The use of permanent maxillary and mandibular canines in sex and age determination in a South African sample

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Declaration

I, Anja Ackermann, hereby declare that this research dissertation is my own work and has not been presented by me for any degree at this or any other University;

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Abstract

Dental anthropologists study the variation around the common shared patterns of teeth. These differences in the development, size and morphology of teeth are often used to help estimate the age and sex of unknown individuals. The aim of the study was two-fold. Firstly, it was determined whether sexually dimorphic characteristics exist in the size of permanent canines of South Africans, and whether these differences are of sufficient magnitude to make them usable as a method to determine sex from unknown remains. For this purpose the mesiodistal and buccolingual crown diameters and the maxillary/mandibular canine index were used. Secondly, the Lamendin technique of age estimation was tested and adapted to a South African sample. Therefore, this study aimed to assess the usability of human permanent canines in the determination of two demographic characteristics, namely sex and age, in a South African sample. A sample of known sex, age and population group was obtained from the Pretoria Bone Collection (University of Pretoria, South Africa) and the Raymond A. Dart Collection (University of Witwatersrand, Johannesburg, South Africa). The canines of 498 skulls were measured from four groups namely, black males, black females, white males and white females. The age of the sample ranged from 20 to 90 years. Using discriminant function analysis, it was possible to differentiate between the sexes with a relatively good accuracy of up to 87%. It was also evident that the two populations differed from one another as far as tooth size is concerned. Lamendin's method of age estimation yielded poor precision and accuracy. Periodontosis was better correlated with age than root transparency, where the highest R^2 value was 0.35. In summary it seems that the dimensions of the canine are useful in estimation of sex, should the population group be known. The Lamendin technique, however, gave relatively poor results even though new population specific formulae were created for the black and white populations of this sample. It could only estimate the age of the sample with an R^2 value of 0.41 and mean errors ranging from 12.02 to 15.76 years.

Key words: canines, mesiodistal, buccolingual, sex, age, Lamendin, discriminant, periodontosis, root height.

Abstrak

Tandheelkundige antropoloë bestudeer die variasie teenwoordig in die gemeenskaplike patroon van tande. Hierdie verskille in ontwikkeling, grootte en morfologie van tande word dikwels in die bepaling van geslag en ouderdom van onbekende individue gebruik. Die doel van hierdie studie is tweevoudig. Eerstens was daar bepaal of seksueel dimorfiese eienskappe in die grootte van permanente oogtande van Suid-Afrikaners bestaan en of hierdie verskille groot genoeg is om bruikbaar te wees as ‘n metode van geslagbepaling in onbekende individue. Vir hierdie doel was die mesiodistale - en buccolinguale kroonafmetings en die maksillêre/mandibulêre- oogtandindekse gebruik. Tweedens was die Lamendin-metode vir ouderdomsbepaling getoets en aangepas tot die Suid-Afrikaanse-studiegroep. Hierdie studie is dus gemik op die assessering van die bruikbaarheid van menslike permanente oogtande in die bepaling van twee demografiese eienskappe naamlik geslag en ouderdom, in ‘n Suid-Afrikaanse-studiegroep. Hierdie studiegroep van bekende geslags-, ouderdoms- en populasiegroep, is verkry vanaf die Pretoria-Beenversameling (Universiteit van Pretoria, Suid-Afrika) en die Raymond A. Dart-Versameling (Universiteit van Witwatersrand, Johannesburg, Suid-Afrika). Die oogtande van 498 skedels van vier populasiegroepe, naamlik swart mans, swart vrouens, wit mans en wit vrouens is gemeet. Die ouderdom van die studiegroep het gestrek van 20 tot 90 jaar. Met die gebruik van diskriminante funksie-analises was dit moontlik om tussen die geslagte met ‘n redelike akkuraatheid van 87% te differensieer. Dit was duidelik dat die twee populasiegroepe van mekaar verskil wat tandgrootte betref. Die Lamendin-metode van ouderdomsbepaling het swak presisie en akkuraatheid opgelewer. Periodontose het beter gekorreleer met ouderdom as wortel-deursigtigheid, met ‘n hoogste R^2-waarde van 0.35. Oorsigtelik blyk dit dat die dimensies van die oogtand bruikbaar is in die bepaling van geslag, mits die populasiegroep bekend is. Die Lamendin-metode, in teenstelling, het gelei tot relatief swak resultate al was daar populasie-spesifieke-formules ontwerp vir die swart- en wit-populasies van die studiegroep. Dit was slegs moontlik om die ouderdom te bepaal met ‘n R^2-waarde van 0.41 en gemiddelde fout van 12.02 tot 15.76 jaar.

Kernwoorde: oogtande, mesiodistale, buccolinguale, geslag, ouderdom, Lamendin, diskriminante, periodontose, wortel-deursigtigheid.

Acknowledgements

Lord, Thou hast given so much to me,
One thing more I ask: a grateful heart.
Not thankful when it pleases me,
As if Thy blessings had spare days;
But such a heart whose every pulse may be:
Thy praise.

George Herbert

I would like to thank my mother, who is my mentor, best friend and confidant; for your input, understanding and support throughout my studies and especially my masters. You've been my rock!!

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Table of Contents

List of Figures

List of Tables

Chapter 1: Introduction

The field of physical anthropology focuses on human biological variation through time and space. The role of physical or biological anthropologists is to study human biological characteristics at population level. When it comes to forensic cases concerning skeletal remains, medico-legal investigators can use forensic anthropologists for their expertise with human bones. The traditional focus of forensic anthropology is the assessment of each aspect of human skeletal material in a medico-legal context, for establishing a biological profile (Figure 1.1) (Eckert, 1997). This entails establishing the demographic characteristics, such as age and sex, of the human remains (Jacobson, 1982; Sengupta *et al.*, 1999).

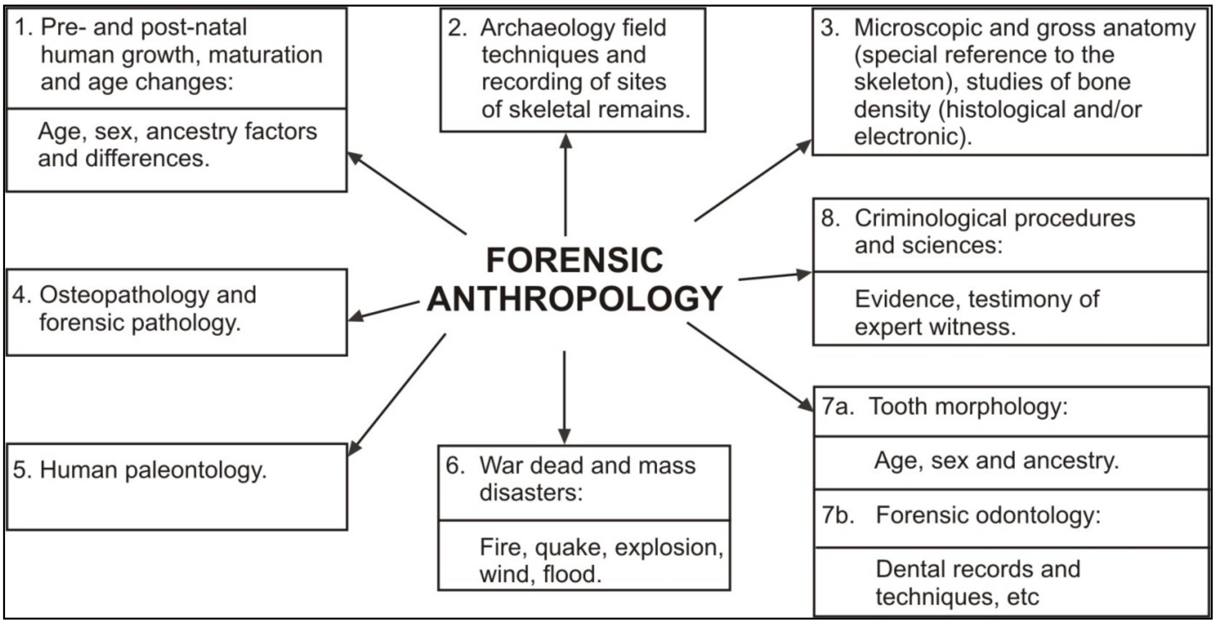


Figure 1.1. The scope of physical anthropology (modified from Krogman and İşcan, 1986).

Dirkmaat *et al.* (2008) added a new perspective on the role of forensic anthropology as the development of DNA analysis techniques modified the classic role of forensic anthropology as a field almost exclusively focused on victim identification. The new questions asked of forensic anthropologists stretch beyond identity and require sound scientific bases which expanded the scope of the field. This required the development of interrelated fields of taphonomy, forensic archaeology, and forensic trauma analysis from the start. These fields are concerned with the reconstruction of events surrounding death. According to Dirkmaat *et al.* (2008) these fields, especially forensic taphonomy, do not only represent an addition of new methodological techniques, but also provide forensic anthropology with a new conceptual framework. This framework is broader, deeper, and more solidly based in the natural sciences. As stated, this new framework represents a true paradigm shift which modifies the way in which classic forensic anthropological questions are answered. It also changes the goals and tasks of forensic anthropologists and their perception of what can be considered a legitimate question or problem to be answered within the field (Dirkmaat *et al.*, 2008).

For most people it will be easy to separate a group of normal humans by sex, age and population group as recognition of morphological characteristics of these categories are considered general knowledge. These morphological characteristics, however, become much more difficult to assess when dealing with defleshed skeletons (Eckert, 1997). Krogman and İşcan (1986) stated that all skeletal analyses should begin with the “big four: stature, age at time of death, sex and race” (p. 8). Thereafter the “accessory information” which include the “weight/body build, duration of internment, cause of death, pathologies, facial reconstruction” etc., should be assessed (Krogman and İşcan, 1986, p. 8). Each of the “big four” characteristics narrows the pool of possible matches to some degree – sex determination alone cuts it by half (Eckert, 1997; Kaushal *et al.,* 2003; Acharya and Mainali, 2009). According to Eckert (1997), these various characteristics can be determined with great accuracy in a complete and undamaged skeleton. Different methods including morphological and metric assessments are implemented to analyze skeletal remains in the identification process of individuals. Teeth are notably the most durable parts of the body which can best withstand the destructive processes that may occur after death and therefore form part of the skeletal remains used for the purpose of identification of individuals (Jacobson, 1982). By using the latest techniques (e.g., pubis, craniofacial morphology and postcranial measurements, DNA analysis, microscopic examinations of teeth) as well as the availability of a complete skeleton, some researchers stated that sex can be determined with near complete certainty (96-100%), age estimated to range within five years and stature approximated with a standard deviation of 3.5 centimetres. The assignment of a population group – Caucasoid, Mongoloid, or Negroid – can reportedly be done with a high degree of certainty in the absence of admixture (Eckert, 1997; Acharya and Mainali, 2009) by looking at various features of the skull as well as different characteristics of the teeth (e.g., Carabelli’s cusp, shovel-shaped incisors, and multi-cusped premolars) (Kaushal *et al*, 2003; Acharya and Mainali, 2009).

Teeth present as one of the most valuable sources of evidence in understanding the biology of ancient communities (archaeology), the course of evolution (fossil studies) and the identification of an individual from their fragmentary remains (forensic cases) (Hillson, 1996). According to Hillson (1996), dental anthropology may be defined as the study of people (and their close relatives) from the evidence that is provided by teeth. This may be seen as a field on its own due to the many contributors from various fields of study which include dentistry, genetics, anatomy and palaeontology; and encompasses a broad range of subjects which in turn have finer levels of specialization.

Through the years studies focused on the developmental aspects of dentition (from tooth germ formation to developmental defects of the crown) (e.g., Cadien, 1972; Kieser, 1990), post-eruptive changes (ordinary crown wear, culturally-prescribed dental modification) (e.g., Bailit, 1975), study of dental pathologies (caries, periapical osteitis, tooth loss patterns, periodontal disease) (e.g., Scott and Turner, 1988; Scott, 1991) and tooth size and morphology (study of elements of human dentition that have an underlying genetic basis) (e.g., Townsend *et al.*, 1994). Teeth have distinct anatomy and physiology which make them unique in the sense that they have their own shape and differ from the rest of the human skeleton. They are also unique amongst the resistant remaining parts of the archaeological and fossil remains which have been exposed to the surface throughout time, since teeth are the hardest substance in the body. The subject of dental morphology and its variations were recorded in literature as early as the time of Aristotle, when he erroneously stated that there are a greater number of teeth in females than in males (Kelley and Larsen, 1991). Observations of teeth, which include numbers of missing teeth, pathology (carious lesions) and periodontal and bone supporting conditions, became more sophisticated and useful as time passed since more knowledge and techniques were obtained (Kelley and Larsen, 1991). As Jacobson (1982) stated, “The value of studying dentition in the light of racial and population differences originates from the generally accepted theoretical consideration that many morphological characteristics of the teeth, such as cusp size and numbers, form and groove patterns, are genetically determined” (p. 1). When comparing these characteristics, it allows reasonable accurate conclusions to be drawn with regard to affinities among the different populations (Jacobson, 1982). Due to teeth’s durability and unique features, dental anthropology can also be used in the living using much of the same techniques employed for ancient remains (Jacobson, 1982; Krogman and İşcan, 1986; Hillson, 1996; Scott and Turner, 1997; Katzenberg and Saunders, 2000).

The human skeleton and dentition with its sexual variation is of great concern for anthropologists and odontologists since the assessment of the variation in dental size gives a clue as to the behaviour of a population as well as the differences between the sexes (Ateş *et al.*, 2006). Sex estimation from human remains forms a fundamental part of forensic medicine and anthropology, especially in cases such as criminal investigations, missing person’s identification and reconstruction of the lives of ancient populations (Vodanović *et al.*, 2006). Although there have been many studies on the sexual dimorphism and its application to human identification in the postcranial skeleton, there are frequent cases where the anthropologist is confronted by isolated teeth and should estimate the sex using teeth only (Scott and Turner, 1997; Katzenberg and Saunders, 2000; Ateş *et al.*, 2006).

Sex estimation is one of the demographic features that is needed to construct a profile of the skeletal remains. Methods in obtaining this feature are based on two primary biological differences between males and females: size and architecture. Generally males are larger than females. The size difference between sexes has been stated to be 8%, meaning females are on average approximately 92% the size of males (Krogman and İşcan, 1986). Therefore, male skeletons are more robust, wider, taller and more rugged than those of females. Architecturally there exist differences that can be used to distinguish between the sexes. For example, the female pelvis accommodates the process of birth. These structures are wider than those of males. Therefore sexing can be done on human bones by knowing how to interpret size and architectural differences (Krogman and İşcan, 1986).

Age estimation is done in different ways depending on the remains present. In children it is easier to estimate the age with very good accuracy. Tooth development and the comparison with developmental charts assist in the estimation of the age. This has accuracy within approximately 1.5 years. Ubelaker, amongst others, developed a chart which graphically illustrates the dental development from five months in utero to 35 years (Prince and Ubelaker, 2002). This chart includes the deciduous, mixed and permanent dentitions (Pretty and Sweet, 2001). As reported by Pretty and Sweet (2001), when the ages of sub-adults are estimated, it should be noted that the eruption times of teeth are highly variable and that the actual developmental stages are more accurate. Other methods of age estimation in juveniles include development of cranial bones and long bone lengths (Hoffman, 1979), and epiphyseal closure (Pyle and Hoerr, 1955; Greulich and Pyle, 1959; Krogman and İşcan, 1986; Albert and Maples, 1995; Austin, 2001; Crowder and Austin, 2005).

