as single 2D MPRs before collating the final estimation, instead of using the multiple views to enhance a single estimation. More experience using multiple MPR data sets and surface rendered images is required so that this technique becomes more familiar to the user, something that can be achieved easily through training and experience. All raters also highlighted resolution and slice thickness as an issue, creating uncertainty in a couple of cases, using the Demirjian method in particular - where the increment between scores is often slight. However, as is well known in clinical practice, PMCT has the advantage over standard radiography, by being able to isolate single thin slices without superimposition of distracting or obscuring structures. This made several cases easier to assess, where using the OPT image it was sometimes difficult to tell whether complete closure of root apices had occurred (Figure 6.14). In OPT, owing to the various positions of teeth within the jaw, roots may not be optimally positioned within the focal trough and therefore their degree of root formation in particular may be adversely interpreted. Jaws can also be digitally reconstructed using PMCT, which means even when severely disrupted, bodies could be scanned in any position and a comprehensive dental examination could still be undertaken. If a full body CT scan is already planned, negating the necessity to take dental OPTs could reduce processing times in forensic investigation. Finally, 3D surface rendering has the potential to further increase the accuracy of estimations and future work by the author would include assessing the impact of this.

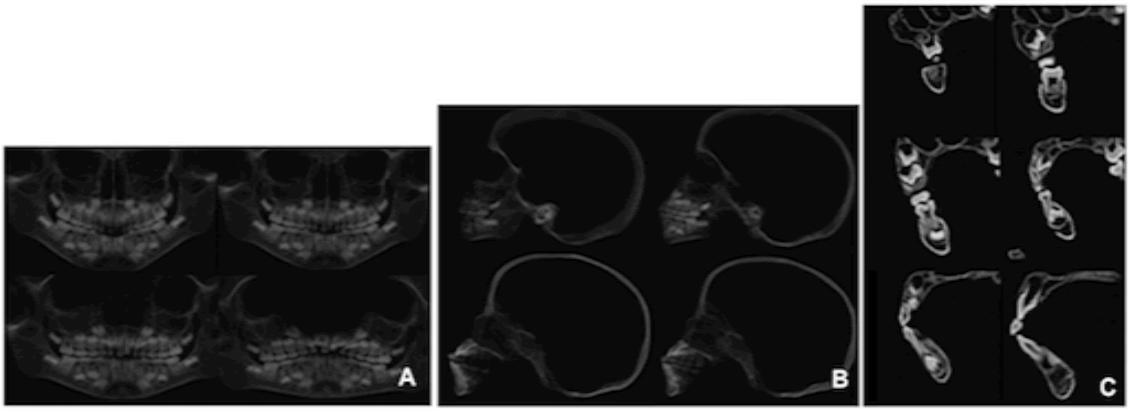


Figure 6.14: Multiple curved MPR views, minimising superimposition. A) Stretched curved MPRs of the entire mandible and maxilla, B) Lateral views of the dentition and skull, C) Perpendicular views along the 3D Bezier path, showing individual teeth, from multiple levels in the series.

Although further research is required in this area before a protocol utilising PMCT can be implemented internationally as part of standard forensic examination this investigation provides evidence that PMCT is a viable imaging technique for the reliable and repeatable assessment of age from juvenile remains. To complete this study and provide further evidence to support this statement in the near future; blind-testing dental PMCT image technique on clinical cases, analysing 3D surface rendered images of each case and exploring the potential application of mandibular measurements, such as maximum ramus height is required.

In conclusion, although this investigation did not prove PMCT to be more accurate at estimating juvenile age at death, it did prove it to be equally as accurate as dental age estimations using conventional OPT images. Therefore, the age estimation process performed on victims following a mass disaster may now be complemented with the use of PMCT, where previously any dental ageing analysis was performed using conventional radiographs, either full mouth periapicals or OPTs. Finally, with 3D surface rendered techniques, the authors believe PMCT has the potential to be more accurate than OPTs for juvenile dental age determination. PMCT also has greater flexibility of measurement and is not restricted to a single image-so the potential to develop new techniques exists.

CHAPTER 7: MINIMUM DATA SET RECORDING FORM

"A picture is worth a thousand words. However, few investigators may realise that a picture may also contain a thousand measurements."

- John H. Garstang.

7.1 Introduction

Currently there is no agreed protocol to create a biological profile from post-mortem computed tomography (PMCT) examination of juveniles or adults. The literature presents a number of studies using single osteological or odontological assessments to assist with biological profiling, but, prior to this thesis, a system compiling all of these assessments into a single protocol had yet to be published or presented.

In 2012, the International Society of Forensic Radiology and Imaging (ISFRI) was formed, with the aim to strengthen and develop the field of forensic radiology and imaging worldwide. This includes promoting best practice and developing international quality standards and guidelines in the field of imaging. Society members have a professional involvement in forensic radiology and imaging. The second congress was held in Zurich, May 2013. During this conference, several important areas of development were identified. As a result, six working groups were formed to tackle issues and produce recommendations regarding (a) Data Acquisition, (b) Reading and Reporting of Images, (c) Education, (d) Certification and Accreditation (e) Networking and (f) Disaster Victim Identification.

This commitment from the ISFRI highlights the significant role radiology now plays in forensic casework and mass fatality incidents. The formation of a dedicated team of experts working towards a common goal emphasizes the urgency and dedication of researchers within this field towards producing a standard approach to reporting, to maintain professional quality and standards.

From the investigations conducted in previous chapters it is evident that;

1) All the measurements required for current age estimation methods are reproducible using PMCT and all morphological assessment markers are also discernible.

2) Studies of maximum clavicle length and dental development found no significant difference between PMCT measurements compared with dry bone and dental OPT's, respectively.

3) Interclass correlation analysis for both investigations also illustrated a near perfect agreement between different observers, with varying degrees of experience.

Therefore, it could be concluded that PMCT measurements are accurate, produce reliable age estimations and are reproducible with little extra training required. With this is mind, the final question to consider in this series of investigations was – what images are necessary to conduct a full anthropological assessment of a juvenile and how can these images be presented in a way that could potentially be integrated into an international DVI reporting form?

With this in mind, the work presented in this chapter attempts to construct a PMCT radiographic protocol for the measurement and assessment of the entire immature human skeleton for biological profiling purposes. Skeletal material from the Scheuer Juvenile collection (see Section 3.2.1) was used primarily to develop the process. This was then 'blind' tested against a known population of fleshed juvenile cadaver PMCT images, from the East Midlands forensic pathology database, spanning the full age range of the developing human.

A template for a worldwide, standard minimum data-set anthropological reporting form will be presented; illustrating how it can be used for fleshed or un-fleshed remains to assist with biological profiling during the identification processes. The resulting minimum data-set form is designed to be used by suitably trained assessors, either at the site of image capture or remotely. The information contained within the form will be gathered by a single lead investigator and available for distributed to subsequent investigators, so that post-processing time is reduced. After initial inspection of the minimum data-set form, supplementary information can be requested, and supplied by the lead PMCT processor. For example, if an area of trauma is indentified as needing further inspection, additional images detailing this region could be supplied upon request. With any examination, there is always a danger of missing something important, that at time of examination is considered irrelevant but becomes relevant. This risk is undoubtedly increased if the data is reduced to begin with into a minimum data set. However, the raw PMCT data can still be made available and revisited indefinitely, if required. So as long as the minimum data set contains enough information to detect these areas, it should prevent important information being missed.

The layout of the minimum data set anthropological recording form has been designed with the intention to facilitate future development, through the addition of more sections. A pathological recording form could be incorporated for example, which could include PMCT full body angiography and post-mortem ventilation images. Furthermore, the simple format and easy to replicate image extraction method has been specifically designed so that it is translational to an international DVI protocol. There is therefore scope in the future to build a PMCT database to encourage data sharing and collaborations between different research facilities. The minimum data set form would provide researchers with an overview of each case, which could be retrieved more rapidly than large quantities of raw PMCT data and provide sufficient information for a full anthropological examination, at least. Such a database would be a valuable teaching and learning resource for researchers in this field.

This novel approach to PMCT biological profiling is an important contribution to the forensic community, particularly forensic anthropology, radiology and the work being undertaken by the ISFRI, by providing a minimum standard of reporting and recording of osteological and odontological identification features. This is an important step for the future development of this rapidly evolving professional body.

7.2 Aims and Objectives

The overall aim of this chapter is therefore to create a 'best code of practice' radiological reporting form for biological profiling of human remains, to promote best practice and develop international quality standards and guidelines in the field of forensic imaging.

This will be achieved by the following two research objectives:

1. Identifying a concise collection of PMCT osteological and odontological images from which an extensive full body examination can be conducted for the purposes of biological profiling, by including:
    - The information requested by Interpol to complete their current DVI form, replicated on PMCT images.
    - The most frequently required measurements for current anthropological biological profiling methods, determined from an extensive literature review.
    - Any supplementary information, which is deemed relevant by a panel of experts, which includes representatives from the fields of radiology, anthropology, forensic pathology and odontology.
2. Developing an appropriate format to presents the PMCT data and to test the reliability of this PMCT minimum data-set form.

7.3 Materials and Methods

At the East Midlands Forensic Pathology Unit, as many forensic cases as possible undergo a PMCT scan. The scan data is then transferred via a secure network to a computer lab, where they are processed using OsiriX imaging software (a program free to download and compatible with any Macintosh computer, although can be upgraded for a small charge to a 64 bit system, which is beneficial for the kind of work presented in this thesis). With this software, as for many image analysis software platforms, MPR reconstructions in any plane, curved MPRs (to replicate dental OPTs) and good quality 3D reconstructions can be produced, which are used to view soft tissue, muscle and bone in a high level of detail and to digitally reconstruct dismembered remains. After image acquisition the files can then be sent to any number of independent forensic practitioners, using, for example, a secure file drop system, to be analyzed (Figure 7.1).

This system has been developed over the course of this project, so that it is now a standard process of analysis for any PMCT scan data. It has been configured in such a way (i.e. using standard clinical scanning protocols, minimal cost software platforms and reproducible image acquisition methods) that it can be easily integrated into other research facilities; as a reporting system that can be integrated internationally has been an important consideration in this thesis.

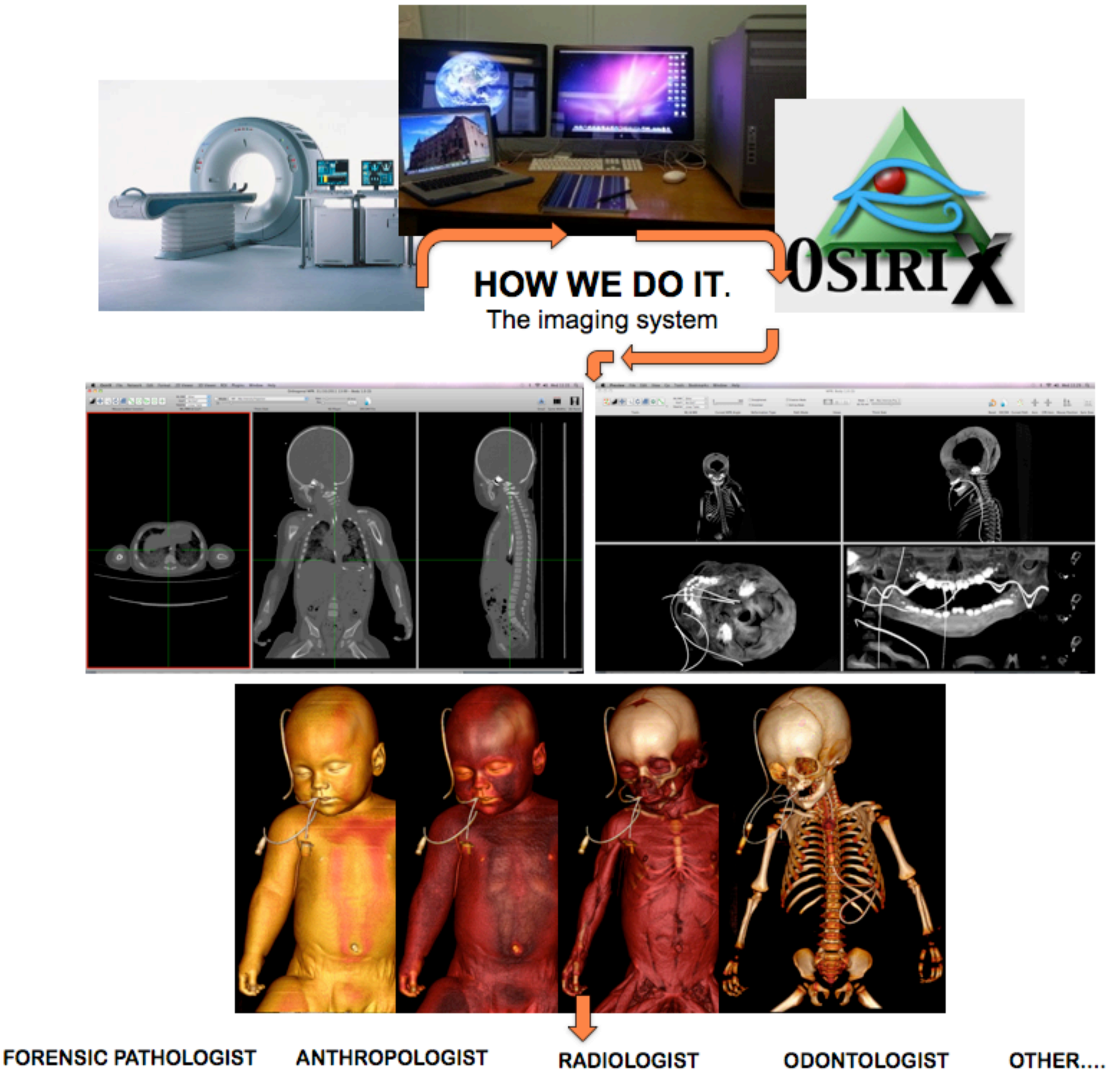


Figure 7.1: Image acquisition procedure used at East midlands forensic pathology unit. Bodies are Scanned, data is transferred to our computer lab and processed by OsiriX; to produce MPRs, Curved MPRs and 3D Images.

Prior to designing and constructing the PMCT form the recommendations of Cunha *et al*. (2009) were assessed against the peer reviewed literature to construct a protocol for PMCT osteological and odontological assessment supported by published peer reviewed literature methods for the developing human (Cunha *et al*., 2009; Rutty *et al*., 2009). This was then assessed against the current Interpol post-mortem DVI form and verified by a panel of experts from the field of anthropology, odontology, radiology and pathology; before the final two-page PMCT minimum data-set anthropological reporting form was produced.

Five observers were then simultaneously sent the minimum data-set form, along with a list of recommended anthropological age determination techniques and a structured form to record their results. Each observer was instructed to use the two-page PMCT minimum data-set form to estimate age-at-death, in a cohort of fleshed juvenile cadavers, of known age at death, spanning the full age range of the developing human (birth-18 years).

7.3.1 Anthroposcopic requirements

Although the accuracy of assigning the correct ancestry and sex to an individual are not being analysed in this investigation, due to the age cohort of the data, inclusion of these features in a biological profiling form is required to ensure the form has a standard format for both adult and juvenile remains.

The visual identification of morphological variations between different ancestral groups is the main method for assessing ancestry to adult remains. As previously discussed (section 1.3.3), the subjective nature of these traits, and because they are not measured on a continuous scale, make them difficult to use. However, they are often the only indication of an individual's provenance and therefore must be considered for inclusion in a biological profiling form suitable for investigating adult remains. Anthroposcopic features that are used to assess ancestry are found predominantly in the cranial skeleton. For this reason, clear images of the lateral and frontal views of the skull will need to be included in the profiling form. This will allow the main characteristics to be viewed from multiple angles, so that they can be thoroughly examined. A summary of the features used for ancestry determination and an example of the image required to capture these is provided in Table 7.1.

Similarly, visual inspection of the skull and pelvis can be used to determine the sex of an individual, albeit with much more accuracy and success than for ancestry. As previously discussed (section 1.3.1), the pelvis and skull are the most sexually dimorphic regions of the adult skeleton and are therefore the most regularly used for anthropometric sex determination. The classic traits used, and an example of the images required to assess these are given in Table 7.1 For fleshed remains, soft tissue features such as the external genitalia and breast tissue, and inspection of internal sex organs can also be used to indicate the sex of the individual. For this reason a full body fleshed representation of the body will also be required for an adequate examination.

7.3.2 Interpol requirements

The pink post-mortem INTERPOL form is designed for listing and recording all obtainable data about a dead body that may assist in its identification; in order to compare that data with the information obtained at the place of residence of a possible victim or missing person and recorded on the counterpart, yellow ante-mortem form. The form has a standard layout to facilitate electronic processing and to make it possible to handle reports complied from around the world (Appendix 4).

By dissecting the INTERPOL DVI form it was evident that the majority of it could be completed using PMCT, as summarised in Table 7.2 This table was then used to help determine which images would be required in the ‘minimum’ data-set recording form, to ensure it contained all the necessary information, whilst remaining succinct.

Table 7.1: Anthroposcopic traits used for ethnicity assessment.

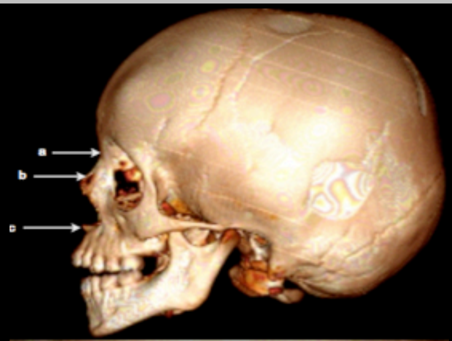
	Structure	Feature	Example Image Required
Sex	Nose	a) Root b) Spine c) Lower border Bridge Width	
	Face	Profile Shape Eye orbits Lower eye border	
	Vault	Browridges Muscle marks Vault sutures Postbregma	
	Jaws and teeth	Jaws Palatal shape Upper incisors Size	
Race	Pelvis	Ilium Pelvic inlet Pubic shape Sub-pubic angle Obturator foramen Greater sciatic notch Pre-auricular sulcus Shape of sacrum	
	Skull	Size Mastoid Browridges Nuchal area Supraorbital margin Chin	

Table 7.2: A summary of INTERPOL requirements.

Sections PMCT could complete	Sections PMCT could not complete
B (22): State of body	**B0:** Checklist of operations for mortuary
B (22A): Important ID information	**D5 (1-4):** Fingerprint information
C (24-25): Clothing and footwear	**E1 (60-65):** Internal examination***
C2+C3 (26-30): Personal effects	**E4:** DNA information
D1 (31-55): Physical description*	
D4 (described in 22 and or/31,53): Body sketch	
E2 (71-75): Medical conclusions**	
E3: Skeletal inventory	
F1 (83-85): Dental findings	
F2 (86-91): Dental inventory	
G (92): Further information	

** (33) Except Weight; **(73) Except Samples taken; ***Although, accompanying 'photo' inventory of internal examination could be completed with PMCT.*

Additional notes: Personal effects; PMCT would be able to identify general items for inventory but for more specific details e.g. travellers cheques, type of credit card, a physical examination of the item would be required. However, in scenarios where the bodies can be scanned in sealed body bag to minimise contamination exposure, PMCT would be able to map the exact location of these items for rapid retrieval. (Please see Appendix 4 for complete INTERPOL post-mortem form).

7.3.3 Metric requirements

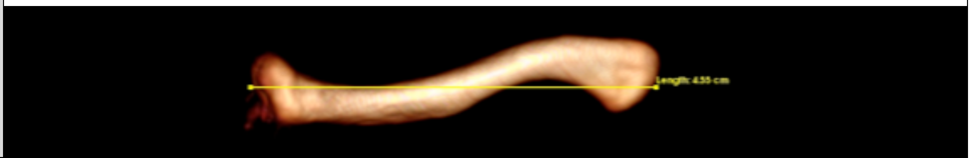
Techniques for the estimation of age using the long bones tend to refer only to the diaphyseal length of the bone. No other methods have been presented and widely accepted using other measurements of the long bones for age determination. Similarly, it is the diaphyseal lengths of the long bones that are generally used for stature estimation. Although other bones and bony structure have been considered for the estimation of stature, including the metacarpals, metatarsals and the vertebral column, none of these studies have resulted in formulas with accuracies comparable to those using the diaphyseal length of the long limb bones (Byers, 2008). Meadows and Jantz (1992) present arguably the most comprehensive data, exploring the relationship between metacarpal length and stature. The authors recorded measurements taken from the middle of the proximal articular surface to the middle of the distal tip and compared the results with known stature. Their standard errors were larger than those used for long bone lengths and therefore should not be used when long bones are present. Although these alternative measurements could be used if the long bones are absent, inclusion of these on a standard recording form is not considered necessary. The reporting practitioner should request these additional measurements, only if necessary.

In a forensic investigation, physical, chemical or biological forces may have damaged bones that require osteological analysis. In some cases, the damage is sufficient to prevent long bone length being measured reliably. Faced with this situation, several researchers have attempted to provide information to establish the relationship between fragmented long bones and their total lengths (Steele, 1970; Krogman, 1962) but unfortunately, the errors associated with fragmented long bone estimations are even larger than those for metatarsals, metacarpals and the vertebral column, and would therefore only be required in exceptional circumstances, if they were the only osteological material available.

To summarise, the diaphyseal lengths of the long bones were measured and recorded within the minimum data-set form, as they are regularly used in a number of age-at-death and stature estimation methods. For adult cases metric measurements of the skull used in cranial discriminate function calculations would also be included for the purpose of sex and ancestry estimation.

The measurements were taken using the osteometric standards reported by Buikstra and Ubelaker (1994). A detailed description and an illustration of how to correctly take each measurement required for the minimum data-set recording form are provided below (Table 7.3).

Table 7.3: Osteometric board maximum length measurements (adapted from Buikstra and Ubelaker, 1994; and Moore-Jansen *et al*., 1994).

BONE	DESCRIPTION OF MEASUREMENT AND DIAGRAM
Clavicle	Maximum distance between most extreme ends of the clavicle. 
Humerus	Direct distance from the most superior point on the head of the humerus to the most inferior point on the trochlea. *Comment: Humerus shaft should be positioned parallel to the long axis of the osteometric board.* 
Radius	Maximum Length: distance from the most proximally positioned point on the head of radius to the tip of the styloid process without regard for the long axis of the bone. 

Ulna	Distance from the most superior point on the olecranon to the most inferior point on the styloid process. 
Femur	Maximum Length: distance from the most superior point on the head of the femur to the most inferior point on the distal condyles. *Comment: Place the medial condyle against the vertical end-board while applying the movable upright to the femoral head.* 
Tibia	Distance from the superior articular surface of the lateral condyle to the tip of the medial malleolus. *Comment: place the tibia on the board, resting on its posterior surface with the longitudinal axis parallel to the instrument. Place the lip of the medial malleolus on the vertical end-board and press the movable upright against the proximal articular surface of the lateral condyle.* 
Fibula	Maximum distance between the most superior point on the fibula head and the most inferior point on the lateral malleolus. 

7.3.4 PMCT Biological profiling form

The form contains 2 pages. The first page consists of a surface image and a skeletal view of the full body. The surface image should be used to record a physical description of the cadaver and may help detect dysmorphic or congenital features, distinguishing marks, clothing, shoes and personal effects (where applicable). The whole body skeletal image allows identification of congenital or acquired bone disease, for example scoliosis. The second page of the form includes: 7 views detailing the complete morphology of the skull; all the long bones and their measurements; a clear view of each joint of the shoulder, elbow, hip and knee; an isolated view of the hand, foot, pelvis and rib ends; the spine, sacrum and axis; and finally, dental OPT reconstructions. These were selected to cover the osteological and odontological features used for biological profiling throughout the developing human. The final profiling form layout is illustrated in Figure 7.2 and Figure 7.3.

Figure 7.2: Two-page minimum data set anthropological reporting form: Page 1; full surface and skeletal 3D images.

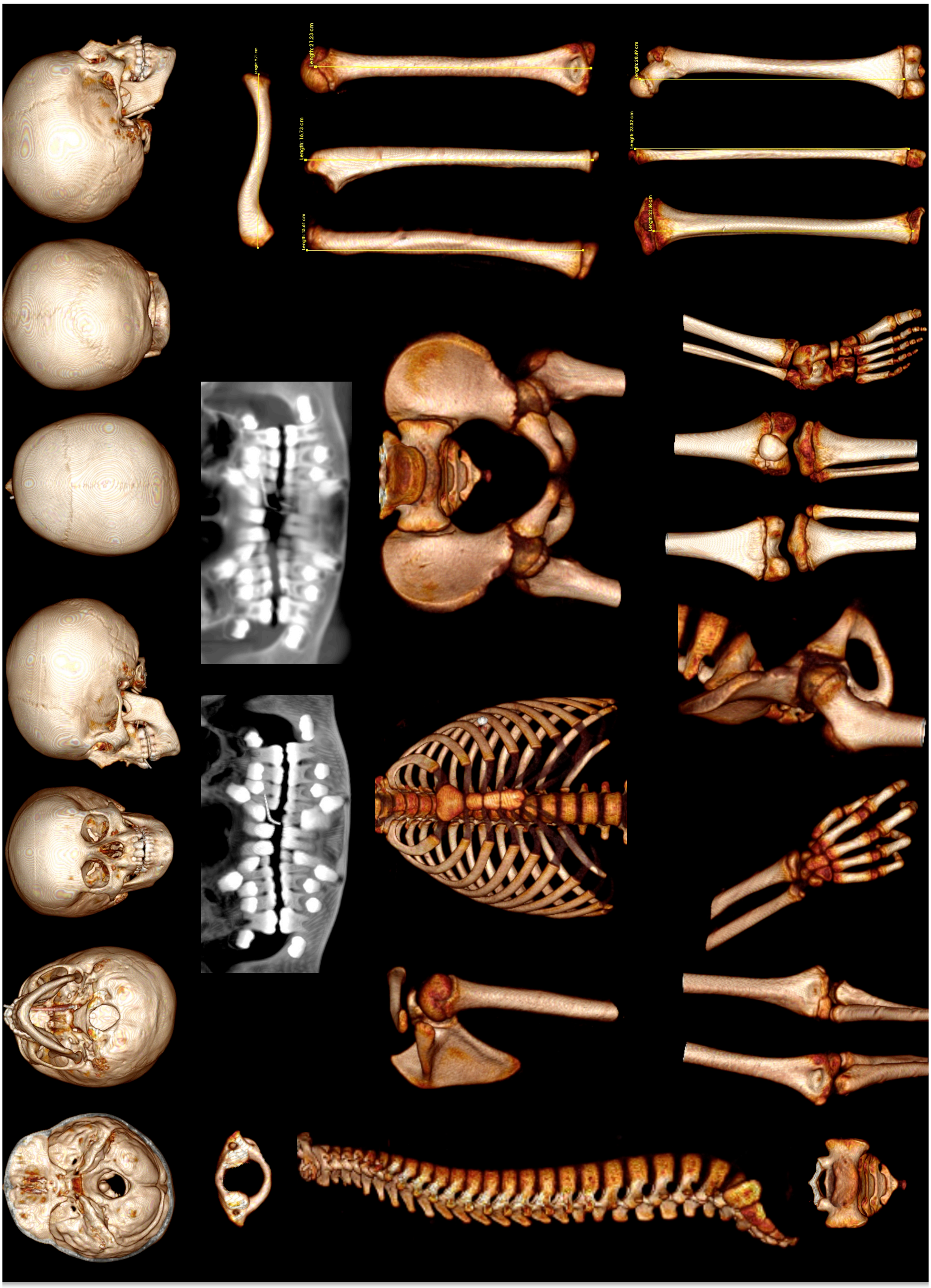


Figure 7.3: Two-page minimum data set anthropological reporting form: Page 2; Isolated regions of interest for anthroposcopic assessment and long bone measurements for metric assessment.

7.3.5 Assessment cohort

Thirty-one cases were selected by a forensic pathologist, who was not part of the assessment team, from the East Midlands Forensic Pathology Units secure PMCT database to represent cadavers spanning the full age range of the developing human (0-20 years). Cases included both natural and un-natural deaths with body injury and cavity disruption. All cases had undergone a forensic pathology autopsy in line with current United Kingdom forensic pathology standards under the legal authority of HM Coroner for the jurisdiction of the where the death had occurred. As part of this examination they had all undergone PMCT. In each case the relevant HM coroner gave written consent for the assessment of each case, in this study. The autopsy reports containing the biological profiling information relevant to each case, as acquired at autopsy, as well as the PMCT images were then made anonymous and given a unique study code.

7.3.6 Case form generation

The PMCT images of all cases were downloaded onto a Macintosh computer processed using OsiriX imaging software (http://www.osirix-viewer.com/. Last visited August 2013). A single individual with both anthropological and PMCT 3D image processing training subsequently produced all images for all cases. For each case, images for a full two-page minimum data set form were produced, where possible. If an image could not be created (i.e. the element was missing or badly damaged) this was clearly stated on the form in place of the image (the minimum data set forms for each case can be seen in Appendix 3).

Each case was assessed during form construction by the OsiriX processor and a biological profile constructed blind to the autopsy findings. Four independent and remote assessors from different professional backgrounds and experience were then simultaneously sent the composite forms for each case by a secure file drop system. These independent observers did not have access to the raw PMCT data and did not need to generate any of the images themselves. This was done in order to simulate how the form could work in a practical situation. In a DVI event, post-processing would be greatly reduced if a single OsiriX processor created the principle form and distributed it to numerous other practitioners; compared with multiple practitioners processing the raw data to produce the same form. In this type of event, this process would also minimize observer subjectivity, as all ensuing examiners would have access to exactly the same information. Each of the four observers were subsequently asked to produce a biological profile for each case, as they would in a real forensic investigation, blind to both the OsiriX assessors results and the autopsy findings. Each assessor was provided with a list of recommended age estimation methods and a reporting form. A list of methods was supplied simply as an aid for practitioners who were not as familiar using anthropological techniques but a "technique" column was included within the reporting form so that observers could detail the exact methods applied. Each observer was asked to provide age estimation, as they would in practice, and where several methods were applied they were asked to provide a breakdown of each methods results followed with an overall age estimation (Figure 7.4). Observers were also asked to note any difficulties they had interoperating the PMCT form and any alterations they would make to improve it. Finally, each observer was also asked to rate - how easy they found the minimum data form to use and how confident they were with the estimations they provided, in the following manner:

- ***Ease of use score***: 1 = extremely easy, 2 = easy, 3 = relatively easy, 4 = difficult, 5 = extremely difficult.
- ***Level of certainty score***: 1 = extremely uncertain, 2 = uncertain, 3 = relatively certain, 4 = certain, 5 = extremely certain.

All results were then collated by the study lead and the autopsy reports were used as a gold standard to assess the results of the independent observers and look at inter-observer variation.

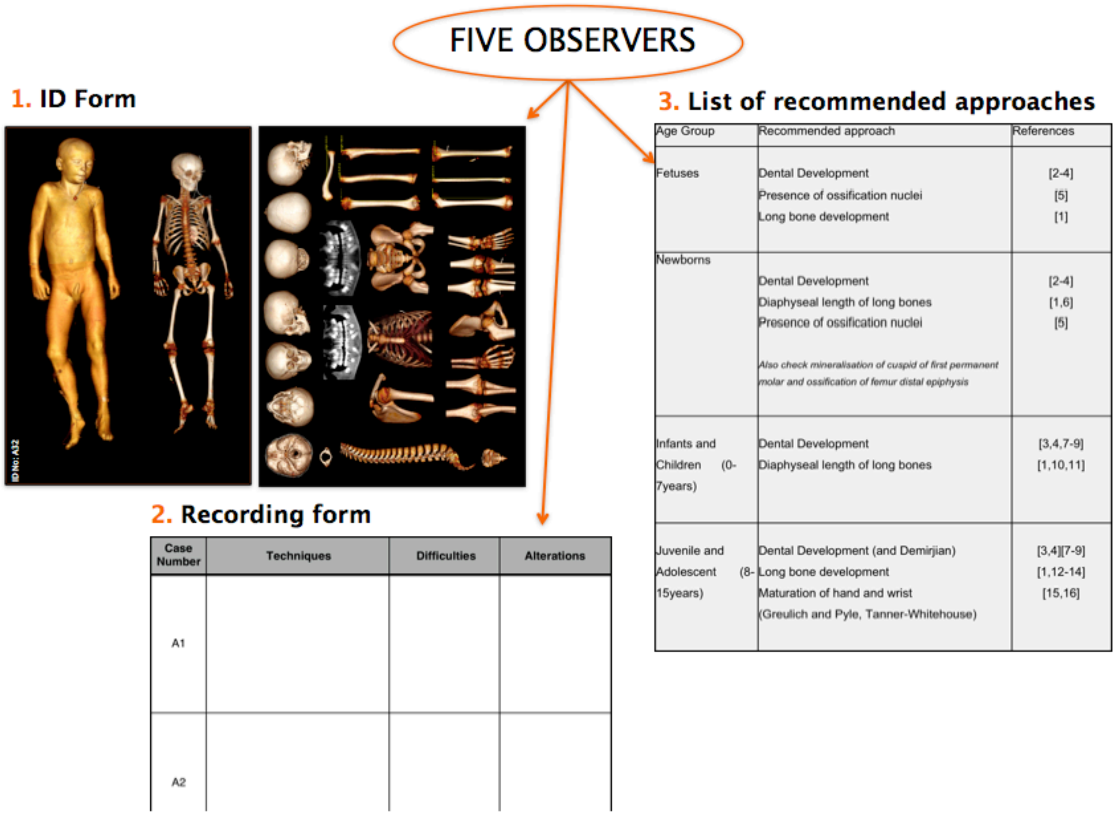


Figure 7.4: Experimental design. Each observer was given 1) the two-page PMCT ID form, 2) recording sheet and 3) list of recommended age estimation techniques.

7.3.7 Statistical analysis

There is no standard margin of error that is considered acceptable for every age estimation method. Previous work by Ritz-Timme *et al*. (2000) suggests that the range of error associated with dental age estimation, using seven different methods for developing dentition, is up to 24 months for a small child (age 0-1 years) and 48 months for the sub-adult range (1-12 years) (Ritz-Timme *et al.*, 2000). Likewise for the diaphyseal length of long bones the majority of observers use the standards created by Maresh (Maresh and Deming, 1939), which give similar ranges. There are many other methods of assessment including the hand and wrist atlas of Greulich and Pyle (Greulich and Pyle, 1971) and ossification center appearance and fusion as detailed by Scheuer and Black (Scheuer and Black, 2000). These methods all report different tolerated error ranges but are broadly consistent with those reported by Ritz-Timme and Maresh (Al Qahtani *et al*., 2010; Dimirjian *et al*., 1976; Dimirjian *et al*., 1973). I have therefore concluded that error ranges of 24 months for 0-1 year and 48 months for 1-12 years are the tolerated error margins typically accepted in anthropological practice and therefore these are used as our standard to be achieved by PMCT based age estimations.