The development of the third molar is used by some researchers (e.g., Hongwei *et al.*, 1991; Mincer *et al.*, 1993; Prieto *et al.*, 2005; Kasper *et al.*, 2009) to establish the age of young adults but doubts about the accuracy of this method were raised by some practitioners due to the variability of these teeth (Thorson and Hägg, 1991; Mincer *et al.*, 1993; Pretty and Sweet, 2001; Willerhausen *et al.*, 2001). For example, an accuracy of approximately 4 years is claimed by those using this method of age estimation.

Estimation of age in adults is more difficult. Methods include cranial suture closure (Meindl and Lovejoy, 1985), changes in sternal ends of ribs (İşcan *et al.*, 1984, 1985) and pubic symphyses (Todd, 1920) as well as dentition. Teeth can be used to obtain relatively good age estimations by looking at the shape, measurements and periodontal defects. Some features, such as periodontal disease, excessive tooth wear and multiple restorative procedures, may point to an older individual. These are highly variable age markers with an accuracy of approximately 10-12 years. Based on the high variability in age estimation with teeth, other researchers such as Gustafson (1950) focused on the histology of the teeth (including tooth wear) and obtained a standard error of approximately 3.63 years. Lamendin's technique estimated the age of a French population with a mean error between the actual and estimated age of approximately 10 years on the working sample and approximately 8.4 years on a control sample. The mean error on the estimation was 8.9 ± 2.2 years in comparison to Gustafson's 14.2 ± 3.4 years (Lamendin *et al.*, 1992). The Lamendin technique is recorded in literature as one of the methods to estimate the age of an individual. This method used single-rooted teeth (anterior teeth) and three measurements were taken namely, the root height, periodontosis and root transparency. Some odontologists make use of aspartic acid racemization and claim an accuracy of approximately 4 years. Other methods include SEM-EDXA (Scanning Electron Microscope with Energy Dispersive X-ray Analyzer to examine dentine in relation to age) and TCA (tooth cementum annulations) (Pretty and Sweet, 2001; Meinl *et al.*, 2008).

The forensic anthropologist does not very often deal with complete skeletons. Often the case consists of partial and fragmented remains. The anthropologist should therefore be prepared to get as much information as possible from each and every bone (El-Najjar and McWilliams, 1978; Eckert, 1997; Katzenberg and Saunders, 2000). It has also been shown repeatedly that each population needs its own population-specific standards since some studies have shown differences between population groups (e.g. Katzenberg and Saunders, 2000; Patriquin *et al.*, 2002; Patriquin *et al.*, 2005; Ateş *et al.*, 2006). New sex and age estimation methods are continuously developed and old methods are tested, fine-tuned and adapted for specific populations. Due to regional variation in dental size, it is essential that there are new studies to estimate sex and the rest of the biological profile which is important in forensic sciences as well as the identification of the human remains. Therefore teeth are of great importance since the individual tooth may present an opportunity to establish the sex or age on its own which is not always possible with the long bones or the cranial parts (Scott and Turner 1997; Katzenberg and Saunders 2000; Ateş *et al.* 2006).

In the current study an attempt will be made to apply and adapt certain methods of estimating sex and age using human teeth. As previous studies indicated that the canines may be one of the best teeth to use for this purpose (e.g., Sengupta *et al.*, 1998; Sarajlic *et al.*, 2006), this study will focus on canines only.

Purpose of study

This study will focus on the permanent canines, and its aim is two-fold. Firstly, an assessment will be made of the sexually dimorphic characteristics of the permanent canines in two South African populations, using the mesiodistal and buccolingual crown diameters and some indices. If significant size differences are found to exist, discriminant function formulae will be developed to estimate the sex of unknown individuals using dimensions of both the upper and lower canines.

The second aim of this study is to test the usability of the Lamendin technique of age estimation, also using canines, in a South African sample. If necessary, the data obtained will be used to adapt the Lamendin formulae to better fit the South African sample so that more accurate age estimates can be obtained.

In assessing the teeth to achieve the aims as explained above, it will also be investigated whether significant differences exist between the teeth of black and white South Africans, and whether it is necessary to use population-specific formulae to assess age and sex from canines in these two groups.

Chapter 2: Literature review

Dental evidence was used in identification profiles dating back as far as 2500 B.C. and possibly further. The science of forensic odontology is based on the facts that teeth, dental restorations, dental prostheses, maxillary sinus configuration, anatomical characteristics of the hard palate, bone trabeculae patterns, bony protuberances, cracks, crevices, and wrinkles in lips, anatomical landmarks, and overall oral and facial morphology exist as a vast number of relatively stable individual characteristics. Each of these features expresses variation in their anatomy as time passes (Woolridge, 1980).

Forensic anthropologists use as many methods as possible to assist law enforcement with the identification process. In cases where only skeletal-dental remains are available, characteristics such as age, sex, stature, population affinity, time since death, pathology or distinctive anatomical features and perimortem damages or changes should be estimated. When medical or dental records (radiographs) are available and obtained, they are used to determine the exact identity of the individual (individuation) (Scott and Turner, 1997). From a historical standpoint, human skeletal remains, as in the other areas of evolutionary biology, provide the only direct link to other past and living populations. The study of the morphology of the tooth has contributed to the resolution of a number of historical problems that have attracted anthropological interest for a long time (Scott and Turner, 1997).

Observations on teeth alone provide extensive information that is relevant to a number of study fields, such as zoology and human biology. The number of journal papers produced reflects the fascination of many scientists with dental dimensions (Kieser, 1990). The length and width of teeth are the most widely documented anthropometric features as it provides significant information on human biological problems such as “genetic relationships between populations and human environmental adaptation” (Kieser, 1990, p. 1). These two measurements can be used as standard in craniofacial dental procedures as well as a resource for answering questions pertaining to comparative anatomy and phylogeny or evolutionary studies. Teeth comprise of the hardest materials in nature, which preserve them to provide evidence of evolutionary changes over time. The high genetic component in their expression implies that they are less affected by environmental factors and can therefore be useful in the study of the establishment of biological relationships between various groups of human and nonhuman primates (El-Najjar and McWilliams, 1978; Kieser, 1990).

Since teeth and bones are very durable, they are the last structures of the human body to disintegrate after death. They provide a lot of evidence for the anthropologist, archaeologist or forensic expert (Kieser, 1990). Therefore dental characteristics, including features such as tooth wear, oral pathology, enamel hypoplasia and other microscopic changes in the tooth structure, give the scientist information which can be used to reconstruct much of the individual’s lifestyle. Generally teeth are used for corroboration rather than diagnostic purposes, such as in the estimation of sex, age and population affinity. There are various ways in which the demographic profile of an individual can be estimated using the dentition. To name a few, the forensic anthropologist uses tooth calcification and eruption sequence as age estimation traits and the morphology of the tooth as a possible sex-size difference. Differences between populations are seen, for example, in the shovel-shaped upper central and lateral incisors that are present in Asian populations, and the lower first permanent molars with five cusps, as well as the Y-shaped groove pattern which are usually found in black populations rather than in the white populations (Krogman and İşcan, 1986). Dental anthropologists focus on the variation found around the common shared patterns of teeth. These patterns are expressed as differences in the tooth size and morphology (Scott and Turner, 1997).

The development of human dentition begins very early in gestation and ends in the third decade of life (Katzenberg and Saunders, 2000). The final shape and size of the tooth crown are determined well before its eruption into the oral cavity (Kieser, 1990). For this reason, any insults that the body and/or dentition have received may be stored permanently in the teeth. Bone, on the other hand, has the potential to remodel during life and therefore any sign of previous insults may be obliterated (Katzenberg and Saunders, 2000). The development of the dental tissues has the ability to withstand endocrine diseases or nutritional variations that other tissues cannot. Therefore when the tooth is fully mineralized and erupted it is a very stable entity. Changes such as developmental and regressive alterations to the tooth are stated to be related to the chronological age of the individual. This makes teeth the most suitable material to be used in identity profiles for example in estimating age (Kaushal *et al.*, 2003 and 2004; Reppien *et al.*, 2006). So the only change in teeth that is possible is the removal or loss of the teeth either through wear, pathology or trauma. Teeth are also the only hard tissue in the body that can be observed directly, i.e. without the help of radiographic or other non-invasive (intra-oral) intervention. By making casts, it provides the physical anthropologist with accurate, permanent and easily obtainable records of the dentitions of populations or individuals. Studies using metric data derived from such collections, contributed to the better understanding of human variation (Kieser, 1990; Scott and Turner, 1997).

To be able to reconstruct the population affinities of groups under examination is important since there is evidence of a genetic basis to much of the variation observed in the tooth size and shape. When the genetics of tooth size and shape are better understood, it will be possible to more accurately determine the affinities of individuals and/or groups (Katzenberg and Saunders, 2000). In the field of Physical Anthropology, tooth size and shape in human populations are of importance and the differences which exist vary within and between populations. There have been numerous studies done which include those of Otuyemi and Noar (1996) and Brooke *et al.* (2009) that have shown that significant differences exist between ancestral groups with regard to tooth size, particularly the mesiodistal and buccolingual crown diameters. It was also stated that apart from the population affinity differences there are other factors contributing to tooth size variability namely sex, hereditary factors, bilateral differences, environmental and secular changes (Otuyemi and Noar, 1996; Kaushal *et al.*, 2003; Hemanth *et al.*, 2008; Brooke *et al.,* 2009). Therefore there is a need to create population specific standards for sex and age determination (Otuyemi and Noar, 1996; Vodanović *et al.*, 2006, Hemanth *et al.*, 2008).

Since the adult human dentition comprises of a complement of 32 teeth, there will at least be a few teeth that are recovered and used in the identification of human remains (Kaushal *et al.*, 2003; Acharya and Mainali, 2009). The canines in particular have been used in a number of studies (Jacobson, 1982; Hillson, 1996; Kaushal *et al.*, 2003 and 2004); therefore the information discussed below will be focused on the human canines.

2.1. Dental anatomy

2.1.1. General dental anatomy

Dental anatomy can be defined as the study of the form and structure of teeth. This also includes topics such as mensuration (the study, act or process of measuring geometric magnitudes such as length; Latin *mensura* = measure), tooth wear, as well as pathology, which have an effect on the shape and size of teeth. All these features contribute to the "look" of the teeth and are used to describe the dentition and much of the lifestyle of populations or individuals (skeletal or living) (Katzenberg and Saunders, 2000).

Tooth morphology is primarily concerned with normative tooth form. The human dental formula of 2:1:2:3 which is shared by all catarrhine primates (Old World monkeys, apes, and humans), represents different types of teeth in each quadrant of the upper and lower jaws. Each person has two sets of dentition. The deciduous dentition or milk teeth are half-formed by birth and then erupts into the oral cavity during the following two years.

The permanent dentition (Figure 2.1) gradually displaces the deciduous dentition. It starts with the first tooth being formed just before birth and ends when the last tooth erupts (usually the third molar) in the late teens. There are four quadrants (Figure 2.2) in each set of dentition: upper left, upper right, lower left, and lower right. The left and right quadrants are separated by the midline of the skull (mid-saggital plane) creating mirror imagery.

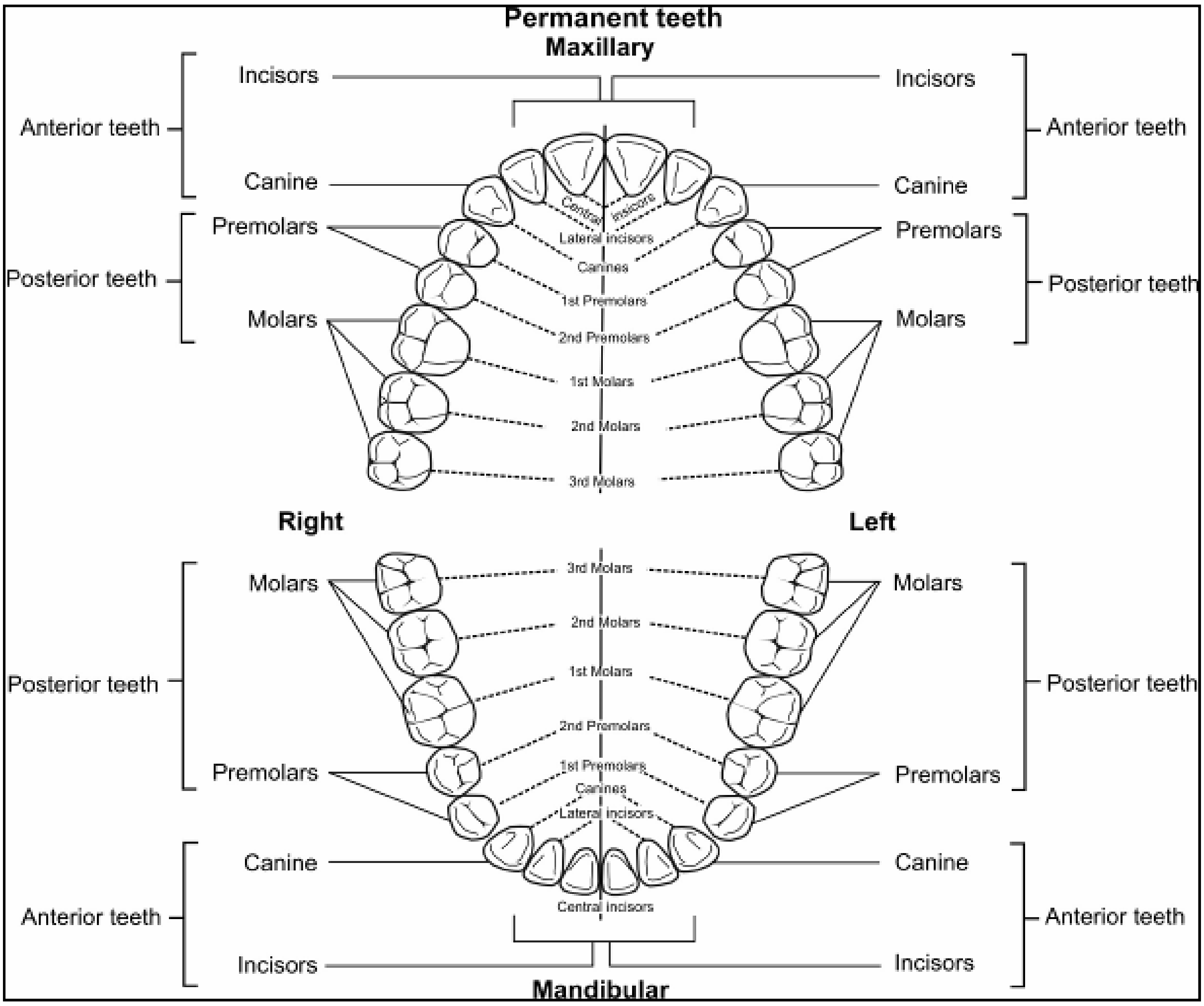


Figure 2.1. The maxillary and mandibular permanent dentition viewed from the occlusal (chewing) surfaces (modified from Scheid, 2007).

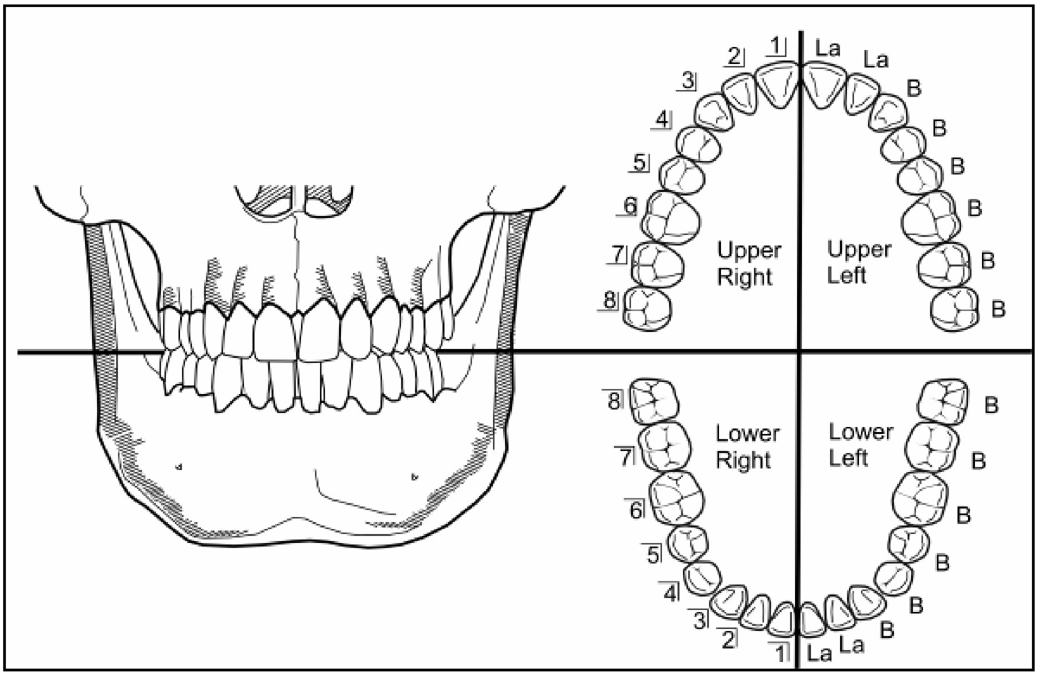


Figure 2.2. The four quadrants of the permanent dentition namely, upper left, upper right, lower left and lower right. La = labial surface, B = buccal surface (modified from Hillson, 1996).

Each quadrant has different classes of teeth (Figure 2.1): incisors, canines, premolars and molars. The incisors and canines are often termed together as anterior teeth, whereas the premolars and molars are called posterior or cheek teeth. Each quadrant of the permanent dentition has two incisors, one canine, two premolars and three molars, whereas the deciduous dentition comprises of two incisors, one canine and two cheek teeth (molars). These deciduous cheek teeth are normally called deciduous molars but some believe that the proper term is deciduous premolars (Hillson, 1996; Scott and Turner, 1997). Although the same tooth in the different jaws differ in size and form, one can characterize the incisors as spatulate and single-cusped, canines as single-cusped and conical, the premolars as bicuspids (two cusps) with three roots and the molars as multi-cusped with two roots. This is the basic blueprint for human dentition but one should be aware of the variety of morphological structures that may be present in some instances (Scott and Turner, 1997).

Each tooth is divided into two compartments, namely the crown and the root (Figure 2.3). The crown is defined as the part of the tooth that projects into the oral cavity and is covered with enamel. This is the hardest biological tissue consisting of 97% inorganic material, made up of hydroxy-apatite crystallites (a calcium phosphate). Since calcium hydroxy-apatite is extremely durable, the teeth are in excellent preservation in most taphonomic contexts. For example, teeth are often the best represented remains in hominid fossil localities and recent archaeological sites. In some of these cases it is not uncommon to find isolated teeth when the rest of the skeleton has already disintegrated (Scott and Turner, 1997). The crowns of the teeth consist of smaller features or structures, namely cusps which are augmented by regularly occurring occlusal and marginal ridges, and grooves or fissures. These grooves or fissures are stated to be of varying depths dividing the tooth into constituent (its different parts) cuspal and ridge components. The root is the part that is embedded in the jaws and is coated with a thin layer of cement. Dentine makes up the core or body of the tooth (Scott and Turner, 1997). The enamel-dentine junction (EDJ), cement-dentine junction (CDJ) and the cement-enamel junction (CEJ) (Figure 2.3) form the boundaries between the abovementioned tissues.

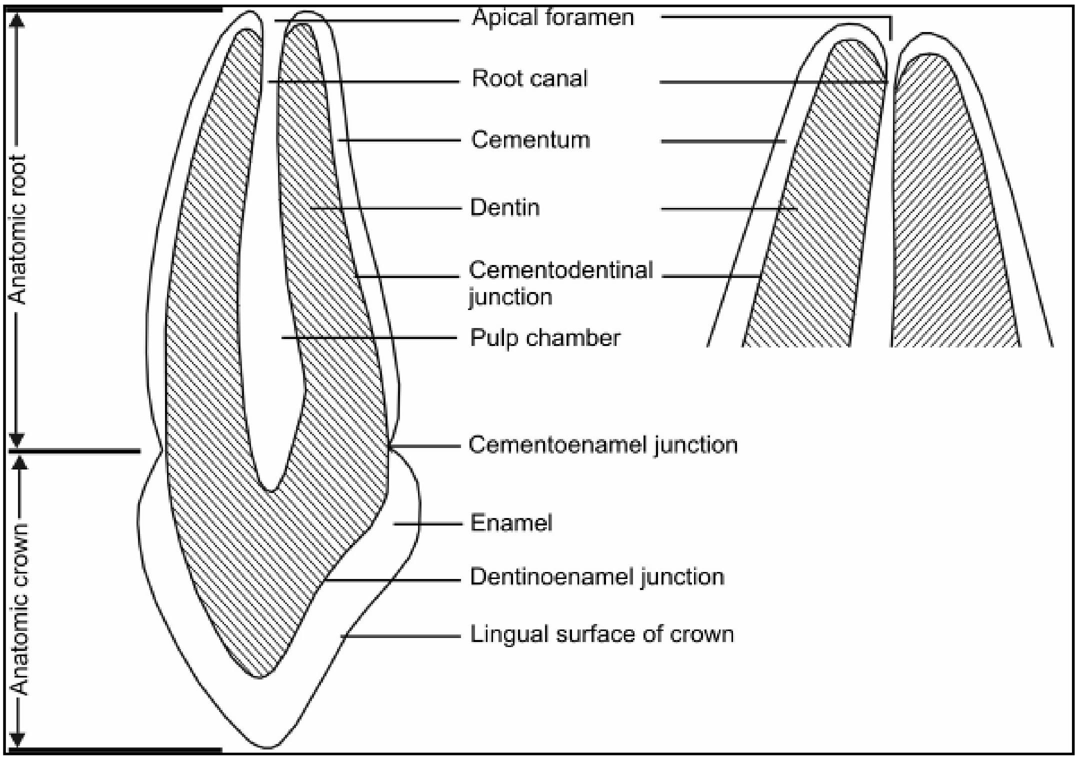


Figure 2.3. Longitudinally sectioned maxillary canine to illustrate distribution of dental tissues and the shape of the pulp cavity which is made up of pulp chamber and root canal. On the right side is a close-up of the apical portion showing the constriction of the root canal near the apical foramen (modified from Scheid, 2007).

The cervix or neck is defined as the meeting point between the crown and the root of the tooth, whereas the cervical margin is the base of the crown (Figure 2.4).

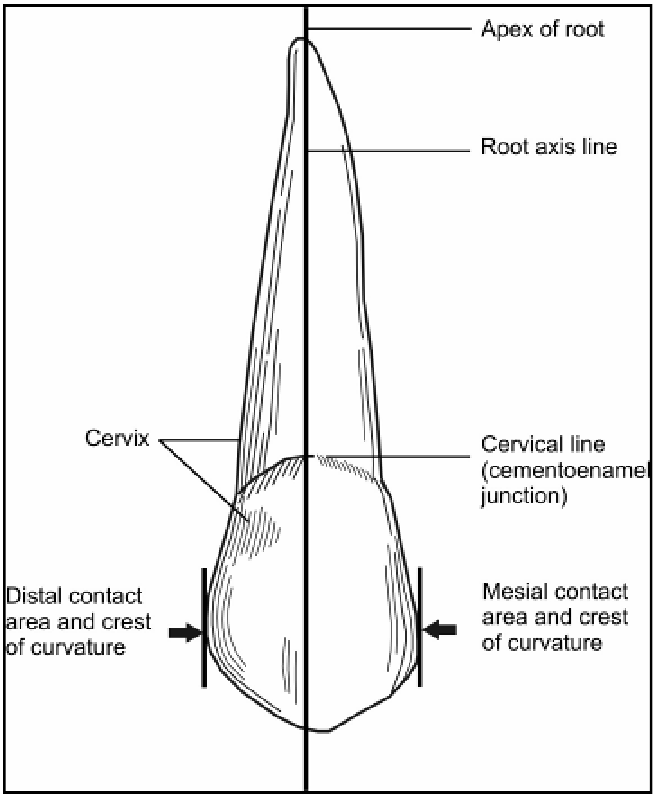


Figure 2.4. Labial surface of the maxillary right canine, showing the mesial and distal contact areas (modified from Scheid, 2007).

The cingulum is a broad bulge on the lingual surface of teeth that girdles the cervical one-third of the crown. The pulp chamber is found inside the tooth and it contains the soft tissue of the pulp and receives blood vessels and neurological supports through the opening of the root. This chamber has conical hollows or horns in its roof and its floor opens into one or more root canals (Figure 2.5).

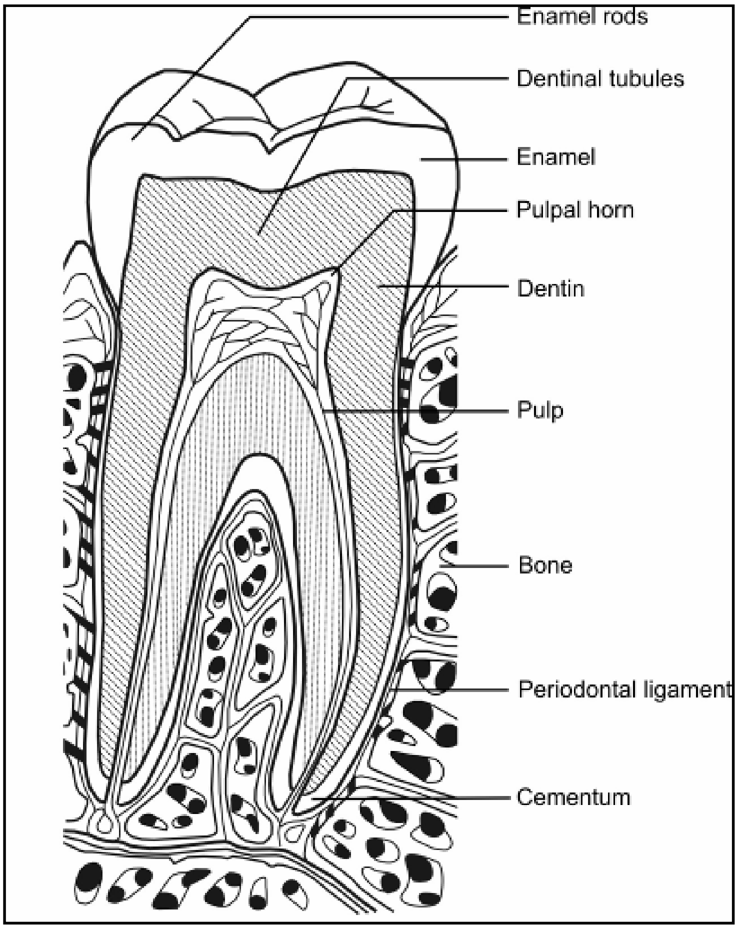
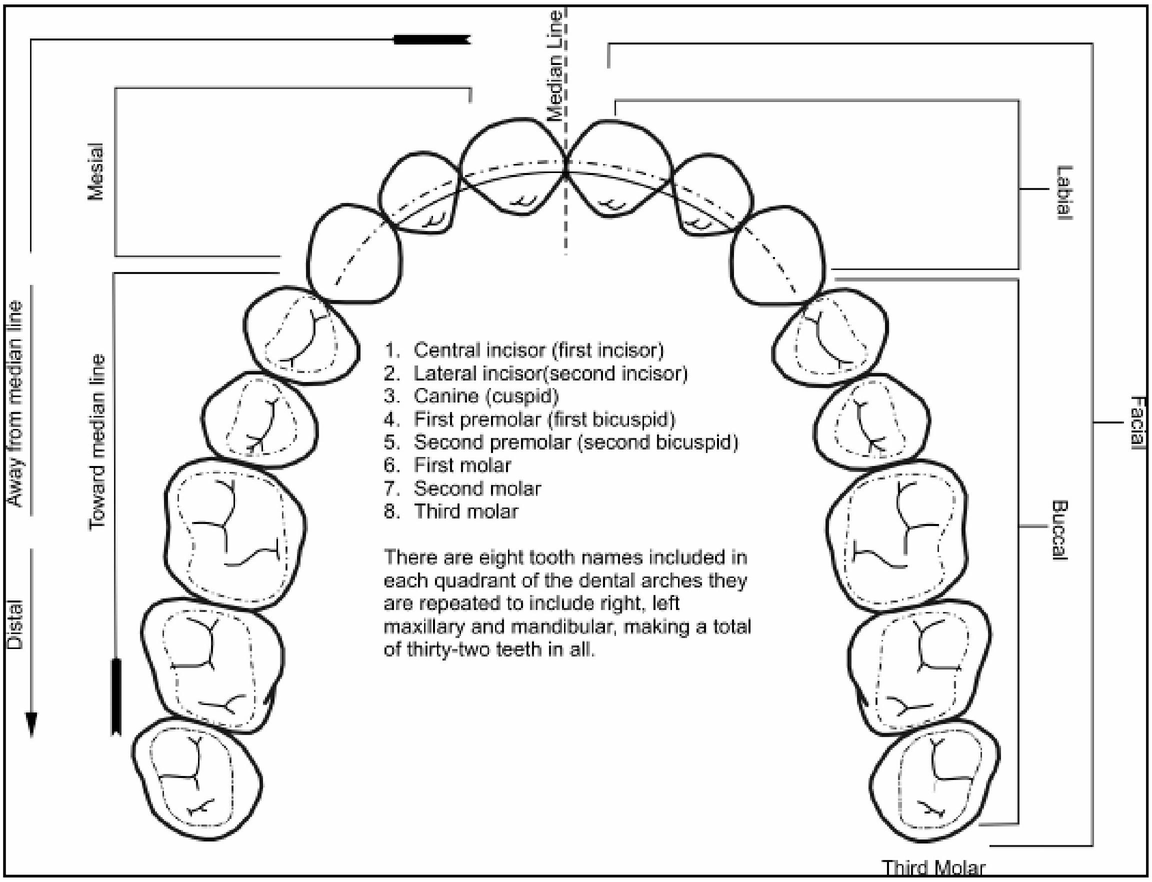


Figure 2.5. The anatomy of a tooth and its surrounding tissue (modified from Krogman and İşcan, 1986).