Mean age estimations are compared to actual age using 95% confidence intervals (CI) and paired Students 't' test. The level of agreement between each observer and documented age was examined using interclass correlation (ICC) statistics. The ICC provides a scalar measurement of agreement, where a value of 1 represents almost perfect agreement and 0 represents no agreement. Measurement Error was calculated and presented using the 'Bland and Altman' plots (Bland and Altman, 1996). To calculate measurement error (within subject standard deviation, wSD), data should either be independent of magnitude (age) or a 'data transformation' is required (Bland and Altman, 1996). In this study however the relationship of error to age was complex and varied depending on the age. Measurement error and 95% confidence intervals were therefore calculated separately for age groups 0–1 year, 1–5 yrs and 5 yrs to adult, as within these ranges the degree of error was independent of magnitude (age).

7.4 Results

Mean observer score for the ease of use of the system was 2.0 (easy) and 3.8 (just under certain) for their level of certainty. This provided useful feedback for the further development of the PMCT ID form.

The results of this study are shown in Table 7.4 and illustrated in Figure 7.5 and 7.6. In all cases errors are within the 'tolerated error margin' based on previous studies (Ritz-Timme *et al*., 2000; Maresh and Deming, 1939).

For ages up to 1 year, there was no significant under or over estimation of age, with a mean error of 0.23 months (95% CI=0.5 months, p=0.4). For ages greater than 1 year there was a mild trend to underestimate age by 3.7 months (95% CI=3.3 months, p=0.03).

Interclass correlation statistics show almost perfect agreement, with interclass correlation coefficients for observers one to five of 0.98, 0.99, 0.98, 0.97, 0.99 respectively.

Observer 3 used only dental information to make their age estimations, which were within the tolerated margin of error (age range) for dental techniques. In 3 cases (5, 12 & 15) the dental images were absent, incomplete or of poor quality due to injury, incomplete scanning or image resolution. In these cases the odontologist gave an estimated age based on non-dental images, and these results are shown but not included in statistical calculations of measurement error, as they would not normally have done this assessment in a forensic investigation. Three cases (29, 30 & 31) had dental anomalies (impacted canines, left/right differential development, premature premolar eruption), which may have benefited from the expertise of an odontologist.

Figure 7.6 shows a Bland Altman plot of measurement error against documented age at death for all observers. Measurement error (within subject standard deviation, wSD) was calculated as 9.6 months overall and at 1.8, 6.3 and 13.8 months for Age groups 0–1 year, 1–5 yrs and 5 yrs-to-adult respectively. Ninety-five percent confidence intervals (1.96* wSD) were therefore 3.5, 12.4 and 27.1 months respectively and 19.0 months overall. Measurement error improves if a mean age estimation value from all observers is used, giving wSD as 6.9 months overall and 0.9, 1.8 and 10.1 months for age groups 0–1 year, 1–5 years and 5 years-adult respectively. Ninety-five percent confidence intervals were therefore 1.8, 3.6 and 19.9 months respectively and 13.5 months overall. This is consistent with the principle that a multi-factorial approach using multiple observers is the standard of good practice (Saunders *et al.*, 1992; Bedford *et al.*, 1993; Fairgrieve and Oost, 1995; Nagar and Hershkovitz, 2004; Franklin, 2010; Garvin and Passalacqua, 2012).

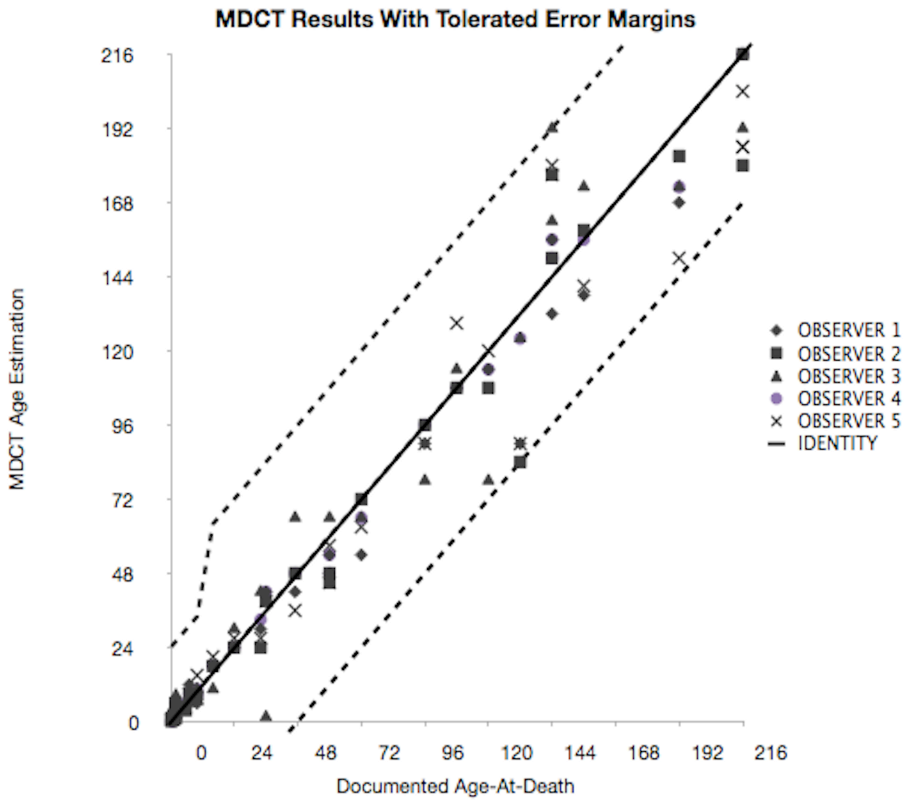


Figure 7.5: Age estimations based on 'PMCT minimum data-set' anthropological reporting form Vs documented age-at-death. Dotted lines: tolerated error margins based on studies by Ritz-Timme *et al*. (2000) and Maresh and Deming (1939).

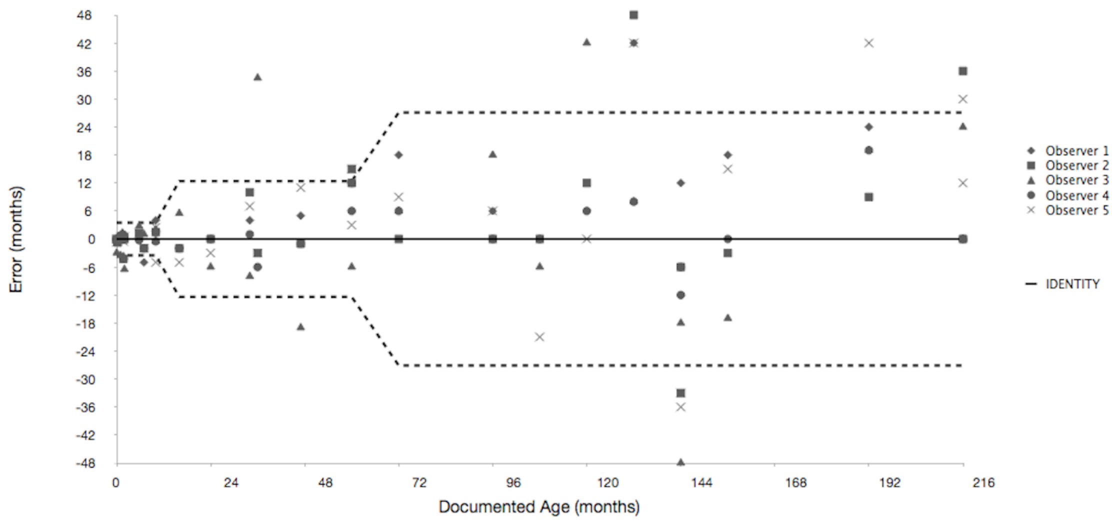


Figure 7.6: Bland and Altman plot; Measurement errors for each observer, against documented age-at-death. Dotted lines: 95% confidence intervals.

Table 7.4: Estimated and documented age-at-death for cases (n=31). Including tolerated margins of error based studies by Ritz-Timme *et al*. (2000) and Maresh and Deming (1939).

Case	Documented Age (mths)	Tolerated error margin (mths)	Age range & error margin (mths)	PMCT Estimated Age (mths)				
				Obs 1	Obs 2	Obs 3	Obs 4	Obs 5
1	0.25	+/- 24	0 - 24	0.25	1	0.25	0.25	0.75
2	144	+/- 48	96 - 192	132	150	192	150	180
3	47	+/- 48	0 - 95	42	48	66	48	36
4	34	+/- 48	0 - 82	30	24	42	33	27
5*	36	+/- 48	0 - 84	42	39	1.5	42	39
6	2	+/- 24	0 - 26	1	2	8.5	2	1.5
7	16	+/- 48	0 - 64	18	18	10.5	18	21
8	108	+/- 48	60 - 156	108	108	114	108	129
9	10	+/- 24	0 - 34	6	8.5	8	8.5	15
10	10	+/- 24	0 - 34	6	8.5	10.5	10.5	7.5
11	1.75	+/- 48	0 - 26	6	6	5.5	6	2.3
12*	216	+/- 48	168 - 264	216	180	192	216	186
13	144	+/- 48	96 - 192	156	177	162	156	177
14	216	+/- 48	168 - 264	216	216	216	216	204
15*	216	+/- 48	168 - 264	216	216	216	216	186
16	1.5	+/- 24	0 - 25.5	1	1.5	0.25	1.5	1.5
17	96	+/- 48	48 - 144	90	96	78	96	90
18	7	+/- 24	0 - 31	12	9	6	9	9
19	120	+/- 48	72 - 156	114	108	78	114	120
20	60	+/- 48	12 - 108	48	48	66	48	45
21	72	+/- 48	24 - 120	54	72	66	66	63
22	1.5	+/- 24	0 - 25.5	0.25	1	0.5	1	1.5
23	24	+/- 48	0 - 72	24	24	30	24	27
24	60	+/- 48	12 - 108	54	45	66	54	57
25	1	+/- 24	0 - 25	0.25	1	4.5	1	0.75
26	0.3	+/- 24	0 - 24.3	0.25	0.25	0.3	0.25	0.75
27	5.75	+/- 48	0 - 30	6	4.5	3	6	4.5
28	1	+/- 24	0 - 25	0.25	1	1.5	0.25	0.5
29	156	+/- 48	108 - 204	138	159	173	156	141
30	192	+/- 48	144 - 240	168	183	173	173	150
31	132	+/- 48	84 - 180	90	84	124	124	90

***Cases with no dental images.**

7.5 Discussion

When estimating age-at-death, forensic anthropologists face a series of methodological choices. These decisions, such as which skeletal region to evaluate, which method to apply, what statistical information to use and how to combine information from multiple methods, ultimately impact the final reported age estimation. A questionnaire conducted by Garvin *et al*. (2011), given to 145 forensic anthropologists, found that the majority of respondents vary their skeletal age estimation process case-by-case and ultimately present officials with both a narrow and a broad possible range. Overall, respondents displayed a very high degree of variation in how they generate their age estimations, and indicate that experience and expertise play a large role in skeletal age estimations.

The choice of method has to take into account the circumstances of each case. This depends in part on the skeletal elements available for analysis: different bones are inherently more resilient than others to damaging taphonomic processes or high velocity impact etc., causing preservation bias. Other considerations are related to the actual methods, which as outlined by Ritz-Timme *et al*. (2000) should fulfill the following specific demands:

1) They must have been presented to the scientific community through peer-reviewed publication.

2) Their accuracy must be tested using valid statistical procedures and described by clearly defined terms.

3) The method must be accurate enough for routine forensic application.

Sex and/or population specificity are also important. As male and female growth trajectories diverge, many current aging methods, formulated using documented reference samples, provide both separate and pooled data. In most forensic cases however, it is highly unlikely that sex would be known, or able to be reliably estimated in the juvenile, especially pre-pubertal skeleton. In this situation pooled age-sex estimation standards are preferable.

Very few attempts have been made to find common standardization, calibration and evaluation procedures, or to develop means of quality assurance for methods of age estimation. The issue of what constitutes an ‘appropriate’ statistical approach for the estimation of skeletal age-at-death has been the subject of considerable debate. One of the primary issues is related to the variation in the statistical assumptions of the different methods that are available, also whether the distribution of data in the original reference sample from which a standard is formulated is the same for an individual outside that reference group.

The errors in estimation age are not always apparent. To evaluate the degree of error associated with dental age estimation, Repine *et al.* (2011) reviewed 51 forensic case files involving unidentified bodies for which positive identification were subsequently obtained. A total of seven different methods were assessed and on the basis of their assessment, Repine *et al*. (2011) concluded that:

> “*in cases with a developing dentition the estimated age range can be narrowed to 2-4 years. Depending on the degree of development the dentition of a small child can be estimated within a range of 2 years and for the sub-adult range of 4 years is more appropriate*” (Repine *et al*., 2011: pg152).

By the accepted definitions referred to by pediatricians a ‘small child’ refers to an infant (from birth to one year) and a ‘sub-adult’ refers to childhood (from one year to puberty, 1-12 years). Applying these tolerated error margins for standard anthropological age estimation methods to the results obtained in this investigation-using a PMCT profiling form show all five of the observers’ results fell within these tolerated error margins. In fact the majority of results are well within the upper and lower values of this error range (Table 7.4, Figure 7.5).

Observers used standards by Maresh (1970) to calculate age using diaphyseal length of long bones. Data from Maresh (1970) are from the university of Colorado longitudinal study and are of bony diaphysis (except between ages 10 and 12, when double sets of figures give lengths with and without epiphyses).

Variance increased as the diaphyseal length (and therefore the age) of the subjects increased. The age ranges using this method are given +/- 24 months for individuals up to 1 year and +/- 48 months for individuals over 1 year. Again, applying these tolerated error margins to the results obtained in this investigation shows all five of the observers' results fell within these tolerated error margins and that the majority of results are well within the upper and lower values of this error range (Table 7.4, Figure 7.6).

7.6 Summary and Conclusion

Previous research suggests that PMCT derived anthropological measurements are accurate (Brough, *et al*., 2012; Brough *et al*, 2013; Brough *et al*., 2014; Cavalcanti *et al*., 2004; Robinson *et al*., 2008; Verhoff *et al*, 2008; Dedouit *et al*., 2007). This study illustrates that PMCT findings can be condensed into a minimum data-set recording form and provide an efficient method of requesting multiple odontological and anthropological age estimations. Feedback from the users suggests the form is easy to use and they felt able to provide estimations with a 'high degree' of certainty. These age estimations were shown to be within margins of error acceptable in anthropological practice. Furthermore, unsurprisingly (Saunders *et al*., 1992; Bedford *et al*., 1993; Fairgrieve *et al*., 1995), averaging multiple age estimations using several techniques produces a lower range of error.

In summary, this investigation has developed a coherent post-mortem radiological recording format, which has accuracies comparable to traditional anthropological methods, can be easily replicated for use in any forensic or mass disaster situation, and promotes international quality control and best practice in the field of radiology and imaging worldwide (Figure 7.7).

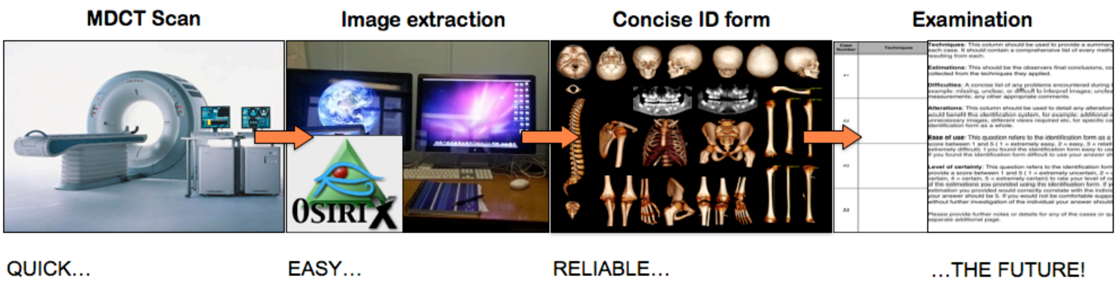


Figure 7.7: Summary of minimum data set approach to anthropological reporting. This method is quick, easy, reliable and undoubtedly the future of forensic investigation.

7.6.1 Key points

1) The 'minimum data-set form' presented in this chapter, provides a minimum standard of radiological reporting.

2) By producing a form for every individual that was CT scanned - forensic practitioners could be confident that all the relevant information required to produce an ID and complete the current INTERPOL DVI pink form was recorded – potentially replacing, dental X-rays and other radiological imaging platforms.

3) The minimum data form is designed to contain the necessary quantity of information whilst remaining succinct and transferable.

4) Although providing adequate information for a general anthropological assessment-if an area of particular interest was identified, additional images could be requested and obtained by the principle investigator, from the raw data, rapidly.

5) The profiling form can be sent simultaneously to multiple practitioners - much faster and easier than transferring large raw data sets for independent image processing.

6) A standard format to build a database for future research.

CHAPTER 8: CASE STUDY

“Richard III: the dead king who brought the world's gaze to a Leicester car park”

- Maev Kennedy, the Guardian.

8.1 Adult Case Example - Richard III

To demonstrate that the biological profiling form developed in the previous chapter was transferable to adult remains, a single case study was conducted.

In August 2012 a skeleton was excavated in Leicester that was identified as Richard III, the last King of England to die in battle, by a collection of evidence including; a DNA match with modern maternal-line relatives, archaeological evidence of the burial structure and location, and osteological analysis of the skeleton which conformed with known descriptions of Richard III.

Along with a traditional osteological analysis of the remains, PMCT scans of the complete skeleton were taken. Using this data, a minimum data-set recording form was completed in line with the method developed in chapter 7 (Figure 8.1 and Figure 8.2).

From the recording form it was evident that the skeleton was that of an adult male, in his late 20s to mid 30s with a gracile build and a severe scoliosis of the thoracic spine. This was consistent with the reports of Richard III age at death and physical description of slender build and raised right shoulder.

A total of 12 peri-mortem injuries were also identified on the minimum-data set form, nine to the skull and three to the postcranial skeleton. This prompted a request for supplementary images, which were provided upon request, for a more comprehensive final anthropological report.

Figure 8.1: Minimum data set anthropological reporting form: Page 1.

Figure 8.2: Minimal data set anthropological reporting form: Page 2.

8.1.1 Biological profiling

The case was reviewed, following the same procedure discussed in chapter 7. Using diaphyseal measurements of the long bones, morphological features of the skull and pelvis, and epiphyseal fusion events; age, sex, stature and ethnicity were estimated. The analysis was done blind to knowledge of the suspected identity of the remains and independent of the traditional osteological investigation.

The results were consistent with those calculated during the traditional osteological assessment. The biological profile produced from the PMCT investigation was as follows:

Age: 25-35 years

Stature: 5 foot 8 inches

Sex: Male

Ethnicity: Caucasian

8.1.2 Trauma

This exercise presented an ideal opportunity to test the logistics of using a minimum data-set anthropological reporting form in a real case. As stated previously, the form provides a minimum standard of osteological reporting. However, if further images are required for a more comprehensive analysis of a particular region, the form creator can upon request produce additional supplementary images as specified by the form examiner. In this particular case, upon initial inspection a number of areas were identified that required a more in-depth examination. These included; areas of anti-, peri-, and post-mortem trauma that could have potentially been injuries procured during battle.

Additional images were requested for the following regions/features of interest on the skull:

1) A keyhole trauma wound on the parietal bones of the skull: Additional images of this wound, including probable direction of impact (determined

by an extensive examination of the 3D skull reconstruction (Figure 8.3: 1a-c)), and measurements (Figure 8.3: 1d-e)) were provided on request. It was subsequently confirmed that the wound was positioned over the sagittal suture, 65mm behind the Bregma, and measured 9x10 mm. The posterior portion was square in outline whilst the anterior portion showed beveling to the outer table. It was obliquely oriented at approximately 60 to 210 from the sagittal plane. This injury was consistent with a typical keyhole injury with penetrating the dipole, and causing the inner table to be pushed inwards towards the location of the brain and meninges. Further images were produced to illustrate the extent of beveling, to determine if the injury could have been fatal (Figure 8.3: 1f).

2) Two superficial grazing injuries on parietal bone (Figure 8.3: 1b): In life, these injuries would have cut the scalp and shaved the bone surface, causing significant blood loss but given the other injuries present these were unlikely to be the cause of death and therefore no other analysis was requested.

3) Two areas of potential trauma (Figure 8.3: 1c; circled in red): These areas were particularly important in this investigation, as they were not identified during the dry bone examination. It was therefore important to establish if these regions were true traumas, which had not been detected during the initial inspection of the skeleton; or resolution anomalies on the PMCT image reconstructions, that were presenting false traumas. The dry skull was subsequently re-examined and these areas were identified as true minor traumas.

   This is an interesting result suggesting that imperfections of the structural integrity of bone, that would perhaps go undetected during a traditional osteological examination, would be distinguishable on PMCT images. In cases, such as child abuse, this could be extremely important, when even minor injuries could be significant.

4) A 10mm obliquely orientated linear incised wound to the right side of the jaw, below and in close proximity to the mental foramen (Figure 8.3: 2a-b)

5) A tool mark with area 50x40mm to the ramus of the mandible, on its anterior aspect (Figure 8.3: 2c)

6) An irregular fracture from the canine/first premolar in a medio-inferior direction towards the mental eminence (Figure 8.3: 3a-c).

7) The absence of a 10mm area of bone from the right maxilla (Figure 8.3: 3d-e).

8) A 14mm long fracture joining a 15x15mm irregular area of bone deficit to the posterior aspect of the maxilla (Figure 8.3: 3d-e).

9) Bone loss of the occipital bone: on the right side this was recorded to measure 65x50mm and on the left side 32x17mm area of bone loss (Figure 8.3: 4a-d).

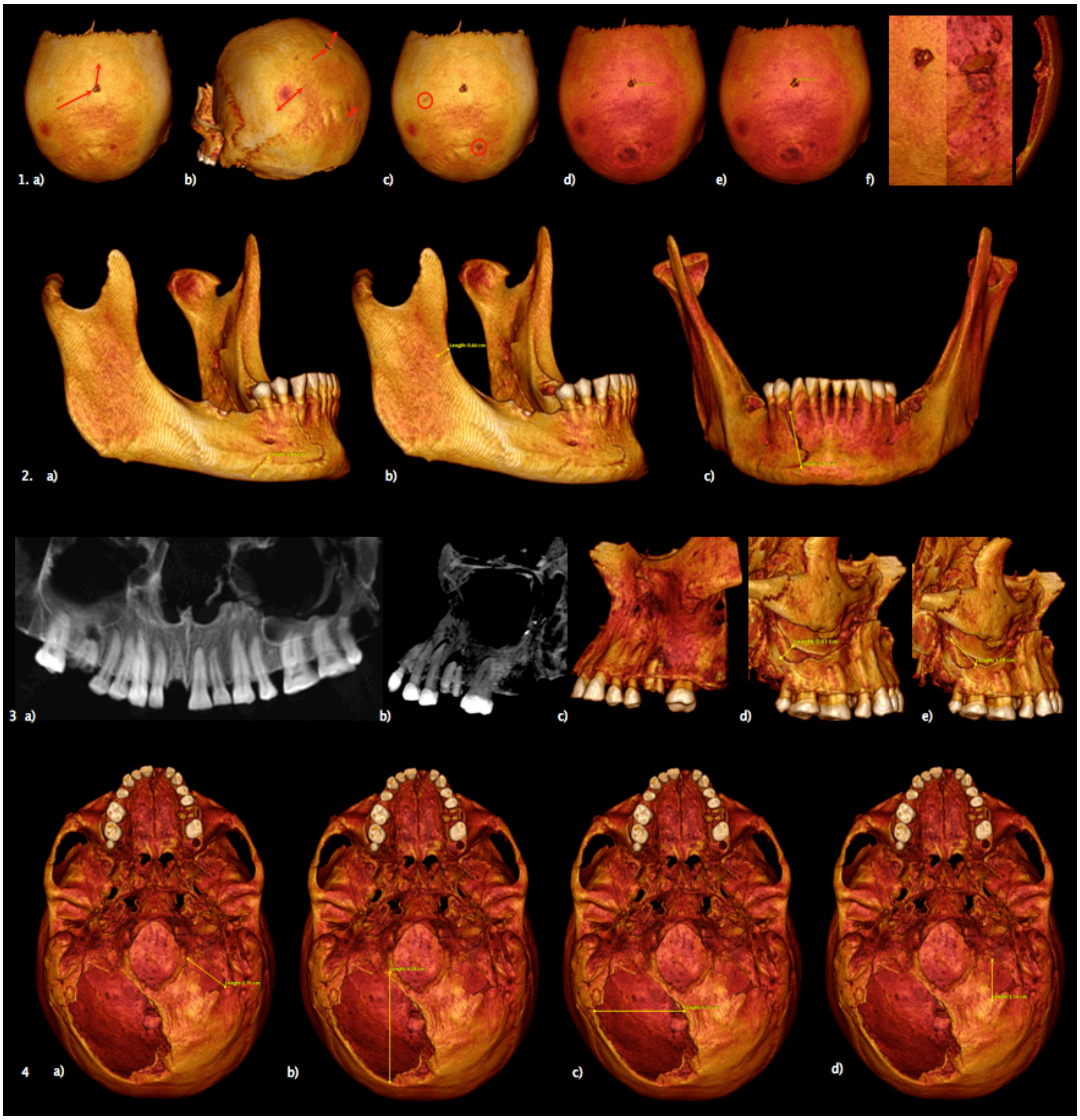


Figure 8.3: Additional images supplied on request, for trauma analysis.
1) Keyhole trauma, superficial grazing injuries, areas of potential trauma. 2) Mandible trauma. 3) Canine fracture and damage to the maxilla. 4) Bone loss to occipital region.

The injuries detected from the PMCT images were all consistent with the injuries recorded during the dry bone examination. PMCT measurements were also all in agreement with those recorded on the dry bone. Importantly, PMCT identified two additional areas of trauma that were overlooked during the dry bone examination. However, PMCT failed to detect a sharp force tool mark of the right tenth rib-even when re-examined.

As the skeleton was of great historical value great care had to be taken during handling. This caused a number of issues when measuring the dry skull. Unlike most forensic cases, where the skeletal elements could be reconstructed using resin or glue to make measurements easier to obtain, in this situation the skull had to be carefully held together by a team of practitioners while the measurements were recorded. This time consuming issue was resolved using PMCT-where the skull segments could be digitally re-constructed rapidly.

CHAPTER 9: FUTURE WORK

"When all is said and done, a bit more is said than done"

- Aesop.

9.1 Future Work

This thesis lays the foundations for a vast number of possible future studies, which can be roughly divided into four smaller projects:

1) *Adult trial of PMCT minimum data-set recording form;*

The next stage of this research project, to develop the minimum data-set recording form further, would involve conducting a similar investigation as discussed in chapter 7, on an adult cohort. Although a single case report has been presented on adult remains in chapter 8, a more exhaustive investigation would be required before the minimum data set form could be officially implemented in a forensic investigation. For this study, I would suggest using a sample size of approximately 30 (comparable to the juvenile trial). Cases would be available from the East Midlands Forensic Pathology Unit database, which already includes a large sample of consented adult cases.

This investigation would determine if it was necessary to include any additional images to help with adult biological identification. For example, it might be necessary to include images of rib-end development, pubic symphysis morphology or additional measurements. Using the feedback from the users, the minimum data set form could be modified accordingly, before further development and the next stage of investigation.

2) Large-scale international Trial of PMCT minimum data-set recording form;

After modifications of the recording form to accommodate adult biological identification, the next stage of research would involve conducting a large scale international trial of this approach. Recent communications with several international forensic centres has suggested that up to 14 countries would be interested in participating in such a trial. If a number of international forensic units integrated the recording form into their standard practice, it would be possible to determine whether or not this method is translational on an international scale and would identify any potential problem or limitations with this technique. It may also identify some areas for future development, so that this method is robust enough to satisfy the needs of different professionals within, pathology, radiology, imaging, anthropology, odontology and law. Further modification of the recording form in line with any feedback from these international facilities would ensure that the form included all necessary information.

3) Produce a 'quick PMCT ID' form;

At present the Netherlands do not use the full INTERPOL DVI form in a MFI. Instead they use a "quick scan ID" form. This is a single-sided leaflet, which includes only a fraction of the information included in the official DVI form. Many forensic experts believe that this is all the information required to make a quick ID of unknown remains. By combining this form with PMCT information, a picture record of the evidence could be accurately recorded.

The results of the research carried out in chapter 7 of this thesis, were recently presented at the annual INTERPOL DVI meeting. As a result, for the first time there is now a representative of forensic imaging on the inner committee for future INTERPOL development. Therefore, if the investigations suggested in this section were conducted, results were positive and the minimum data-set form was developed into a 'quick ID PMCT form' this could potentially be considered for inclusion in the next INTERPOL update, in four years.

4) Development of a PMCT teaching and learning database;

Having developed an appropriate PMCT minimum data-set recording format there is scope to develop an online anthropological teaching and learning database. This database could be operated similarly to the DNA database and allow the open sharing of skeletal anthropological information between different research facilities. A digital collection offers a number of advantages over a physical one, namely; it reduces the need to handle the sometimes-fragile bone samples and requires less physical storage. In addition, a digital database offers the opportunity to measure many properties that are difficult to quantify on a physical bone sample, but that are easily calculated numerically on the computer, such as; volume, surface area, and surface curvatures.

There are already a number of digital human anatomical databases. Probably the most well known of these is the visible human project from the U.S. National Library of Medicine (US National Library, 2014). It consists of full-body CT, MRI, and digital photographs of cross-sections of a human male and a human female subject. Similar projects exist in China (Zhang *et al*., 2004) and Korea (AUSM, 2014). Although, these projects only provide raw image and volume data and leave all image reconstruction to the user. Digital databases of individual bones also seem to be quite common for specific studies (Daruwalla *et al*., 2010; Kurazune, 2009); however, to my knowledge, very few of these databases are openly available for other researchers (see, for example, Moore *et al*., 2007 collection).

In contrast, a PMCT database could be constructed to allow open access to participating research facilities as an educational and teaching resource. Additionally, PMCT scans could not only be made available remotely on a secured network but also downloaded onto external hard drives so that data could be revisited indefinitely.

In order to maximize the effectiveness of a database, it would be necessary to ensure data is uploaded in a compatible format, so that different institutes and countries can contribute. The PMCT minimum data-set recording form developed in this thesis could be used as a formatting guideline to ensure that data is entered into the database in a compatible manner. By uploading a minimum data set form for each case, in addition to the raw data, researchers would have rapid access to sufficient information for biological profiling and could use it as an overview of each case in order to determine whether they require supplementary information by downloading the large raw data set.

For completeness, the PMCT images could be encoded with the known demographics for each case so that the database could be used as a valuable digital skeletal collection that would be available, indefinitely, for future research. This could aid research to update existing anthropological techniques, using a modern population; to develop new methods, using up-to-date software programmes; or to enhance traditional methods, using PMCT specific adaptations. In particular, an online database could be used to provide further evidence regarding the accuracy and reliability of PMCT measurements and could potentially be used to develop PMCT specific measurements and methods. This would be an invaluable resource for any researcher that is not available anywhere else at present.

The easily replicated, simple recording format would enable international data sharing, offering an opportunity to produce up-to-date ethnicity studies which are generally difficult to conduct due to issues with data sharing and access to ethnic minority skeletal data.

There is also scope for archaeologists and oesteo-archaeologists, with an interest in comparative osteology and skeletal pathology to create an online library of images, which could be a valuable study aid and reference collection.

The development of a digital system would of course raise critical issues concerning ownership and open access to the data. There would need to some discussion regarding ownership issues, in order to protect the intellectual property of the research facilities and of scientists who generate the scanned data on the PMCT database. A possible solution to these concerns might be to suggest that organisations such as the ISFRI, who aim to facilitate data sharing among researchers, are given control over access and ownership. This would also allow the system to be continually updated and upgraded depending on the developing needs of the collective membership to produce a database that is beneficial to the maximum number of researchers.

The scope of future work in this area is extensive, due to the constantly evolving nature of forensic casework and the rapid progression of technology. The work carried out in fulfilment of this thesis is a small stepping stone to aid the future development of forensic anthropology and forensic science in general, towards a future that is undoubtedly rooted by PMCT.

CHAPTER 10: DISCUSSION AND CONCLUSIONS

"To think and think for months and years. Ninety nine times, the conclusion is false. The hundredth time I am right."

- Albert Einstein.

10.1 Discussion

This thesis provides evidence that PMCT derived anthropological measurements are accurate (Brough *et al*., 2012; Brough *et al*., 2013; Brough *et al*., 2014) and has produced sufficient evidence to support the implementation of PMCT for anthropological examinations in routine forensic and disaster victim identification (DVI) events. Previous literature (Cavalcanti *et al*., 2004; Robinson *et al*., 2008; Verhoff *et al*., 2008; Dedouit *et al*., 2007) has tended to concentrate on isolated skeletal elements and has used raw PMCT data. Although providing useful results, these studies were not extensive enough to provide guidelines regarding what measurements are required, how to acquire these and how to best record information; so results can be shared internationally between numerous investigators.

The current official DVI Interpol form provides guidelines for the correct handling of remains and evidence in a disaster event. To date, this 'recording' form does not contain a PMCT section. On inspection of the current Interpol form, it appears that the majority of information required can be retrieved from data derived from PMCT. Over the last decade, the frequency of PMCT scanning has increased exponentially. To ensure that PMCT is considered for the next DVI Interpol update it is essential to develop an adequate PMCT recording format, which includes an anthropological reporting section.

In a DVI event, although it is possible to transfer large quantities of raw PMCT data between different countries (Rutty *et al*., 2009); this process takes approximately 20 minutes per case (or longer, if there are security measures such as firewalls in place), requires a large computer memory and storage facility, and post-processing of the raw data can be labour intensive (depending on the case). In addition, despite it being possible to transfer the data, there is no guarantee that the recipient has access to sufficient post-processing facilities or adequate knowledge of how to use these imaging systems. Using a minimum 'data-set' recording form, completed by a central investigator, which can be rapidly sent to numerous practitioners for independent analysis, would therefore be considerably beneficial in these situations. A standard PMCT reporting form should also ensure that an adequate amount of information about each case was recorded in a standard format; for multiple practitioners, using numerous anthropological identification techniques to use rapidly and remotely.

From the very initial stages of this project it became evident that there were no guidelines for anthropological PMCT reporting. There was no published literature regarding what anthropological measurements could be accurately recorded, what images were required for an adequate anthropological assessment, or how to appropriately process and record PMCT data to aid an anthropological assessment. As a consequence, the aim of this thesis was to answer these questions, resulting in the production of a minimum data set approach to anthropological biological profiling using PMCT. Juvenile age estimation is used as an example throughout the series of investigations presented in this thesis, to demonstrate the practical application of the suggested measurement procedure and final recording form.

When investigating human remains, forensic anthropologists face methodological choices; such as which skeletal region to evaluate, which method to apply, what statistical information to use and how to combine information from multiple methods to give a final estimated age, sex, stature and ethnicity to build an accurate biological profile. A questionnaire conducted by Garvin *et al*. (2012), given to 145 forensic anthropologists, found that the majority of respondents vary their skeletal estimate process case-by-case and ultimately present officials with both a narrow and a broad possible ranges. Overall, respondents displayed a very high degree of variation in how they generate their estimations, and indicate that experience and expertise play a large role.

The choice of method has to take into account the individual circumstances of each case. This depends in part on the skeletal elements available for analysis: different bones have variable resilience to events such as damaging taphonomic processes or high velocity impact, leading to preservation bias. Other considerations are related to the actual methods, which should fulfill the following specific demands (Ritz-Timme *et al*., 2000):

- They must have been presented to the scientific community through peer-reviewed publication;
- Their accuracy must be tested using valid statistical procedures and described by clearly defined terms; and
- The method must be accurate enough for routine forensic application.

The main objective of this thesis was to produce a minimum 'data-set' anthropological reporting form, however; including enough information to satisfy practitioners from different professional backgrounds and experience; using a range of anthropological methods depending on the case, was also a major consideration.

The preliminary research highlighted the broad scope of this project. There was very little evidence regarding the accuracy of PMCT measurements. Using the clavicle and the dentition, I was able to show that anthropological standard measurements could be replicated accurately using PMCT data. I was then able to identify a select number of images that provide sufficient information to conduct a full anthropological examination. By formatting these images onto a two page minimum 'data-set' form I have produced a minimum standard for radiological anthropological reporting, which can be easily replicated. This coherent post-mortem radiological recording form, which has been used to produce age estimations with accuracies comparable to traditional anthropological methods, can be used in any forensic or mass disaster situation, and promotes international quality control and best practice in the field of radiology and imaging worldwide.

10.1.1 Scope of this project

It appears that as a profession, forensic science is in danger of relying on obsolete technologies. Some areas in the field, such as genetics, toxicology and crime science investigation have experienced a number of revolutionary advances; but in contrast forensic pathology often still uses time-honoured, evidenced based methods that were developed centuries ago; namely, the physical dissection of a corpse, an oral description and then a written report of the findings. This has been slightly updated in the past decades by the addition of photography, but remains generally unchanged. Although conventional X-rays have become routinely implemented in forensic practice, the adoption of newer, clinically established methods, such as CT and MRI seems to be lagging behind. This conservative attitude towards new technologies is surprising, in a profession that is inherently linked with others (genetics and toxicology etc.) that have embraced novel methods. However in the UK reforms to the Coroners and Justice Act 2009, implemented in July 2013, pave the way for alternative methods to the classical autopsy for victim identification and post-mortem autopsy, making the inclusion of imaging technology into post-mortem investigation a very real possibility.

As well as the scientific potential of this research, it also has a significant potential public impact, with social applications, as continuing development should ultimately change the way post-mortem identification and autopsies are conducted, and the way the public perceive them.

I believe the future of forensic pathology, odontology and anthropology depends on encouraging research to exploit these new technologies to ensure criminal justice is not jeopardised by outdated technologies. Although CT has been used in medicine and forensic investigations for decades it is only recently, that PMCT's vast potential has truly been appreciated. However, the majority of journal articles on this subject are anecdotal case reports, or speculative analysis of CT as a modern alternative to traditional invasive post-mortems, and as a result, many important questions remain unanswered. Such as, what PMCT measurements would be the most useful to record, how to record these, and how to appropriately report the PMCT findings? However a new flourish of activity has begun regarding 'near virtual autopsy' techniques that could provide innovative solutions to enhance forensic casework.

Several institutes have already introduced PMCT into their investigations. For example, the universities of Copenhagen (Denmark) and Linkoping (Sweden) have started scanning forensic cases on a large scale and every corpse entering the Victorian Institute of Pathology (Sydney, Australia) undergoes a CT scan prior to autopsy. A group from the Office of Armed Forces Medical

Examiner (Armed Forces Institute of Pathology, Washington, D.C.) also perform CT scans on a routine basis to evaluate high velocity gunshot wounds of military personnel killed in combat. Other institutes routinely using CT include a French anthropology group who have used CT to analyse burned bodies with promising results, and the Institute of forensic Medicine of Bern, Switzerland, who began the Virtopsy project in 2000.

However, despite a number of publications from these centres, research and development in this area is generally basic. Most publications are of a speculative nature in relation to anthropology and the majority have been driven by specific requirements, on a case-to-case basis. For this reason, the research presented in this thesis has attempted to be translational research - where the method has been developed and refined using archive cases, and then tested by applying the methods to a randomly selected cohort of juvenile remains from a modern population; with its practical application being assessed by a number of practitioners from various professional backgrounds. As the nature of forensic science is complex, and covers a wide variety of topics, it is undoubtedly important for future development that research efforts are concentrated towards innovations that have the potential to link multiple disciplines and translate well into the working forensic environment.

The research presented in this thesis covers multiple research topics, including; radiology, imaging, forensic anthropology, and forensic odontology. If the minimum data set recording form presented in this thesis was included in the next update of the Interpol DVI forms, this research could potentially have an international impact. The standardisation of forensic radiological reporting would have important implications for future research, development and teaching. As well as spanning the fields of radiology, odontology, pathology and anthropology - there is an opportunity to build working relationships and collaborations with police forces, to ensure evidence is collected, processed and presented to a minimum standard of best practice. Furthermore, this research has social implications, potentially reducing the number of invasive autopsies required.

10.1.2 Future of forensic science in the UK

Forensic science service provision in the UK has undergone considerable changes during the last decade, culminating with the government announcing their decision to close the forensic science service (FSS) in 2011. The FFS was a government-owned company in the United Kingdom, which provided forensic science services to the police forces and government agencies of England and Wales, as well as to the Crown Prosecution Service, HM Revenue and Customs, HM Coroners' Service, Ministry of Defense Police, British Transport Police and worldwide forensic services. The organisation is probably best known for establishing the world's first DNA database in 1995, but also provided a source of training, consultancy and scientific support to numerous other organisations. There has been much debate about the impact the closure of this important resource would have on service delivery, impartiality of evidence and future research and development in the field of forensic science. Closure of the FSS consequently means that funding for forensic research is now even more difficult to acquire, which could make it challenging for the UK to sustain its reputation in forensic science and fall behind on the capitalisation of new research and technologies. Today, the priority for UK forensic science is therefore to ensure that this does not happen and to continue to provide high quality forensic services to local police forces and to the criminal justice system. Therefore, despite a chaotic landscape, great efforts have been made in the field of forensic science over the following two years to establish collaborations between researchers, practitioners, policy makers and those responsible for delivering improved crime prevention and detection. This has generated a new flourish of research activity, particularly surrounding new technologies such as PMCT, to help find innovative solutions to meet the UK's many and varied future forensic service provision needs.

I believe the future of forensic science in the UK relies on looking forward to identify potential problems and seizing the opportunity now, to develop revolutionary solutions to strengthen and enhance forensic casework, at every level. I think PMCT provides a prime opportunity to achieve this objective and to propel the UK to the forefront of forensic science research. Considering the early introduction of CT into clinical medicine, its use in forensic practice has been slow to follow. As a result, there are many prospective applications of this technology that still need to be explored and the potential for further development is vast. The progression of forensic science will undoubtedly involve the integration of digital imaging systems into routine practice, in the very near future. A mortuary based PMCT imaging facility would without a doubt enhance forensic casework.

At the Forensic Horizons Conference, which I attended in 2013, Andrew Miller MP, chair of the House of Commons Science and Technology branch asked the question: "*DNA fingerprinting was revolutionary - where [is] the next revolution coming from, and will we be at the forefront?"*

In response to this question, my answer would be that the next revolution in forensic science will almost certainly involve medical imaging, most probably PMCT, and if the UK continues to support research, such as the work carried out in fulfilment of this theses-then we will undoubtedly be at the forefront.

10.2 General Conclusions

- It is apparent that in the UK and also internationally, forensic practice procedures for the identification and autopsy of individuals are progressing rapidly in a 'digital' direction.

  This is evidenced in the UK by the introduction of cross-skill training programmes, such as a new teaching module at Leicester Medical School, that are being initiated to ensure radiologists can interpret forensic images and that forensic pathologists, anthropologists and odontologists, who will undoubtedly be involved in digital autopsies in the future, are trained in radiology, to ensure an appropriate standard of image interpretation.

- Despite an initial lag, forensic radiology is finally catching up with other areas of forensic science, such as genetics and crime scene investigation that have benefited from the advanced technologies developed for DNA and fingerprint analysis, through the exploitation of innovative applications of medical imaging, predominantly PMCT.

The formation of the International Society of Forensic Radiology and Imaging, a dedicated group of professionals working towards the common objective of developing the field of forensic radiology and imaging worldwide, is evidence of this progression. Members of this society include some of the most influential and well-respected professionals involved in forensic radiology and imaging worldwide.

- The scope for further research relating to PMCT is vast and covers multi-disciplines. As well as potentially affecting many scientific subject areas, the outcomes of future research in this area will also influence legal practice and will, undoubtedly, have a huge social impact.

  The far reaching interest in the development of PMCT in forensic practice is particularly evident when considering the varied professional backgrounds of the ISFRI membership, which includes; experts from forensic and general pathology, medical examiners and Coroners, autopsy technicians, radiology and radiologic technologists, medical and forensic photographers, MR physicists, surveying specialists, and computer scientists.

10.3 Final Conclusions

- The measurements and information required for the majority of methods used in current forensic anthropological practice can be extracted from PMCT data.

  Evidence of this was presented in Chapter 4, through an extensive review of the literature, using samples from the Scheuer juvenile skeletal collection as a study aid.

- PMCT derived anthropological measurements are accurate and repeatable, by multiple practitioners of various professional backgrounds and experiences.

  This is evidenced by the results of the investigations conducted in Chapter 5 and 6 of this thesis on the clavicle and dentition, respectively. A statistical analysis of the results illustrated that there was no significant difference between PMCT measurements and those recorded on dry bone or dental OPT's, respectively. Using juvenile age as a working example, the estimations from five independent researchers were shown, using interclass correlation calculations, to be in 'almost perfect agreement' with each other. Each observer was also found to be in 'almost perfect agreement' with actual documented age. This technique could therefore be used interchangeably with traditional methods, to produce as accurate and reliable results.

- The information required to conduct a comprehensive anthropological examination can be condensed into a concise two-page 'minimum data-set' form.

  Chapter 7 demonstrates, using juvenile age determination as an example, how this form could be used in any mass disaster or forensic scenario for anthropological, biological profiling. Using a single case example, chapter 8 also illustrates that this minimum data set approach to recording is transferrable to adult remains.

- This work conducted in this thesis has a practical application within current UK and international forensic science practice.

  As PMCT is undoubtedly becoming more prominent in standard forensic practice, as illustrated by the literature review and introduction chapters, the development of an appropriate recording format, to ensure data is collected and presented to a minimum standard, is vital.

This PhD presents a minimum standard of PMCT forensic anthropological reporting and provides a strong platform for future research on this topic.

APPENDIX 1: SCHEUER COLLECTION INVENTORY

APPENDIX 2: DENTAL OPT AND CURVED MPR DATA

APPENDIX 3: MINIMUM DATA SET FORMS

(PLEASE SEE ATTACHED CD)

APPENDIX 4: INTERPOL DVI FORMS

DISASTER VICTIM IDENTIFICATION (DVI)

HOW TO USE THE PINK POST-MORTEM (PM) FORM

Please write legibly.

I. RULES TO BE OBSERVED ON THE DISASTER SITE

No body should be moved before its location has been recorded.

All personal effect that undoubtedly belonged to a deceased individual should be collected and kept with the body or parts of the body of that individual. Any other effects should be recorded as unidentified and kept separately in the first instance.

A moisture resistant number card should be attached to each body or unidentified part of a body to ensure that it cannot get lost.

II. GENERAL INSTRUCTIONS

The PM form is designed for listing all obtainable data about a dead body that may assist in its identification in order to compare that data with the information obtained at the place of residence of the possible victim or missing person and recorded on the yellow ante-mortem form.

IMPORTANT: Record all data that can be obtained, since it is impossible to know what information will be supplied at the victim's place of residence for comparison purposes.

The layout of the form is intended to correspond to the actual sequence of events, and allows a simultaneous examination of effects, body, and teeth.

Where provided, use the appropriate figures for description.

EXAMPLE: Section C1: Fill in the figures "0203" in the "No." column at item 24 to designate a pullover and describe the material, etc. in the space provided for this information.

Wherever appropriate, boxes that can simply be marked with a cross are provided. Please use as many of them as possible. This will facilitate electronic processing of the information and also make it possible to handle reports compiled in a foreign language without translation (the Interpol Member States all use the same forms). For this reason, the layout is the same for the AM and PM forms.

III. SPECIFIC INSTRUCTIONS

Section B	Recovery of body from site: Fill in this form during recovery from the site of the disaster and add the number from the number-board attached to the body or part of the body.
Sections C1 to C3	Photograph the body first, then remove any clothing and jewellery from the body. C1 - clothing and shoes C2 - personal effects C3 - jewellery
Sections D1 to D5	While the effects are examined and described. D1 to D3 - physical description of the dead body. D4 - record any distinguishing marks (tattoos, etc.) D5 - record any fingerprint information.
Sections E1 to F2	a medical examination is performed E1 to E3 - list all data obtained by an internal examination that may assist in identification. E4 - DNA profiles F1 & F2 - dental data (cf. instructions on the back of Section F1)
Sections G	Record any further information that may assist in identification, and/or continue with your description from a previous section (C to F) if there is not enough space.

If an identification is made, complete a "Victim Identification Report" in accordance with the instructions.

P~ost~M~ortem~ (pink) **VICTIM IDENTIFICATION FORM** **B0**

DEAD BODY

Nature of disaster :	____________________	No: ____________ Barcode
Place of disaster :	____________________	Male ☐ Female ☐ Sex unknown ☐
Date of disaster :	☐☐ Day ☐☐ Month ☐☐☐☐ Year	

CHECKLIST OF OPERATIONS IN THE MORTUARY			Date	Remarks
Photographs	Full size - front, back	☐ With clothes ☐ Without clothes		
	Head	☐ Front ☐ From left ☐ From right		
Fingerprints	Finger	☐ No ☐ Not Possible ☐ Yes		
	Palm of the hand	☐ No ☐ Not Possible ☐ Yes		
Autopsy	Medicolegal examin.	☐ No ☐ Yes		
	Full autopsy	☐ No ☐ Yes ☐ X-rays ☐ Photo		
	Pathologist name Address/Phone			
Dental examination	Completed	☐ No ☐ Yes ☐ X-rays ☐ Photo		
	Jaws removed	☐ No ☐ Yes ☐ X-rays ☐ Photo		
	Odontologist name Address/Phone			
Samples		☐ Taken ☐ Sent for analysis ☐ Result enclosed		
	(cf. E2 item 73)	☐ DNA profiles ordered		

CHECK LIST OF CONTENTS		Enclosed complete	Enclosed in part	Issued to Name	Date	Returned Date	Remarks
B	Recovery from scene						
C1	Clothing and Foot wear						
C2	Personal Effects						
C3	Jewellery						
D1	Physical description						
D2	Physical desc. cont.						
D3	Physical desc. cont.						
D4	Body sketch						
D5	Fingerprint information						
E1	Internal examination						
E2	Medical conclusions						
E3	Skeleton sketch						
E4	DNA						
F1	Dental findings						
F2	Dental findings cont.						
G	Further information						

P~ost~M~ortem~ (pink) **VICTIM IDENTIFICATION FORM** **B**

DEAD BODY **No:** ____________ Barcode

Nature of disaster : ______________________

Place of disaster : ______________________

Date of disaster : [ ][ ] Day [ ][ ] Month [ ][ ][ ][ ] Year — Male [ ] Female [ ] Sex unknown [ ]

a = Data not available b = Photo c = Further information on page G

RECOVERY OF BODY FROM SCENE									a	b	c
20	**Apparent age**	*Min* ______ / *Max* ______		*Txt:* ______							
21	**Date -** **and place where the body was found**	[ ][ ] *Day* [ ][ ] *Month* [ ][ ][ ][ ] *Year*									
	01 Map reference/GPS	*Coordinates:* ______ / ______									
	02 Photographs	1 [ ] *No*	2 [ ] *Yes*	3 [ ] *Digital*	4 [ ] *Film*	5 [ ] *Other /Specify:*					
22	**State of the body**	*Complete* 1 [ ]	*Incomplete* 2 [ ]	*Presentable* 3 [ ] *No*	4 [ ] *Yes*	*Body part(describe)* 5 [ ] ______					
		1 *Damaged*	2 *Burnt*	3 *Decomp.*	4 *Skelet.*	5 *Missing*	6 *Loose*				
	01 Head										
	1A Neck / Throat										
	02 Right arm										
	03 Left arm										
	04 Right hand										
	05 Left hand										
	06 Body front										
	07 Body back										
	08 Right leg										
	09 Left leg										
	10 Right foot										
	11 Left foot										
22 A	**Important ID information**										
23	**Person - finding the body** If an ID-team is involved - name officer in charge										
	Any other person - Name, Address, Phone/E-mail, Occupation										

Registered by		
Duty Title :		Signature / Date
Name :		
Address :		
Phone/E-mail :		

[(GB) Version 2008]

PostMortem (pink) **VICTIM IDENTIFICATION FORM** **C1**

DEAD BODY

Nature of disaster :		**No:** ______ Barcode
Place of disaster :		Male ☐ Female ☐ Sex unknown ☐
Date of disaster :	☐☐ Day ☐☐ Month ☐☐☐☐ Year	

a = Data not available b = Photo c = Further information on page G

CLOTHING AND FOOT WEAR

		No:	1 Material	2 Colour	3 Type	4 Label	5 Size	a	b	c
24	**Clothing Items** **01 Head and neck** 0101 Hat 0102 Scarf 0103 Tie 0199 Other **02 Upper part of the body and arms** 0201 Overcoat 0202 Coat 0203 Pullover 0204 Shirt 0205 Waistcoat 0206 Vest 0207 Dress 0208 Cardigan 0209 Blouse 0210 Petticoat 0211 Chemise 0212 Brassiere 0213 Braces 0214 Gloves 0215 Jacket 0299 Other **03 Lower part of the body and legs** 0301 Trousers (men) 0302 Underpants 0303 Trousers (women) 0304 Skirt 0305 Panties 0306 Girdle 0307 Corset 0308 Stockings 0309 Tights 0310 Socks 0311 Belt 0312 Belt buckle 0313 Shorts 0314 Swimming attire 0399 Other **04 The whole of the body** 0401 Flying suit 0402 Boiler suit 0403 Trouser suit 0499 Other In case of using "xx99 Other" describe the kind of item in column "3 Type".									

		No:	1 Material	2 Colour	3 Type	4 Label	5 Size	a	b	c
25	**Foot wear** 01 Shoes 1A Open footwear 03 Boots 99 Other Describe the kind of Foot wear in column "3 Type", eg Sport shoes Sandals									

Registered by	Duty Title : Name : Address : Phone/E-mail :	Signature / Date

[(GB) Version 2008]

P ost M ortem (pink) **VICTIM IDENTIFICATION FORM** **C2**

DEAD BODY

No: ____________
Barcode

Nature of disaster : ______________________

Place of disaster : ______________________

Date of disaster : [ ][ ] Day [ ][ ] Month [ ][ ][ ][ ] Year

Male [ ] Female [ ] Sex unknown [ ]

a = Data not available b = Photo c = Further information on page G

PERSONAL EFFECTS			a	b	c
26	**Watch** 00 Wearing watch	1 [ ] No 2 [ ] Yes			
	01 Digital 02 Analog 03 Digital/Analog	No: 1 Material / 2 Colour / 3 Design / 4 Brand / 5 Inscription			
	04 If wrist watch worn on	Left 1 [ ] Right 2 [ ] Outside 3 [ ] Inside 4 [ ]			
	05 Watch strap/chain	Leather 1 [ ] Metal 2 [ ] Other (specify): 3 [ ]			
	06 Watch, other type	Where worn:			
27	**Glasses** 00 Wearing glasses	1 No 2 Yes			
	01 Frame	1 Material / 2 Colour / 3 Design / 4 Brand / 5 Inscription			
	02 Lenses (glass)	Tinted 1 [ ] No 2 [ ] yes (specify): Strength - Left/Right 3 [ ] L 4 [ ] R			
	03 Lenses/Shape	Round 1 [ ] Oval 2 [ ] Square / Half 3 [ ] 4 [ ] Rimless 5 [ ]			
	3A Lens type	Glass 1 [ ] Polycarbonate 2 [ ] Bi-focal 3 [ ]			
	04 Contact lenses	1 [ ] No 2 [ ] yes (colour?): Strength - Left/Right 3 [ ] L 4 [ ] R			
28	**Identity Papers** 00 Carrying ID-papers	1 [ ] No 2 [ ] Yes			
	01 Passport 02 Driving licence 03 Credit cards 04 Identity card 05 Donor card 06 Travellers cheques 07 Personal cheques 08 Health card 99 Other	No: 1 Type / 2 Photograph / 3 Fingerprint / 4 Blood type			
29	**Effects** 00 Carrying effects	1 [ ] No 2 [ ] Yes			
	01 Wallet 02 Purse 03 Money belt 04 Badges/keys 05 Currency 06 Mobile phone 07 PDA 08 Sim card 09 Ticket 10 Camera/Video 99 Other	No: 1 Material / 2 Colour / 3 Design / 4 Brand / 5 Markings			

Registered by Duty Title :
Name :
Address :
Phone/E-mail :

Signature / Date

[(GB) Version 2008]

PostMortem (pink) **VICTIM IDENTIFICATION FORM** **C3**

DEAD BODY

No: ____________
Barcode

Nature of disaster : ____________________

Place of disaster : ____________________

Date of disaster : ☐☐ Day ☐☐ Month ☐☐☐☐ Year

Male ☐ Female ☐ Sex unknown ☐

a = Data not available b = Photo c = Further information on page G

JEWELLERY | a | b | c

30 **Rings, chains etc.**

01 Wedding ring
02 Other finger rings
03 Earrings
04 Earclips
05 Neck chains
06 Necklace
07 Bracelets
08 Other chains
09 Pendant on chain
10 Piercing trinkets
11 Nose ring
12 Anklet
99 Other

In case of using "99 Other" describe the kind of item in column "3 Design".

No:	1 Material	2 Colour	3 Design	4 Inscription	5 Where worn	a	b	c

Registered by
Duty Title :
Name :
Address :
Phone/E-mail :

Signature / Date

[(GB) Version 2008]

P ost M ortem (pink) **VICTIM IDENTIFICATION FORM** **D1**

DEAD BODY

Nature of disaster : ____________________

Place of disaster : ____________________

Date of disaster : ☐☐ Day ☐☐ Month ☐☐☐☐ Year

No: ____________ Barcode

Male ☐ Female ☐ Sex unknown ☐

a = Data not available b = Photo c = Further information on page G

PHYSICAL DESCRIPTION (at mortuary)									a	b	c
31	**State of the body**	Complete 1 ☐	Incomplete 2 ☐	Presentable 3 ☐ No	4 ☐ Yes	Body part(describe) 5 ☐					
		1 Damaged	2 Burnt	3 Decomp.	4 Skelet.	5 Missing	6 Loose				
	01 Head										
	1A Neck / Throat										
	02 Right arm										
	03 Left arm										
	04 Right hand										
	05 Left hand										
	06 Body front										
	07 Body back										
	08 Right leg										
	09 Left leg										
	10 Right foot										
	11 Left foot										
		Indicate specific details on body sketch, page D4.									
31	**Estimated age**	Min /	Max	Method used ?							
32	**Height**	Min/cm /	Max/cm	Method used ?							
33	**Weight**	Min/kg /	Max/kg	Method used ?							
34	**Build** 01 Bodily constitution	Light 1 ☐	Medium 2 ☐	Heavy 3 ☐							
	02 Head form, front	Oval 1 ☐	Pointheaded 2 ☐	Pyramidal 3 ☐	Circular 4 ☐	Rectangular 5 ☐	Quadrangular 6 ☐				
	(02-03 see Silhouette sketch) 03 Head form, profile	Shallow 1 ☐	Medium 2 ☐	Deep 3 ☐							
35	**Race** 01 Group	Caucasoid 1 ☐	Mongoloid 2 ☐	Negroid 3 ☐	Type:						
	02 Complexion	Light 1 ☐	Medium 2 ☐	Dark 3 ☐							
36	**Hair of the head** 01 Type	Natural 1 ☐	Artificial 2 ☐	Hair-piece 3 ☐	Wig 4 ☐	Braided 5 ☐	Implanted 6 ☐				
	02 Length	Short<5cm 1 ☐	Medium<12cm 2 ☐	Long>12cm 3 ☐	Shaved 4 ☐						
	03 Colour	Blond 1 ☐	Brown 2 ☐	Black 3 ☐	Red 4 ☐	Grey 5 ☐	White 6 ☐				
	04 Shade of colour	Light 1 ☐	Medium 2 ☐	Dark 3 ☐	Turning grey 4 ☐	Dyed 5 ☐	Streaked 6 ☐				
	05 Thickness	Thin 1 ☐	Medium 2 ☐	Thick 3 ☐							
	06 Style	Straight 1 ☐	Wavy 2 ☐	Curly 3 ☐ /	Parted 4 ☐ Left	5 ☐ Right	6 ☐ Middle				
	07 Baldness	Beginning 1 ☐	Advanced 2 ☐	Total 3 ☐ /	Forehead 4 ☐	Sides 5 ☐	Tonsure 6 ☐				
	08 Other	(specify):									

Registered by		Signature / Date
	Duty Title :	
	Name :	
	Address :	
	Phone/E-mail :	

[(GB) Version 2008]

P ost M ortem (pink) **VICTIM IDENTIFICATION FORM** **D2**

DEAD BODY

No: ____________________
Barcode

Nature of disaster : ____________________

Place of disaster : ____________________

Date of disaster : ☐☐ Day ☐☐ Month ☐☐☐☐ Year

Male ☐ Female ☐ Sex unknown ☐

a = Data not available b = Photo c = Further information on page G

PHYSICAL DESCRIPTION (cont.)			a	b	c
37	**Forehead** 01 Height / Width (01-02 see Silhouette sketch)	*Low* 1 ☐ *Medium* 2 ☐ *High* 3 ☐ / *Narrow* 4 ☐ *Medium* 5 ☐ *Wide* 6 ☐			
	02 Inclination	*Protruding* 1 ☐ *Vertical* 2 ☐ *Receding/slightly or clearly* 3 ☐ S 4 ☐ C			
38	**Eyebrows** 01 Shape / Thickness	*Straight* 1 ☐ *Arched* 2 ☐ *Joining* 3 ☐ / *Thin* 4 ☐ *Medium* 5 ☐ *Thick* 6 ☐			
	02 Peculiarities	*Plucked* 1 ☐ *Tattooed* 2 ☐			
39	**Eyes** 01 Colour	*Blue* 1 ☐ *Grey* 2 ☐ *Green* 3 ☐ *Brown* 4 ☐ *Black* 5 ☐			
	02 Shade	*Light* 1 ☐ *Medium* 2 ☐ *Dark* 3 ☐ *Mixed* 4 ☐			
	03 Distance between eyes	*Small* 1 ☐ *Medium* 2 ☐ *Large* 3 ☐			
	04 Peculiarities	*Cross-eyed* 1 ☐ *Squint-eyed* 2 ☐ *Artificial eye* 3 ☐ Left 4 ☐ Right			
40	**Nose** 01 Size / Shape	*Small* 1 ☐ *Medium* 2 ☐ *Large* 3 ☐ / *Pointed* 4 ☐ *Roman* 5 ☐ *Alcoholics* 6 ☐			
	02 Peculiarities	*Marks of spectacles* 1 ☐ No 2 ☐ Yes *Misshapen* 3 ☐ *Other(specify):* 4 ☐ ____________			
	(03 see Silhouette sketch) 03 Curve / Angle	*Concave* 1 ☐ *Straight* 2 ☐ *Convex* 3 ☐ / *Turned down* 4 ☐ *Horizontal* 5 ☐ *Turned up* 6 ☐			
41	**Facial hair** 01 Type	*No beard* 1 ☐ *Moustache* 2 ☐ *Goatee* 3 ☐ *Whiskers* 4 ☐ *Full beard* 5 ☐			
	02 Colour	*Blond* 1 ☐ *Brown* 2 ☐ *Black* 3 ☐ *Red* 4 ☐ *Grey* 5 ☐ *White* 6 ☐			
42	**Ears** 01 Size / Angle	*Small* 1 ☐ *Medium* 2 ☐ *Large* 3 ☐ / *Close-set* 4 ☐ *Medium* 5 ☐ *Protruding* 6 ☐			
	(02 see Silhouette sketch) 02 Ear lobes / Pierced	*Attached* 1 ☐ No 2 ☐ Yes / *Pierced - specify number of piercings* 3 ☐ Left ______ 5 ☐ Right ______			
43	**Mouth** 01 Size / Other	*Small* 1 ☐ *Medium* 2 ☐ *Large* 3 ☐ / *Other (specify):* 4 ☐			
44	**Lips** 01 Shape / Other	*Thin* 1 ☐ *Medium* 2 ☐ *Thick* 3 ☐ / *Made up* 4 ☐ *Other (specify):* 5 ☐			
45	**Teeth** (cf.page F1/F2) 01 Conditions	*Natural* 1 ☐ *Untreated* 2 ☐ *Treated* 3 ☐ *Crowns* 4 ☐ *Bridges* 5 ☐ *Implants* 6 ☐			
	02 Gaps / Missing teeth	*Gaps between front teeth* 1 ☐ Upper 2 ☐ Lower / *Missing teeth* 3 ☐ Upper 4 ☐ Lower *Toothless* 5 ☐ Upper 6 ☐ Lower			
	03 Dentures	*Part.upper* 1 ☐ *Part.lower* 2 ☐ *Full upper* 3 ☐ *Full lower* 4 ☐ *ID-number(specify):* 5 ☐ ____________			
46	**Smoking habits** 01 Stains found	*No* 1 ☐ *Teeth* 2 ☐ / *Lips* 3 ☐ *Moustache* 4 ☐ *Finger / Hands* 5 ☐ Left 6 ☐ Right			

Registered by
Duty Title :
Name :
Address :
Phone/E-mail :

Signature / Date

[(GB) Version 2006]

P ost M ortem (pink) **VICTIM IDENTIFICATION FORM** **D3**

DEAD BODY

No: ____________
Barcode

Nature of disaster : ____________________

Place of disaster : ____________________

Date of disaster : ☐☐ Day ☐☐ Month ☐☐☐☐ Year

Male ☐ Female ☐ Sex unknown ☐

a = Data not available b = Photo c = Further information on page G

PHYSICAL DESCRIPTION (cont.)			a	b	c
47	**Chin** 01 Size / Inclination	Small 1☐ Medium 2☐ Large 3☐ / Receding 4☐ Medium 5☐ Protruding 6☐			
	02 Shape	Pointed 1☐ Round 2☐ Angular 3☐ Cleft chin 4☐ Groove 5☐			
48	**Neck** 01 Length / Shape	Short 1☐ Medium 2☐ Long 3☐ / Thin 4☐ Medium 5☐ Thick 6☐			
	02 Peculiarities	Goitre 1☐ Prominent Adams apple 2☐ Collar / Shirt No 4☐ Circumference 6☐ in cm			
49	**Hands** 01 Shape / Size	Slender 1☐ Medium 2☐ Broad 3☐ / Small 4☐ Medium 5☐ Large 6☐			
	02 Nail length	Short 1☐ Medium 2☐ Long 3☐			
	03 Peculiarities	Bitten short 1☐ Manicured 2☐ Painted 3☐ Artificial 4☐ Nicotine 5☐ Left 6☐ Right			
50	**Feet** 01 Shape / Size	Slender 1☐ Medium 2☐ Broad 3☐ Flatfooted 4☐ Arched 5☐ / Length in cm 6☐			
	02 Condition / Nail	Bunion 1☐ Corn 2☐ / Painted 3☐ Defective 4☐			
	03 Peculiarities	(Specify): ____________________			
51	**Body hair** 01 Extent	None 1☐ Slight 2☐ Medium 3☐ Pronounced 4☐			
	02 Colour	Blond 1☐ Brown 2☐ Black 3☐ Red 4☐ Grey 5☐ White 6☐			
52	**Pubic hair** 01 Extent	None 1☐ Slight 2☐ Medium 3☐ Pronounced 4☐ Shaved 5☐			
	02 Colour	Blond 1☐ Brown 2☐ Black 3☐ Red 4☐ Grey 5☐ White 6☐			
53	**Specific details** 01 Head, 1A Neck / Throat, 02 Right arm, 03 Left arm, 04 Right hand, 05 Left hand, 06 Body - front, 07 Body - back, 08 Right leg, 09 Left leg, 10 Right foot, 11 Left foot	No: | 1 Scars/Piercing | 2 Skin marks | 3 Tattoo marks | 4 Malformations | 5 Amputations — Indicate specific details on body sketch, page D4.			
54	**Circumcision**	1☐ No 2☐ Yes			
55	**Other peculiarities**				

Registered by Duty Title :
Name :
Address :
Phone/E-mail :

Signature / Date

[(GB) Version 2008]

PostMortem (pink) **VICTIM IDENTIFICATION FORM** **D4**

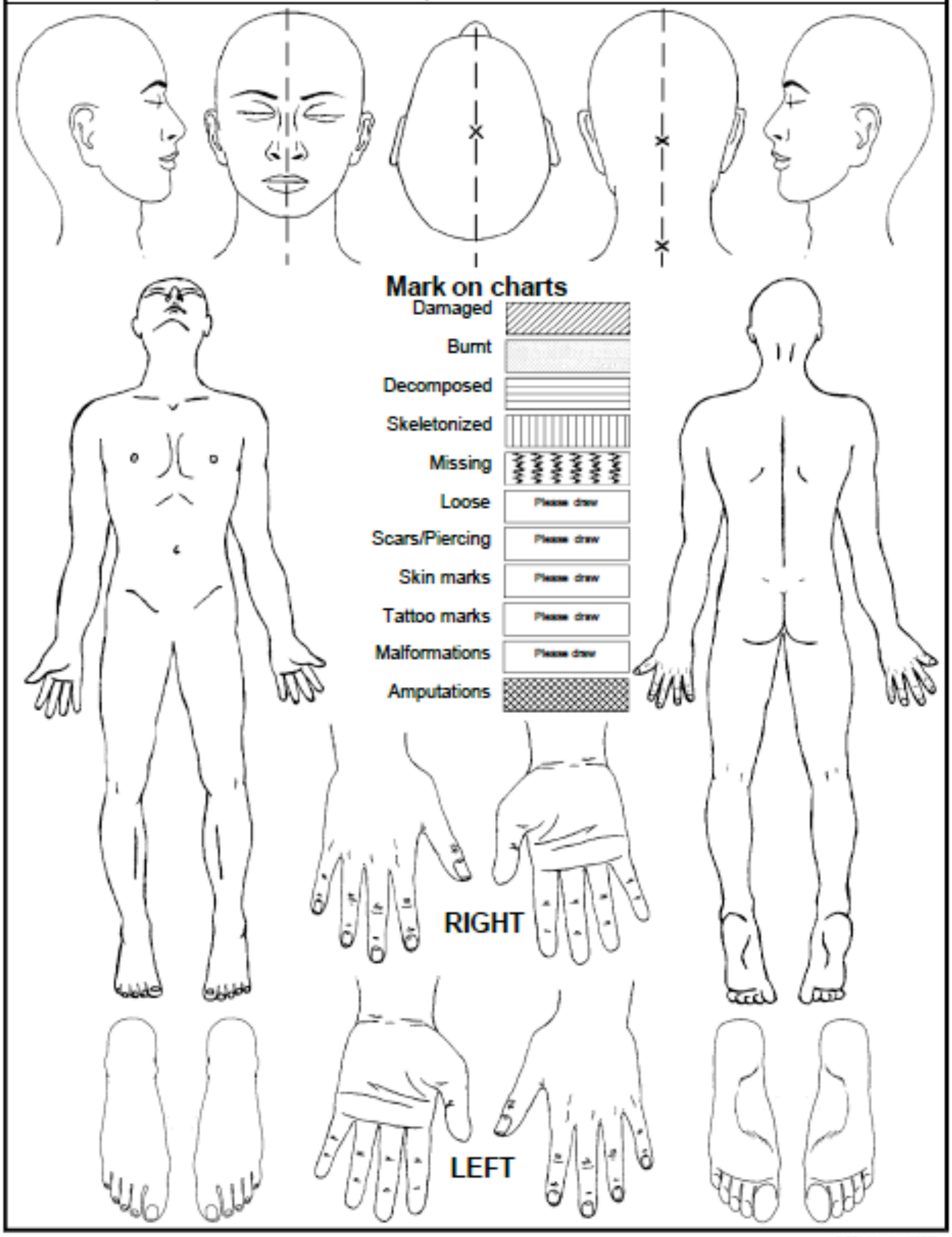
DEAD BODY

Nature of disaster : ____________________ **No:** ____________ Barcode

Place of disaster : ____________________

Date of disaster : ☐☐ Day ☐☐ Month ☐☐☐☐ Year Male ☐ Female ☐ Sex unknown ☐

BODY SKETCH (described in item 22 and/or 31, 53)