A tooth may also have more than one root, each with a root canal. The point at which the roots divide is called the root fork or furcation (Krogman and İşcan, 1986; Hillson, 1996; Katzenberg and Saunders, 2000). The alveolar bone and periodontal ligament help to keep the tooth in place (El-Najjar and McWilliams, 1978). Each tooth displays the following surfaces (Figure 2.6) (Hillson, 1996; Katzenberg and Saunders, 2000):

- Occlusal aspect (*facies occlusalis*/closed up face): It is the aspect of the crown that faces the teeth in the opposing jaw when the mouth closes. The molars and premolars have broad crown surfaces that meet when the jaws shut; therefore it can be called the occlusal surfaces. The incisors and canines are tall and spatulate with high crowns that do not meet edge-to-edge but rather overlap. Therefore, it is better to call the occlusal surface of the crown of these teeth the incisal edge (*margo incisalis*).
- Apical aspect: This is the opposite of the occlusal aspect. It describes the tips of the roots (apex).

- Mesial (Greek: *mesos* - middle) surface: This surface faces along the dental arcade towards the median saggital plane.
- Distal surface: This surface faces along the arcade away from the median saggital plane.
- Approximal surfaces: When looking at two neighbouring teeth of the same jaw, their adjoining sides are called approximal surfaces.
- Lingual (*facies lingualis*/tongue face) surface: It is the part of the tooth that faces the tongue. In the upper jaw, where it faces the palate, this surface is called the palatal surface.
- Buccal (*facies buccalis*/cheek face), Labial (*facies labialis*/lip face) or Vestibular (space between the teeth, lips and cheeks) surface: This is the surface that faces outside the dental arcade, towards the cheeks and lips. The term “labial” is used for the incisors. Some authors also use this term for the canines. “Buccal” is generally used for the canines, premolars and molars, whereas the term “vestibular” accounts for all teeth.



1. Central incisor (first incisor)
2. Lateral incisor(second incisor)
3. Canine (cuspid)
4. First premolar (first bicuspid)
5. Second premolar (second bicuspid)
6. First molar
7. Second molar
8. Third molar

There are eight tooth names included in each quadrant of the dental arches they are repeated to include right, left maxillary and mandibular, making a total of thirty-two teeth in all.

Figure 2.6. Application of the nomenclature. The tooth surfaces related to the tongue is called lingual, cheek is buccal, lips is labial, and face is facial. This applies to all four

quadrants. Teeth and their surfaces may also be described as being away from the midline (distal) or toward the midline (mesial) (modified from Ash and Nelson, 2003).

2.1.2. Specific canine features

The crowns of the permanent canines are broadly spatulate with a single main, central cusp. This cusp has ridges running down the incisal edge to the mesial and distal sides where the mesial ridge is usually shorter than the distal. A prominent buttress runs down the lingual surface joining the cingulum bulge of the tuberculum (Figure 2.7) (Hillson, 1996).

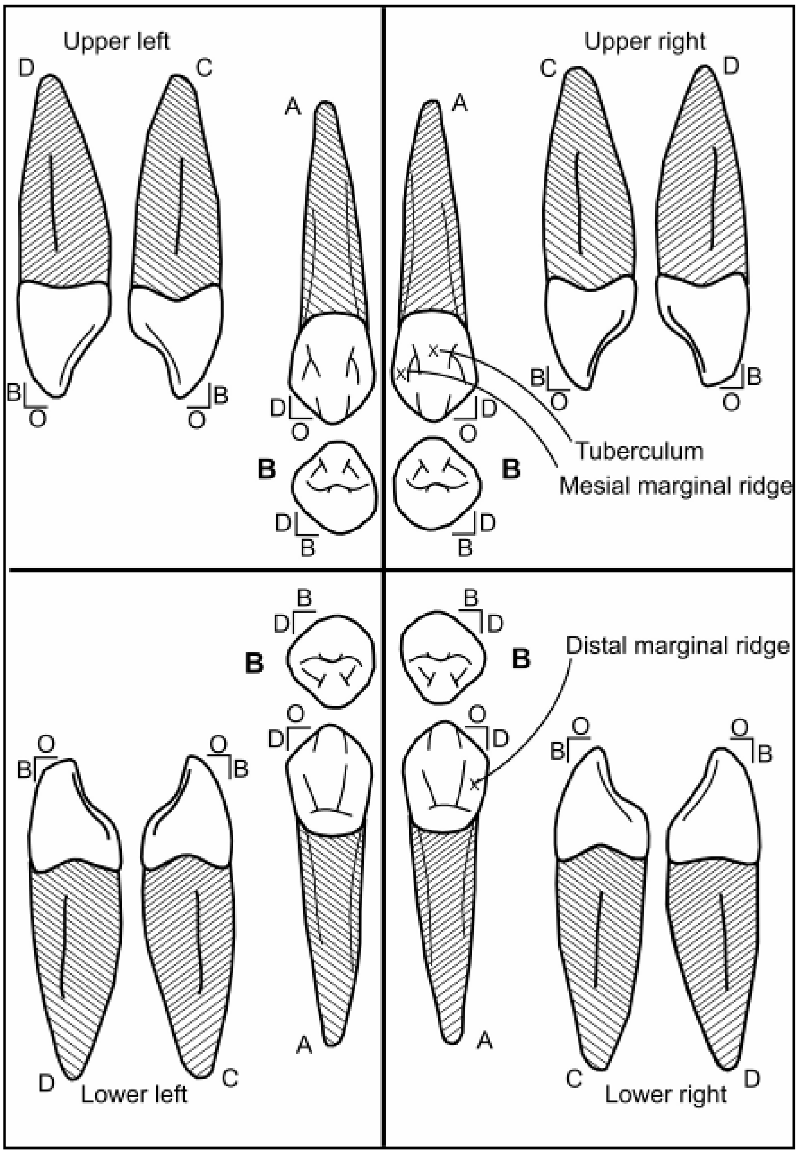


Figure 2.7. The permanent upper and lower, left and right canines in the four quadrants. A: lingual aspect, B: incisal aspect, C: mesial aspect, D: distal aspect (modified from Hillson, 1996).

The marginal ridges, which are outlined by grooves, are prominently developed on the lingual surface. The mesial marginal ridge is bulkier and more vertically arranged than the distal, and it reaches slightly higher to the occlusal surface. Therefore, the crowns of most canines are asymmetrical in the buccal or lingual outline and have a more vertical mesial and a more bulging distal side. All canines present with a cervical margin which curves down smoothly to the apex on the buccal and lingual sides and up to the occlusal surface on the mesial and distal sides. Here the mesial curve is also more prominent (Hillson, 1996).

Permanent canines usually have a long root with broad, shallow grooves running down the mesial and distal sides and an apical third which curves distally (quite variable). The pulp chamber is found inside the cervix with a large diverticle which corresponds to the main cusp and it grades into the root canal. There is usually one large canal inside the root, but at times it is possible to find a canine where the canal divided within the root into two canals (buccal and lingual canals) (Hillson, 1996).

The wear of a canine starts at the tip of the main cusp of the canine where a dot of dentine is exposed (Figure 2.8). Thereafter the mesial and/or distal ridges of the incisal edge start to wear and a line of dentine is exposed. Further wear leads to the main labial bulge of the central cusp element to form into a worn facet. Together with the lingual buttress and marginal ridges, these progress to form an area of dentine with an outline. When the wear approaches the cervix of the tooth, the dentine area becomes a diamond-shaped worn area which is bounded by a higher rim of enamel. At the final stages of wear, this rim is breached as the wear progresses down the root. Approximal wear is found at the contact points of the canines on the most prominent mesial and distal bulges of the crown sides. This type of wear proceeds slowly at these points, but eventually forms facets (Hillson, 1996).

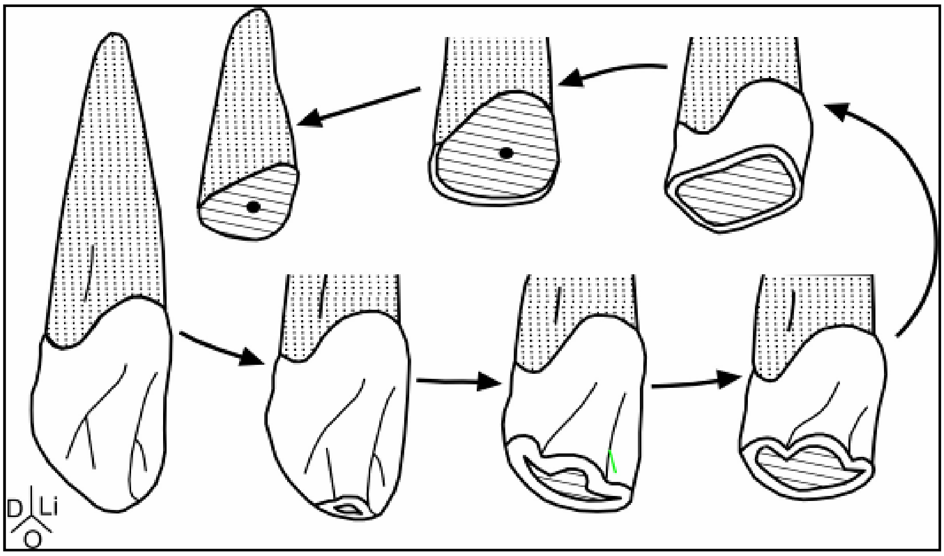


Figure 2.8. The stages of wear in the canine. The permanent upper left canine is viewed from the distal-lingual-incisal corner (modified from Hillson, 1996).

2.2. Dental measurements

2.2.1. Introduction

Dental measurements in the assessment of size form an integral part of anthropology for studying sexual dimorphism, trends toward tooth and jaw size reduction in Late Pleistocene/Early Holocene humans, and differences between past and present human populations (Hillson *et al.*, 2005). The uniqueness of teeth being comparable between skeletal samples and living populations, lies in the fact that casts of the dentition of living individuals can be used to measure teeth accurately. This can also be done for archaeological specimens. By using standardized measurement techniques, it allows accurate comparisons of individual teeth as well as those of populations and various subgroups. Caution should still be taken since there is a wide range of variability in tooth size inter-populationally and intra-populationally (Katzenberg and Saunders, 2000). These authors stated that as years passed, various arguments were made which supported the use of measurements as sources of taxonomic information. The theoretical basis of metric analyses lies in the precision and repeatability of measurements, the conservative nature of the continuous variation, the direct link with the past as well as the demonstration of a heritability component for this category of biological variation. It is stated that when looking from a statistical and mathematical standpoint, the continuous and correlated nature of measurements makes them highly suited for the application of multivariate statistical procedures (Katzenberg and Saunders, 2000).

The mesiodistal and buccolingual crown diameters are routinely measured and these definitions were only established approximately a century ago, but reassessed by researchers such as Goose (1963); Tobias (1967); Kieser (1990) and Hillson (1996) to name but a few. Hillson *et al.* (2005) stated that the definitions given by Moorrees and Reed (1964) are the most widely followed. According to these authors, the mesiodistal diameter is defined as the largest mesial-to-distal dimension parallel to the occlusal surface, whereas the buccolingual diameter is taken as the greatest distance between the buccal/labial and lingual/palatal surfaces perpendicular to the mesiodistal measurement (Figure 2.9). Although the system of measurements centres on the axis of the mesiodistal crown diameter, the line of the axis itself is still not clearly defined. In 1963 Goose suggested that the mesiodistal diameter axis should be between the contact points (Figure 2.9) of the tooth with its neighbours in normal occlusion. When malocclusion is present, it is said that the positions on the crown at which the contact points would have been in normal occlusion are used instead. In the case of unworn incisors and canines the definitions of the mesiodistal crown diameter are the same (Hillson *et al.*, 2005).

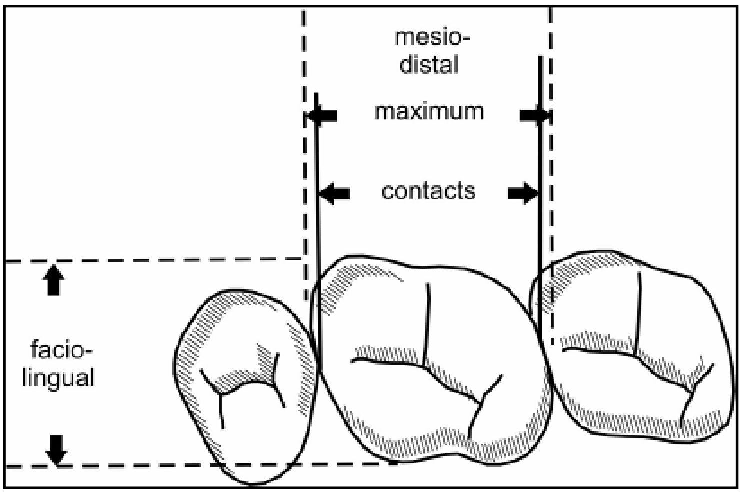


Figure 2.9. The dimensions which are included in the two principal methods of determining the mesiodistal diameters (contact points or maximum diameter). The buccolingual (faciolingual) measurement is obtained by holding the calliper beaks perpendicular to the mesiodistal measurement and parallel to the occlusal plane (modified from Katzenberg and Saunders, 2000).

As Hillson *et al.* (2005) stated, any change theoretically, in the axis of the mesiodistal crown diameter would lead to a change in the measurement axis of the buccolingual crown diameter. This buccolingual axis should be perpendicular to the mesiodistal crown diameter (Figure 2.10), but in no way could the angle of it be checked in practice.

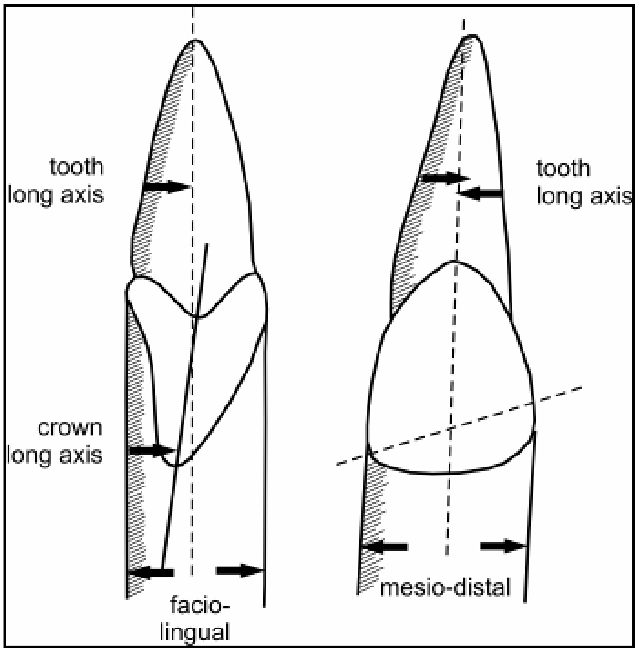


Figure 2.10. Landmarks used for measuring the anterior teeth. Note should be taken that the long axis of the crown may not be the same as the long axis of the tooth. The mesiodistal diameter is determined through the contact points and is perpendicular to the long axis of the tooth (modified from Katzenberg and Saunders, 2000).