[(GB) Version 2008]

Post**M**ortem (pink) **VICTIM IDENTIFICATION FORM** **D5**

DEAD BODY

Nature of disaster : _______________________

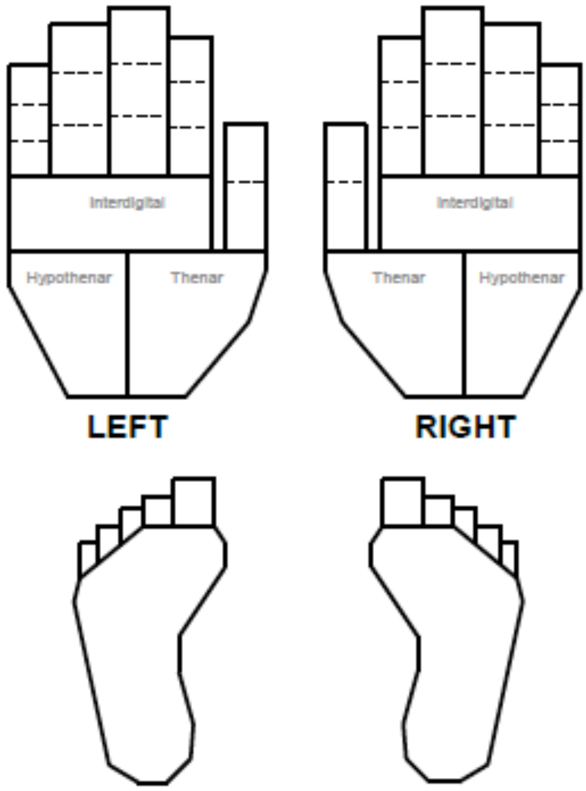
Place of disaster : _______________________

Date of disaster : ☐☐ Day ☐☐ Month ☐☐☐☐ Year

No: ____________
Barcode

Male ☐ Female ☐ Sex unknown ☐

a = Data not available b = Photo c = Further information on page G

FINGERPRINT INFORMATION			a	b	c
01	Skin type fingerprints retrieved from	1 ☐ EPIDERMIS 2 ☐ DERMIS			
02	Fingerprint development technique	1 ☐ Boiling water technique 2 ☐ Epidermal glove 3 ☐ Casting agent, eg Microsil, Aquasil 4 ☐ Other:			
03	Fingerprints recorded using	1 ☐ Black powder 2 ☐ Ink 3 ☐ Photograph 4 ☐ Other: ________			
04	Prints retrieved from	 SHADE AREAS PRINTS RETRIEVED FROM			

Registered by		Signature / Date
	Duty Title :	
	Name :	
	Address :	
	Phone/E-mail :	

[(GB) Version 2008]

P ost M ortem (pink) **VICTIM IDENTIFICATION FORM** **E1**

DEAD BODY

Nature of disaster : ____________________ **No:** ________ Barcode

Place of disaster : ____________________

Date of disaster : ☐☐ Day ☐☐ Month ☐☐☐☐ Year   Male ☐ Female ☐ Sex unknown ☐

a = Data not available / indefinable b = Photo c = Injuries and further information on page G d = X-rays

INTERNAL EXAMINATION - Full autopsy ☐ No ☐ Yes - autopsy No:

		No:		a	b	c	d
60	**Head** 01 Head 1A Skull 1B Brain 02 Neck						
61	**Chest** 01 Thorax/Ribs/Sternum 02 Lungs 03 Heart/Vessels						
62	**Abdomen** 01 Stomach 02 Intestines 03 Appendix						
63	**Other internal organs** 01 Adrenals/pancreas /Spleen 02 Liver/Gall bladder 03 Kidneys/Ureters/Bladder 04 Genitalia-male 05 Genitalia-female 06 Hysterectomy						
64	**Skeleton/Soft tissue** 01 Vertebral column 02 Pelvis 03 Limbs-right arm 04 Limbs-left arm 05 Limbs-right leg 06 Limbs-left leg 07 Other Bones 08 Soft tissue, other locations						
65	**Various** 01 Pregnancies 02 Healed fractures 03 Operations 04 Demonstrable pathological condition (e.g. heart disease, cancer etc.)						
	IMPLANT: 05 Intrauterine contra-ceptive devices	Metal 1 ☐	Plastic 2 ☐ Describe:				
	06 Other implants	Metal 1 ☐	Plastic 2 ☐ Describe:				

Continued item no 71 (Item 66 - 70 in form AM only)

Registered by Duty Title :
Name :
Address :
Phone/E-mail :

Signature / Date

[(GB) Version 2008]

P ost M ortem (pink) **VICTIM IDENTIFICATION FORM** **E2**

DEAD BODY

No: ____________
Barcode

Nature of disaster : ____________________

Place of disaster : ____________________

Date of disaster : ☐☐ Day ☐☐ Month ☐☐☐☐ Year

Male ☐ Female ☐ Sex unknown ☐

MEDICAL CONCLUSIONS

71	**Sex**	Male 1 ☐	Female 2 ☐	Undetermined 3 ☐	Reason of decision
72	**Estimated age**	Min ____ / Max ____			Method used

73	**Samples taken**			Purpose	Place of storage	Result
	01 Stomach contents	1 ☐ No	2 ☐ Yes			
	02 Urine	1 ☐ No	2 ☐ Yes			
	03 Blood-heart	1 ☐ No	2 ☐ Yes			
	04 Blood-peripheral	1 ☐ No	2 ☐ Yes			
	05 Blood-elsewhere	1 ☐ No	2 ☐ Yes			
	06 Bile	1 ☐ No	2 ☐ Yes			
	07 Vitreous humour L	1 ☐ No	2 ☐ Yes			
	08 Vitreous humour R	1 ☐ No	2 ☐ Yes			
	09 Other fluids	1 ☐ No	2 ☐ Yes			
	10 Symphysis pubis	1 ☐ No	2 ☐ Yes			
	11 Hair	1 ☐ No	2 ☐ Yes			
	12 Tissue dry	1 ☐ No	2 ☐ Yes			
	13 Tissue in formalin	1 ☐ No	2 ☐ Yes			
	14 DNA-specimens	1 ☐ No	2 ☐ Yes			
	Where were the DNA samples taken from	Specify:				
	Number of DNA samples taken	Specify:				

74	**Other clues for identification**	1 ☐ No 2 ☐ Yes (describe)
75	**Other medical findings**	

Continued item no 83 (Item 76 - 82 in form AM only)

Registered by	Duty Title : Name : Address : Phone/E-mail :	Signature / Date

[(GB) Version 2008]

Post**M**ortem *(pink)* **VICTIM IDENTIFICATION FORM** **E3**

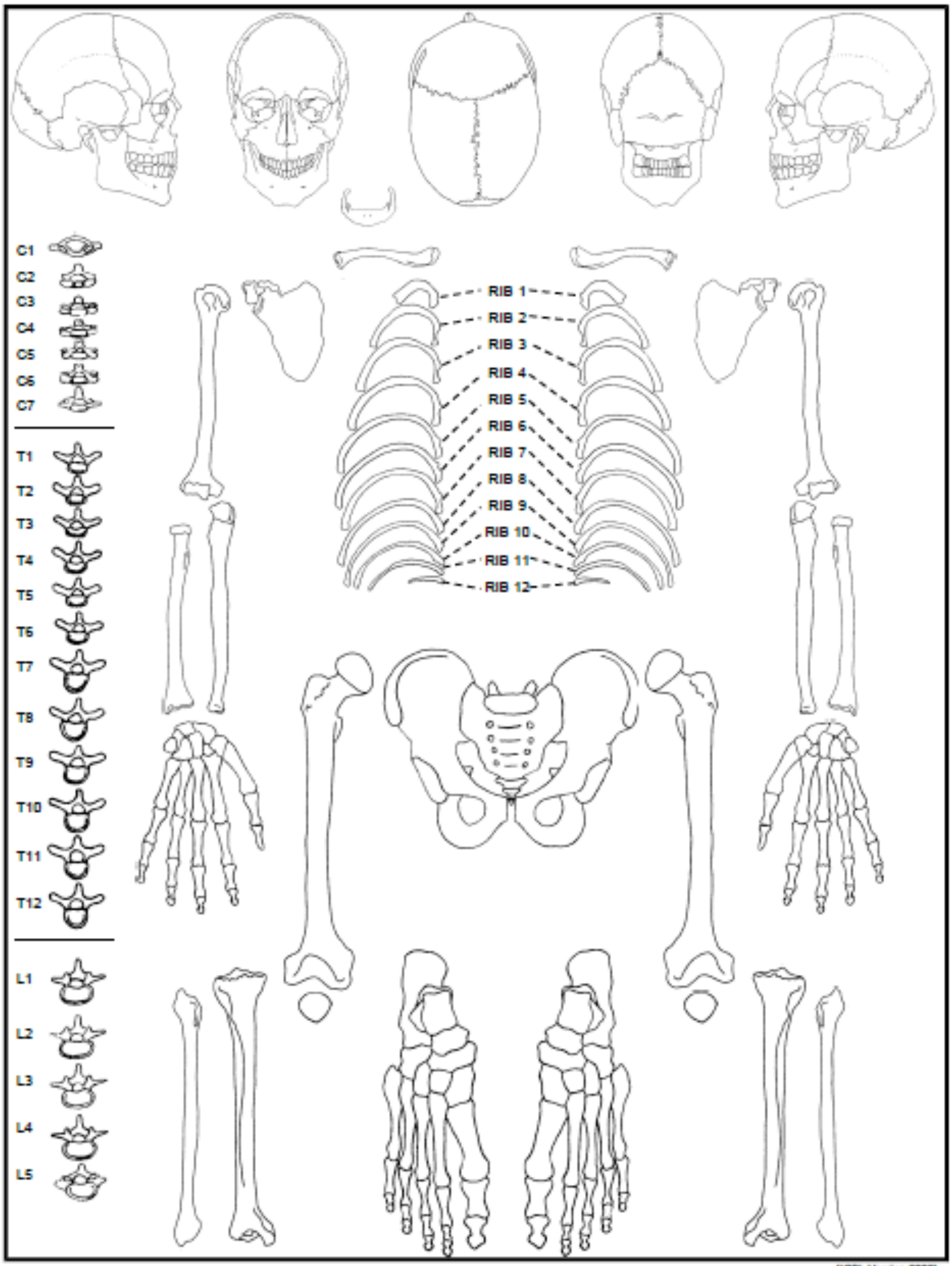
DEAD BODY

Nature of disaster : ____________________ **No:** ______________ Barcode

Place of disaster : ____________________

Date of disaster : ☐☐ *Day* ☐☐ *Month* ☐☐☐☐ *Year*

Male ☐ *Female* ☐ *Sex unknown* ☐



[(GB) Version 2008]

P ost M ortem (pink) **VICTIM IDENTIFICATION FORM** **E4**

DEAD BODY

No: ______________
Barcode

Nature of disaster : ____________________

Place of disaster : ____________________

Date of disaster : ☐☐ Day ☐☐ Month ☐☐☐☐ Year

Male ☐ Female ☐ Sex unknown ☐

c = Further information on page G

DNA			c
93	1. Sample	Received (date):	
		Label:	
		Type:	
		Condition:	
	2. Sample	Received (date):	
		Label:	
		Type:	
		Condition:	
	3. Sample	Received (date):	
		Label:	
		Type:	
		Condition:	
	4. Sample	Received (date):	
		Label:	
		Type:	
		Condition:	

94	DNA profiles	1. Sample		2. Sample		3. Sample		4. Sample		
	D3S1358									
	VWA									
	D16S539									
	D2S1338									
	Amelogenin									
	D8S1179									
	D21S11									
	D18S51									
	D19S433									
	TH01									
	FGA									
	TPOX									
	CSF1PO									
	D13S317									
	D7S820									
	D5S818									
	Penta D									
	Penta E									
	FES									
	F13A1									
	F13B									
	SE33									
	CD4									
	GABA									
95	Checked by	Date:				Signature:				

Registered by	Duty Title : Name : Address : Phone/E-mail :	Signature / Date

[(GB) Version 2008]

P ost M ortem (pink) **VICTIM IDENTIFICATION FORM** **F1**

DEAD BODY

Nature of disaster : ____________________

Place of disaster : ____________________

Date of disaster : ☐☐ Day ☐☐ Month ☐☐☐☐ Year

No: ____________

Barcode

Male ☐ Female ☐ Sex unknown ☐

DENTAL FINDINGS		
83	**Recovery** Site of recovery Recovery No. Date Police Agency Address Phone/E-mail DENTAL EXAMINATION Requested by (date) Performed at (date)	

84	**Remains recovered**	PM material present for examination, describe specimens collected.		
	Exhibits:	*Check*	*Specimen taken*	*Stored at*
	01 Jaws with teeth	*Upper:* ☐ *Lower:* ☐		
	02 Jaws without teeth	*Upper:* ☐ *Lower:* ☐		
	03 Teeth only	*FDI #s:*		
	04 Fragments			
	05 Other			

85	**Supplementary details** Condition of the body Condition of the jaws Injuries to - oral soft tissue - jaws - teeth Possible cause(s) of injuries Other details	

Registered by Duty Title : Name : Address : Phone/E-mail :	Signature / Date

[(GB) Version 2006]

The INTERPOL Victim Identification Form, Sections F1 and F2

GENERAL INFORMATION

The INTERPOL Victim Identification Form consists of several sections - divided into two groups:

1) YELLOW FORMS for listing latest known data concerning a mission person;
2) PINK FORMS for listing all findings concerning a dead body.

Identification of a dead body may become possible if data listed on the pink forms concerning this body can be compared with, and shown to match, data listed on the yellow forms concerning one particular missing person. If an identification is made, the experts involved will complete an Identification-Report - as a prerequisite to issuing a death certificate and releasing the body for burial.

The identification of a dead body may be accomplished in several ways, depending upon the type of data used. The INTERPOL Victim Identification Form has been set up in such a way, that sections listing the same type of data are marked with the same capital letter in the upper right-hand corner. For dental identification, the forms to use are Sections F1 and F2 (yellow), and Sections F1 and F2 (pink); because of the specialized vocabulary, they must be filled in by a forensically trained dentist.

INSTRUCTIONS FOR USE - SECTION F1 AND F2 PM (pink)

These forms are designed for listing all dental information collected during the dental examination of an unknown dead body (or remains thereof).

In Section F1, make sure that the reference number is clearly shown - and that the sex is clearly indicated (boxes at the top). Fill in all the details requested further down. Under "Supplementary Details", list any information at hand that may serve to explain the results obtained from the dental investigation, eg. where and when the body was found (co-ordinates), its condition (drowned, burned, skeleton), your own working conditions, presumed identity.

In Section F2, all dental findings related to the dead body must be listed. After having established full access to both jaws and cleaned all remaining teeth, describe in the spaces provided - tooth by tooth, at the right upper jaw with tooth 18, ending in the right lower jaw with tooth 48 - all treatment and other conditions found. Indicate surfaces by using Capital-Letter System: M = mesial, O = occlusal, D = distal, V = vestibular, L = lingual; if other abbreviations are used, please explain them in one of the boxes further down. (NOTE: there must be a notation for every tooth (or corresponding jaw area) recovered as part of the body!)- Next, sketch on the dental chart the location and extent of all fillings and other conditions found. For color distinction, use black for amalgam, red for gold, and green for tooth-colored material. For teeth missing ante mortem, put large cross (X) over the appropriate tooth square; for teeth missing postmortem (open socket), encircle the tooth number over/under the corresponding tooth square; for jaws sections not recovered, leave unmarked. Make sure that sketch and text tally. All X-rays taken in connection with the oral autopsy must be listed (type, date of exposure, teeth concerned). Supplementary examination may include photographic, microscopic, scanning electron microscopic (SEM), or metallographic examination of teeth and/or restoration removed from the body. Finally, and evaluation of age should always be given, either your own clinical estimate or, if teeth have been removed for this purpose, the method used and the result.

Once Section F2 has been completed, type your name, address and telephone number (or use your professional stamp) in the box at the bottom of Section F1. Finally, enter the date of completion above your personal signature. Remember - this is a legal document, so keep a full copy for your own file.

[(GB) Version 2008]

P ost M ortem *(pink)* **VICTIM IDENTIFICATION FORM** **F2**

DEAD BODY

No: ____________
Barcode

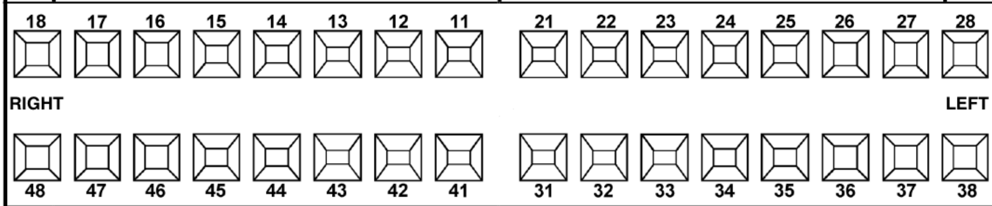
Nature of disaster : ____________________

Place of disaster : ____________________

Date of disaster : ☐☐ *Day* ☐☐ *Month* ☐☐☐☐ *Year*

Male ☐ *Female* ☐ *Sex unknown* ☐

86	**DENTAL FINDINGS for permanent teeth (Note primary teeth specifically)**		
11			21
12			22
13			23
14			24
15			25
16			26
17			27
18			28



48			38
47			37
46			36
45			35
44			34
43			33
42			32
41			31

87	**Specific description of** Crowns, bridges, dentures and implants	
88	**Further findings** Occlusion, attrition, anomalies, smoker, periodontal status, supernumeraries, etc.	
89	**Radiographs taken of** Type and region	
90	**Supplementary examination**	
91	**Estimated age**	*Min* ________ / *Max* ________ *Method used ?*
96	**Quality Check** FOd 1 FOd 2 (If required)	*Date:* *Signature:* *FOd 1 Name:* *Date:* *Signature:* *FOd 2 Name:*

F2 Prepared by	Duty Title : Name : Address : Phone/E-mail :	Signature / Date

[(GB) Version 2008]

PostMortem (pink) **VICTIM IDENTIFICATION FORM** **G**

DEAD BODY

No: ____________
Barcode

Nature of disaster : ______________________

Place of disaster : ______________________

Date of disaster : ☐☐ Day ☐☐ Month ☐☐☐☐ Year

Male ☐ Female ☐ Sex unknown ☐

FURTHER INFORMATION (If referring to data given on a previous page, please indicate item number)

92	

[(GB) Version 2006]

VICTIM IDENTIFICATION FORM

SILHOUETTE SKETCH

Please choose the appropriate sketches and mark items on D1 and D2

34 **02 Head form, front (Shape of head from front)**

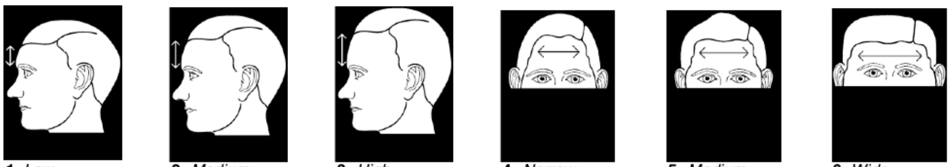
1 Oval · *2 Pointheaded* · *3 Pyramidal* · *4 Circular* · *5 Rectangular* · *6 Quadrangular*

03 Head form, profile (Shape of head from side)

I<->I I<--->I I<----->I

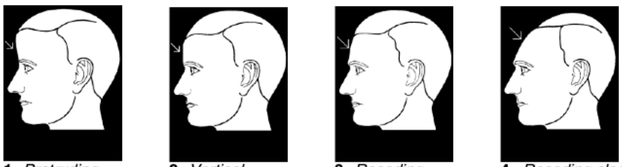
1 Shallow · *2 Medium* · *3 Deep*

37 **01 Forehead - Height/Width**



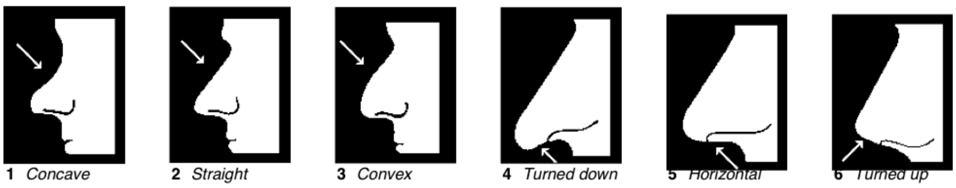
1 Low · *2 Medium* · *3 High* · *4 Narrow* · *5 Medium* · *6 Wide*

02 Forehead - Inclination



1 Protruding · *2 Vertical* · *3 Receding* · *4 Receding clearly*

40 **03 Nose - Curve/Angle**



1 Concave · *2 Straight* · *3 Convex* · *4 Turned down* · *5 Horizontal* · *6 Turned up*

42 **02 Ear lobes**

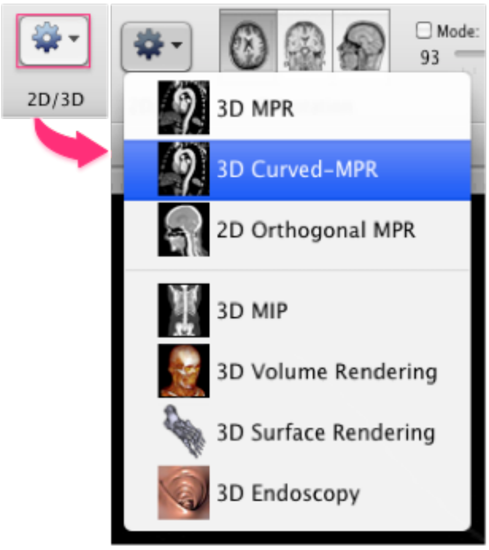
1 Not attached · *2 Attached*

[(GB) Version 2008]

APPENDIX 5: CLAVICLE MEASUREMENT PROTOCOL

OsiriX 3D Curved MPR Viewer: <u>Maximum clavicular length</u>

- Open the *3D Curved MPR* Viewer from the 2D Viewer window.



- Figure 11.1: toolbar command to open Curved MPR Viewer.

The Curved MPR viewer displays the image data in three windows, showing three orthogonal MPR planes, which can be precisely orientated in relation to each other.

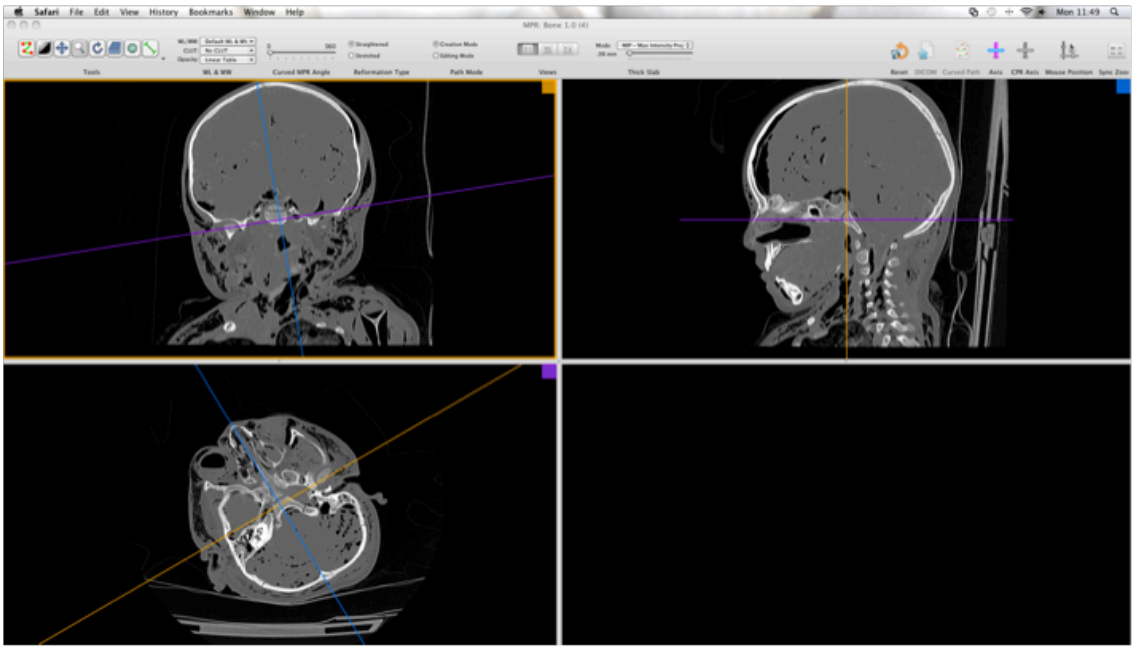


Figure 11.2: 3D Curved MPR Viewer Window.

- Use the *axis gridlines* to move the image into the required plane (i.e. to move the clavicle into its normal anatomical position).

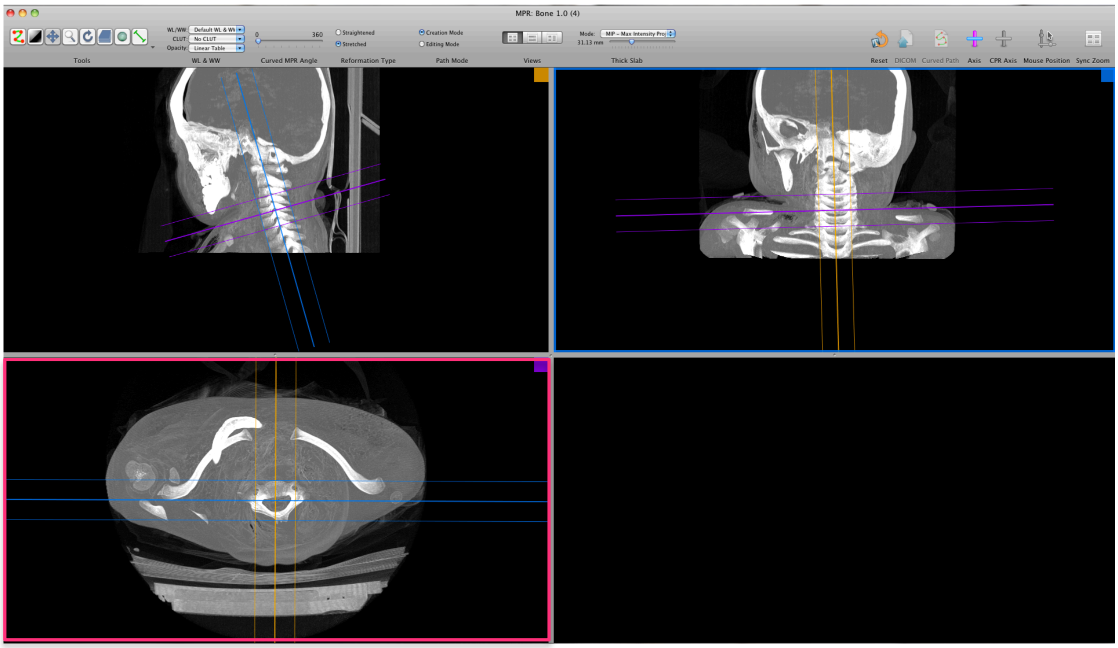


Figure 11.3: The *Axis* command on toolbar and the correct position and planes to view the clavicle (Highlighted, bottom left image).

- Select *Maximum Intensity Projection (MIP)* rendering algorithm from pull down menu on toolbar.

This algorithm extracts and displays the maximum intensities of all pixels.

- Control the number of slices by adjusting the thickness of the slab with the slider.

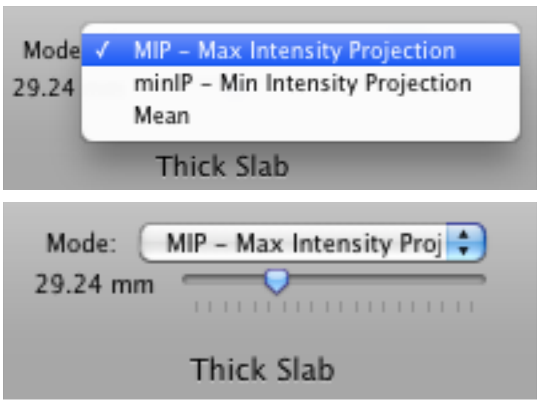


Figure 11.4: *Rendering Algorithm* and *Thick Slab* command on toolbar.

N.B. I found that this setting, and a slice thickness of 20-30mm, produced a clearer sharper image of the clavicle, which facilitated further analysis.

- Double click bottom left image to make it full page.
- Use the Move, Zoom, and Rotate commands on toolbar to orientate the clavicle horizontally within the workspace (i.e. how it would lie on the base plate of an osteometric board).

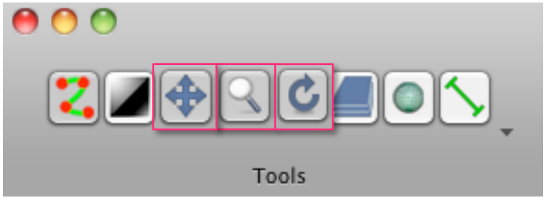


Figure 11.5: Move, Zoom and Rotate commands, respectively

Figure 11.6: How the bone should be positioned on screen.

- Use the *Manual Contrast* command on the toolbar to enhance the bone outline

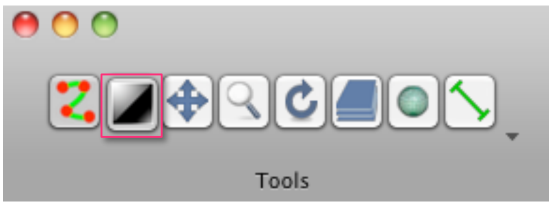


Figure 11.7: Manual Contrast commands on toolbar.

Figure 11.8: Clavicle in the correct position and optimum contrast, ready for measuring.

N.B. The contrast can also be changed using preset algorithm windows, but I generally found that the manual tool gave the user better control and the ability to optimize viewing for their personal requirements.

- Select the *measurement* tool from the toolbar.

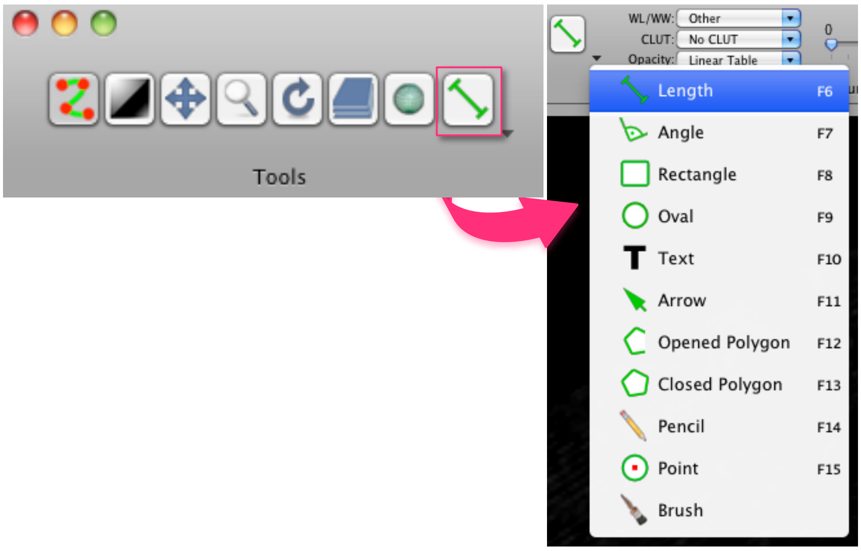


Figure 11.9: The measurement command and drop drown menu choices.

- Use the *length tool* to define the most extreme point of the bone by drawing parallel end plates, to replicate the end plates of an osteometric board.

Figure 11.10: The most extreme point of the bone as defined by virtual end planes (Measurement command > length tool).

- Propagate these regions of interests (ROIs) throughout the entire series.

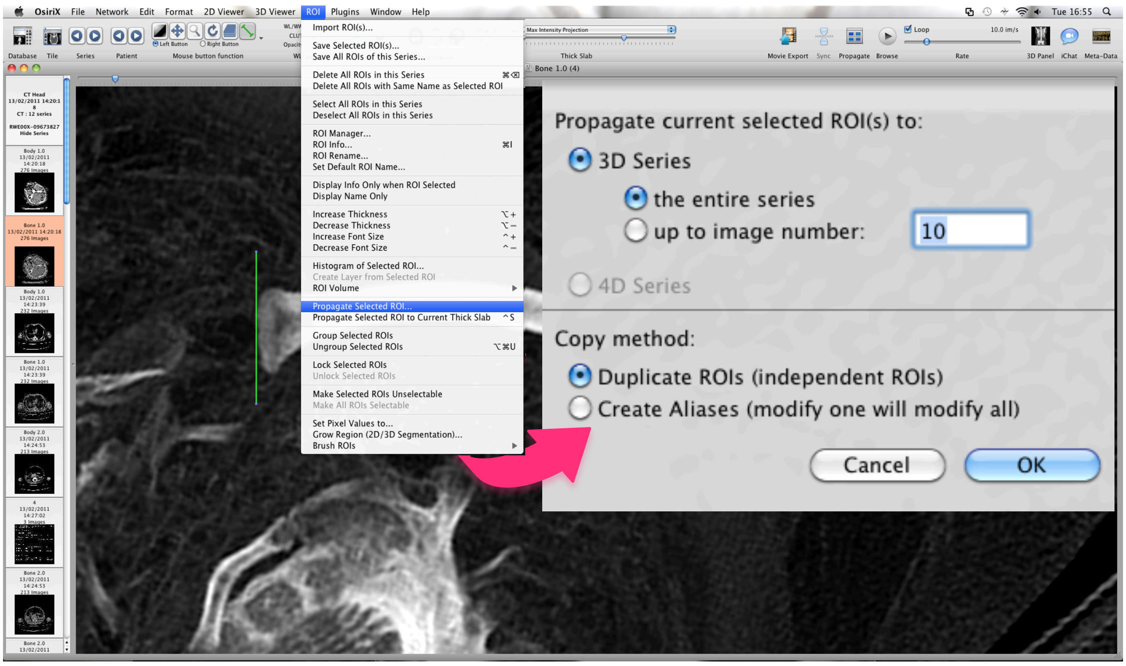


Figure 11.11: Select; *3D series* from toolbar > check entire series > OK.

- Use the scroll command from the toolbar to check the end plates are definitely at the most extreme points of the bone.

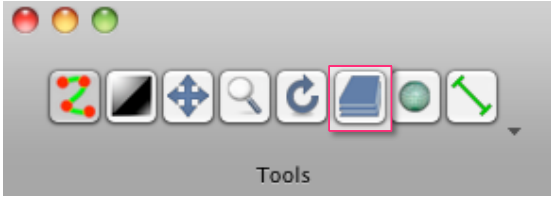


Figure 11.12: Scroll command on toolbar.

N.B. If the plates are not at the most extreme points, move them by deleting the ROI > adding a new end plate with length measurement tool > propagating through series > checking.

- When certain the plates are at the most extreme points, use the length tool to measure the distance between the plates.

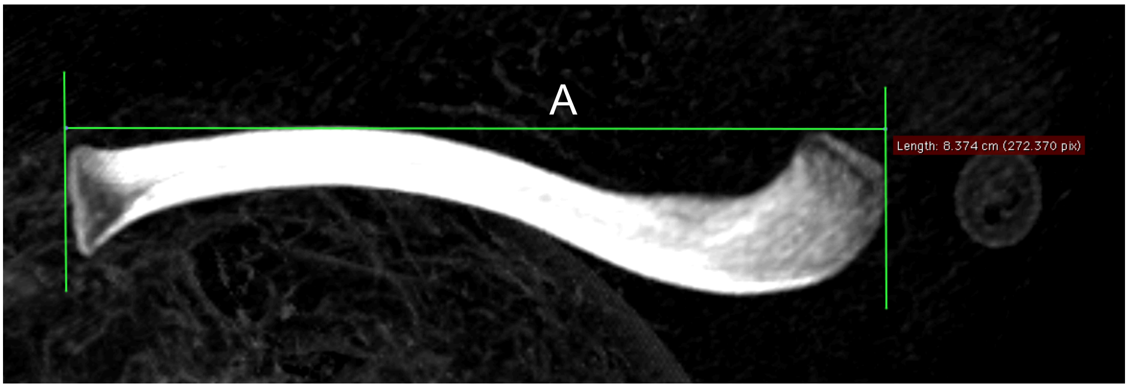


Figure 11.13: A) The maximum diaphyseal length of the clavicle (i.e. the distance between the two previously defined end plates).

Cortical thickness

USE THE SAME STEPS AS MAXIMUM LENGTH PROTOCOL, THEN:

- Calculate the mid-point of the diaphysis, by dividing the maximum length by two.
- Draw a line from one end plate to the calculated mid-point, using the length measurement tool.

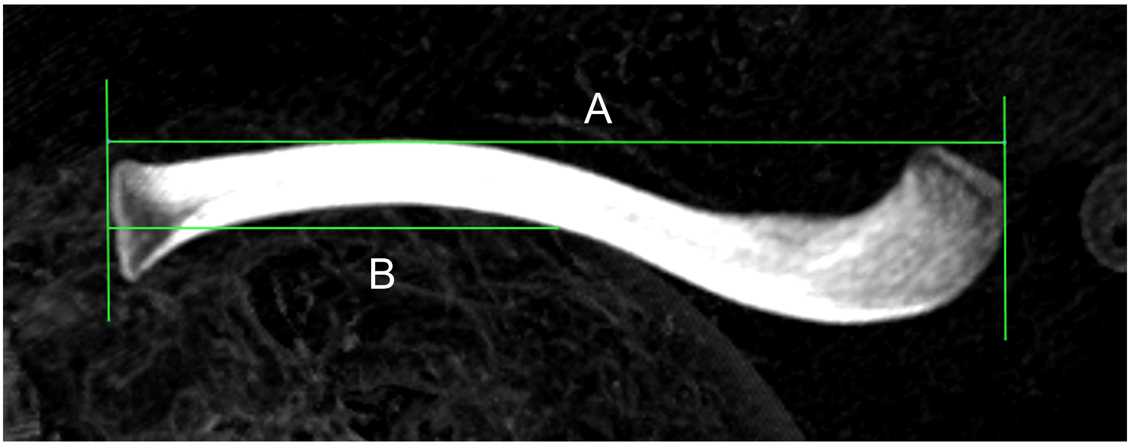


Figure 11.14: A) Maximum diaphyseal length. B) Mid-point marker (maximum length/2)

- Use the *angle measurement tool* (from the measurement command drop down menu) to draw a line tangent to the curvature at the mid-point, and a second line dissecting the shaft of the clavicle at 90° angle from this tangent.

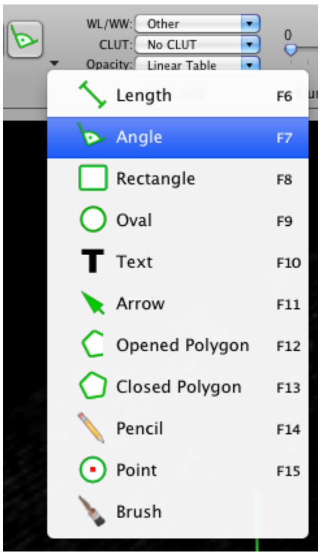


Figure 11.15: Angle measurement tool

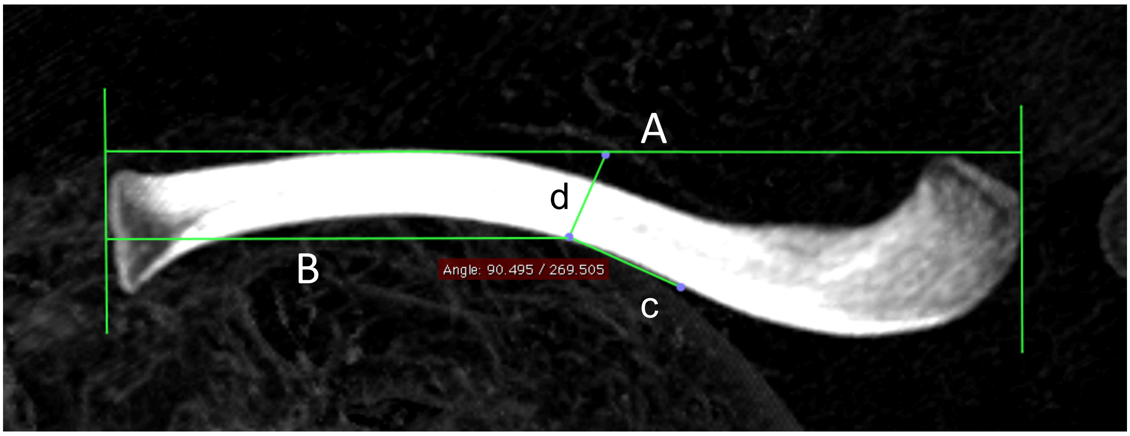


Figure 11.16: A) Maximum diaphyseal length. B) Mid-point marker. c) Tangent line, adjacent to curvature at mid-point. d) Plane dissecting the shaft of the clavicle, 90° from tangent line.

- Adjust the contrast using the *manual contrast* command (see Figure 11.7), until the bone cortex is clearly delineated and separated by the medullary cavity.

- Measure the cortical thickness on both sides of the medullary cavity, along the mid-shaft dissecting plane (d)

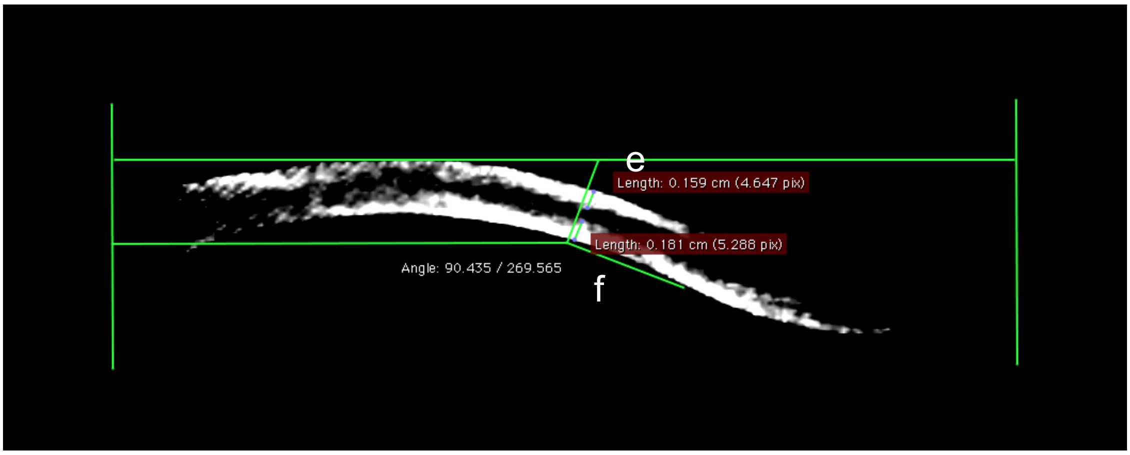


Figure 11.17: e, f) Cortical bone thickness, measured along the dissecting shaft plane.

The cortical thickness is calculated by adding the length of e and f. This total value can then be used in the cortical bone index calculation to estimate age of the individual.

Additional measurements

USE THE SAME STEPS AS FOR MAXIMUM LENGTH AND CORTICAL THICKNESS PROTOCOL, THEN:

- Using the length measurement tool, measure bone diameter at its medial, central and lateral regions.

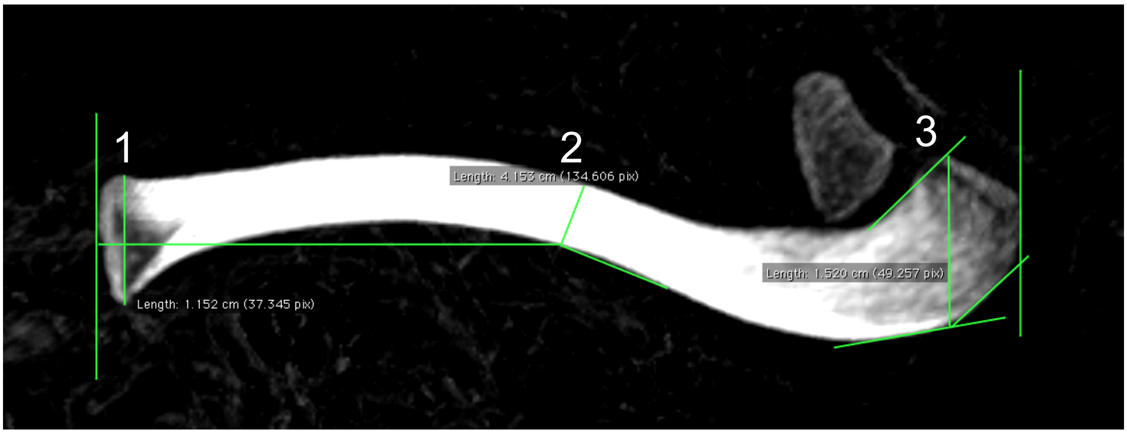


Figure 11.18: Maximum bone diameter measurements at; 1) Medial, 2) Central, and 3) Lateral points.

N.B. I found that using additional length planes, to ensure that the maximum diameter was definitely being recorded at each of these points, extremely helpful. It was also important to ensure that the bone was in the correct anatomical position (i.e. as it would be lying on an osteometric bone) to maintain experimental consistency. Particularly as the additional measurements were not standard measurements and there were therefore no official guidelines for recording these.

APPENDIX 6: DENTAL IMAGE ACQUSITION PROTOCOL

- Open the *3D Curved MPR* Viewer from the 2D Viewer window.

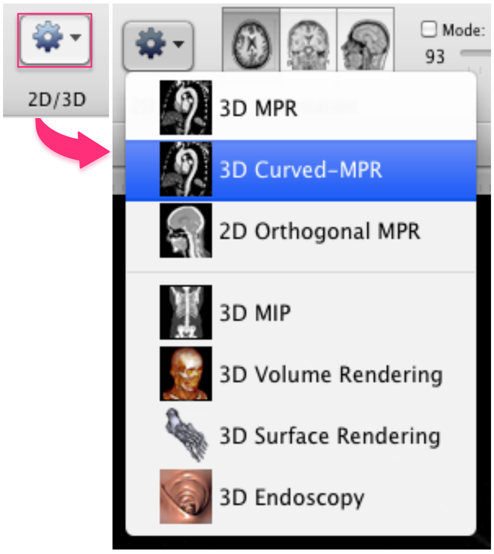


Figure 11.1: toolbar command to open Curved MPR Viewer.

This viewer produces a Curved MPR rendered image from a 3D dataset. The Curved MPR plane can be defined in any direction and any angle on the original dataset. This plane is called the 3D Bezier path. Although the final product is strictly speaking a 2D image, it has been created using the full volume dataset, to optimize data output.

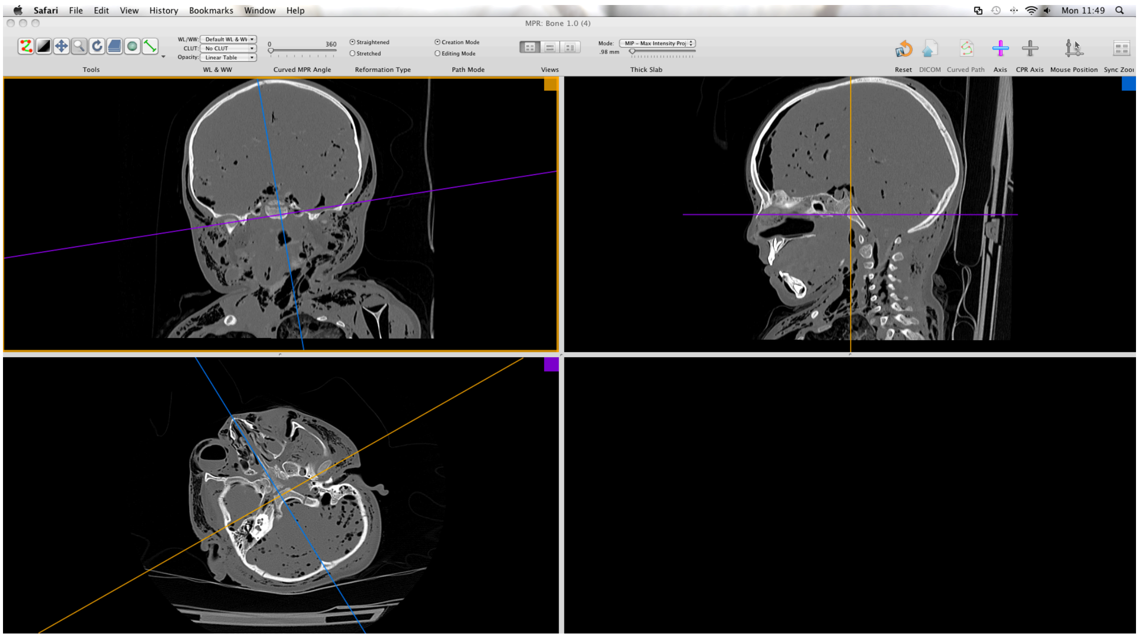


Figure 11.2: 3D Curved MPR Viewer Window

- Four views are displayed in this window. The first three views show an orthogonal MPR plane in relation to the other views. These allow the user to move in a precise manner and to place and edit the 3D Bezier path. The DICOM images are reformatted as the user alters the planes on the orthogonal views.

- Use the *axis gridlines* to move the image into the required plane (i.e. to move the dentition into their normal anatomical position).

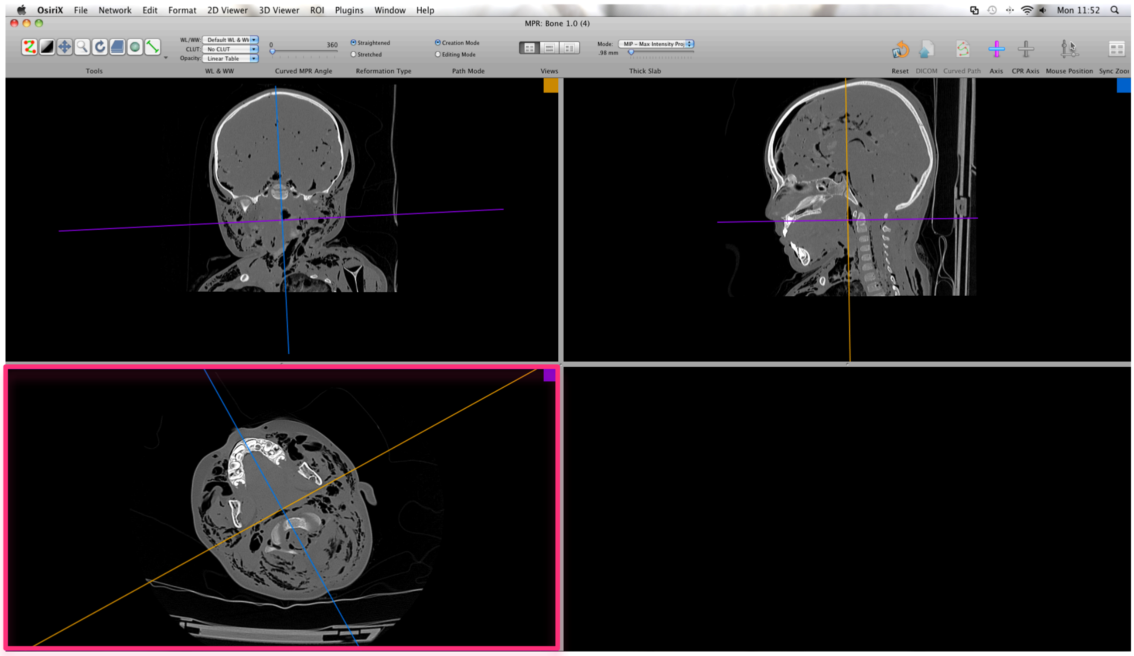


Figure 11.3: The correct position and planes for dental reconstruction (Highlighted, bottom left image).

- Select the “curved path” tool from the toolbar.

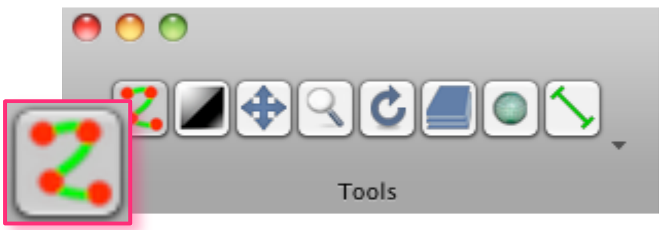


Figure11.4: *Curved path* tool command.

- On the bottom left image, place the 3D Bezier path as a curved plane following the arch of the dentition.

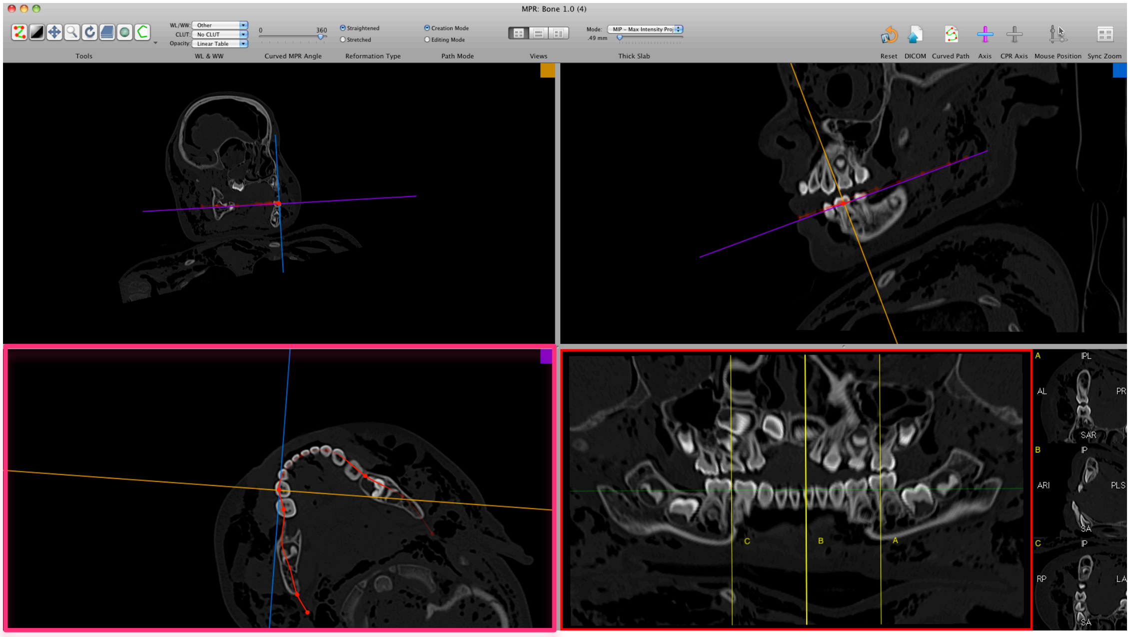


Figure 11.5: The curved 3D Bezier path (bottom left) and curved MPR view and the three orthogonal views (A, B, C).

- The lower right view displays the 3D Bezier path as a *straightened* or *stretched* curved MPR image, which can be selected by the user depending on requirements.

Figure 11.6: *Straightened* and *stretched* toolbar commands.

Straightened Rendering: This type of curved planar reformation (CPR) fully straightens the curves structure. This technique is generally used to view vessels and tubular structures of varying diameter. The advantage of this method is the elimination of curvature of the central-axis so that the changes in diameter are more noticeable. The disadvantage is the inability to measure distance in a non-orthogonal direction: you can only measure distance along the length or in the perpendicular direction, corresponding to the length and diameter of the structure, respectively. This is probably not the best function to use for dental reconstructions, encase future development of the technique involves measurements in the non-orthogonal direction.

Stretched Rendering: The main advantage of this type of CPR is the preservation of isometry (resolution along the X and Y axis are equal, so that distance can be measured), which could be important for a more comprehensive and more accurate odontological assessment; you can measure structures in any direction in this CPR mode.

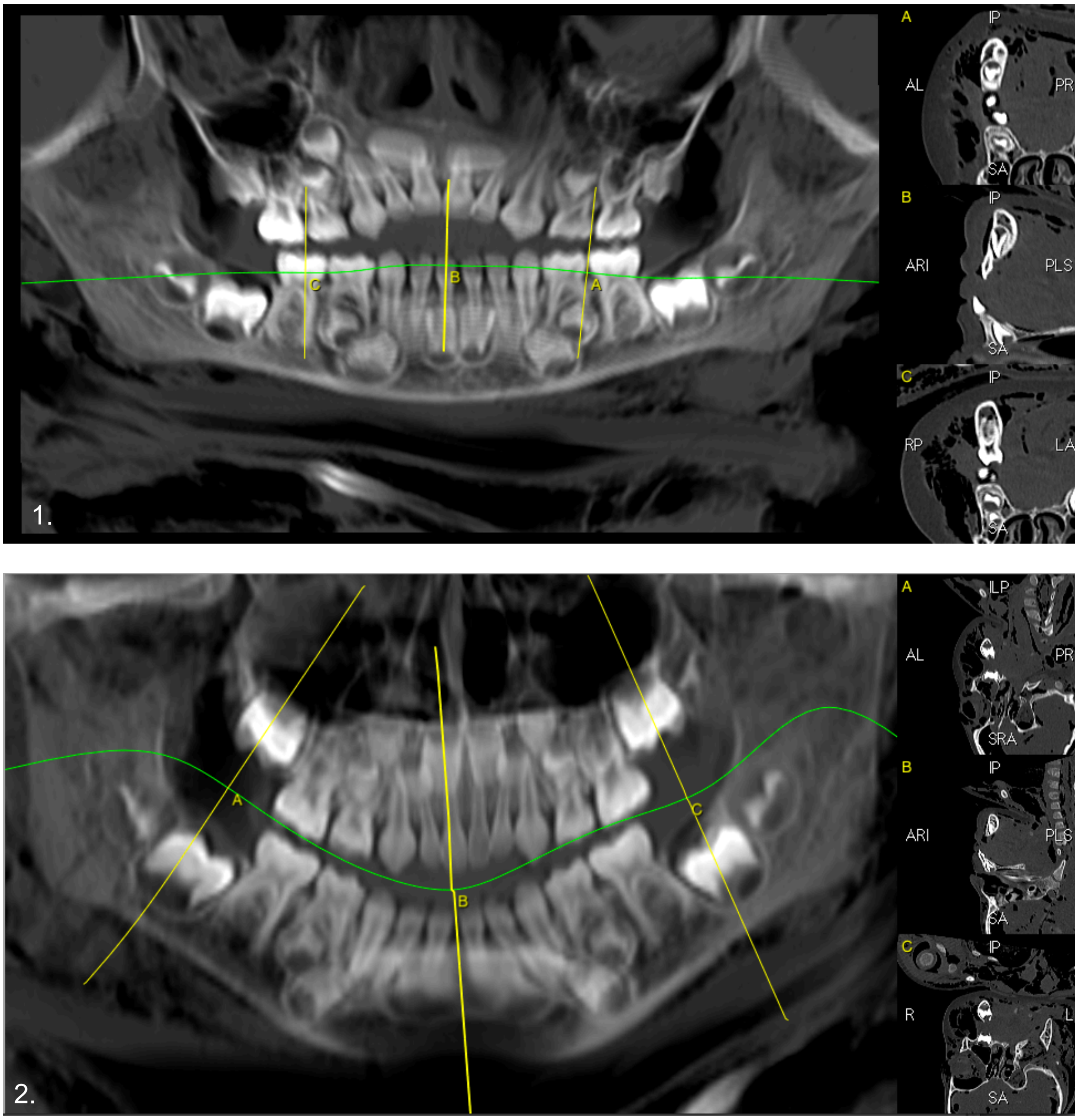


Figure 11.7: The two different renderings of the same curved Path and dataset.
1) Straightened, 2) Stretched.

The three views on the right side of the curved MPR (A, B, and C), correspond to three perpendicular views along the 3D Bezier path. These three views are strictly perpendicular to the Bezier path, but are not necessarily parallel to each other's, if the 3D Bezier path is not in a straight line.

- The user can rotate around this 3D Bezier path, using the yellow perpendicular view lines, and display three perpendicular views from anywhere along this path (A, B, and C).

This function is particularly useful when a more thorough inspection of a particular tooth is required. If there are metal artefacts for example, the surrounding teeth may be obscured-and so the perpendicular views allow the tooth to be viewed individually with less surrounding 'noise'.

- Double click any of these views to display it as full screen. Double-click again to reduce it to normal size.

By default the curved MPR view is rendered as a thin slice with a thickness identical to the original data set.

- Increase this thickness by using the toolbar *Thick Slab* item (see below).

Figure 11.8: *Thick Slab* command on toolbar.

By adjusting the cursor position in the thick slab item, you can modify the slice thickness in millimetres. Three rendering algorithms are available:

- **Mean**: all slices are added and the mean is computed.
- **MIP**: maximum intensities of all pixels are extracted and displayed.
- **MinIP**: minimum intensities of all pixels are extracted and displayed.

You control the number of slices with the slider.

N.B. For dental analysis I found that using these algorithms interchangeably produced the best results. The *Mean* rendering function-produced images most comparable to traditional OPTs, however root formation and degree of mineralisation was often easier to establish using either the *MIP* or *MinIP* mode.

- Adjust the WL/WW (image contrast and intensity) to highlight specific ranges of grey-levels only.

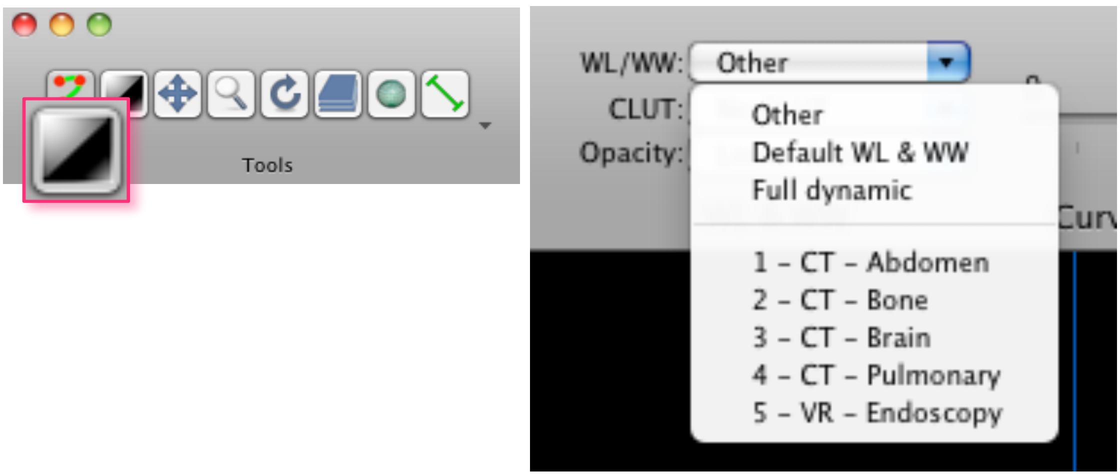


Figure 11.9*: Manual contrast* tool and *preset WL/WW* windows.

- Use either the preset WL/WW windows or the *manual contrast* tool from the toolbar.

This can be used to enhance details in the lighter regions of the image at the expense of detail in darker regions (or vice versa). This is particularly beneficial for dental imaging as the difference between the soft tissue HU value and enamel HU value is large. Therefore, this function can be exploited to investigate the dentition, with minimal loss of important detail.

- When the image is to your satisfaction it can be exported as an individual TIFF or JPEG image or in DICOM format and added to the database as a new series of images. These images can then be exported to a PACS or another DICOM compliant workstation for further analysis if required.

- If exporting as an individual JPEG or TIFF - remove the annotations first.

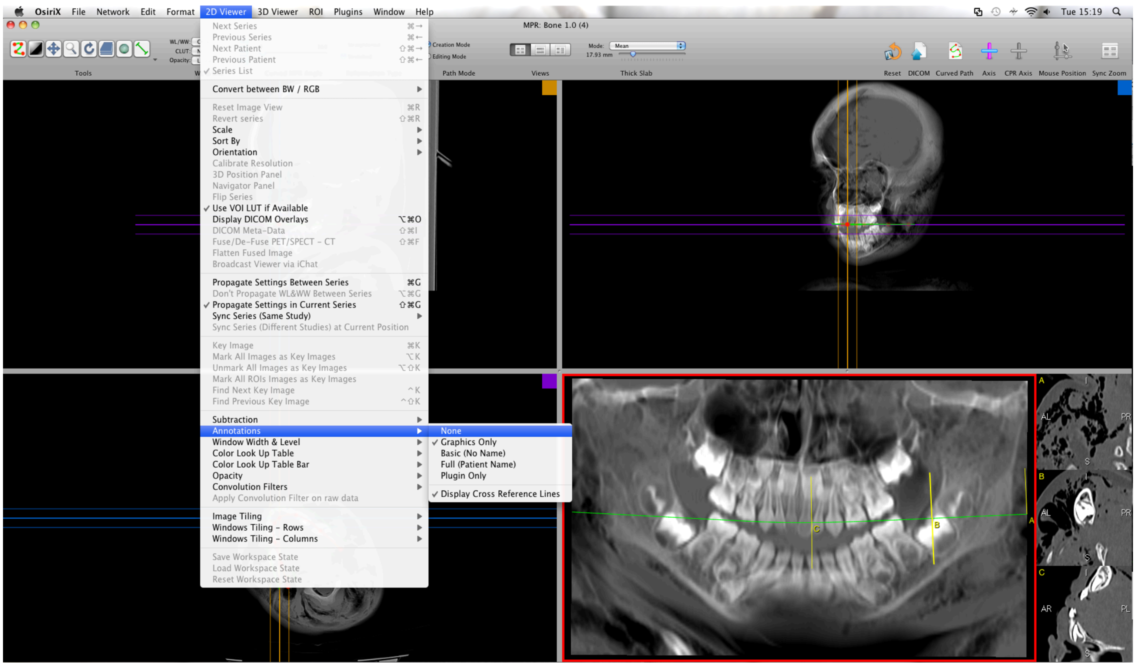


Figure 11.10: Commands to switch off annotations (2DViewer > Annotations > None).

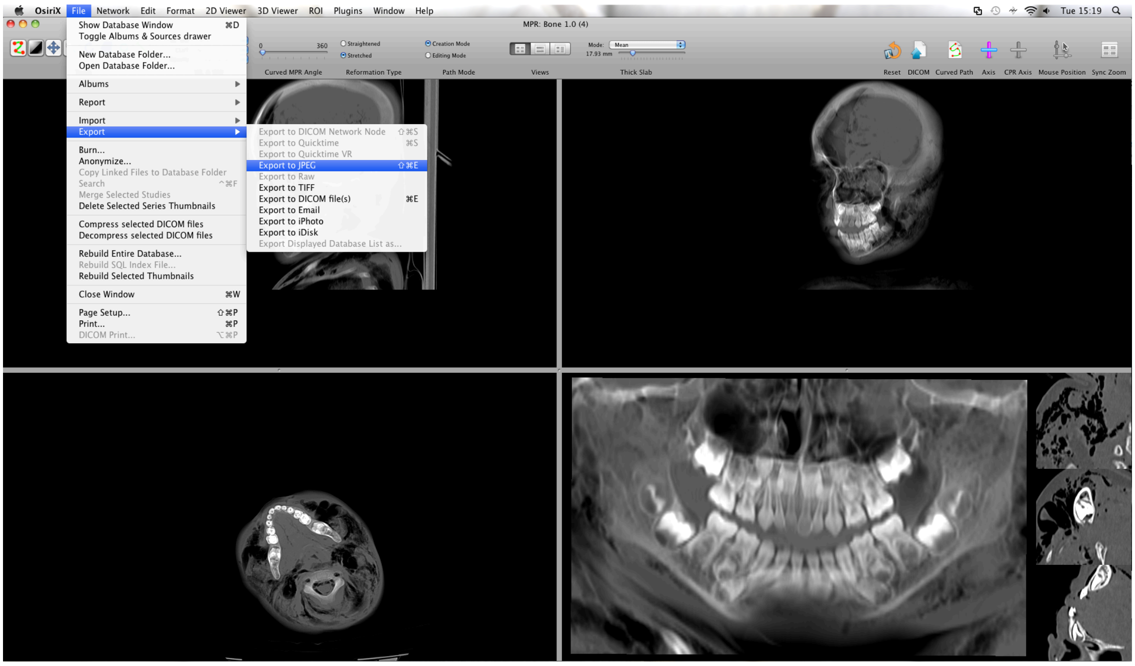


Figure 11.11: Commands to export image in JPEG or TIFF file format (File > Export > Export to JPEG or Export to TIFF).