Therefore, the easiest way of taking this measurement (buccolingual) on the incisors, canines and premolars is to find the maximum diameter from the buccal/labial to the lingual crown area. This is done by slightly rotating the tooth crown to get the maximum and by repeating the calliper readings. This will give an average diameter which may not be the actual perpendicular reading to the mesiodistal diameter or to the occlusal surface. Therefore, most observers consciously or unconsciously ’’bend’’ the rules of measurement to some extent (Hillson *et al.*, 2005). According to Hillson (1996) the measurements of the crown (mesiodistal and buccolingual) are part of the most valuable measurements for identification purposes. The vast majority of literature which include studies by Otuyemi and Noar (1996); Katzenberg and Saunders (2000); Muller *et al.* (2001); Hanihara and Ishida (2005); Hemanth *et al.* (2008); and Brooke *et al.* (2009) to name a few, indicated that these traditional measurements of teeth are still the most useful, since several indices were based on these to describe tooth crown proportions and basic shapes. This does not suggest that there are only two measurements; there are other methods that can be used for descriptive purposes such as diagonal crown measurements reported by Rai and Anand (2007). Teeth from archaeological and fossil origin are often heavily worn which limits the usefulness of dental crown diameters since these are usually defined as the widest points of the crown. Alternative measurements which are much less affected by wear were proposed and these include measurements at the cervix of the tooth (where the crown joins the root) and measurements along the diagonal axis. This will result in a wider range of specimens to be included into the sample as well as allow little-worn teeth of juveniles to be compared directly with well-worn adult teeth (Hillson *et al.*, 2005). Hillson *et al.* found that the cervical (Figure 2.12 and 2.13) and diagonal measurements are just as reliable as the usual crown dimensions. This was seen in the buccolingual cervical diameter, showing a strong correlation with the normal buccolingual crown diameter (all the teeth) as well as the mesiodistal cervical diameter which presented a high correlation with the normal mesiodistal crown diameter in the incisors and canines (less in the premolars and molars). They, and Rai and Anand (2007), concluded that although the usual maximum crown diameters have been used for a long time, the alternative and/or diagonal dental measurements are just as reliable, record similar information regarding crown size and would be the preferred way of measuring worn teeth from archaeological and fossil samples (Hillson *et al.*, 2005).

For the purposes of age estimation, a different set of measurements is used. Lamendin *et al.* (1992) defined three measurements on the tooth, namely periodontosis, root transparency and root length. Periodontosis (P) (gingival regression) is due to the soft tissue degeneration which surrounds the tooth and it progresses from the neck to the apex of the root. It is stated that it appears as a smooth yellowish area below the enamel which is darker but still lighter than the rest of the root. Tartric deposits are often found at this level. Therefore, this feature is measured on the labial surface as the maximum distance between the cementoenamel junction and the line of soft tissue attachment and is less susceptible to be influenced by pathologic factors (infections). Root transparency (RT) is a physiologic feature which is said to never appear before the age of 20 years, but it appears to become more common with advancing age. This is due to crystals of hydroxy-apatite deposits within the dentin tubuli. This transparency can be present on the entire root of the tooth and can be seen with the help of a light source like a negatoscope. It is measured as the maximum height from the apex of the root to the visible transparency seen within the root (Figure 2.11). This measurement is also taken on the labial surface, since transparency is usually the highest at this surface. Root height (RH) is the distance between the apex of the root and the cemento-enamel junction (Lamendin *et al.*, 1992).

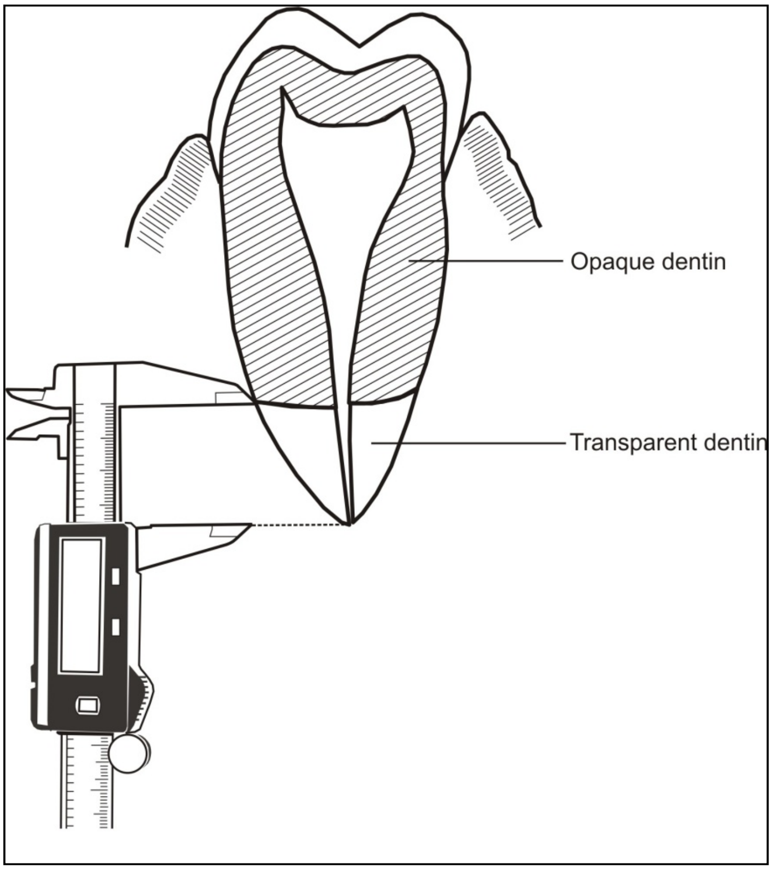


Figure 2.11. The measurement (in millimetres) of the transparent dentin carried out by sliding a calliper in front of a constant light source (modified from İşcan, 1989).

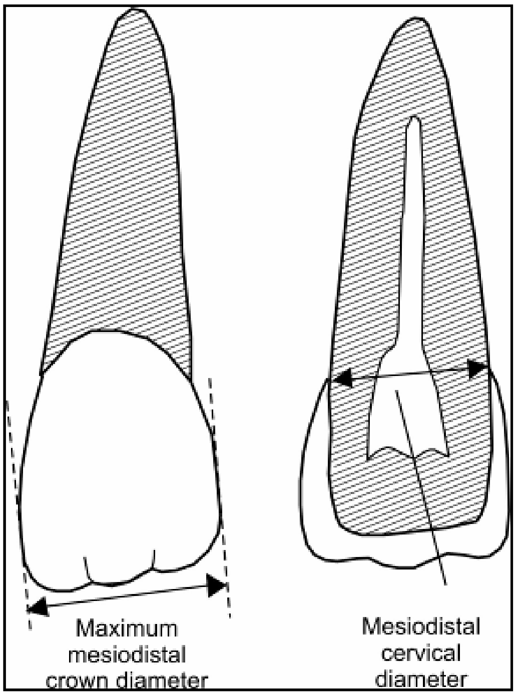


Figure 2.12. A labial view of the mesiodistal section of the anterior teeth showing the crown and cervical diameter measurements (modified from Hillson *et al.*, 2005).

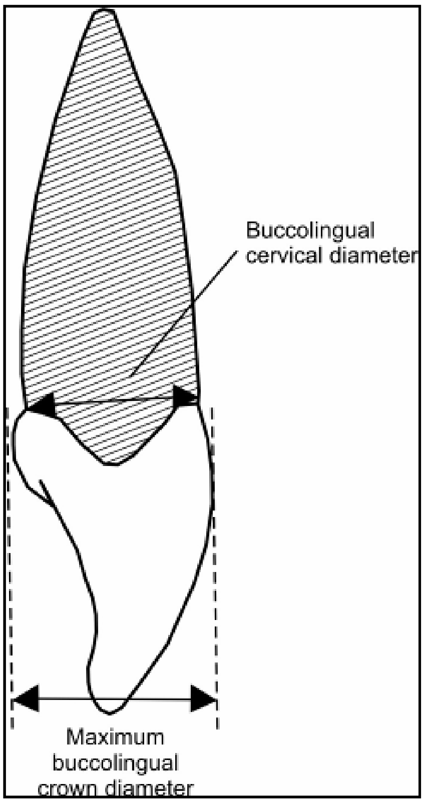


Figure 2.13. The mesial view of the crown diameter and cervical diameter measurements in the anterior teeth (modified from Hillson *et al.*, 2005).

2.2.2. Measurement difficulties

Some difficulties exist when measuring teeth. Measuring the mesiodistal diameter becomes difficult when the teeth are in situ. They are held in the jaw and fit tightly against the neighbouring teeth in such a way that the calliper points cannot be placed on the maximum convexity of the mesial and distal crown ends. Some observers use callipers with needle points whereas some slightly move the teeth a little in the jaw so as to allow the measurement to be taken. This latter method of measurement is rarely possible since the delicate specimen can easily be damaged. The complex shape of teeth, for example the molar teeth which have cusps of varying sizes and surfaces of varying curvatures, may be difficult to measure which leads to errors. Teeth are part of those anatomical structures expressed bilaterally which show complete symmetry. But sometimes asymmetry can be observed in the crown and root trait expression. These asymmetries have developmental as well as methodological implications (Scott and Turner, 1997). Dental wear is also one of the difficulties found in measuring tooth crowns, but it can be identified. The only effect that wear has on the buccolingual diameter is when most of the crown has been lost through occlusal attrition, whereas the mesiodistal diameter is strongly affected by approximal (interproximal/ interstitial) attrition, even at the earliest stages. By measuring unworn teeth, these measurement landmarks are infinitely small points at the maximum convexity of the curving area. Progression of the approximal attrition leads to the formation of two flat wear surfaces which lies tangential to the curve of the crown side and thus the smallest amount of wear can have a large effect which can be doubled due to the measurement being reduced at both ends. Teeth with advanced wear, which makes measuring difficult, are excluded by most studies, since it is apparent that there is no clear limit as to classify a tooth as too worn. The teeth mostly affected are the incisors and canines, but the effects on the rest of the teeth can still not be ignored. This may ultimately lead to the exclusion of some teeth, thereby reducing the sample size which is needed to make a study reliable. With this said, it is also not possible to compare the mesiodistal diameter for example, of the less heavily worn teeth of children with the more heavily worn teeth of adults (Jacobson, 1982; Hillson, 1996; Hillson *et al.*, 2005).

Measuring errors may also occur in studies, therefore it is necessary to include intra-an inter-observer error tests into any study. Intra-observer error is when one observer, repeating a certain measurement a few times on the same teeth, is liable to obtain a range of different results whereas an inter-observer error is when different observers repeating a measurement may produce an even greater range. The usual procedure of measuring to the nearest 0.1 millimetres (mm), reflects the expected level of error (Hillson, 1996). Jacobson

(1982) mentioned that Shaw (1931) found that even with exact definitions of the measurements, it was impossible to obtain a greater accuracy than 0.5 mm. Jacobson's (1982) study corroborated Goose's claim (1963) that an accuracy of 0.1 mm is readily attainable.

Hillson (1996) stated that measurements can be taken both on the original teeth and models prepared from dental impressions. Although Hillson (1996) stated that casting introduces errors, other researchers like Hunter and Priest (1960) found less than 0.1 mm difference between casts and the originals. Therefore, some measurements taken from plaster casts may not necessarily correspond with those made on the original teeth. This can be directly linked to the differing techniques used and may bring about measuring errors (Jacobson, 1982). It has also been suggested that dental calculus, which is the hard, mineralized, accumulation of variable thickness found above or below the gingivae, may affect the mesiodistal and buccolingual diameters of the anterior teeth (Kieser, 1990). This feature may add up to 5 mm to a tooth's buccolingual diameter. This was also one of the reasons why some investigators chose to omit the buccolingual measurements of the upper and lower anterior teeth from their odontometric studies unless there are no calculus deposits on these teeth. Restorations, caries, crowns or any mechanical intervention by dentists also contribute to the errors in measurements. Therefore measurements should not be attempted on carious, restored or fractured teeth, especially for identification purposes or population studies (Kieser, 1990). Although there are limitations to measurements of teeth, it still remains the only option for attempting to construct a profile when there are no differences in the basic anatomical features. One of the advantages of the metrical method in sex and age determination is that it offers a source of accurate and objective data which can be subjected to statistical analyses (Jacobson, 1982).

2.2.3. Mesiodistal and Buccolingual diameters

The mesiodistal diameter of the crown has many definitions, but as stated before the most often quoted is the greatest mesiodistal dimension. This is the dimension taken parallel to the occlusal and labial surfaces of the tooth crown (Moorrees *et al.*, 1957). If this definition is applied strictly, a problem arises when malocclusion causes the teeth to rotate or be displaced out of the dental arcade. When this occurs, the most mesial and most distal points are in different positions on the crown. Therefore they are no longer homologous points (Hillson, 1996). Other investigators defined the mesiodistal diameter as the distance between the contact points of the tooth (Figure 2.9). Contact points are the areas where the tooth contacts its neighbour and these areas may become larger with interproximal attrition (the wear of the enamel that occurs when the adjacent teeth move against each other during mastication). When this method is used, the calliper is held parallel to the occlusal plane of the tooth and the beaks of the calliper are placed on the mesial and distal contact points of the crown. The one advantage of this method of measurement is that the researcher will be able to sum all the mesiodistal measurements and derive the length of the dental arcade (Katzenberg and Saunders, 2000). Jacobson (1982) stated that this measurement represents the greatest dimension of the crown of the tooth between the adjacent contact points or areas in the case of cheek teeth and the maximum mesiodistal dimension in the case of incisors and canines. This method is satisfactory since the contact points are a natural anatomical feature and when rotation is present, the mesiodistal diameter may have a larger value (Jacobson, 1982). There will be instances where the different measurement methods use the same landmarks and derive the same values, therefore it is important to decide before the study commences which will be used. Landmarks may also vary considerably depending on the tooth type being measured (Katzenberg and Saunders, 2000). Disadvantages include, in cases of any inter-proximal attrition, a reduction in the mesiodistal diameter and smaller sample sizes due to the number of rejected measurements (mesial or distal attrition) (Katzenberg and Saunders, 2000).

Another method of measuring the mesiodistal diameter is by taking the maximum width of the crown in the mesiodistal plane (Figure 2.9). This measurement will be wider than the distance between contact points and may be easier to apply. Katzenberg and Saunders (2000) stated that this measurement provides, theoretically, a bit more information about the tooth development due to the fact that it measures the largest amount of growth in the mesiodistal plane. Other authors used a definition that allows for the possibility by measuring the diameter between the mesial and distal contact points of each crown as they would be if the crown were in normal occlusion. Hillson (1996) stated that this method of measuring is currently the recommended procedure. Care must be taken, however, to determine the definition used when interpreting published reports (Hillson, 1996). Approximal or inter-proximal wear, which produces a facet on which no clear measurement point is defined, and occlusal attrition, both reduce the original mesiodistal diameter. Therefore most researchers would exclude teeth with marked approximal wear as well as heavily worn teeth (Hillson, 1996).