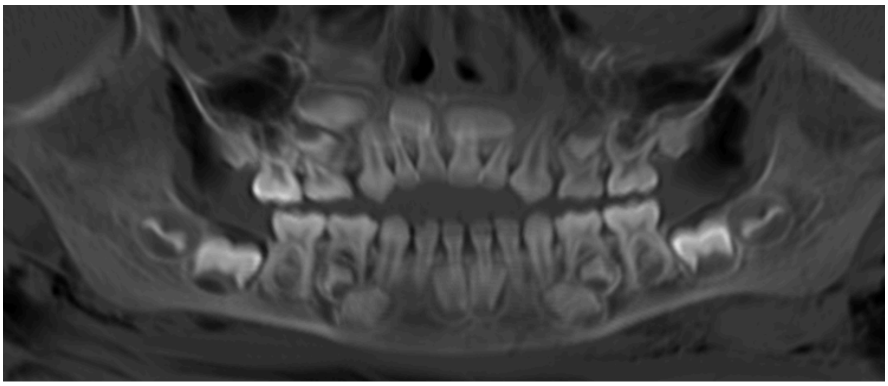


Figure 11.12: Resulting JPEG image.

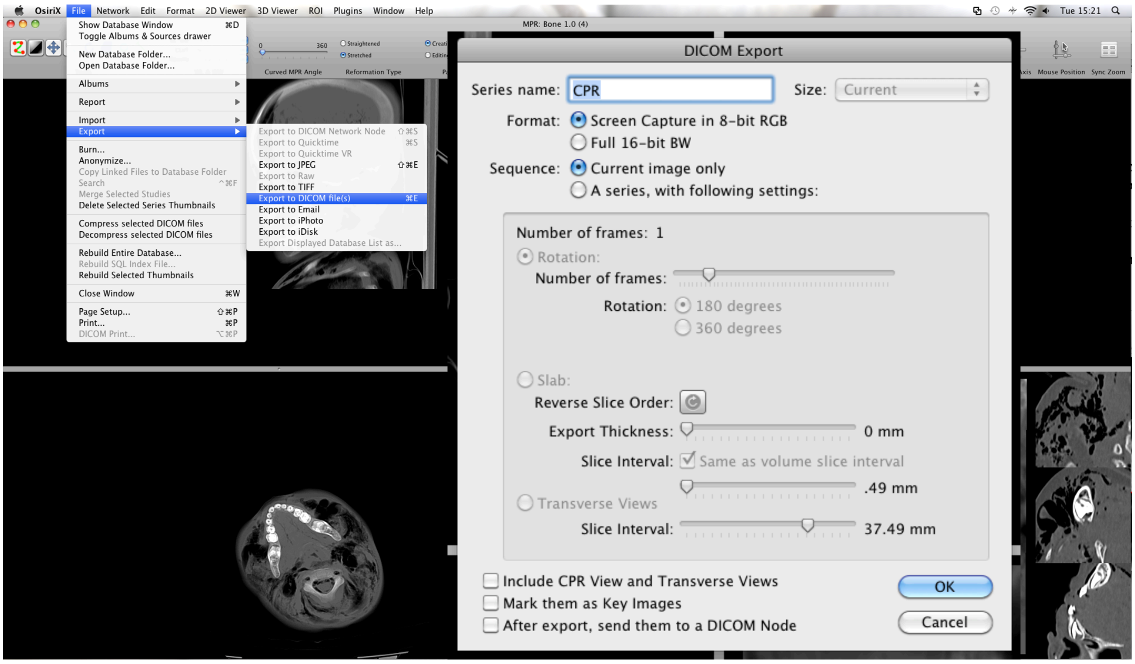


Figure 11.13: Commands to export as a DICOM series (File > Export > Export to DICOM file(s) > [tick] Full 16-Bit).

N.B. By exporting a series in 16-bit, you can create a new series without losing dynamic information and you can keep diagnostic quality.

APPENDIX 7: PUBLICATION

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REVIEW

Post-mortem computed tomography and 3D imaging: anthropological applications for juvenile remains

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Abstract Anthropological examination of defleshed bones is routinely used in medico-legal investigations to establish an individual's biological profile. However, when dealing with the recently deceased, the removal of soft tissue from bone can be an extremely time consuming procedure that requires the presence of a trained anthropologist. In addition, due to its invasive nature, in some disaster victim identification scenarios the maceration of bones is discouraged by religious practices and beliefs, or even prohibited by national laws and regulations. Currently, three different radiological techniques may be used in the investigative process; plain X-ray, dental X-ray and fluoroscopy. However, recent advances in multi-detector computed tomography (MDCT) mean that it is now possible to acquire morphological skeletal information from high resolution images, reducing the necessity for invasive procedures. This review paper considers the possible applications of a virtual anthropological examination by reviewing the main juvenile age determination methods used by anthropologists at present and their possible adaption to MDCT.

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Keywords Forensic · Anthropology · Multi-detector computed-tomography · Virtual · Imaging

Introduction

Forensic anthropology was primarily considered to be a tool to aid in the analysis and investigation of skeletonised remains; but more recently it has developed to encompass the assessment of decomposed, burned or fully fleshed remains and evaluation or verification of identity in the living. Establishing the biological profile of an individual after death, such as age, sex, stature and ethnicity, is of fundamental importance in a forensic investigation. In some instances identification by DNA analysis may have replaced the need for forensic anthropology and odontology, but these techniques have not been completely superseded. Even when recoverable, DNA analysis is constrained by the need for appropriate comparisons to arrive at a positive identification. DNA may also fail to answer other forensic questions that may be amenable to anthropological techniques. For example, in a mass fatality incident (MFI), remains may be disturbed and co-mingled and anthropological techniques can play an invaluable role [1], particularly in relation to anatomical identification of body parts present.

In forensic practice, the examination of de-fleshed bones remains the "gold standard" for anthropological assessment and osteometry [2]. However, the removal of soft tissue from bone is an invasive, time consuming and unpleasant procedure and can damage underlying bone and lead to the loss of potentially vital evidence. It is therefore only undertaken when there is little information to be gained from the soft tissues, and their removal is absolutely necessary for identification purposes [3]. Additionally, the maceration of remains is not acceptable to some cultures and religions, and may even be prohibited by national laws and regulations [4]. As a result, non-invasive alternative approaches that utilize medical imaging for identification offer considerable practical and aesthetic benefits.

At present, radiographic techniques, such as plain film radiography (X-rays), dental X-rays, and fluoroscopy, are used routinely in forensic investigative processes [5–9] and their value is widely recognized [10]. Recently there has been gradual acceptance of computed tomography (CT) as an alternative to the more traditional imaging modalities [2, 11–26]. This has been driven by the increasing availability and affordability of CT; but also by the development of "spiral" and "multiple detector" CT (MDCT). This has improved scanning speed and spatial resolution in the longitudinal axis, allowing high quality image reconstruction in any plane [multiple plane reconstruction (MPR)] and 3D modeling of images. MDCT therefore allows the measurement of bones in any plane avoiding the need for invasive procedures. MDCT has demonstrated superiority to other imaging techniques in this regard, and is generally considered the most effective imaging technique for post-mortem examination [27]. The basic principles of MDCT permit the acquisition of "volume" data, allowing both external and internal structures to be visualized in any imaging plane or in a three-dimensional form [28]. Possibly the most notable quality of CT is its capacity to display information from a wide variety of tissues of differing densities with minimal superimposition of overlying structures [29]. Image acquisition time is also just a matter of seconds, although other factors such as image reconstruction can prolong the ultimate process. Rapid acquisition is a primary benefit making CT particularly useful in a MFI [21] and with the development of mobile CT technology, scanning can take place almost anywhere where the scanner can be driven or airlifted to, including the scene of the incident itself. Most of the current mobile CT scanners are also equipped with telecommunications facilities for the transmission of data to remote locations [20]. In a MFI, MDCT also has the potential to replace X-ray, dental X-ray and fluoroscopy reducing the number of on-site personnel required, which may overcome problems associated with contamination and judicial requirements [30] and compared with fluoroscopy, MDCT helps to minimize radiation exposure to staff. MDCT techniques, particularly using mobile CT platforms, will also be extremely valuable if chemical, biological, radiological or nuclear (CBRN) materials are involved. In this scenario, traditional post-mortem examination in situ is hazardous due to the high risk of contamination to the investigative teams. Field based exercises have shown that bodies can be CT scanned on-site, in a sealed body bag by trained operators in protective CBRN suits, and that image data can then be sent to remote sites via tele-radiology, for reporting by radiologists, odontologists and anthropologists [2, 30]. From an anthropological perspective, this accelerates the process of data acquisition, reduces risk to personnel and removes the necessity to clean the soft tissue from the bones. This "virtual anthropology" uses CT information to replicate traditional anthropological methods, such as osteometry, by creating three-dimensional virtual representations of bone [31]. Although modern CT scanners have accurate scaling of images, allowing accurate measurements to be made of calibration phantoms as part of quality control, there is currently little evidence regarding the application and repeatability of metric analyses using CT to assess biological profiles.

This article reviews one of the key questions for anthropological investigation; juvenile age determination. The traditional methods of age determination are reviewed and the feasibility of using MDCT as an equivalent is considered. Using MDCT should prove to be quicker, cheaper and culturally more acceptable in some situations than more traditional anthropological methods, particularly when processing the remains of deceased children.

Juvenile age determination using traditional anthropological methods

For adults, preliminary determination of age-at-death, racial/ethnic affiliation, living stature and sex are the first steps towards arriving at a positive identification. When dealing with juvenile remains age-at-death is the factor that can be most reliably determined [32]. Sex and race/ethnicity evaluation cannot be achieved with any degree of reliability from the skeleton until post puberty. Therefore age estimation is of primary importance. Before the cessation of growth in height, a child's age can be estimated using numerous markers from the teeth and skeleton. Techniques generally utilize the dentition, diaphyseal length of the long bones, and the appearance and fusion of ossification centers. Research dating back to 1939 [33] demonstrates that the relationship between growth and maturity is strong but not linear, displaying periods of both acceleration and stasis. As such, the timing of these growth spurts must be considered in any attempt to evaluate the relationship between skeletal (or biological) and chronological age. Children of the same chronological age can have markedly different biological ages and because of this, assessment of age remains something that, while more accurate in juveniles than adults, still has a significant error range. Females' exhibit advanced skeletal maturation compared to males and therefore it is beneficial to investigations if the sex of the individual is known, but this is not always possible. This review paper will therefore consider the possible applications of a "virtual anthropological" examination, as well as the advantages and limitations of this approach when dealing with juvenile remains.

There is a considerable range of well established techniques, which have been presented and reviewed in the literature for age determination in the juvenile. Each of these techniques has limitations and may not be applicable to all cases. At present there is no standardized approach to age estimation in forensic cases and there is no consensus concerning which method is the most appropriate and practical [32]. Juvenile age determination is generally only discussed perfunctorily in general forensic anthropology textbooks [1, 34]. However in 1987 Ubelaker presented a literature review and recommendations for estimating juvenile age at death, which has since been updated [35–38]. Contributions by Cunha and colleagues [38] provided tables of recommended techniques for specific age groups including fetuses, newborns, infants and children, adolescents and transitory groups. This general overview of this area is a useful summary for both students and practicing anthropologists.

The accuracy of age determination in the juvenile is dependent upon the preservation and availability of skeletal elements [39]. The most reliable methods in children utilize morphological and developmental changes to the hard tissues of bone and teeth [1]. Dental age estimations are predominantly determined from the degree of mineralization or eruption of the teeth, whereas skeletal age estimations are established from the developmental stage of primary and secondary ossification centers or the size and diaphyseal length of the long bones. These characteristics have a reasonably predictable rate of progression and a well documented pattern of development. As such, age may be estimated relatively accurately, from a multiplicity of markers [39]. It is frequently observed that dental age estimates are closer to actual chronological age than those of skeletal age as they are less affected by environmental insults that can delay maturational processes [32]. This is probably due to the fact that the development of all the deciduous dentition and part of the permanent dentition takes place before birth in a relatively protected environment whereas skeletal growth and development, although having a strong genetic basis, is influenced for a longer time by external factors such as nutrition and disease burden [40, 41].

Juvenile age determination using MDCT "virtual" anthropology

Possibly the first attempt to determine the repeatability of osteometric measurements using CT was undertaken in 1990 by Hildebolt et al. [29]. The small pilot study obtained comparative measurements to test the accuracy of a 3D-CT technique compared with osteometric methods. They used standard spreading calipers to measure five adult skulls, using a number of cranial landmarks already well established in the literature. The results of the study suggested that there was no significant difference between CT measurements from 3D image reconstruction and osteometric measurements. The authors concluded that measurements using 3D reconstructions were superior to those obtained directly from CT images, but the sample size was small ($n = 5$) and CT technology has changed since 1990.

A more recent study in 2004 by Cavalcanti et al. [42] employed more modern CT scanner and analysis methods. Their study produced similar results and concluded that a high degree of precision and accuracy could be achieved. Nevertheless, their sample size was also small ($n = 5$) and did not use standard anthropological measurements generated via an osteometric board. Recently Robinson et al. [2] compared MDCT generated measurements of the lower limb with anthropological measurements obtained using an osteometric board after the bones were defleshed. The results of this study indicated that there was no significant difference between the measurements determined using CT and those recorded using an osteometric board. Imaging took approximately 5 min per limb, and image transfer via the internet and then analysis (tele-anthro-radiology) took between 20 and 30 min, depending on internet speed and operator experience. These results show that the use of MDCT would accelerate the process of anthropological measurements by removing the necessity to clean bones and prompts the question of whether this "virtual technique" should be the standard for future anthropological examination. Robinson et al. [2] also determined that sex assessment using CT was close to the reliability of direct anthropological measurements, using "cut off" points described in previous studies of calcaneal and talar length. Grabherr et al. [43] have also studied the feasibility of MDCT for age and sex determination. The study group was an adult population which represented ages between 17 and 92 years at the time of death. The results of their study clearly illustrate that the estimation of both age and sex is possible using MDCT and they proposed that these techniques would be useful in MFI investigation [25].

None of the previous studies established a standardized protocol for this technique, which is a necessary step before this method can be internationally adopted for disaster victim identification or nationally accepted as a forensic standard. Standardizing image processing and measurements will therefore be extremely important for the development and acceptance of this technique.

Other previous reports in the area of anthropological computed tomography have been "case-report" oriented and typically based on qualitative techniques that compare morphological traits as opposed to quantitative metric anthropological techniques. Although morphological approaches are commonly used (and often preferred) by forensic anthropologists, they rely more heavily on subjective opinion and experience and are therefore not as judicially sound as metric methods that are more objective and therefore facilitate greater inter-operator reliability testing.

The above studies have provided an evidence base to support the implementation of MDCT into routine adult forensic investigations. However, none of these studies extend to juvenile age determination, where the techniques used vary from those employed for adult remains. Juvenile age techniques utilize the appearance and fusion of ossification sites, which are not applicable to adult remains. There is currently no scientific literature to validate whether osteological measurements required for juvenile age determination are reproducible on MDCT images, or indeed how accurate this "virtual approach" might prove to be.

Using the review provided by Cunha et al. [38] as a guide for age determination of juvenile remains as summarized in Table 1, each of the suggested methods were considered as to whether MDCT acquired images can provide equivalent anthropological assessment. A sample (n ~ 40) of the Scheuer juvenile skeletal collection, housed at the University of Dundee, was scanned using a truck mounted SOMATOM® Emotion 16-detector CT scanner (Siemens AG Medical Solutions). Scans involved helical acquisition using a 0.75 mm slice thickness, 120kVp, and 100 mA with bone and soft tissue reconstructions at 1.25 mm. Data were stored on compact disc and transferred to the image archive at East Midlands Forensic Pathology Unit. Multi-plane reconstructions (MPR) and 3D surface rendered images were then created for analysis using a comprehensive image software program (OsiriX version 3.7.1; distributed freely as open-source software under the GNU licensing scheme at the following Web site: http://homepage.mac.com/rossetantoine/osirix).

Dental development

From the early work of Logan and Kronfeld [44], Schour and Massler [45] produced an atlas of dental development which is the most regularly cited in dental and forensic literature and is still widely used today. By matching radiographs of preferably the entire maxilla and mandible to diagrams depicting the stage of development of the dentition that can be expected at each year in the life of a child, the estimated age of the child is assigned in accordance with the associated diagram. Images of comparable quality to the radiographic images (oral pan tomogram or OPT) traditionally used for this method of age determination are obtainable from MDCT data (Figs. 1, 2). CT has a major disadvantage in the adult jaw due to artefacts produced from dental amalgam in fillings. Other metal work causes fewer problems. This is clearly less common in younger age groups. An advantage of CT over standard

Table 1 Modified table from Cunha et al. [38] reviewing the reproducibility of juvenile age determination methods on MDCT data

Age group	Recommended approach	Reproducible by MDCT
Fetuses	Dental development	Yes
	Presence of ossification nuclei	Yes
	Long bone development	Yes
Newborns	Dental development	Yes
	Diaphyseal length of long bones	Yes
	Presence of ossification nuclei	Yes
	Also check mineralisation of cusp of first permanent molar and ossification of femoral distal epiphysis	
Infants and children (0–7 years)	Dental development	Yes
	Diaphyseal length of long bones	Yes
Juvenile and adolescent (8–15 years)	Dental development	Yes
	Long bone development	Yes
	Maturation of hand and wrist	Yes
Transition and adult (16–25 years)	Third molar development	Yes
	Fusion of spheno-occipital synchondrosis, iliac crest and vertebral ring	Yes
	Maturation of hand and wrist	Yes
	Fusion of medial end of clavicle	Yes

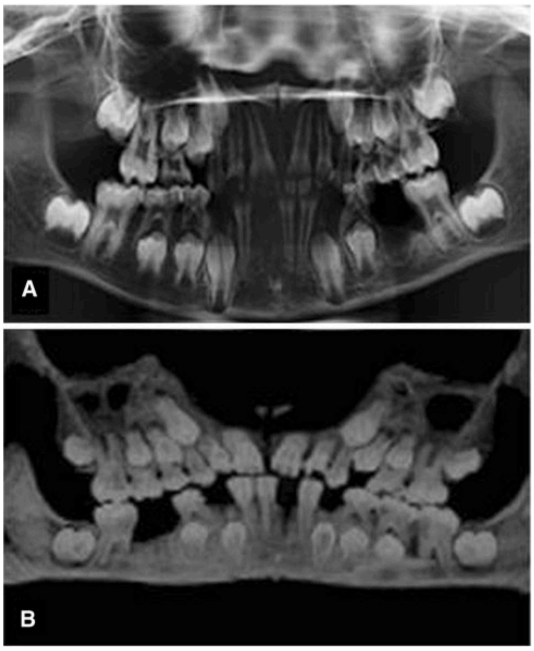

Fig. 1 a Radiographic images traditionally used for comparison in Schour and Massler method [52]. b MDCT images created using OsiriX software package. It is evident from these images that a comparable level of detail is achieved using MDCT to carry out this technique successfully

radiographic techniques however is the ability to define soft tissues as well as bone. This would theoretically allow stages of clinical emergence to be better determined [32].

Scoring systems such as those by Demirjian et al. [46] for the permanent dentition and by Mincer et al. [40] for third molar development, that are also widely used in forensic investigations are reproducible using MDCT data. The clear images and 3D representations of the dentition allow each tooth to be individually studied in all planes in order to accurately score its developmental stage (Fig. 3).

Presence of ossification nuclei

Age evaluation of a fetus places a significant emphasis on the location and number of ossification nuclei present [47]. These appear in a relatively well defined sequence and knowledge of the centers present compared with those yet to form, allows the investigator to assign a possible age range to the individual. The utility of dry bone analysis of ossification centers is variable, depending on the age of the individual and the skeletal element being examined. This is because ossification centers are often initially small, non-specific nodules and are therefore unlikely to be recovered or be of particular significance until they have developed

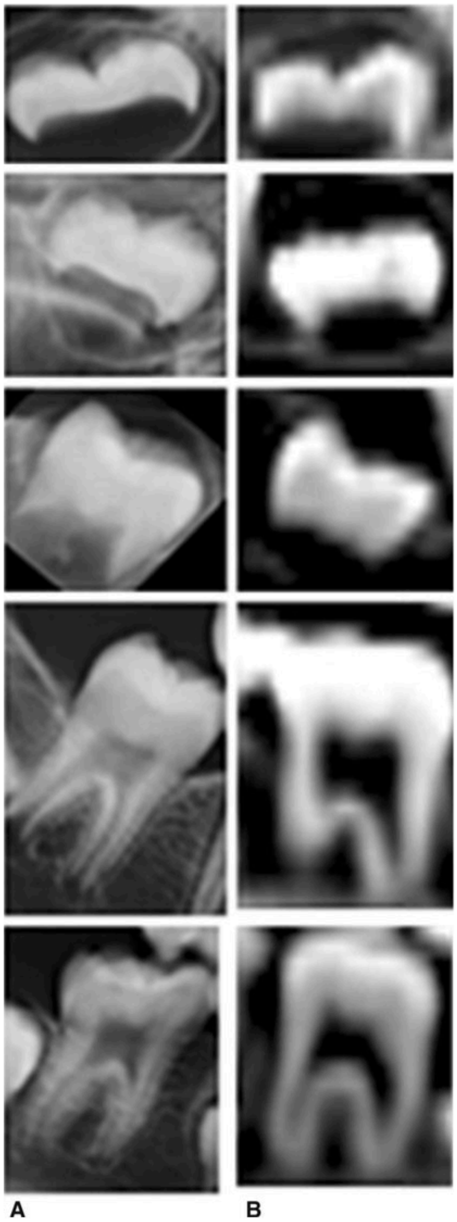

Fig. 2 a Radiographic images traditionally used for comparison for the Demirjian method [53]. b MDCT images created using OsiriX software package. It is evident from these images that a sufficient amount of detail is achieved using MDCT to carry out this technique successfully

into a recognizable morphology. Clinically these images are produced by ultrasound as it does not pose a radiation risk to the unborn fetus. In forensic cases radiographic imaging is generally utilized. However, radiography has

Fig. 3 3D representations of the dentition can also be produced, allowing each tooth to be individually studied in all planes in order to more accurately score there developmental stage. Different reconstruction windows can be used to maximise the level of detail

Fig. 4 a MDCT reconstruction of the lower limb. Ossification sites can be seen clearly. b X-ray image. Ossification centres are visible, but not as clear as on MDCT image

significant limitations as the appearance of ossification centers precedes radiographic visualization, which depends on calcification (mineralization). MDCT is able to replicate images produced by radiography but with increased contrast resolution of non ossified structures, albeit at lower spatial resolution (Fig. 4). In practice the lower spatial resolution is only significant for detecting subtle fracture such as metaphyseal fractures which are better detected by a plain radiograph (Table 2).

Table 2 Advantages and limitations of a "virtual" anthropological examination using MDCT

Advantages	Even if there are no ante-mortem records the amount of soft tissue and bone information may achieve a positive identification of an unknown body
	CT is not restricted to a single area of the body
	No need for lengthy preparations of bones
	Less chance of information or evidence being lost
	Fast scan and analysis time
	Expert analysis can occur remotely and at any time
	MDCT is non-invasive. Virtual anthropology procedures are likely to be culturally more acceptable
	A virtual anthropology database could be created improving on traditional anthropological tables
Limitations	Access to MDCT scanners may be limited
	Equipment is expensive

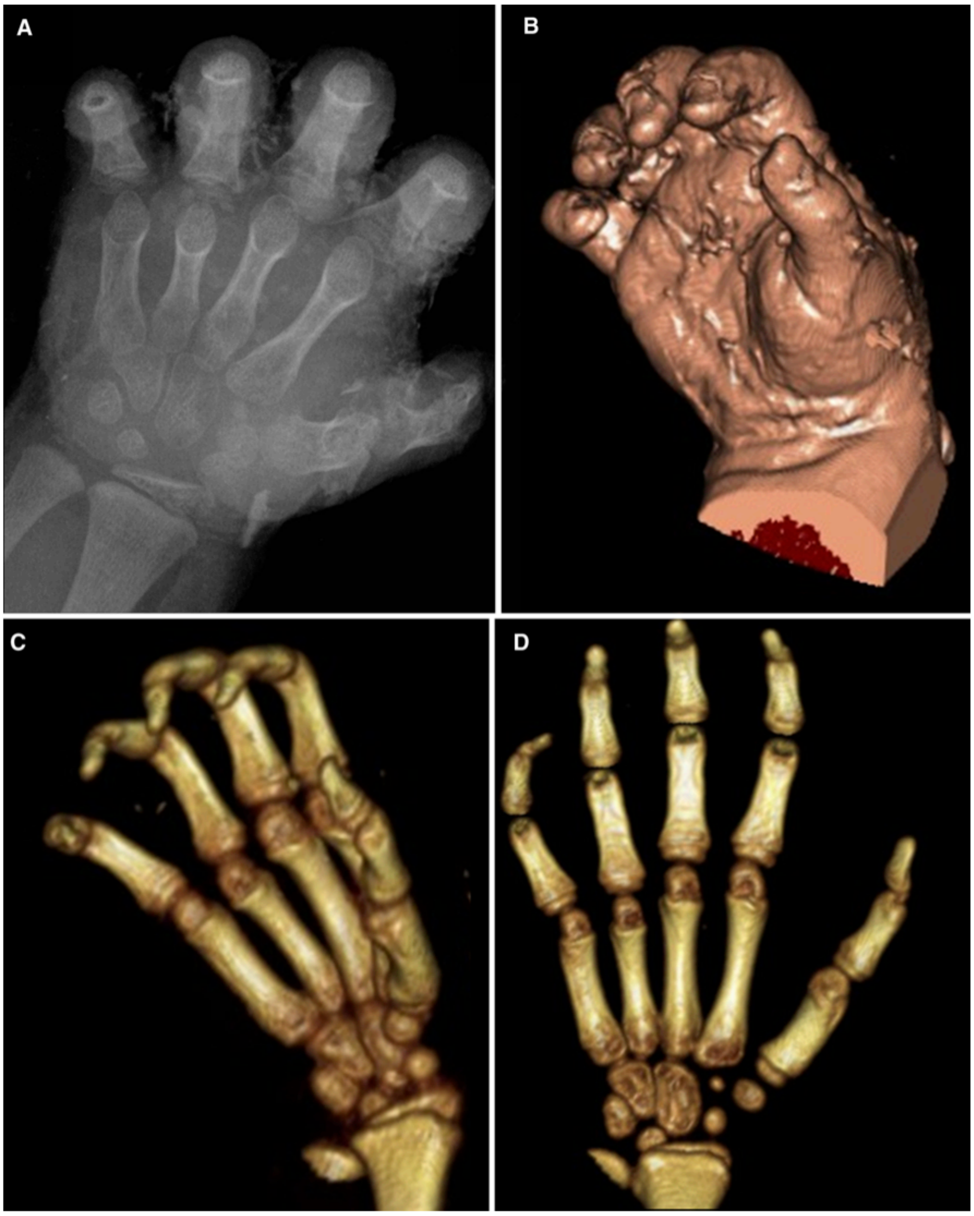

Fig. 5 a Radiograph—difficult to get the hand straight post-mortem, particularly in contractures that may occur due to fire. b Virtual representation of hand from CT. c 3D reconstruction of the hand that can be view in any plane. d Manipulation of 3D reconstruction to open out hand

Length and development of long bones: measurement accuracy

As ossification progresses, bones become amenable to recording length, width and morphology which can be used to assess the age and maturity of the individual. As above, the developing secondary centers of ossification can be assessed using MDCT.

Common models of longitudinal growth changes [48, 49] of the humerus, radius, femur and tibia, measure all diaphyseal long bone lengths parallel to the bone's long axis, from the most proximal to the most distal extremity. Robinson et al. [2] have shown that there is no significant difference between longitudinal diaphyseal measurements made by MDCT or traditional osteometry.

Maturation of hand and wrist

The hand-wrist atlas of Greulich and Pyle [50] and the Tanner-Whitehouse [51] scoring system both use radiographic images which can be produced using MDCT data. For these methods it is important that the radiographs are taken in the appropriate plane of view for comparison with the standard image so that all points of interest (ossification centers) can be viewed simultaneously. Although straight forward in clinical practice this can be difficult for post-mortem assessment due to rigor or contractures. MDCT volume rendered images can be sectioned in any plane and rotated in space to better conceptualize the underlying anatomy and therefore could create an appropriate image regardless of the body position during scanning (Fig. 5). In addition Aaron et al. [11] have shown no significant loss in accuracy when using CT instead of radiographs for the Tanner-Whitehouse system.

Similar issues apply to assessment of fusion of spheno-occipital synchondrosis, iliac crest and vertebral ring and fusion of the medial end of the clavicle. These morphological changes, as with the hand and wrist, should also be accurately assessable by using MDCT.

Summary/discussion

Radiological imaging modalities have been used in forensic practice as an adjunct to traditional anthropological techniques for many years. Recently with the advancement of computer technology and MDCT in particular, there has been an international interest in a near virtual approach to forensic investigation. However, at present there is little anthropological research carried out in this area and currently there are no established protocols outlining which virtual measurements would be the most valuable to obtain a positive identification and the best imaging system to use. This may cause judicial problems were virtual anthropology to be implemented as part of a forensic or disaster victim identification protocol.

This review highlights how MDCT is a suitable imaging modality to perform all the major age determination methods for juveniles as stated by Cunha et al. [38]. MDCT also offers further advantages, first validation studies of anthropologic measurements to predict biologic profiles are easier than by traditional methods, as data acquired for clinical medical reasons could be used and second, novel more complex measurements are possible that may more consistently correlate with the biological profile of an individual.

The testing of MDCT measurements against validated anthropological collections would help support this claim and groups including the Developing Human Research Group based at the Universities of Leicester, Dundee and Cardiff are currently conducting research to encourage a shift in anthropological practice to MDCT.

Key points

1. There has been a gradual acceptance that a "near virtual autopsy" using CT could aid more traditional imaging and dry bone investigative methods, driven by its increasing availability and affordability, as well as the development of spiral and multiple detector CT, which has improved scanning speeds and resolutions.
2. Multi-detector computed tomography allows morphological skeletal information to be obtained from high quality image reconstructions in any plane and 3D modeling of images such that bones and teeth can be viewed and measured in any plane without invasive procedures, offering considerable practical and aesthetic benefits.
3. This review highlights the suitability of MDCT to perform all the age determination methods for juveniles as suggested in a recent review by Cunha et al.
4. Greater flexibility of measurements afforded by MDCT and 3D surface rendered techniques suggests MDCT has the potential to develop new methods specifically designed for CT analysis which could improve the efficiency of the age screening process on victims following a mass disaster.

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TECHNICAL NOTE

ANTHROPOLOGY

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Anthropological Measurement of the Juvenile Clavicle Using Multi-Detector Computed Tomography—Affirming Reliability

ABSTRACT: Currently, there is no standardized protocol for multi-detector computed tomography (MDCT) measurement of juvenile remains. Using 33 juvenile clavicles, this paper investigates a protocol to allow MDCT measurements, comparable or supplemental with traditional osteometric measurements, to be acquired for application to previously published algorithms. The results illustrate that there is no significant difference between MDCT measurements and those taken by direct osteometric methods. By presenting such a protocol, this paper takes the first steps toward validation of the process of conversion from measurement of dry juvenile bone to MDCT compatibility and allows the forensic world to take a step forward in standardizing the way MDCT is used for forensic practice. This paper assesses the limitations and potential applications of this virtual approach and offers some suggestions for where further work might progress the conversion of these new approaches into legally admissible anthropological techniques of age estimation.

KEYWORDS: forensic science, forensic anthropology, multi-detector computed tomography, virtual, juvenile, clavicle

The anthropological examination of defleshed bones is routinely undertaken in medico-legal and disaster victim identification (DVI) investigations as an aid to the accurate determination of all four principal parameters of biologic identity: age, sex, stature, and ethnicity, and to assist in establishing personal indicators of identity. In forensic practice, the examination of defleshed bones remains the "gold standard" for obtaining accurate and repeatable osteometric measurements (1) as original algorithms are derived from dry bone studies. However, the removal of soft tissue from bone can be a time-consuming and invasive procedure which can damage underlying bone and therefore should only be undertaken when there is little to gain from retaining the covering soft tissues and when there is no viable alternative (2). Additionally, the maceration of bones may be contrary to religious practices and beliefs, or even prohibited by national laws and regulations (3).

The radiographic techniques of plain X-ray, dental X-ray, and fluoroscopy are frequently used to gain additional information (4–8). Where it may not be permissible or possible to remove soft tissue (9), these imaging modalities offer some indication of features that are of value in the identification of the deceased (10). Recently, however, there has been a trend toward the use of multi-detector computed tomography (MDCT) in forensic anthropological research that is increasingly in competition with more traditional imaging modalities for cadaveric investigation (1,11–15). In light of the recent Law Commission report on expert evidence (16), there is an increasing demand within the judicial system that the reliability, accuracy, repeatability, and validity of all forensic science techniques and methodologies be rigorously assessed before consideration for admissibility.

MDCT can reconstruct an image data set in any plane without significant loss of resolution and therefore provides the opportunity to measure and view bones in three dimensions without defleshing. In addition, by offering an alternative to traditional imaging techniques, this modality helps to minimize radiologic exposure, reduce the number of on-site personnel, and overcome problems associated with contamination and judicial requirements (17). However, in order for MDCT examinations to become routinely implemented for human osteological identification by metric evaluation, evidence is required that this virtual approach has comparable accuracy and repeatability to traditional techniques where measurements may be directly recorded from the dry bone.

This project uses remains from the Scheuer juvenile skeletal collection, which is believed to be the only active repository exclusively for juvenile skeletal remains held anywhere in the world. The collection contains the skeletons of over 100 individuals, from archaeologic, forensic, and historic anatomic sources and is held in the Centre for Anatomy and Human Identification at the University of Dundee, Scotland. Many of the demographics for these remains are documented, but for those where information is unknown, each skeleton has been assessed by trained

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forensic anthropologists using standard anthropological techniques (18). During the curatorial process of the collection, and as a mechanism of internal audit, this approach was also performed on the material of documented demographics to ensure consistency of approach and was found to be highly accurate. To the best of the authors' knowledge, this is the first series of MDCT studies of this nature to be undertaken exclusively on juvenile skeletal remains.

As the primary demographic feature used in the identification of juvenile remains is age estimation (18,19), determining the accuracy and repeatability of measurements derived by traditional and MDCT measurements is a vital preliminary step in validating the admissibility of MDCT analysis for forensic and DVI investigations. This would accelerate the process of anthropological assessment and remove the necessity to deflesh remains, which may be considered less distasteful and more morally acceptable, particularly when dealing with young victims.