The buccolingual diameter can be ascertained when the mesiodistal measurement is determined. This measurement is defined as the greatest distance between the buccal and lingual surfaces of the crown, taken at right angles to the plane in which the mesiodistal diameter was taken (Figure 2.9) (Jacobson, 1982; Hillson, 1996; Katzenberg and Saunders, 2000). According to Katzenberg and Saunders (2000), this measurement is subject to error since the most protruding portion of the facial aspect of the molars will be towards the mesial and the corresponding point for the lingual will be toward the distal side of the crown. Therefore the callipers should be carefully positioned so that it is not anything other than being perpendicular to the mesiodistal axis. It should also be noted that the two points will probably not be in the same plane occluso-cervically. The callipers should also be parallel to the occlusal plane when measuring the buccolingual diameter. In the case of the anterior teeth, the buccolingual diameter may be difficult because the heights of the contour of the buccal and lingual surfaces are so different (Figure 2.10). Therefore it is important to ensure that the calliper beaks are parallel to the long axis of the tooth and not the crown, since the long axis of the crown is often not in the same plane as the axis of the entire tooth. It is also stated that it is important to ascertain that the heights of contour on the cingulum area of the incisors and canines are reachable with the calliper points. It is also possible that the cingulum area, which is the portion of the lingual surface that forms a protuberance in the cervical third of the crown, may be obscured by the lingual plate of bone if the tooth is partially erupted (Katzenberg and Saunders, 2000). The buccolingual diameter is unaffected by approximal wear, but can be influenced by marked occlusal attrition (Hillson, 1996).

Hillson (1996) suggested that for the measurements not affected by wear, the mesiodistal and buccolingual diameters at the cervical margin should be taken, since these give similar results to the normal crown diameters. The mesiodistal and buccolingual diameters are used to construct indices that describe features such as proportions of the tooth, and the approximate area of the occlusal surface. These dental indices include:

- Crown module: Average diameter of the crown in a particular tooth class (mesiodistal diameter + buccolingual diameter, then divided by 2). It was claimed to express the mass of the crown and not the shape, but others said it is rather imperfect and has fallen into disuse (Jacobson, 1982; Hillson, 1996; Katzenberg and Saunders, 2000).
- Crown index: Buccolingual diameter expressed as a percentage of the mesiodistal diameter (buccolingual diameter divided by mesiodistal diameter, multiplied by 100). This index displays the ratio between the two measurements and will illustrate the shape of the crown. This index was used, for example, by Rosenzweig in 1970 to study sexual dimorphism and population differences of the Middle Eastern groups (Rosenzweig, 1970; Hillson, 1996; Katzenberg and Saunders, 2000).
- Robustness index/Crown area/Crown robustness: Area that would be enclosed by the occlusal surface if it were a perfect rectangle (the mesiodistal diameter is multiplied by the buccolingual diameter). This was used by Lukacs (1988) in the

study of early agriculturists, for example, and was found to be an accurate representation of a rectangle into which the tooth could be fitted. This was an effective method to calculate tooth size for comparative purposes even though it did not take into account the differences in shape (Lukacs, 1988; Hillson, 1996; Katzenberg and Saunders, 2000).

It is expected that for one population and a single sex, the mesiodistal and buccolingual diameters of each tooth have normal (Gaussian) distributions, since this is usual for the dimensions of the anatomical structures in adults as well as skeletal measurements. Archaeological and museum collections may deviate from the ideal of normality, since this is true for small numbers of individuals and their uncertain derivation (Hillson, 1996). It was stated that the living *Homo* shows a moderate correlation between the mesiodistal and buccolingual diameters of the same crown. The correlations are slightly greater in females than in males, in the upper than the lower teeth as well as in the cheek than anterior teeth. The mesiodistal diameter on average is larger than the buccolingual in the upper incisors, but the opposite in the lower incisors. The canines present with diameters which are approximately equal. All the cheek teeth, except the lower molars, have larger buccolingual diameters than mesiodistal ones. Both dentitions (deciduous and permanent) have moderate correlations for the diameters between the different teeth in the same jaw. Therefore, if the tooth of one part of the jaw is large then the teeth from the other parts are most probably also large. This is different when the anterior teeth are taken as a group, and then their crown diameters are inversely related to those of the cheek teeth which will be treated as the other group. This means that when the individual present with larger than normal anterior tooth diameters, the cheek teeth are smaller than normal and *vice versa* (Hillson, 1996). There is therefore variability amongst the tooth diameters and the teeth which are the most variable in their dimensions are the upper second incisors, third molars and second premolars. The least variability is seen in the upper first molars, first incisors and canines. The same cannot be said about the lower dentition due to it showing a less consistent pattern, but the premolars are often the teeth showing the most variability (Hillson, 1996).

2.3. Estimation of sex

2.3.1. Introduction

Sexual dimorphism can be defined as the differences seen in the size, structure and form between the male and female of the same group or population. This can be applied to the dental identification process since no two individuals have identical oral cavities and features (Kieser, 1990; Kaushal *et al.*, 2003).

Sex estimation is not always a simple process. The male and female attributions, e.g. the morphologic contributions and metric measures in the skeleton, exist in a continuum. There are some bones that serve as better indicators of sex than others but none will be as exclusive as the differences seen between fleshed individuals. Remains such as clothing, hair, jewellery and artifacts; may in some cases help in the corroboration of a positive identification. These features can also be misleading since long hair, for example, is not always a female feature. Therefore care must be taken when studying remains, especially in cases of mass disasters (comingled remains) (Loth and İşcan, 2000).

Before the process of sex estimation from the skeleton is started, it is useful to look for some clues as to what population group the individual belonged to since regional and population variations exist in the development of the sexual characteristics in the skeleton. The accuracy of sex estimation is also dependent on the availability of the bones, since some bones show more sexual differences than others. Most of the sexual criteria observed in bones are expressed as a matter of degree rather than an absolute difference. Therefore, it is essential to be thoroughly acquainted with a fairly typical male and a fairly typical female. This is acquired through the handling and experience, since without it, it will be impossible to be accurate to any degree due to the fact that absolute criteria seldom exist. It is also important to be aware of the degree of variation found in each trait as well as the variation from one population to another (El-Najjar and McWilliams, 1978).

The distribution of skeletal features between males and females used in the estimation of sex considerately overlap and some of these are generally taken as more reliable than others. The degree of sexual dimorphism in the skeletal characteristics is stated to vary among the human populations. It is highly valued that the more bones included in the study, the better as well as the multivariate statistical techniques which are used to characterize the size and shape of these bones or skeletal features (Katzenberg and Saunders, 2000).

To understand the meaning and origin of the differences between male and female organisms of a given species, sexual dimorphism has been divided into three levels according to Kieser (1990). The first and most fundamental level is the primary sexual characters which distinguishes the sexes – the testes in the males and the ovaries in females. This is essential to the process of sexual reproduction. The second level of dimorphism is found in the external features that distinguishes males from females. These features function as releasers of social reactions which are intimately connected to courtship and mating, and thus are called secondary sexual characters. For example, the large canines found in the males of many animal species, function as visual sexual signs of dominance and rank. Sexual dimorphism also involves the differences in the size between the males and females - the third level of dimorphism (Kieser, 1990). According to Kieser (1990), when considering the levels and patterning of sexual dimorphism in human tooth size, the measurement of dimorphism is best starting point for one's study. Numerous other studies (Seipel, 1946, Moorrees, 1959, Garn *et al.*, 1964) reported that the dental dimensions of males are considerably larger than those in females and that the largest differences were found in the canines in spite of the reduction in the tooth size of humans (Kieser, 1990).

The degree of accuracy of sexing unknown material becomes decreased by factors such as the often fragmentary or isolated nature of the remains which are available for the study, the age at death of the remains as well as the intrinsic variability and population specific standards. Part of this problem is the subjectivity versus the objectivity, description versus measurement and experience versus statistical standardization. Although previous as well as current studies of sex differences in the skeleton, which include skull and pelvis, centre on the morphological traits assessed in a descriptive manner, morphometry (discriminant function analysis) which is done in a quantitative and statistical sense, is also incorporated (Krogman and Işcan, 1986). Many studies have been done concerning the sexual differences in the human skeleton of different populations (e.g., Kieser, 1990; Hillson, 1996; Scott and Turner, 1997; Katzenberg and Saunders, 2000; Ates *et al.*, 2006; Vodanović *et al.*, 2007). The two most studied skeletal elements are the pelvis and the skull. The unique features of teeth and the jaws have also been used often in the identification of humans, since jaw and tooth dimensions and morphology show dimorphism (Kieser, 1990; Hanihara and Ishida, 2005; Vodanović *et al.*, 2007). Sex estimation becomes complicated when the remains are fragmented or poorly handled. Morphometry or methods using measurements are especially helpful in such cases, and has become increasingly utilized in the diagnosis of sex with the advancement of modern statistical techniques (Vodanović *et al.*, 2006 and 2007).

Sexual dimorphism of tooth size has been the subject of many studies in various populations (Garn *et al.*, 1964; Garn and colleagues, 1967, 1977 and 1979; Garn *et al.*, 1967b; Jacobson, 1982; Kieser, 1990; Hillson, 1996; İşcan and Kedici, 2003; Kaushal *et al.*, 2003; Ates *et al.*, 2006; Vodanović *et al.*, 2007b; Acharya and Mainali, 2008; Hemanth *et al.*, 2008; Acharya and Mainali, 2009). Katzenberg and Saunders (2000) reported that both dentitions show statistically significant sex differences even though these differences are small, where the permanent canines displayed a 5-6% difference and the rest of the teeth about 2-4%. They stated that these percentages correlate with those of other studies. Another study done by Scott and Turner (1997) also found that there are consistently low levels of sexual dimorphism in the human crown dimensions where male teeth are 2-6% larger than female teeth. They also found that discriminant function analysis of tooth size can correctly differentiate between the sexes in about 86% of the cases. Sexual dimorphism found in the mesiodistal diameter of the lower canine is said to be among the highest of all tooth dimensions in the modern humans (Scott and Turner, 1997). Therefore, it is possible to use odontometrics in the process of sex determination (Vodanović *et al.*, 2007). In the process of identifying the sex of an individual, one must take into account that these differences in male and female odontometric features differ among and within populations. Therefore it is needed to determine specific population values to make a possible identification based on dental measurements (Vodanović *et al.*, 2007).

2.3.2. Odontometric studies

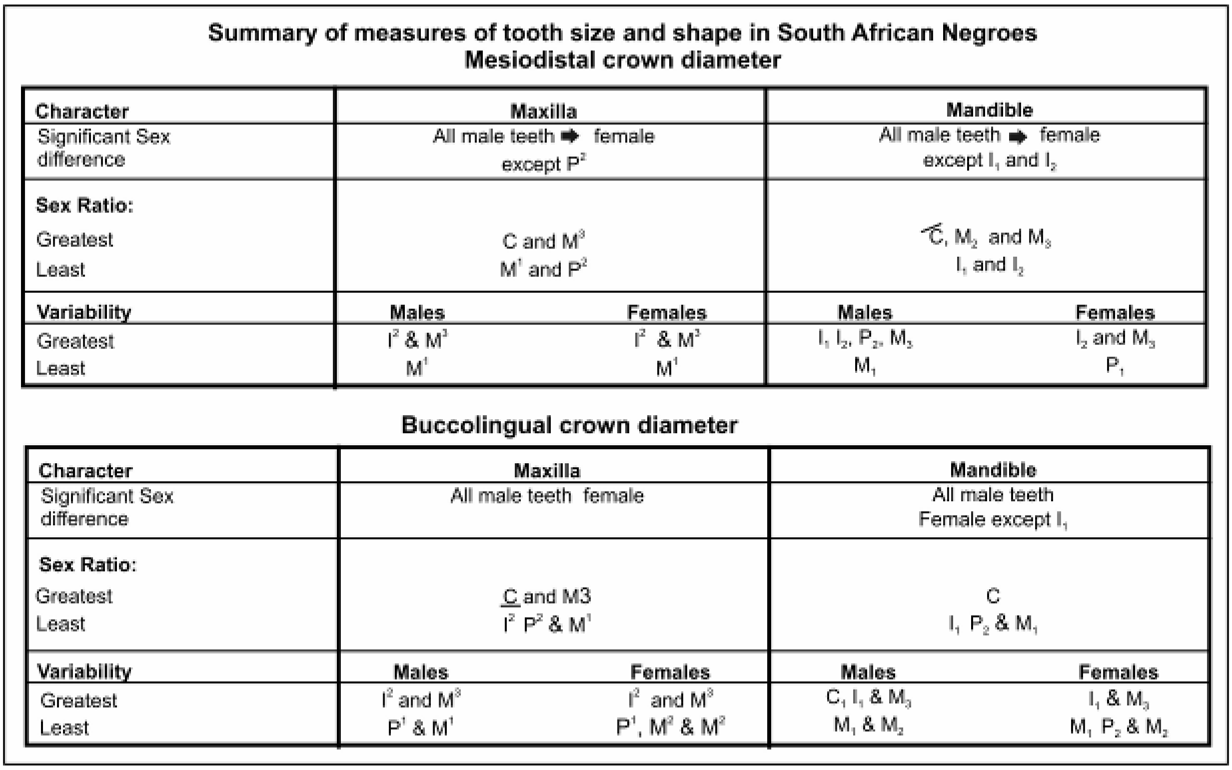
Many studies have found that odontometrics, especially the mesiodistal and buccolingual diameters, are useful in sex determination (Garn *et al.*, 1964; Garn *et al.*, 1967, 1977, 1979; Moss, 1978; Jacobson, 1982; Kieser, 1990; Hillson, 1996; Otuyemi and Noar, 1996; Yuen *et al.*, 1997; İşcan and Kedici, 2003; Kaushal *et al.*, 2003; Moss *et al.*, 2005; Potter *et al.*, 2005; Ates *et al.*, 2006; Vodanović *et al.*, 2007b; Acharya and Mainali, 2008; Prabhu and Acharya, 2009). Hanihara and Ishida (2005) have also done a study on the metric dental variation of major human populations which have used the mesiodistal and buccolingual crown diameters of teeth in 72 major human populations. They analyzed seven geographical groups and found that odontometric variation is an effective tool to study the variation pattern among modern human populations on a larger, worldwide scale. Although their study focused more on the population differences using the two crown measurements, it served a purpose in that they confirmed the fact that odontometric variation is an effective tool for assessing differences in tooth size within (sex estimation) and between populations.

All tooth types have been tested in studies of sexual dimorphism. Hemanth *et al.* (2008); Suazo *et al.* (2008) and Pettenati-Soubayroux *et al.* (2002); found that the premolars, molars and incisors also show significant sex differences (80% accuracy), even though the canine almost always shows the greatest dimensional differences between sexes. Although the present study focuses on the permanent dentition, a worldwide survey of the mesiodistal tooth crown dimensions of primary dentition found that, with regard to the mesiodistal crown dimensions, the canine and first molar are more sexually dimorphic than the other tooth types (p=0.0002) (Harris and Lease 2005).

2.3.3. Canines and Odontometrics

Some researchers, like Jacobson (1982), Kieser (1990), Hillson (1996), Otuyemi and Noar (1996), Yuen *et al.* (1997), Kaushal (2003), Ates *et al.* (2006), and Vodanović *et al.* (2007b) to name a few, stated that the canines are the most suitable teeth for sex estimation. They also have a low prevalence of ante- or post-mortem loss (Vodanović *et al.*, 2007b).

Several authors worldwide, as well as studies done on South African populations (e.g. Table 2.1), for example Garn *et al.* (1964); Garn and colleagues (1967, 1977 and 1979); Garn *et al.* (1967b); Ditch and Rose (1972); Anderson and Thompson (1973); Moss (1978); Jacobson (1982) (Table 2.1); Kuwana (1983); Rao *et al.* (1986 and 1989); Kieser (1990); Minzuno (1990); Hillson (1996); Otuyemi and Noar (1996); Pettenati-Soubayroux *et al.* (2002); Yadav *et al.* (2002); Işcan and Kedici (2003); Kaushal *et al.* (2003); Kaushal *et al.* (2004); Potter *et al.* (2005); Ates *et al.* (2006); Acharya and Mainali (2007); Vodanović *et al.* (2007b); Acharya and Mainali (2008); Hemanth *et al.* (2008); and Acharya and Mainali (2009) used the mesiodistal and buccolingual crown diameters of the canines as indicators of sex and reported successful results with accuracies ranging from 58% to 94.1% (Vodanović *et al* 2007b). Some of them used both the maxillary and mandibular canines whereas some reported a better result with either the buccolingual or the mesiodistal crown diameter. From this it is evident that there are odontometric data of many population groups including the South Africans available, but discriminant functions based on the South African population groups are lacking and needs further attention. Some of these studies also stated that these sex estimation methods are population specific (Otuyemi and Noar 1996; Kaushal *et al.,* 2003; Vodanović *et al.,* 2006; Vodanović *et al.,* 2007b; Brooke et al 2009).



Summary of measures of tooth size and shape in South African Negroes
Mesiodistal crown diameter

Character	**Maxilla**		**Mandible**	
Significant Sex difference	All male teeth ➡ female except P^2		All male teeth ➡ female except I_1 and I_2	
Sex Ratio:				
Greatest	C and M^3		$\bar{C}$, M_2 and M_3	
Least	M^1 and P^2		I_1 and I_2	
Variability	**Males**	**Females**	**Males**	**Females**
Greatest	I^2 & M^3	I^2 & M^3	I_1, I_2, P_2, M_3	I_2 and M_3
Least	M^1	M^1	M_1	P_1

Buccolingual crown diameter

Character	**Maxilla**		**Mandible**	
Significant Sex difference	All male teeth female		All male teeth Female except I_1	
Sex Ratio:				
Greatest	$\underline{C}$ and M3		C	
Least	I^2 P^2 & M^1		I_1 P_2 & M_1	
Variability	**Males**	**Females**	**Males**	**Females**
Greatest	I^2 and M^3	I^2 and M^3	C_1 I_1 & M_3	I_1 & M_3
Least	P^1 & M^1	P^1, M^2 & M^2	M_1 & M_2	M_1 P_2 & M_2

Table 2.1. Summary of the mesiodistal and buccolingual diameters of Jacobson's study on the South African black population (modified from Jacobson, 1982).

2.3.4. Statistical analyses

In earlier studies the mesiodistal and buccolingual measurements were used to create indices to differentiate between the sexes of a group (Rao *et al.* 1986; 1989). Since then more sophisticated methods (multiple discriminant function analyses) were created and applied. Discriminant function analyses became the method of choice since it includes more parameters and provides better separation. Discriminant analysis utilizes interrelationship between all teeth within the dentition whereas univariate analysis does not utilize these tooth correlations and this may lead to a loss of information. Discriminant function analysis can be used for the purpose of determining the sex of an individual since it can classify the individual into two or more different classes, for example male and female (Vodanović *et al.,* 2006; 2007). Ditch and Rose (1972) published one of the first studies utilizing discriminant function analyses to determine the sex of their sample and reported an accuracy ranging from 88% to 95.5%

To make discriminant function analysis work, a large, random sample of individuals from each of the classifying groups or populations must be available. The researcher will then be able to build a discriminant function that can be used in the study to classify others into one of the groups (Vodanović *et al.*, 2006). The discriminant function analysis is done by statistical software, for example the SPSS package. This will calculate the within-group correlation matrix for the analyzed variables, eigenvalue, canonical correlation, Wilks' lambda, chi-square and significance level for the derived discriminant function. It will also provide values for the standardized and unstandardized coefficients, structure matrix and the accuracy of the functions. The eigenvalue indicates the ratio of importance of the dimensions which classify cases of the dependent variable. The eigenvalues assess the relative importance because they reflect the percentage of variance explained in the dependent variable, cumulating to 100% for all functions. The canonical correlation is a measure of the association between the groups formed by the dependent and the given discriminant function. When the canonical correlation ends in a value of zero, there is no relation between the groups and the function. When the canonical correlation results in a large value, there is a high correlation between the discriminant functions and the groups. Wilks' lambda will test the significance of the discriminant function, specifically the significance of the eigenvalue for a given function. The Chi-squared test can be used to test the significance of the discriminant function as a whole. The standardized discriminant coefficients (standardized canonical discriminant function coefficients) compare the relative importance of the independent variables, whereas the unstandardized discriminant coefficients are used in the discriminant formula for making the classifications of new cases. The structure matrix is a table of structure coefficients of each variable with each discriminant function and the structure coefficients are the correlations between a given independent variable and the discriminant scores associated with a given discriminant function. These can be used to indicate how closely a variable is related to each function in the discriminant analysis. The accuracy of the derived discriminant functions is validated on the original and cross-validated samples. Cross-validation is done where each case is classified by the functions derived from all cases other than that case. This is reinforced by the thought that it gives a better estimate of what classification results would be in the general population (Vodanović *et al.*, 2006).

In discriminant function analysis a single variable may be used, but most often a combination of measurements is chosen from each bone to maximize the sex estimates. These are freely available for many bones in the form of discriminant function statistics. Du Jardin *et al.* (2009) pointed out three main assumptions that must be met when using discriminant analysis: (i) the observed variables within each sample or population must follow a multivariate normal distribution; (ii) the variance-covariance matrices of the groups must be equal e.g. the variance of each variable must be similar in each group; (iii) the correlation between the variables must be as low as possible. It was also stated that discriminant analysis is generally easy to use and very popular amongst anthropologists (Du Jardin *et al.*, 2009).

Other researchers focused on indices to establish the sex of remains. Rao *et al* (1989)'s index, the Mandibular Canine Index (MCI), uses the relationship between mesiodistal crown measurement and the intercanine distance. With this index it was possible to achieve good results – the accuracy of sex estimation was 84.3% in the male group and 87.5% in the female group. This method was deemed as a simple and inexpensive method for establishing sex (Rao *et al.*, 1989). Kaushal *et al.* (2003, 2004) applied Rao *et al.*'s method and found that the mandibular canine exhibits statistically significant sexual dimorphism and could detect the sex of a North Indian population with an accuracy of 75%. Muller *et al.* (2001) and Kaushal *et al.* (2004), both stated that this method requires correct dental alignment and can be influenced by cultural, environmental and "racial" factors.

2.4. Estimation of age

2.4.1. Introduction

Age estimation using the human skeleton is mostly a difficult task and the accuracy thereof decreases with the advancing age of the individual. There are also different parts of the skeleton that are more informative at different ages (El-najjar and McWilliams, 1978; Eckert, 1997; Katzenberg and Saunders, 2000). An accurate estimation of age at death is one of the most important aspects of a biological profile. The term 'accuracy' involves estimating the age as close as possible to the actual chronological age at death and to realistically convey the probabilities associated with the estimate. This process gets complicated due to many individual techniques available that have been developed by different researchers using diverse samples. Currently there are many techniques available that reflect the age changes in different populations at different periods in history (Katzenberg and Saunders, 2000).

2.4.2. Methods

Katzenberg and Saunders (2000) stated that the decision of which method to use in the estimation of age is based on the relative accuracy of the method, the experience of the investigator, the available equipment as well as the preservation of the skeletal remains. There are a number of methods available and it is advisable to use as many methods as possible to narrow down the estimate of age at death. Some of the most accurate approaches for adult age estimation include assessment of the postcranial elements (pelvis and ribs - İşcan *et al.*, 1984; Brooks and Suchey, 1990; Oettlé and Steyn, 2000), the skull (Meindl and Lovejoy, 1985; Mann et al., 1987; Gruspier and Mullen, 1991; Buikstra and Ubelaker, 1994), or the microscopic examination of the long bone cortical microstructure (histological approach) (Katzenberg and Saunders, 2000; Reppien *et al.*, 2006). Due to the vast amount of literature, this review will only focus on adult age estimation using the dentition.

Age assessment in adults is more complicated than in juveniles, due to lack of changes in the mature skeleton. It is also important to remember that, especially in a forensic context, a relatively wide range of the estimated age of the individual should be given as not to mislead the authorities - this takes into account much of the imprecision that goes with adult age estimation (Katzenberg and Saunders, 2000; Reppien *et al.*, 2006). The best results can be achieved when using a combination of methods to estimate the age of the individual (Kilian and Vlček, 1989). It is said that results are considered very good when the difference between the actual and estimated age is approximately 5 years, it is satisfactory at approximately 10 years and unsatisfactory when it is more than approximately 10 years (Kilian and Vlček, 1989; Smith, 1991). Therefore, to ensure the successful use of these methods, it is necessary to observe the following general requirements (Kilian and Vlček, 1989):

- The investigator using the methods for age estimation should have the required theoretical knowledge of the problems surrounding age determination from teeth as well as practical experience
- Availability of as many as possible teeth of the same individual (incisors and canines)
- The method need to be as simple and fast as possible
- Well defined, easy evaluated and quantitative criteria should be used in the determination of age as well as to avoid the use of single criterion as a sole indicator of age estimation
- The use of more criteria and methods will give a more accurate estimate.

Adult age estimation from teeth has been of concern for anthropologists, especially in cases where there is only a skull available. The first attempts were mainly based on the assessment of the tooth attrition. Many factors play a role in dental wear, however, and culture, diet, pathology should all be taken into account (Krogman and İşcan, 1986). There are age assessment methods that use the dentition, to name a few, these include the:

- Observation of the removal of enamel (dental wear) and measurement of changes in the crown dimensions for the occlusal and interproximal surfaces (Hinton, 1982);
- Examination of the height of the cementoenamel junction above the inferior dental canal through radiographic images together with the radiographic or half tooth section examination of the changes to the pulp chamber through secondary dentin deposition (Boyuan and associates, 1983; Solheim, 1992; Foti *et al.*, 2001);
- Histological studies of the dentin and cementum (Gustafson, 1950; Vlček and Mrklas, 1975);
- Root transparency as an age indicator which formed the main focus of various studies (Miles, 1963; Johnson, 1968; Bang and Ramm, 1970; Vasiliadis *et al.*, 1983; Bang, 1989; Lorentsen and Solheim, 1989; Drusini *et al.*, 1991; Thomas *et al.*, 1994; Whittaker and Bakri, 1996; Sengupta *et al.*, 1999; Katzenberg and Saunders, 2000).

The histology of dentition provides a variety of methods for age estimation in adult remains. These methods compete with those that are based on the macroscopic examination of the skeleton and dentition (Hillson, 1996). More sophisticated methods, which allow a closer estimated or real age, have also been investigated. These methods include aspartic acid racemization (Ogino and Ogino, 1985; Ohtani and Sugimoto, 1995) or the ratios of measurements of pulp/root and pulp/tooth from radiographs (e.g., Foti *et al.*, 2001). These methods, however, do not fall into the scope of this study.

2.4.2.1. Gustafson's method

Gustafson was one of the first researchers to use a combination of characteristics seen on a dental section to estimate age, after which several modifications and additions followed. Gustafson (1947) published a paper on an age estimation method which uses six features on the dental microstructure. These are gingival attachment level, root apex transparency, occlusal wear, amount of secondary dentine, cementum apposition, and root resorption (Gustafson, 1947). Ground sections are used for the scoring of several characteristics (Metzger *et al.*, 1980). Due to Gustafson's great success with this method, he devised a system in 1950 where the emphasis has been on the many different changes on an anterior teeth and that was based on six age-related factors which included dental attrition (A), periodontosis (P), secondary dentine deposition (S), cement apposition (C), root resorption (R) and root transparency (T) (root dentine sclerosis). The sample comprised of only 37 teeth from northern Europeans aged 11 to 69 years (Gustafson, 1950; Krogman and İşcan, 1986; Hillson, 1996). Gustafson (1950) also included the closing of the orifice of the root as an additional factor, but found that it is more influential in badly maintained teeth. The dentine translucency was determined from 1.0 mm thick ground sections, while the other factors were determined from 0.25 mm thick sections. It was stated that most of these factors have a pathologic basis, but they also correlate well with age. Each of the six characteristics is scored with a value ranging from 0 to 3 (Figure 2.14), and the values were summed (Table 2.2) to give an overall score (Total scale points = $A_n + P_n + S_n + C_n + R_n + T_n$).