This study concentrates on the examination of the juvenile clavicle as it exhibits the longest period of growth-related activity of all the long bones of the human skeleton. It displays a high level of sexual dimorphism in the adult (20,21), exhibits some characteristics of ethnicity (22), can be of value in stature estimation, and may even assist in the identification of hand dominance. However, the protracted age range for both diaphyseal growth and epiphyseal union means the clavicle is arguably of greatest value in the estimation of age at death (19) and is regularly utilized in the evaluation of age from the living (23). Due to the extremely early attainment of adult morphology, changes in length and width are the most meaningful measurements for age determination of juvenile specimens. Studies on the fetal growth of the clavicle both by direct examination of the bone and by ultrasound (24), have shown that the clavicle grows at a virtually linear rate of approximately 1 mm per week throughout fetal life (19). Despite this, there have been few studies that have investigated the relationship between clavicle length and age in older juveniles (25), most probably due to the universal paucity of juvenile skeletal remains of documented age at death. Thus, due to its potential use for human biologic profiling and the current lack of knowledge in this area, the clavicle was chosen as the first bone to study from the Scheuer collection.

The objectives of this study were twofold: (i) to ascertain whether an MDCT measurement protocol produces equivalent measurements to those recorded from dry bone, and (ii) to determine whether anthropological experience is relevant to the reliability and accuracy of the recording process.

Materials and Methods

The entirety of the Scheuer collection was scanned using a truck mounted SOMATOM® Emotion 16-detector CT scanner (Siemens Healthcare, Surrey, U.K.). Scans involved helical acquisition using a 0.75 mm slice thickness, 120kVp, and 100 mA with bone and soft tissue reconstructions at 1.25 mm. Data were stored on compact disk and transferred to the image archive at East Midlands Forensic Pathology Unit. Image analysis was performed using image analysis software (OsiriX version 3.7.1; distributed freely as open-source software under the GNU licensing scheme at the following Web site: http://homepage.mac.com/rossetantoine/osirix). Multi-plane reconstructions (MPR) from the data set were then created to allow measurements to be taken in any plane.

Thirty-three clavicles from 20 individuals were selected for further analysis. MDCT measurements on the clavicles were

(a)

(b)

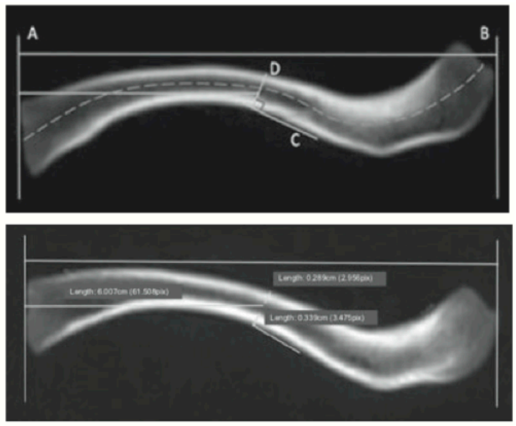

FIG. 1—*(a) Reconstructed MPR image illustrating the measurement method for length, diameter, and cortical thickness. Orthogonal boundaries A and B were drawn at the most extreme points of each clavicle to enable the maximum clavicular length (AB) to be measured. A further line was also generated along the mid-shaft of the bone (dotted line) to give the true length. A line at a tangent to the clavicular curvature at the midpoint (C) was generated as a baseline from which a line (D) at a 90° angle determined the mid-shaft diameter. The base plane of the reconstructed 3D clavicle was defined by identification of the most inferior parts of the bone, which would ordinarily lie in contact with the horizontal surface of an osteometric board. Two additional boundaries, orthogonal to this plane (A, B), were generated to simulate the sliding planes of an osteometric board and define the most extreme points. By propagating these boundaries through the entire series of images and scrolling through the contiguous slices, maximum precision could be achieved. (b) Reconstructed MPR illustrating the measurement method for cortical thickness (26). To measure the cortical thickness of the bone, the mid-shaft was determined as occurring at half of the maximum length measurement. A line at a tangent to the curvature at this point was then created, and the maximum diameter of the bone was given by the width at a 90° angle to this line. Using this as a baseline, the cortical was measured on each side of the medullary cavity, and the total cortical thickness was calculated by the addition of these two measurements.*

performed by a forensic anthropology graduate (ALB), a forensic pathologist (GNR), and a medical student (JB) under the supervision of a radiologist (BM). Osteological measurement of the clavicle using MDCT has not been reported previously, and thus there is no defined protocol available. The method therefore involved taking measurements in planes based on how the bone would lie on a traditional osteometric board (Fig. 1*a*). The forensic pathologist and the medical student were given instructions by the anthropologist on how to achieve MPR comparable with standard osteological measurement (as outlined in Fig. 1*a*). With experience, each clavicle could be measured in less than 10 min. The medical student had little experience with either MDCT or anthropological measurements. Measurements were undertaken blind to the knowledge of other practitioners' results. Previous studies have demonstrated that inter-observer errors between different professional groups for osteological measurements using MDCT are extremely low (1). Therefore, although all measurements were overseen by the appropriate professionals (BM, SB), it was not considered necessary for each measurement to be recorded by all study participants.

Measurements

The "osteological" maximum length of the dry bone clavicles was recorded by the anthropologist using sliding calipers with measurements recorded to the nearest 0.1 mm. This measurement was recorded five times for each bone, and the intra-observer variability calculated. The MDCT maximum clavicle length was recorded to the nearest 0.1 of a millimeter (25) by the anthropologist and the pathologist from reconstructed images (Fig. 1*a*).

Additional measurements recorded from the MDCT image included as a means of future proofing for possible studies in age evaluation from these dimensions: Cortical thickness (AB) was measured using the method described by Kaur and Jit (26) (Fig. 1*b*). The midpoint of the shaft was identified (point C), and the cortical index was calculated as the ratio of the combined cortical thickness to the maximum diameter of the bone (26) at this point. Also recorded were the maximum bone diameter at its medial, central, and lateral regions and a true length of the bone (following the "S" shaped curvature, as opposed to the distance between the proximal and distal extremities). Measurements were repeated three times, and the mean was recorded.

Where multiple measurements were compared, analysis of agreement was performed using a method described by Bland and Altman (27). First, the effect on measurement magnitude of intra-observer variability was assessed and then the within-subject standard deviation (wSD) was calculated. The difference between a subject's measurement and the true value would be expected to be less than 1.96 wSD for 95% of observations. The 95% repeatability was calculated ($\sqrt{2} \times 1.96 \times$ wSD), that is, the range within which 95% pairs of measurements would be expected to lie. Where two different measurement approaches are compared, the mean difference and 95% confidence intervals were calculated to identify any significant measurement bias for each technique. The wSD and 95% repeatability were also assessed for MDCT measurements compared with osteological measurements. The inter-observer error was then calculated by comparing the measurements obtained by GNR and those by ALB to determine whether the accuracy of measurements was affected by professional experience of the observer. The consistency of different individuals using MDCT was also analyzed by calculating the average "scaled error index," which permits the comparison of measurements regardless of scale (28).

Results

Data for the repeated osteological and MDCT measurements are given in Tables 1 and 2. The wSD is extremely low for both repeat osteometric and MDCT measurements, which indicates that each measurement did not deviate far from the mean (0.08 and 0.49 mm, respectively). Comparison of mean osteological and MDCT measurements is given in Table 3 and plotted in Fig. 2. There is no evidence that MDCT consistently either over or under measures compared with osteometric measurements, with an average difference of 0.005 mm. Also, there is no evidence of error being proportional to the magnitude of the measurement. The wSD for intra-observer variation between osteometric and MDCT measurements was 0.39 mm. The inter-observer error is also minimal, indicating there is no statistical significance between measurements taken by individuals with different professional experience. The statistical results are presented in Table 4, along with the mean measurements obtained by each observer and their associated absolute error. The low error rates signify that the results are consistent.

TABLE 1—*Osteometric measurement of the maximum length of the clavicle.*

	Repeat Measurement (mm)							
Skeleton	1	2	3	4	5	Mean (mm)	wSD	95% Repeatability
1a	120.1	120.2	120.2	120.1	120.1	120.1		
1b	123.8	123.7	123.7	123.5	123.6	123.7		
2a	44.8	45.0	44.9	44.9	45.0	44.9		
2b	45.0	45.0	44.9	45.0	45.0	45.0		
3a	93.7	93.7	93.7	93.8	93.7	93.7		
3b	92.5	92.6	92.5	92.5	92.5	92.5		
4a	80.6	80.7	80.6	80.7	80.7	80.7		
4b	78.5	78.5	78.4	78.6	78.6	78.5		
5	144.2	144.2	144.2	144.1	144.1	144.2		
6a	85.9	85.8	85.8	85.9	85.9	85.9		
6b	80.7	80.8	80.8	80.6	80.8	80.7		
7	80.5	80.4	80.4	80.5	80.3	80.4		
8	106.2	106.1	106.0	106.2	106.1	106.1		
9a	145.9	145.9	146.0	145.9	146.0	145.9		
9b	143.2	143.3	143.2	143.2	143.3	143.2		
10a	83.7	83.6	83.6	83.5	83.5	83.6		
10b	83.5	83.4	83.5	83.5	83.5	83.4		
11a	54.2	54.1	54.2	54.2	54.2	54.2		
11b	54.9	54.9	54.9	54.9	54.9	54.9		
12	82.7	82.7	82.7	82.8	82.8	82.7		
13a	78.0	78.0	77.6	77.6	77.9	77.8		
13b	76.8	76.8	76.7	76.8	76.8	76.8		
14a	124.2	124.3	124.2	124.0	124.2	124.2		
14b	124.7	124.7	124.8	124.7	124.7	124.7		
15a	40.0	39.9	39.8	40.0	40.0	39.9		
15b	39.8	39.8	39.8	39.7	39.8	39.8		
16a	47.4	47.5	47.6	47.6	47.4	47.5		
16b	49.1	49.1	49.2	49.0	49.2	49.1		
17	121.4	121.5	121.5	121.5	121.4	121.5		
18a	108.5	108.5	108.3	108.6	108.6	108.5		
18b	111.2	111.2	111.2	111.2	111.2	111.2		
19	139.1	139.3	139.1	139.0	139.2	139.1		
20	123.9	123.9	123.8	123.8	123.8	123.8		
							0.08 mm	0.22 mm

Discussion

Radiologic imaging modalities have been used in forensic practice as an adjunct to traditional anthropological techniques for many years. Recently with the advancement of computer technology and MDCT in particular, there has been an international interest in a near virtual approach to forensic investigation, with some studies producing promising results (29,30) to support the implementation of a virtual anthropological investigation in routine forensic practice instead of the traditional invasive approach. However, at present, there is a deficiency in the amount of anthropological research carried out in this area; such that there is currently no established protocol outlining which virtual measurements would be the most valuable to obtain a successful identification and the best imaging system to use. This is the first such study to the authors' knowledge that provides direction to further this area of research.

There is a growing anthology of literature relating to morphological changes at the medial end of the clavicle (31–35). Although medial epiphysis morphology is of considerable value in estimating age at death in the postpubertal period, more attention to changes in diaphyseal length is required in current research to facilitate the identification of juvenile individuals. To supplement or replace traditional anthropological techniques in a forensic investigation, the practicality of a "virtual" MDCT approach for anthropology is clear. This could be of particular use in contaminated mass fatality incidents where the presence

TABLE 2—*MDCT measurements of the maximum length of the clavicle.*

Skeleton	Repeat Measurement (mm) 1	2	3	Mean (mm)	wSD	95% Repeatability
1a	120.1	120.1	120.4	120.2		
1b	123.3	122.8	123.0	123.0		
2a	45.3	44.4	45.3	45.0		
2b	45.2	45.3	44.7	45.1		
3a	94.4	94.2	93.5	94.0		
3b	92.9	92.9	92.5	92.7		
4a	79.9	79.9	79.9	79.9		
4b	77.2	78.1	78.0	77.8		
5	144.2	144.4	144.4	144.3		
6a	86.1	86.0	85.8	86.0		
6b	80.3	80.7	80.7	80.6		
7	81.4	81.4	81.3	81.3		
8	105.2	105.2	105.3	105.2		
9a	145.4	145.5	145.0	145.3		
9b	143.8	143.8	144.0	143.8		
10a	82.8	82.9	83.0	82.9		
10b	83.9	83.2	83.8	83.6		
11a	54.8	54.6	54.8	54.8		
11b	55.4	55.0	55.2	55.2		
12	83.0	83.2	83.0	83.0		
13a	78.1	78.4	77.9	78.1		
13b	76.5	76.8	77.0	76.7		
14a	123.5	124.0	123.7	123.7		
14b	124.1	123.3	124.7	124.0		
15a	40.0	39.9	40.3	40.0		
15b	40.3	40.1	39.8	40.1		
16a	48.1	48.1	48.4	48.2		
16b	49.7	49.4	49.3	49.5		
17	121.9	122.0	122.7	122.2		
18a	109.0	109.0	109.3	109.1		
18b	111.1	110.5	111.2	110.9		
19	139.4	139.5	139.2	139.4		
20	122.9	123.0	123.1	123.0		
					0.49 mm	1.36 mm

TABLE 3—*MDCT, Osteometric board analysis using maximum length of the clavicle measurements.*

Skeleton	MDCT	Osteometric Board	Difference	wSD	95% Repeatability
1a	120.20	120.14	0.06		
1b	123.04	123.66	−0.62		
2a	45.00	44.92	0.08		
2b	45.07	44.98	0.09		
3a	94.01	93.72	0.29		
3b	92.74	92.52	0.22		
4a	79.88	80.66	−0.78		
4b	77.77	78.52	−0.75		
5	144.34	144.16	0.18		
6a	85.95	85.86	0.09		
6b	80.56	80.74	−0.18		
7	81.34	80.42	0.92		
8	105.24	106.12	−0.88		
9a	145.30	145.94	−0.64		
9b	143.84	143.24	0.60		
10a	82.90	83.58	−0.68		
10b	83.62	83.48	0.14		
11a	54.76	54.18	0.58		
11b	55.18	54.90	0.28		
12	83.02	82.74	0.28		
13a	78.12	77.82	0.30		
13b	76.73	76.78	−0.05		
14a	123.73	124.18	−0.45		
14b	124.04	124.72	−0.68		
15a	40.04	39.94	0.10		
15b	40.06	39.78	0.28		
16a	48.19	47.50	0.69		
16b	49.46	49.12	0.34		
17	122.19	121.46	0.73		
18a	109.10	108.50	0.60		
18b	110.91	111.20	−0.29		
19	139.35	139.14	0.21		
20	122.98	123.84	−0.86		
				0.36 mm	0.99 mm
Mean difference		0.005			
Standard deviation		0.51			
95% CI		0.17			

of chemical, biologic, radiologic, or nuclear contaminants could render the process of defleshing bones for anthropological assessment unfeasible. For MDCT, the scan and image processing time is rapid, allowing practical visualization and interaction with the three-dimensional image and consistent measurements. The clavicle is an appropriate bone to study for application to MDCT virtual dissection as it often survives inhumation more successfully than other long bones (19) and is reported to show a linear trend of growth for age estimation certainly in the younger years. However, to date, the clavicle has not been extensively (35) examined by MDCT throughout the full ontogenetic spectrum. Comparisons of the results obtained in this study are similar to those presented by Black and Scheuer (25), which, to the authors' knowledge, is the only previously published set of results for this bone. There is therefore supporting evidence to suggest that MDCT measurements closely parallel the dry osteological data available from existing published communications.

Using the clavicle, this paper investigates a protocol to allow MDCT measurements, comparable or supplemental with traditional osteometric measurements, to be acquired for application to previously published algorithms. By presenting such a protocol (Table 5), it allows the forensic world to take a step forward in standardizing the way MDCT is used for forensic practice. Comparison to dry bone is essential as all accepted current methodologies are based on dry bone evaluation. However, there is a significant paucity of juvenile skeletal remains of documented age, and so it is vital that the association between dry bone and MDCT compatibility is initially confirmed, to permit reassessment of existing algorithms and the design and development of new ones.

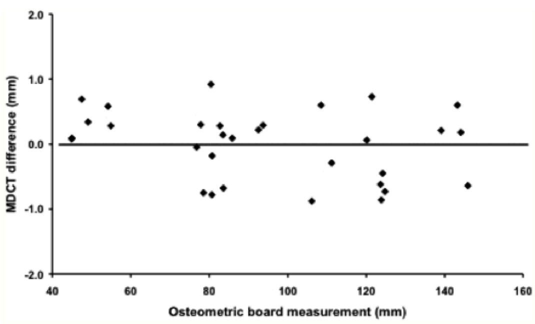

FIG. 2—*Difference of MDCT measured maximum length compared with length of dry bone measurements via the osteometric board.*

TABLE 4—*Inter-observer error of maximum length of the clavicle.*

	Observer						
	Measurement (mm)						
Skeleton	GNR	ALB	Average (mm)	Error (mm)	SD	VAR	95% CI of Difference
1a	119.7	120.1	119.9	0.4	0.3	0.1	1.1
1b	123.4	123.3	123.4	0.1	0.1	0.0	
2a	45.2	45.3	45.3	0.1	0.1	0.0	
2b	45.2	45.2	45.2	0.0	0.0	0.0	
3a	93.3	94.4	93.8	1.1	0.8	0.6	
3b	92.0	92.9	92.4	0.9	0.6	0.4	
4a	79.6	79.9	79.7	0.3	0.2	0.0	
4b	77.8	77.2	77.5	0.6	0.4	0.2	
5	143.1	144.2	143.7	1.1	0.8	0.6	
6a	85.3	86.1	85.7	0.8	0.5	0.3	
6b	80.0	80.3	80.2	0.3	0.2	0.0	
7	80.9	81.4	81.1	0.5	0.3	0.1	
8	105.4	105.2	105.3	0.2	0.1	0.0	
9a	145.2	145.4	145.3	0.2	0.2	0.0	
9b	143.6	143.8	143.7	0.2	0.1	0.0	
10a	81.2	82.8	82.0	1.6	1.1	1.3	
10b	82.9	83.9	83.4	1.0	0.7	0.5	
11a	53.3	54.8	54.1	1.5	1.1	1.1	
11b	53.8	55.4	54.6	1.6	1.2	1.3	
12	81.4	83.0	82.2	1.6	1.1	1.2	
13a	78.3	78.1	78.2	0.2	0.2	0.0	
13b	75.9	76.5	76.2	0.6	0.4	0.2	
14a	123.7	123.5	123.6	0.2	0.2	0.0	
14b	124.5	124.1	124.3	0.4	0.3	0.1	
15a	39.4	40.0	39.7	0.6	0.4	0.2	
15b	39.8	40.3	40.0	0.5	0.3	0.1	
16a	48.1	48.1	48.1	0.0	0.0	0.0	
16b	48.1	49.7	48.9	1.6	1.1	1.2	
17	121.6	121.9	121.8	0.3	0.2	0.1	
18a	108.5	109.0	108.8	0.5	0.4	0.1	
18b	111.0	111.1	111.1	0.1	0.1	0.0	
19	139.6	139.4	139.5	0.2	0.2	0.0	
20	122.6	122.9	122.8	0.3	0.2	0.0	
			Mean	0.6	0.4	0.3	

TABLE 5—*Summary of measurement protocol.*

	Virtual Osteometric Board
Step 1 (Base plane)	Generate base plane of the reconstructed bone by identifying the most inferior part of the bone, which would ordinarily lie in contact with the horizontal surface by scrolling through the contiguous image slices.
Step 2 (Sliding planes)	Create two additional boundaries orthogonal to this base plane to simulate the sliding planes of an osteometric board and define the most extreme points.
Step 3	By scrolling through the contiguous slices, maximum precision can be achieved.

The results of this study illustrate that there is no significant difference between the measurements taken by MDCT and those by the direct osteometric method although the MDCT measurements have slightly higher intra-observer variability. This may be expected as the methodology for virtual calliper placement has more opportunity for operator variation but does not imply it is less accurate. MDCT has greater flexibility of measurement and is not restricted to physical landmarks.

A number of problems were encountered during the MDCT investigation which must be considered. The main problem was the lack of any previous standards relating to MDCT measurement of the clavicle. The current MDCT protocol proved repeatable by a variety of practitioners from different backgrounds (Table 4).

In light of the National Academy of Sciences report on forensic expert evidence and the most recent Law Commission report for England and Wales (16), it is essential that all existing methodologies be tested on appropriate samples and that protocols be developed for new approaches that are based on robust principles and sound scientific analysis.

This paper sets the first steps toward validation of the process of conversion from measurement of dry juvenile bone to MDCT compatibility and offers some suggestions for where further work might progress. The clavicle has been shown to be a valid model for these purposes, but it cannot be assumed that this will hold true for all other bones in the juvenile skeleton until these areas are also tested, and so this will also require further detailed investigation.

The paper presents the results from the assessment of the clavicle only. Further MDCT measurement protocols are being considered for other bones to search for measurements that may prove to be of added value in the estimation of biologic indicators such as sex and age. The clavicle was chosen as the first bone to study from the Scheuer collection by the Developing Human Research Group (based at the University of Leicester). This group is currently carrying out a series of similar studies that will eventually provide guidance on the collection of metric anthropological data via MDCT for the entire juvenile skeleton.

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ORIGINAL ARTICLE

Postmortem computed tomography age assessment of juvenile dentition: comparison against traditional OPT assessment

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Abstract Age estimation is one of the primary demographic features used in the identification of juvenile remains. Determining the accuracy and repeatability of age estimations based on postmortem computed tomography (PMCT) data compared with those using conventional orthopantomography (OPT) images is important to validate the use of PMCT as a single imaging technique in forensic and disaster victim identification (DVI). In this study, 19 juvenile mandibles and maxilla of known age underwent both OPT and PMCT. Three raters then estimated dental age using the resulting images and 3D reconstructions. This assessment showed excellent agreement between the age estimations using the two techniques for all three observers. PMCT also offers a greater range of measurements for both the dentition and the whole human skeleton using a single image acquisition and therefore has the potential to improve both the speed and accuracy of age estimation.

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Introduction

Under the United Nations Universal Declaration of Human Rights, retaining an identity after death represents a basic human right [1]. When dealing with juvenile remains, age-at-death is an important criterion, and odontological examination is arguably the most rapid and practical method available for this purpose. Dental age markers are reported to correlate more strongly with chronological age than skeletal markers, being less affected by the environment [2]. Dental age estimation techniques are predominantly based on the degree of mineralization and/or eruption of the dentition and involve matching radiographs to atlases of dental development or assigning formative stages of mineralization by scoring individual teeth. Although development of the third molar is regularly used as a quick indication of sub-adult age [3], this tooth displays the highest degree of variation amongst individuals, and therefore a technique using multiple tooth development is preferred, where possible [4, 5]. The most regularly utilised methods for odontological age-at-death estimation in UK forensic practice at present are the 'London Atlas of Human Tooth Development and Eruption (QMUL) [6, 7] and Demirjian's method [8, 9] of formative tooth maturity scores'. The QMUL method is an adaptation of Ubelaker's 1978 atlas [10], using data from modern UK dental collections, illustrating dental structures including tooth roots, with a more comprehensive range of age stages and where eruption refers to emergence from alveolar bone.

An orthopantomogram (OPT) captures a full dental arcade in a single image using a revolving X-ray tube and is traditionally used in forensic practice. However, OPTs may be difficult to obtain in a forensic context, due to a lack of access to equipment or difficulties with the revolving nature of the image acquisition due to severe trauma or rigor mortis [11, 12]. Recently, there has been gradual acceptance that postmortem computed tomography (PMCT) could aid, and potentially replace, this more traditional imaging method by both reproducing and augmenting the information available from an OPT. This has been driven by increasing availability and affordability, and the development of spiral and multiple detector CT (MDCT), which has improved scanning speeds and resolution, allowing high quality image reconstructions in multiple planes and 3D modelling of slices [13]. A single image acquisition using PMCT allows teeth and bones to be assessed in any plane without invasive procedures, offering considerable practical and aesthetic benefits. Used in conjunction with a teleradiology system, such as the FiMag system [14], it would also allow secure global distribution and evaluation of images used for identification purposes. Although the availability of PMCT for forensic examinations is not widely spread, if a 'one stop' protocol could be produced for the examination and identification of the entire human skeleton using PMCT, this could replace all existing image techniques currently used. A particular advantage of PMCT dental evaluation is that it does not need the placement of image receptors into the mouth, required for intra-oral radiography, nor manipulation of the head to align the X-ray beam as required for OPT [15]. However, there are significant disadvantages due to increased sensitivity to metal artefact [16, 17]. CT reconstruction algorithms can now accommodate for metal implants such as titanium, but high-density metals such as gold or mercury in dental restorations are still a problem [18]. Cone beam CT (CBCT) is also being increasingly used in dental practice and gives similar (but not superior) 3D dental assessment. However, CBCT cannot be practically used to image the whole skeleton and so could not provide a 'one stop' protocol.

For PMCT to become routinely implemented in forensic practice, evidence is required that age estimations using this approach have comparable accuracy and repeatability to methods using conventional OPTs. The aims of this investigation were therefore (1) to determine if age estimations based on PMCT data were in agreement with those made on OPTs, using the London Atlas of Human Tooth Development and Eruption (QMUL) and Demirjian's dental age estimation method; (2) to determine whether prior knowledge of software had any effect on precision of measurement; and (3) to asses intra- and inter-rater variability between three observers of varying experience with dental PMCT age estimation.

Materials and methods

Selection of cases

This project utilised human remains from the Scheuer Juvenile Skeletal Collection, believed to be the only active repository exclusively for juvenile skeletal remains. The collection contains the skeletons of over 100 individuals, from archaeological, forensic and historical anatomical sources and is held in the Centre for Anatomy and Human Identification at the University of Dundee. Nineteen mandibles and maxillae from individuals of known age (between 0 and 18 years) were selected randomly from the collection, by an independent practitioner who had no further involvement or knowledge of this investigation and CT scanned by a trained dental radiographer. PMCT dental images were acquired using a truck mounted SOMATOM® Emotion 16-detector CT scanner (Siemens AG Medical Solutions). Scans involved helical acquisition using a 0.75 mm slice thickness, 120 kVp, and 100 mA with bone and soft tissue reconstructions at 1.25 mm. Data were stored on compact disc and transferred to a workstation with image analysis software (OsiriX version 3.7.1; distributed freely as open-source software under the GNU licensing scheme at the following Web site: http://homepage.mac.com/rossetantoine/osirix).

Imaging protocol

Image analysis for the PMCT data was undertaken using software (OsiriX version 3.7.1; distributed freely as open-source software under the GNU licensing scheme at the following Web site: http://homepage.mac.com/rossetantoine/osirix). Curved multi-plane reconstructions (curved MPRs) from the data set were created to allow dental analysis to be undertaken in any plane. Typical OPT and OsiriX curved MPRs are shown in Fig. 1.

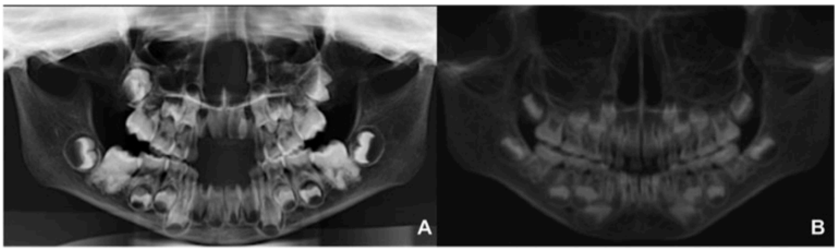

Fig. 1 **a** Conventional orthopantomography (OPT) image and **b** multidetector computed tomography curved MPR image

Table 1 Comparison of predicted age in months obtained using the QMUL method and OPT and PMCT images

Case	Documented age (months)	QMUL method						Average
		Rater 1		Rater 2		Rater 3		
		OPT	PMCT	OPT	PMCT	OPT	PMCT	
1	228	282	270	288	276	282	270	278.0
2	96	114	115.5	114	120	114	102	113.3
3	144	144	150	144	150	150	150	148.0
4	168	174	160	180	160	150	162	164.3
5	228	282	234	288	240	282	234	260.0
6	228	222	246	222	252	234	234	235.0
7	228	186	222	180	186	198	216	198.0
8	Perinate	1.5	2.25	1.5	2.25	1.5	2	1.8
9	2	1.5	3	1.5	3	1.5	2	2.1
10	18	18	18	18	18	18	18	18.0
11	18	24	30	30	30	30	24	28.0
12	96	90	97.2	90	98.4	90	102	94.6
13	144	150	162	150	162	150	162	156.0
14	60	78	78	78	81	78	78	78.5
15	96	168	174	168	174	168	162	169.0
16	216	198	198	204	205.5	210	210	204.3
17	144	96	92.4	90	88.8	114	102	97.2
18	132	66	42	66	42	66	42	54.0
19	216	72	168	72	168	78	168	121.0

Image analysis

The PMCT and OPTs were reviewed independently by three raters: two dental practitioners with experience in dental age estimation and one forensic anthropologist with PMCT data analysis experience. Observers were unaware of the age of each individual. The raters applied the QMUL method to each case, followed 1 week later by the Demirjian method (where possible—see below). The PMCT images were assessed 2 months after the OPTs. The age assigned through both methods and both media were then compared with the original age assigned for each individual. Age was only revealed after all age estimations were completed. If there was any uncertainty over age estimation, the practitioner was asked to comment and provide a reason for this uncertainty.

Statistical analysis

PMCT age estimations are compared to actual age using 95 % confidence interval (CI) and paired Student's t test (statistical

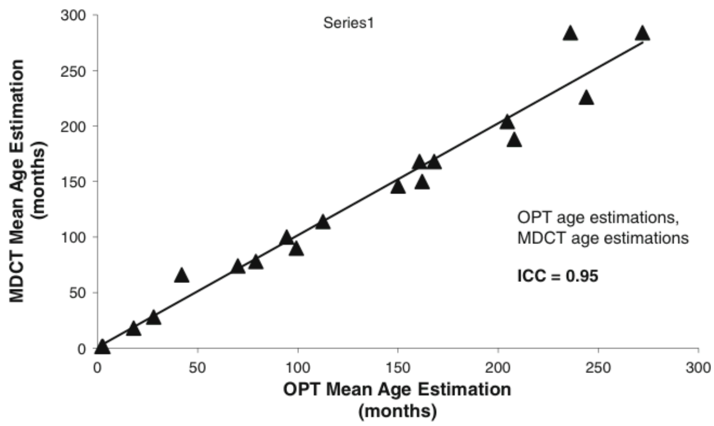

Fig. 2 Mean age estimations using PMCT images versus mean age estimations using conventional OPTs

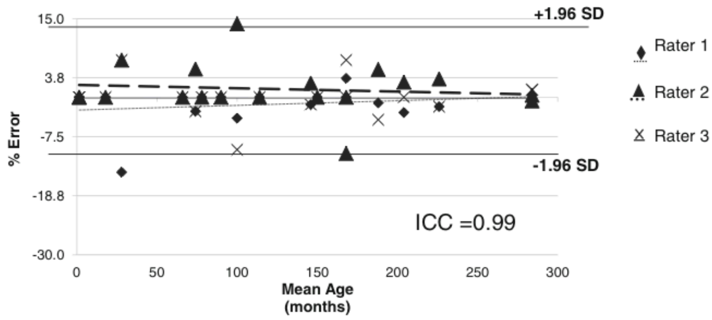

Fig. 3 The *diamond shaped* and *triangular shaped markers* correspond to the two dental practitioner observers. These data points are extremely close for almost every case compared with the inexperienced observers (*cross shaped marker*) that are clearly more scattered, with two points lying outside the confidence intervals

significance was assumed if $p<0.05$), after a Kolmogorov–Smirnov test using SPSS determines the data was normally distributed. Measurement error is calculated and presented using the 'Bland and Altman' plots [16, 19]. Statistical analysis of agreement is performed using interclass correlation (ICC) to measure the level of agreement between observers, methods and data sets. Initially, a two-way analysis of variance was performed, and if the column mean sum of squares (between methods of measurements or raters) is not significantly greater than the residual error, concordance is assumed. The ICC for the relationship provided a scalar measurement of agreement, where a value of 1 represented perfect agreement, and 0 was interpreted as a lack of any agreement. ICC has advantages over correlation coefficient analysis, as it adjusts for the effects of scale magnitude and can represent the agreements for more than two observers.

Results

Comparison of the predicted ages using OPT and PMCT are given in Table 1 and Fig. 2. Using the paired Student's t test method, no significant difference was detected between the predicted ages for OPT and PMCT for each rater (p values of 0.45, 0.56 and 0.83 for raters 1, 2 and 3 respectively). No significant inter- or intra-rater variation was found. Almost perfect agreement was illustrated, both between observers and between repeat estimations by the same raters, for OPT age estimations and PMCT age estimations, using both the QMUL and Demirjian methods with an ICC coefficient of 0.99 calculated for both (Figs. 3 and 4). Applying the QMUL method obtained almost perfect agreement between mean estimated age using PMCT, mean estimated age using OPTs and original age assignment age (Fig. 5). Age estimations using OPTs and age estimations using PMCT were also in almost perfect agreement (Fig. 5).

Unfortunately, since the Demirjian technique is only valid for individuals aged 3–16 years, the sample size was too small for a similar analysis to be performed. However, as the agreement between observers for this technique was almost perfect (ICC 0.98), our results suggest that in cases where this technique is possible, dental age estimations could potentially be improved by PMCT.

Discussion and conclusion

Orthopantomography is the standard imaging method for dental evaluation in forensic investigations. Current standards for dental identification are therefore based on this method, and there are no recognised standards for PMCT dental evaluation. This study has followed the protocols implemented for OPTs as closely as possible with slight modification where PMCT has offered improved measurement opportunities. This

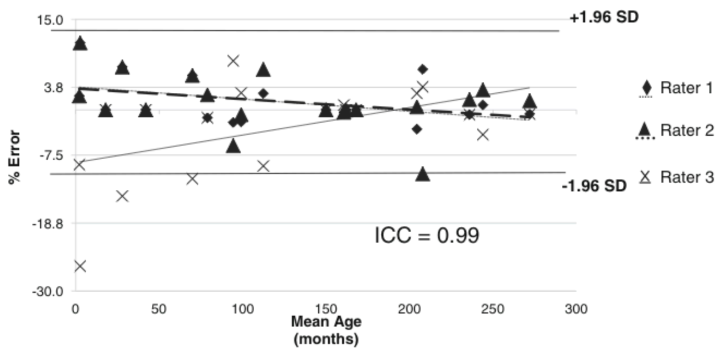

Fig. 4 For PMCT estimations, the results of the inexperienced observer are much less scattered than those using traditional OPT images, suggesting that this technique is more reproducible by individuals with no previous dental experience

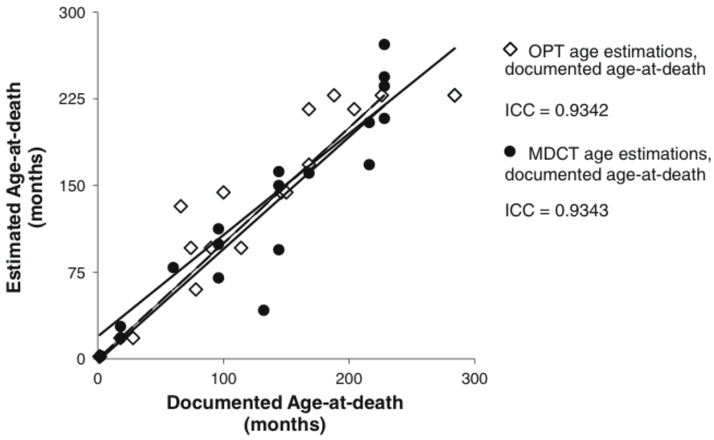

Fig. 5 Age estimations using OPTs versus age estimations using PMCT, in almost perfect agreement

includes the ability to scroll through multiple images to remove superimposition and cross-check results with high quality 3D representations. This image interpretation protocol proved repeatable when used by a variety of practitioners from different professional backgrounds and levels of experience. The figures show that the least experienced rater had greater errors in age prediction, although these were less using PMCT compared with OPT.

The authors were unable to demonstrate the increased measurement opportunities afforded by PMCT-improved age estimation. Difficulties readjusting to the multiple 2D data set and issues relating to resolution and slice thickness highlighted additional training requirements. Feedback from the raters suggested dental practitioners found it difficult to readjust to the multiple 2D data set—numerous still shots of the curved MPRs were provided to each practitioner to ensure all dental features were clearly represented. However, one rater, despite clear instructions, still viewed the images as single 2D MPRs before collating the final estimation, instead of using the multiple views to enhance one single estimation. More experience using multiple MPR data sets and surface rendered images is required so that this technique becomes more familiar to the user, something that can be achieved easily through training and experience. Resolution and slice thickness were also highlighted by all raters as an issue, creating uncertainty in a couple of cases, using the Demirjian method in particular —where the increment between scores is often slight. However, as is well known in clinical practice, PMCT has the advantage over standard radiography, by being able to isolate single thin slices without superimposition of distracting or obscuring structures (Fig. 6). This made several cases easier to assess, where using the OPT image, it was sometimes difficult to tell whether complete closure of root apices had occurred. In OPT, owing to the various positions of teeth within the jaw, roots may not be optimally positioned within the focal trough and therefore their degree of root formation in particular may be adversely interpreted. Jaws can also be digitally reconstructed using PMCT, which means even when severely disrupted, bodies could be scanned in any position, and a comprehensive dental examination could still be

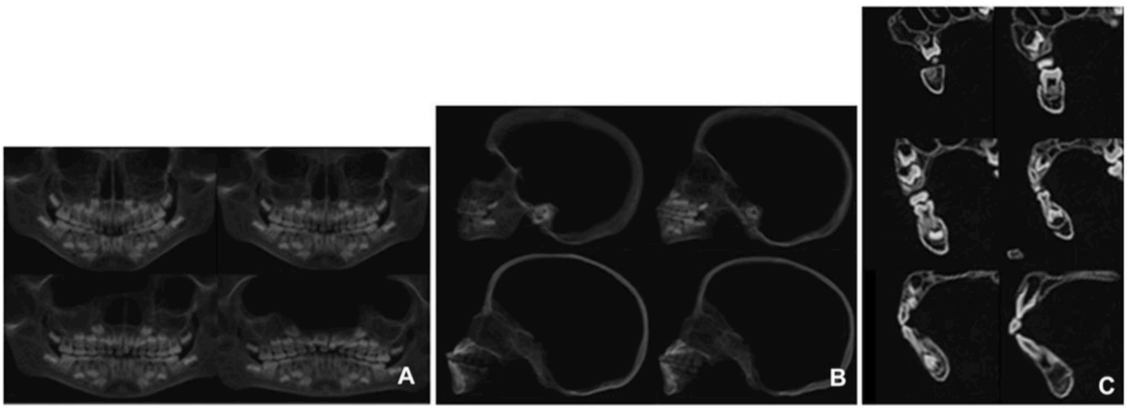

Fig. 6 Series of curved MPR views (**a**) and transverse views (**b** and **c**) of a complete dental arcade using PMCT data. By producing a series of images, problems associated with superimposition are overcome

undertaken. If a full body CT scan is already planned, processing times in forensic investigation could be reduced by negating the necessity to take dental OPTs. Finally, 3D surface rendering has the potential to further increase the accuracy of estimations and future work by the author would include assessing the impact of this.

Although further research is required in this area before a protocol utilising PMCT can be implemented internationally as part of standard forensic examination, this technical report provides evidence that PMCT is a viable imaging technique for the reliable and repeatable assessment of age from juvenile remains. To complete this study and provide further evidence to support this statement in the near future; blind-testing dental PMCT image technique on clinical cases, analysing 3D surface rendered images of each case and exploring the potential application of mandibular measurements, such as maximum ramus height is required.

In conclusion, although this investigation did not prove PMCT to be more accurate at estimating juvenile age at death, it did prove to be equally as accurate as dental age estimations using conventional OPT images. Therefore, the age estimation process performed on victims following a mass disaster may now be complemented with the use of PMCT, where previously any dental ageing analysis was performed using conventional radiographs, either full mouth periapicals or OPTs. Finally, with 3D surface rendered techniques, the authors believe PMCT has the potential to be more accurate than OPTs for juvenile dental age determination. PMCT also has greater flexibility of measurement and is not restricted to a single image, so the potential to develop new techniques exists.

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We would first like to take this opportunity to say how interesting and relevant we as a group found this publication, particularly within the current landscape of forensic radiology.

We agree with the authors that there needs to be more guidelines regarding the correct procedures for the collection, processing and presentation of PMCT acquired anthropological measurements. We also think an 'off-set plane' technique of measuring against anatomical reference planes is an interesting suggestion, and the correct approach for future investigations.

However, we would like to correct a statement made by the authors regarding a recent publication by our research group [1]. The authors state that we used "2D axial and lateral views rather than 3D isosurface mesh models". This is factually incorrect. We used a 3D MPR function on OsiriX. The resulting images are 2D but the 3D function allowed the bone to be viewed in the x- y- and z- planes. This is an important distinction as it subsequently means that our investigation was not limited by "position and orientation of the bone, constrained slice angle, or image distortion," as suggested by the authors.

In addition, we are concerned that specifically recommending software packages such as Amira and Geomagic Design X, for a 'standardized;

protocol may limit the wide-scale application of the suggested technique. These programs are relatively expensive and may not be available to every institute. We believe the measurements and techniques discussed in this publication can be achieved on a number of different software packages (such as OsiriX or Voxar etc.). Therefore, we think that specific reference to Amira and Geomagic Design X features should be removed from the 'standardized' protocol. We acknowledge that the authors make reference to the fact that their technique is transferrable to other software packages, but if a 'standardized' protocol is possible using other software, then this should be clear. The standard then becomes the actual measurement protocol using *appropriate* software.

We agree documented skeletal collections are scarce, but they do exist for example, the Scheuer Juvenile Skeletal Collection, housed at the University of Dundee. A large PMCT database would indeed provide a valuable modern population data set. We would also like to clarify that PMCT is not used in the official Interpol DVI protocol at present, as a pre-screening tool or otherwise. In 2012, the International Society of Forensic Radiology and Imaging (ISFRI) was formed, with a dedicated DVI sub committee to promote the development of 'position' statements from the society. The first DVI statement was published in JoFRI [2] in 2013. It was presented to the Pathology Advisory committee for the Interpol Standing Committee on Disaster Victim Identification, May 2013.

We also question the reasoning behind using a juvenile population for measurements of the skull. Although we appreciate that skull measurements can be used in discriminate function methods for sex and ancestry determination, these techniques are generally used on adult remains, as the majority of sex and ethnicity characteristics do not develop until after puberty. Whilst this is a minor concern, we feel that it limits the practical application of the method described.

This being said, it is great to see such a prominent journal within the field of forensic radiology publishing articles on 'precision testing' of PMCT anthropological measurements, for the advancement of 'virtual anthropology'.

This will ensure that PMCT derived data is robust enough to meet the requirements of the judicial system. We therefore look forward to reading further similar publications in future volumes of the Journal of Forensic Radiology and imaging.

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Original article

A minimum data set approach to Post-Mortem Computed Tomography reporting for anthropological biological profiling

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Conflict of Interest
The preliminary results of this investigation were presented as a non-published, oral presentation at the ISFRI conference, Zurich 2013.

Abstract

Anthropological examination of bones is routinely undertaken in medico-legal investigations to establish an individual's biological profile, particularly age. This often requires the removal of soft tissue from bone (de-fleshing), which especially when dealing with the recently deceased, is a time consuming and invasive procedure. Recent advances in multi-detector computed tomography (MDCT) have made it practical to rapidly acquire high-resolution morphological skeletal information from images of 'fleshed' remains. The aim of this study was to develop a short standard form, created from post-mortem computed tomography (PMCT) images, that contains the minimum image-set required to anthropologically assess an individual. The proposed standard forms were created for 31 juvenile forensic cases with known age-at-death, spanning the full age range of the developing human. Five observers independently used this form to estimate age-at-death. All observers estimated age in all cases, and all estimations were within the accepted ranges for traditional anthropological and odontological assessment. This study supports the implementation of this approach in forensic radiological practice.

Keywords;

Forensic radiology, anthropology, odontology, computed-tomography, post-mortem, identification

Introduction

There is good evidence showing that post-mortem computed tomography (PMCT [1]) derived anthropological measurements are accurate [2-7]. This supports the implementation of PMCT for the anthropological examination of human remains in routine forensic and disaster victim identification (DVI) events. However, guidelines regarding the measurements required, how to acquire these and how to record the information would be very helpful, particularly for international sharing of data between numerous investigators.

We believe that the majority of information required for the current official DVI Interpol form can be retrieved from a combination of PMCT and an external examination [8,9]. As the use of PMCT scanning is becoming routine for DVI in many countries, we propose it would be helpful to develop a recording format for PMCT derived data, including an anthropological reporting section, for inclusion at the next DVI Interpol update.

In a DVI event, although it is possible to transfer large quantities of raw PMCT data between different countries [10]; image transfer takes approximately 20minutes per case [10] (or longer, if there are security measures such as firewalls in place), requires a large computer memory and storage facility and from the experience of these authors, post-processing of the raw image data can be labour intensive (depending on the case). In addition, there is no guarantee that the recipient has access to sufficient post-processing facilities, or adequate knowledge of how to use these imaging systems. Creating a form containing the minimum image-set required to anthropologically assess an individual, which can be rapidly sent to numerous practitioners for independent analysis, would therefore be very useful. This standard minimum image-set should provide the necessary information about each case for multiple remote practitioners to rapidly use multiple anthropological identification techniques.

An individual's biological profile i.e. age, sex, stature and ethnicity, if not already known, is traditionally established at autopsy by osteological and odontological examination of defleshed bones. However, when dealing with the recently deceased the removal of soft tissue from bone can be an extremely time consuming procedure [11]. In addition, due to its invasive nature, the maceration of bones is often discouraged by religious practices and beliefs, or even prohibited by national laws and regulations. Therefore, PMCT analysis offers a number of practical and aesthetic benefits.

When dealing with juvenile remains age-at-death is the only factor that can be reliably determined and is therefore the most vital criteria for identification purposes [12]. Sex, stature, and race/ethnicity cannot be estimated with any degree of reliability from the skeleton until post puberty [12]. For this reason, age estimation will be used as a working example in this investigation. In the case of the developing human there is published guidance regarding the most appropriate assessment method to be applied for specific age groups [13]. However, when estimating age forensic anthropologists face a number of methodological choices; such as which skeletal region to evaluate, which method to apply, what statistical information to use and how to combine information from multiple methods to give the final estimated age. The choice of method has to take into account the individual circumstances of each case. This depends in part on the skeletal elements available for analysis: different bones have variable resilience to events such as damaging taphonomic processes or high velocity impact, leading to preservation bias. A questionnaire conducted by Garvin et al. [14], given to 145 forensic anthropologists, found that the majority of respondents vary their skeletal age estimate process case-by-case, and ultimately present officials with both a narrow and a broad possible range. Respondents displayed a very high degree of variation in how they generate their age estimations, and indicate that experience and expertise play a large role in skeletal age estimations. Other considerations are related to the actual methods, which should fulfill the following specific demands [15]:

1. They must have been presented to the scientific community through peer-reviewed publication;
2. Their accuracy must be tested using valid statistical procedures and described by clearly defined terms; and
3. The method must be accurate enough for routine forensic application.

Although the literature presents a number of studies using single osteological or odontological measurements to assist with biological profiling using PMCT, a simple recording format that allows all of these assessments to be made from a standard minimum image-set has yet to be presented, to our knowledge.

The aim of this investigation was therefore to develop and test the reliability of a single PMCT anthropological identification form, constructed from a concise collection of osteological and odontological images, which contains the minimum necessary information to perform adequate anthropological biological profiling. The minimum data-set form had to include enough information to satisfy practitioners from different professional backgrounds and experience, using a range of anthropological methods depending on the scenario. This image-set approach therefore does not dictate method, and allows forensic anthropologists to use their method of choice. The resulting form is designed to be used by suitably trained assessors, either at the site of image capture or remotely. Age estimation is the only reliable biological profile that can be made from osteological or odontological assessment of the juvenile skeleton and is therefore the only parameter used in this study to demonstrate the practical application of the suggested form. This study uses traditional osteological or odontological measurements reproduced using PMCT images, and makes no attempt to develop a new method of juvenile age estimation.

We present this PMCT anthropological reporting form, illustrating how it can be used for 'fleshed' or de-fleshed remains to assist with biological profiling during identification processes. We believe this novel approach to anthropological biological profiling using PMCT is an important contribution to the forensic community, particularly forensic radiology, by providing a minimum standard for reporting and recording osteological and odontological identification features.

Materials and methods

We developed our proposed PMCT image capture protocol and minimum image-set form using: peer-reviewed literature for osteological and odontological assessment of the developing human, including the recommendations of Cunha et al. [13]; measurement techniques previously developed by the authors [6,7] for the clavicle and dentition and validated using skeletal material from the Scheuer Juvenile skeletal collection, University of Dundee, United Kingdom; and by taking into account the information required in the current version of the Interpol post-mortem DVI forms (http://www.interpol.int/INTERPOL-expertise/Forensics/DVI-Pages/Forms. Last visited August 2013). The resulting minimum image-set form was then blind-tested using a known population of 'fleshed' juvenile cadaver PMCT forensic cases, spanning the full age range of the developing human.

The form is intended to complement the current Interpol post-mortem DVI form. The minimum data set PMCT form presented in this publication can be used to answer the large majority of questions in section B to G of the current official DVI Interpol form (http://www.interpol.int/INTERPOL-expertise/Forensics/DVI-Pages/Forms. Last accessed 26/01/2014) (with the exception of supplementary biopsy, autopsy and DNA information) and could potentially replace compulsory x-ray and dental orthopantomograph (OPT's) evidence.

PMCT minimum data set recording form

The form contains 2 pages and is illustrated in Figure 1. The first page consists of a surface image and a skeletal view of the full body. The surface image should be used to record a physical description of the cadaver and may help detect dysmorphic or congenital radiologically identifiable features, distinguishing marks, clothing, shoes and personal effects (where applicable). The whole body skeletal image allows identification of congenital or acquired bone disease, for example scoliosis. The second page of the form includes: seven views detailing the complete morphology of the skull; all the long bones and their maximum length measurement (as detailed by Buikstra and Ubelaker [16]); a clear view of each joint of the shoulder, elbow, hip and knee; an isolated view of the hand, foot, pelvis and rib ends; the spine, sacrum and axis; and finally, dental OPT reconstructions. These were selected to cover the osteological and odontological features used for biological profiling throughout the developing human.

Assessment cohort

Thirty-one cases were selected, by an independent forensic practitioner who was not involved in data analysis, from the East Midlands Forensic Pathology Units secure PMCT database. One case was used to develop and finalise the form design, before a further 30 cases were selected, which gave a total study sample of 31. Inclusion criteria were cases referred from the HM Coroner for post-mortem investigation, with known age-at-death between 0-20 years, who had whole body PMCT using multi detector CT (MDCT). Cases included both natural and unnatural deaths. The only exclusion criteria applied was individuals over 20 years of age. Cases were not actively selected based on sex or age (within the 0-20 year age range), and therefore these criteria were not assessed in this investigation as they did not effect data handling. In each case the HM coroner gave written consent for the inclusion and assessment of the case in this study. The autopsy reports containing the biological profiling information relevant to each case as well as the PMCT images where then made anonymous and given a unique study code.

Case form generation

The PMCT images of all cases were processed using OsiriX imaging software (Pixmeo, Switzerland). A single individual (AB) with both anthropology and PMCT image processing training created all of the images required for the minimum data set forms of each case. The same observer (AB) then used these images to create the two-page minimum data-set form (Figure 1) for each of the 31 cases. Long bone measurements were made and repeated, as per best practice [16], before the mean value was captured with the 3D image of each bone. If for some reason part of the recording form could not be completed (e.g. missing bone, not scanned or unclear), this was made evident on the form so that observers were aware the information was not available. To clarify, AB (the form creator) was the only observer that had access to the raw PMCT data. Previous research has demonstrated [3, 6, 7] that inter-observer error for PMCT long bone measurements is not statistically significant therefore, the other observers in this investigation used the long bone measurements captured by AB, in order to minimise post-processing and data transfer times.

Each case was assessed by AB during form construction to estimate age, blind to the autopsy findings. Four further independent and remote observers (total number of observers = 5) from different professional backgrounds that represented odontology, radiology, forensic pathology and forensic anthropology were then simultaneously sent the 2-page form for each case, by a secure file drop system. The observers professional experience ranged from 5 years - 20+ years. They were asked to estimate age for each case independently and without knowledge of autopsy findings. Each observer was provided with a list of recommended published age estimation methods (table 1) and the image-set form. The list of methods was supplied as an advisory aid for the observers who were less practiced in anthropological ageing techniques. A "technique" column was included within the reporting form so that each observer could detail the exact methods they applied. Each observer was asked to estimate age, as they would in their normal practice, and where several methods were applied they were asked to produce a breakdown of the results from each method, followed by their final age estimation.

Observers were also asked to note any difficulties they had interpreting the PMCT anthropological reporting form and any alterations they would suggest to improve it. For future development, each observer was asked to rate the 'ease of use' for the form and their 'level of certainty' of the age estimation they provided, in the following manner:

- Ease of use score: 1 = extremely easy, 2 = easy, 3 = relatively easy, 4 = difficult, 5 = extremely difficult.
- Level of certainty score: 1 = extremely uncertain, 2 = uncertain, 3 = relatively certain, 4 = certain, 5 = extremely certain.

All results were then collated. The known age-at-deaths were used as a gold standard.

Assessment methods

A list of recommended methods were supplied to each observer table 1[17-26]. This list was created in line with the guidelines published by the Study Group on Forensic Age Diagnosis (AGFAD), using the recommendations by Cunha et al [13]. There is no standard margin of error that is considered acceptable for every age estimation method. Previous work by Ritz-Timme et al. [15] suggests that the range of error associated with dental age estimation, using seven different methods for developing dentition, is up to 24 months for a small child (age 0-1yrs) and 48 months for the sub-adult range (1-12 yrs) [15]. Likewise for the diaphyseal length of long bones the majority of observers use the standards created by Maresh [22], which give similar ranges. There are many other methods of assessment including the hand and wrist atlas of Greulich and Pyle [23] and ossification center appearance and fusion as detailed by Scheuer and Black [18]. These methods all report different tolerated error ranges but are broadly consistent with those reported by Ritz-Timme and Maresh [17, 23, 27]. We have therefore concluded that error ranges of 24 months for 0-1yrs and 48 months for 1-12 yrs are the tolerated error margins typically accepted in anthropological practice and therefore these are used as our standard to be achieved by PMCT based age estimations.

Statistical analysis

Mean age estimations are compared to actual age using 95% confidence intervals (CI) and paired Students 't' test, after a Kolmogorov-Smirnov test using SPSS determined the data was normally distributed. The level of agreement between each observer's age estimation and documented age was calculated using interclass correlation (ICC) statistics. The ICC provides a scalar measurement of agreement, where a value of 1 represents almost perfect agreement and 0 represents no agreement. Measurement error is calculated and presented using the 'Bland and Altman' plots [28]. To calculate measurement error (within subject standard deviation, wSD), data should either be independent of magnitude (age) or a 'data transformation' is required [28]. In this study however the relationship of error to age was complex and varied depending on the age. Measurement error and 95% confidence intervals were therefore calculated separately for age groups 0–1 year, 1–5 yrs and 5 yrs to adult, as within these ranges the degree of error was independent of magnitude (age).

Results

The results of this study are shown in table 2 and illustrated in figure 2 and 3. For ages up to 1 year, there was no significant under- or over- estimation of age, with a mean error of 0.23 months (95% CI=0.5 months, p=0.4). For ages greater than 1 year there was a statistically significant trend to underestimate age by 3.7 months (95% CI=3.3 months, p=0.03) using the minimum data set PMCT recording forms.

Interclass correlation statistics show almost perfect agreement, between each observer using the PMCT recording form and actual age, with interclass correlation coefficients for observers one to five of 0.98, 0.99, 0.98, 0.97, 0.99 respectively. Figure 3 shows a Bland Altman plot of measurement error against documented age at death for all observers. Measurement error (within subject standard deviation, wSD) was calculated as 9.6 months overall and at 1.8, 6.3 and 13.8 months for Age groups 0–1 year, 1–5 yrs and 5 yrs-to-adult respectively. Ninety-five percent confidence intervals (1.96* wSD) were therefore 3.5, 12.4 and 27.1 months respectively and 19.0 months overall. Measurement error improves if a mean age estimation value from all observers is used, giving wSD as 6.9 months overall and 0.9, 1.8 and 10.1 months for Age groups 0–1 year, 1–5 yrs and 5 yrs-to-adult respectively. Ninety-five percent confidence intervals were therefore 1.8, 3.6 and 19.9 months respectively and 13.5 months overall. This is consistent with the principle that a multi-factorial approach using multiple observers is the standard of good practice [14, 29-33].

Mean observer score for the ease of use of the system was 2.0 (easy) and 3.8 (just under certain) for their level of certainty.

Discussion

This study presents a minimum image-set reporting form based on PMCT for anthropological examination and illustrates that these condensed PMCT anthropological findings provide sufficient information to perform multiple odontological and anthropological age estimations. Feedback from the users suggests the form is easy to use and they felt able to provide estimations with a subjective 'high degree' of certainty. This provides useful feedback for the further development of the PMCT anthropological reporting form.

Although the trend to underestimate the age of individuals older than 1 year, using the PMCT reporting forms, is statistically significant in terms of measurement magnitude; proportionally to age, a 3.7 month underestimation is not significant, as age estimations are generally stated as a range spanning years. 3.7 months is also significantly lower than the 48 month average range of error as reported by Ritz-Timme et al. [15] and Maresh [22], associated with numerous dental and diaphyseal length ageing methods, respectively. In all cases errors are within 'tolerated error margins' based on margins of error that are acceptable in previous anthropological studies [15, 22]. Furthermore, unsurprisingly [29-30], averaging multiple age estimations using several techniques produces a lower range of error.

Observer 3 used only dental information to make their age estimations (in line with their normal professional practice) and their results were still within the tolerated margin of error (age range) for dental techniques. This is an important finding in this investigation as it illustrates that even if the minimum data set form is not used to its full potential, it can still generate age estimations in almost perfect agreement with those observers using multi-factorial methods. In 3 cases (5, 12 & 15) the dental images were absent, incomplete or of poor quality due to injury, incomplete scanning or poor image resolution. In these cases the odontologist gave an estimated age based on non-dental images, and these results are shown but not included in statistical calculations of measurement error, as they would not normally have done this assessment in a forensic investigation. Three cases (13, 28 & 29) had dental anomalies (impacted canines, left/right differential development, premature premolar eruption), which may have benefited from the expertise of an odontologist. The information contained within the form will be gathered by a single lead investigator and available for distributed to subsequent investigators, so that post-processing time is reduced. This investigator therefore has a key role as the form creator - to detect features similar to those dental anomalies discussed, during images acquisition and form construction - to enable a decision to be made regarding the most appropriate professional to send the minimum data set form to for further examination.

After initial inspection of the minimum data-set form, supplementary information can be requested, and supplied by the lead PMCT processor. For example, if an area of trauma is indentified as needing further inspection-additional images detailing this region could be supplied upon request. With any examination, there is always a danger of missing something important, that at time of examination is considered irrelevant but becomes relevant. This risk is undoubtedly increased if the data is reduced to begin with into a minimum data set. However, the raw PMCT data can still be made available and revisited indefinitely, if required. So as long as the minimum data set contains enough information to detect these areas, it should prevent important information being missed.

The layout of the minimum data set anthropological recording form has been designed with the intention to facilitate future development, through the addition of more sections. A pathological recording form could be incorporated for example, which could include PMCT angiography [34] and ventilated PMCT post-mortem images [35-38]. Furthermore, the simple format and easy to replicate image extraction method has been specifically designed so that it is translational to an international DVI protocol. There is therefore scope in the future to build a PMCT database to encourage data sharing and collaborations between different research facilities. The minimum data set form would provide researchers with an overview of each case, which could be retrieved more rapidly than large quantities of raw PMCT data and provide sufficient information for a full anthropological examination, at least. Such a database would be a valuable teaching and learning resource for researchers in this field.

This novel approach to PMCT biological profiling is an important contribution to the forensic community, particularly forensic anthropology, radiology and the work being undertaken by the International Society of Forensic Radiology and Imaging (ISFRI) (http://www.isfri.org/), by providing a minimum standard of reporting and recording of osteological and odontological identification features [39]. This is an important step for the future development of this rapidly evolving professional body.

In summary, this study proposes a minimum image-set approach to biological profiling using PMCT images to create a specially designed two-page form. It can be easily replicated for use in any forensic or mass disaster situation, and promotes the pursuit of international quality control and best practice in the field of radiology and imaging worldwide. We do not provide a new method for anthropological or odontological assessment, but rather propose a process that allows a variety of accepted and regularly used methods of anthropological and odontological assessments to be applied rapidly and easily.

Key Points

1. There is good evidence showing that post-mortem computed tomography (PMCT) derived anthropological measurements are accurate

2. To date, their are no published guidelines regarding which images would be required for a full anthropological assessment

3. A minimum data set PMCT form for anthropological, biological profiling would ensure sufficient data was recorded for a comprehensive assessment and would facilitate data sharing between professionals by providing a structured format to record radiological evidence

4. The practical application of a minimum data set PMCT form has been illustrated in this publication, using juvenile age estimation as an example, by accurately estimating the age of 31 juveniles using only the PMCT minimum data set

5. 5 practitioners, from different professional backgrounds, with varying levels of experience, using different anthropological methods attained results that were in near perfect agreement with each other, and with actual documented age

6. It is suggested that the minimum data set form presented in this publication be used by all forensic practitioners in the future so that sufficient anthropological information is recorded from PMCT scans; and can be transferred rapidly to numerous professionals, worldwide, for assessment.

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Table 1. Methods used for Age Estimations

Age Group	Recommended approach	References
Fetuses	Dental Development Presence of ossification nuclei Long bone development	[17,18] [19,20] [21]
Newborns	Dental Development Diaphyseal length of long bones Presence of ossification nuclei Also check mineralisation of cuspid of first permanent molar and ossification of femur distal epiphysis	[17,18] [21] [20]
Infants and Children (0-7years)	Dental Development Diaphyseal length of long bones	[17,18] [21]
Juvenile and Adolescent (8-15years)	Dental Development (and Demirjian) Long bone development Maturation of hand and wrist (Greulich and Pyle, Tanner-Whitehouse)	[17,18] [22] [21,23] [24,25]
Transition and adult (16-25years)	Third molar development (Mincer) Fusion of spheno-occipital/basilar Synchondrosis, iliac crest and vertebral ring Maturation of hand and wrist (Greulich and Pyle, Tanner-Whitehouse)	[26] [27] [24, 25]

Table 2. Estimated and documented ages for the 31 cases, with tolerated margins of error based on previous studies [9, 10].

		Long Bones and Dentition		Estimated Age using Profiling Forms (Months)				
Case	Documented Age (months)	Tolerated margin of error (months)	Age range including error margin (months)	Observer 1	Observer 2	Observer 3	Observer 4	Observer 5
1	0.25	+/- 24	0 - 24	0.25	1	0.25	0.25	0.75
2	144	+/- 48	96 - 192	132	150	192	150	180
3	47	+/- 48	0 - 95	42	48	66	48	36
4	34	+/- 48	0 - 82	30	24	42	33	27
5*	36	+/- 48	0 - 84	42	39	1.5	42	39
6	2	+/- 24	0 - 26	1	2	8.5	2	1.5
7	16	+/- 48	0 - 64	18	18	10.5	18	21
8	108	+/- 48	60 - 156	108	108	114	108	129
9	10	+/- 24	0 - 34	6	8.5	8	8.5	15
10	10	+/- 24	0 - 34	6	8.5	10.5	10.5	7.5
11	1.75	+/- 48	0 - 26	6	6	5.5	6	2.3
12*	216	+/- 48	168 - 264	216	180	192	216	186
13	144	+/- 48	96 - 192	156	177	162	156	177
14	216	+/- 48	168 - 264	216	216	216	216	204
15*	216	+/- 48	168 - 264	216	216	216	216	186
16	1.5	+/- 24	0 - 25.5	1	1.5	0.25	1.5	1.5
17	96	+/- 48	48 - 144	90	96	78	96	90
18	7	+/- 24	0 - 31	12	9	6	9	9
19	120	+/- 48	72 - 156	114	108	78	114	120
20	60	+/- 48	12 - 108	48	48	66	48	45
21	72	+/- 48	24 - 120	54	72	66	66	63
22	1.5	+/- 24	0 - 25.5	0.25	1	0.5	1	1.5
23	24	+/- 48	0 - 72	24	24	30	24	27
24	60	+/- 48	12 - 108	54	45	66	54	57
25	1	+/- 24	0 - 25	0.25	1	4.5	1	0.75
26	0.3	+/- 24	0 - 24.3	0.25	0.25	0.3	0.25	0.75
27	5.75	+/- 48	0 - 30	6	4.5	3	6	4.5
28	1	+/- 24	0 - 25	0.25	1	1.5	0.25	0.5
29	156	+/- 48	108 - 204	138	159	173	156	141
30	192	+/- 48	144 - 240	168	183	173	173	150
31	132	+/- 48	84 - 180	90	84	124	124	90

***Cases with no dental images.**

Fig 1 Two page minimum data set form for case 9, 10 month old female. Remains that are disarticulated or have been scanned in several separate anatomical regions are first digitally reconstructed to produce full surface and skeletal 3D images (page 1 of profiling form). Specific regions are then isolated and captured (from multiple angles if required); and all long bones are measured, using a dedicated software application and included in captured image of each (page 2 of profiling form). The complete form can then be sent to numerous forensic practitioners simultaneously for examination

Fig 2 Age estimations based on MDCT using the profiling form Vs actual age at death. All age estimations are within the tolerated error margins (dotted lines), based on studies of the most frequently used age estimation methods [12,13]

Fig 3 Bland and Altman plot showing measurement errors for each observer, plotted against documented age-at-death for each case with 95% confidence intervals (dotted lines)

Figure 1

Figure 2

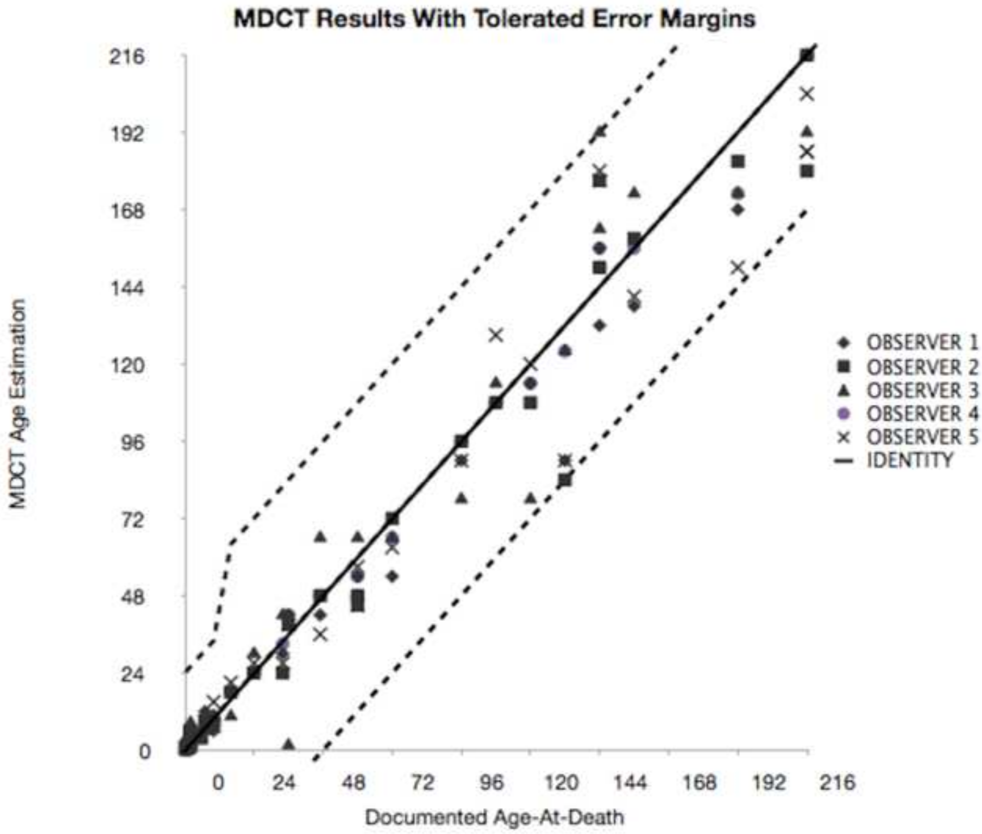
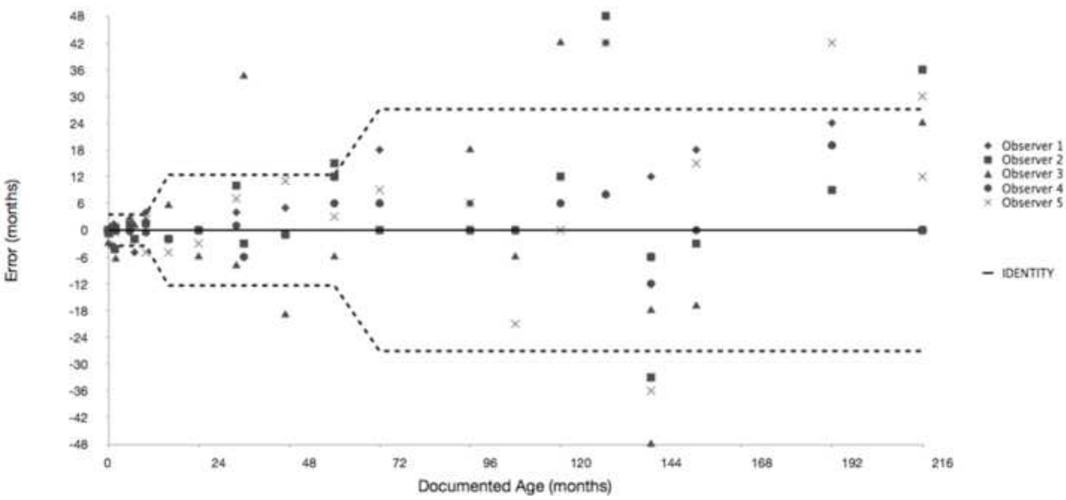


Figure 3



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