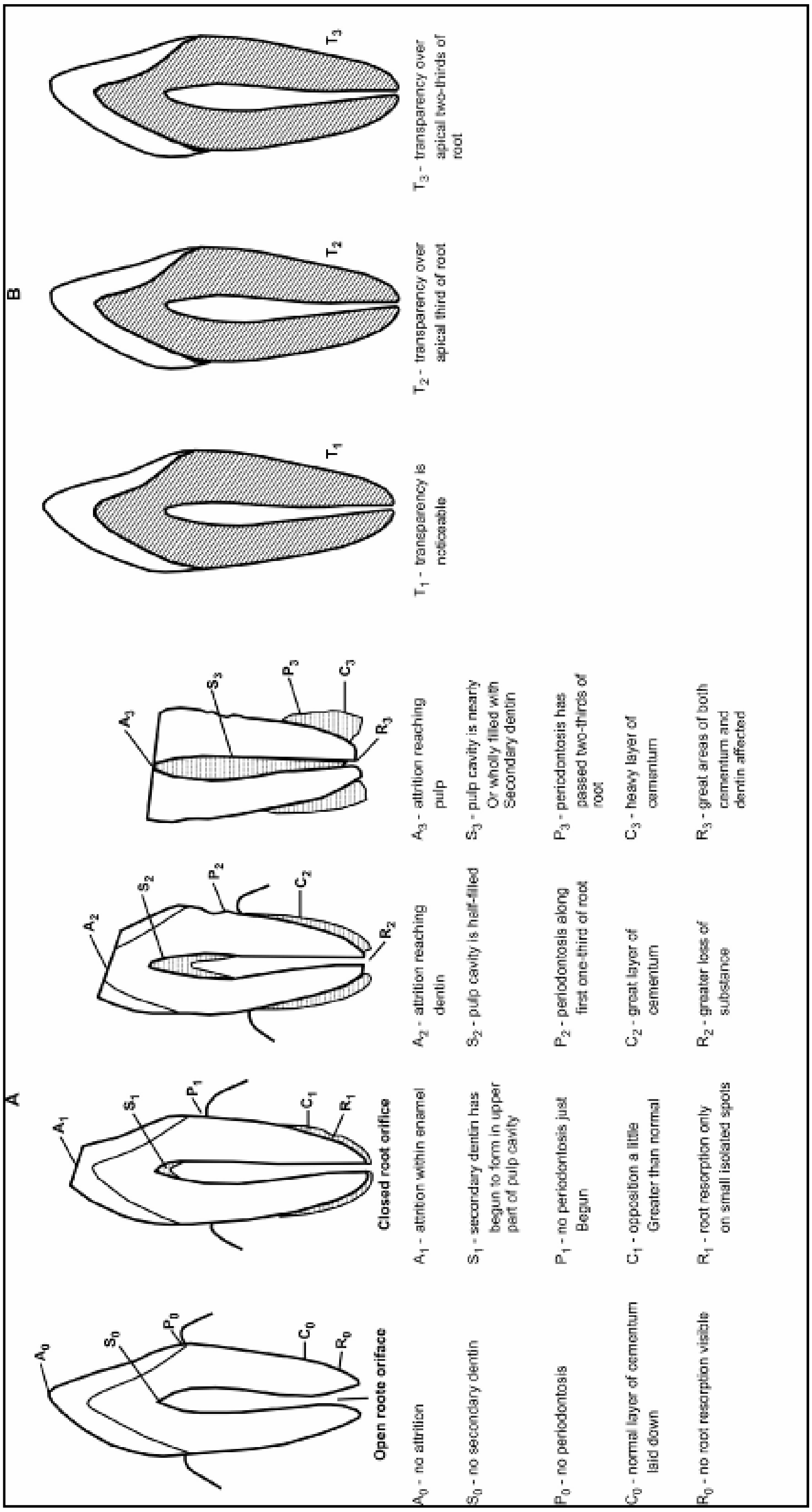
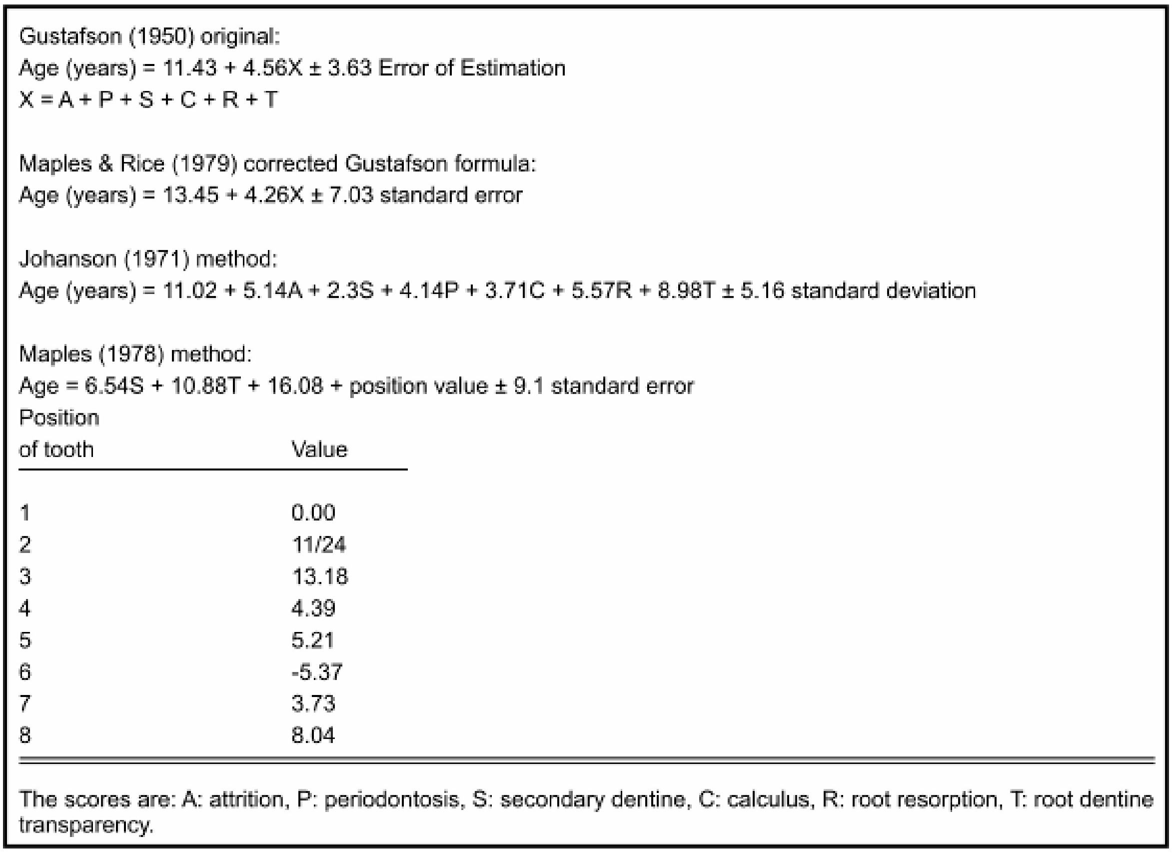


Figure 2.14. Gustafson's method: Point scoring system. A = the scores for attrition (A), secondary dentine (S), periodontosis (P), cementum apposition (C), root resorption (R); B = the scores for root dentine transparency (T) (modified from Hillson, 1996).



Gustafson (1950) original:
$\text{Age (years)} = 11.43 + 4.56X \pm 3.63$ Error of Estimation
$X = A + P + S + C + R + T$

Maples & Rice (1979) corrected Gustafson formula:
$\text{Age (years)} = 13.45 + 4.26X \pm 7.03$ standard error

Johanson (1971) method:
$\text{Age (years)} = 11.02 + 5.14A + 2.3S + 4.14P + 3.71C + 5.57R + 8.98T \pm 5.16$ standard deviation

Maples (1978) method:
$\text{Age} = 6.54S + 10.88T + 16.08 + \text{position value} \pm 9.1$ standard error

Position of tooth	Value
1	0.00
2	11/24
3	13.18
4	4.39
5	5.21
6	-5.37
7	3.73
8	8.04

The scores are: A: attrition, P: periodontosis, S: secondary dentine, C: calculus, R: root resorption, T: root dentine transparency.

Table 2.2. The regression formulae for the Gustafson method and the modifications made by subsequent researchers (modified from Hillson, 1996).

The scale for each characteristic is described as follows:

- Attrition is the wearing down of the incisal or occlusal surface (A0=no attrition; A1=attrition with enamel; A2=attrition reaching dentin; A3=attrition reaching pulp)
- Periodontosis is the loosening or continuous eruption of the tooth (P0=no periodontosis; P1=periodontosis just begun; P2=periodontosis along first one-third of root; P3=periodontosis has passed away two-thirds of root)
- Secondary dentin is the development of the dentin in the pulp cavity (S0=no secondary dentin; S1=secondary dentin has begun to form in the upper part of the pulp cavity; S2=pulp cavity is half filled; S3=attrition reaching pulp)
- Cementum apposition is the deposition of the cementum at the root (C0=no normal layer of cementum laid down; C1=apposition a little greater than normal; C2=great layer of cementum; C3=heavy layer of cementum)

- Root resorption (R0=no root resorption visible; R1=root resorption only on small isolated spots; R2=greater loss of substance; R3=great areas of both cementum and dentin affected)
- Root transparency (T0=no detectable transparency; T1=transparency is noticeable; T2=transparency over apical third of root; T3=transparency over apical two-thirds of root)

The scores were highly correlated (R=0.98) with the known age and a regression curve was constructed with the total point values of each tooth and the known ages of the individuals. In turn this curve was used to determine the age of unknown bodies of forensic investigations. The total score obtained was applied to a regression formula (Estimated age [years] = 11.43 +4.56 * [total points], S.E. (standard error) = 3.63) (Metzger *et al.*, 1980; Krogman and İşcan, 1986; Hillson, 1996). Therefore, if the total point score is 9, then the estimated age is about 52.47 years ± 3.63 years. This method also assumes that sex and ancestry do not affect the result. Gustafson stated that teeth not properly taken care of may appear to be older than the chronological age. Therefore some adjustment should be made in the final age estimate (Gustafson, 1950; Krogman and İşcan, 1986).

Since the 1950's this method was tested by several researchers (e.g., Balogh, 1957; Nalbandian and Soggnaes, 1960; Miles, 1967; Johanson, 1971; Behrend, 1977; Azaz *et al.*, 1977; Maples, 1978; Tomenchuk and Mayhall, 1979; Metzger *et al.*, 1980; Charles *et al.*, 1986; Costa, 1986; Solheim, 1988; Molleson, 1989; Nkhumeleni *et al.*, 1989; Richards and Miller, 1990; Solheim, 1990; Drusini, 1991; Drusini *et al.*, 1991; Whittaker, 1992; Lamendin *et al.*, 1992; Solheim, 1992; Kvaal *et al.*, 1994a; Kvaal *et al.,* 1994b; Kvaal *et al.*, 1995; Hillson, 1996; Sengupta *et al.*, 1998; Sengupta *et al.*, 1999; Katzenberg and Saunders, 2000; Foti *et al.*, 2001; Beyer-Olsen *et al.*, 2005), assessing the objectivity and applicability to contemporary and prehistoric populations. These researchers' reported a standard error ranging from 7.9 to 11.46 years. The root translucency showed in most types of teeth the closest correlation to age. The original Gustafson study was criticized due to sample size, subjective scoring as well as poor statistics and replicability. Johanson (1971) revised Gustafson's scoring system and assigned different weights to each factor (included all the features in the formula with different coefficients) (Table 2.2; Figure 2.15). He found that the root dentine transparency (T) showed the highest correlation with known age, secondary dentine deposition (S), attrition (A), and then cement apposition (C). The periodontosis (P) and root resorption (R) poorly correlated with age (Hillson, 1996).

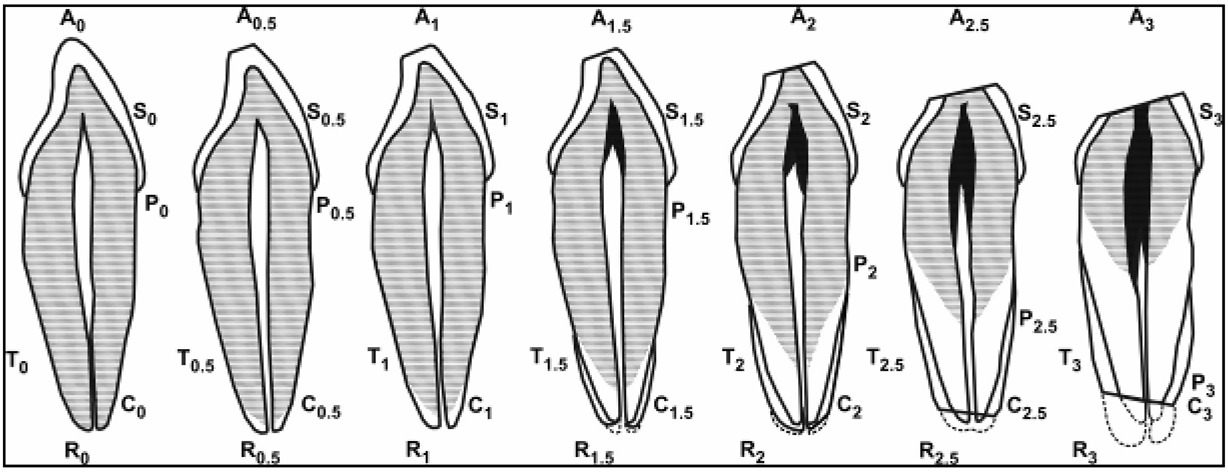


Figure 2.15. Johanson's method: Point scoring system (modified from Hillson, 1996).

Each factor is multiplied by a constant which expressed its correlation with age. An error of ±5 years in 78.3%, ±10 years in 95.7% and ±15 years in 97.8% of all estimations were found. Gustafson's method also underwent significant improvements (e.g. Metzger *et al.*, 1980) such as number of variable reduction, multiple regression analysis, and index values via actual physical measurements, but it still remained the reference dental method of age estimation by most forensic science textbooks.

2.4.2.2. Lamendin's method

Lamendin *et al.* (1992) created a method based on Gustafson's technique and focused their study on two dental features: root transparency (RT) and periodontosis (P), on single rooted teeth (incisors, canines and premolars) which are both measured and expressed as an index value by relating these measurements to a fixed tooth measurement - root height (RH). They applied multiple regression analysis to the variables and it resulted in the formula: A (age) = (0.18 x P) + (0.42 x RT) + 25.53; where P = periodontosis height x 100/root height, RT = root transparency x 100/root height. The advantage of this adaptation of the Gustafson method is that it is not necessary to section the tooth.

The regression formula provided by Lamendin *et al.* (1992) is suitable for both sexes and it ought to be applicable to all types of single rooted teeth (Meinl *et al.*, 2008). The value of 25.53 is the constant of the equation and this makes the equation useless for individuals under the age of 25 years, but this is reported to be the age at which root transparency usually appears. The correlation coefficient of the multiple regressions (R-squared) was found to be 0.33. The accuracy of this resulting equation was tested on a forensic sample and by comparing the outcomes to the original Gustafson method. Lamendin *et al.* (1992) used single rooted teeth, free of restoration, of known age (22-90 years), sex (135 males and 73 females), and ancestry (198 whites and 10 blacks). They estimated the age of each tooth in the sample by using the equation, and found that the mean error between the actual and estimated age was approximately 10 years (Table 2.3) on their working sample and approximately 8.4 years on the control sample (forensic cases).

Age interval (years)	**26-29**	**30-39**	**40-49**	**50-59**	**60-69**	**70-79**	**80-89**	**Total**
Number of teeth	5	42	39	90	65	46	19	306
ME (years)	24.8	15.5	9.9	7.3	6.3	11.6	18.9	10

Table 2.3. Lamendin *et al.*'s mean error (ME) between the actual and estimated age using Lamendin's two criteria dental method (taken from Lamendin *et al.*, 1992).

The upper incisors of their sample gave the best precision and they stated that the accuracy was not sex related. When compared to the Gustafson method, the Lamendin technique resulted in a better mean error of estimation (Gustafson: 14.2 ± 3.4 years, Lamendin: 8.9 ± 2.2 years). They reported inter-observer errors of 9 ± 1.8 and 10 ± 2 years from two independent observers, but still found that this technique worked well with the French population (Lamendin *et al.*, 1992, Prince and Ubelaker, 2002). Lamendin *et al.* argued that the method can be of practical use in forensic setting since it is fast, easy to use, no preparation or destruction of teeth is needed and it is reasonably accurate (except in cases of individuals under the age of 40 where other methods should be preferred). One of the limitations included the fact that large errors were found in some individuals, especially those under 40 and over 80 years of age (Lamendin *et al.*, 1992).

Foti *et al.* (2001) stated that many factors, independent of age, can influence the attachment level of the gingival tissue and the cause of it retracting may be pathological. These factors include bad dental hygiene, physical, chemical, or mechanical irritation. Predisposing factors may also play a role in the attachment level of the gingival tissue and they include specific morphology, systemic diseases and drug treatments. It is said that the recession of the periodontal ligament cannot be used on its own as an indicator of age (Foti *et al.*, 2001).

Although Lamendin *et al.* (1992) found that their method worked well with the French sample, they did not test it outside of the French sample. A few studies have been done on the Lamendin method (Foti *et al.*, 2001; Prince and Ubelaker ,2002; Sarajlic *et al.*, 2006; Megyesi *et al.*, 2006; Martrille *et al.*, 2007; González-Colmenares *et al.*, 2007; Meinl *et al.*, 2008), but very few could reproduce their results (usually resulted in wider standard error of estimates). In general, most researchers found that the Lamendin method is more accurate in the middle to older aged groups (mid-30 age group) and that this method over- and under-estimate older and younger individuals, respectively. The maxillary and mandibular canines (R=0.731 and 0.706) were the subject of one of these studies and obtained the best correlation coefficient in the 40-49 year age group.

Prince and Ubelaker (2002) applied Lamendin's method to their sample which were bigger and not of French origin. Their results suggested that Lamendin's method estimates age fairly accurately outside the French sample which yielded a mean error of 8.2 years, standard deviation 6.9 years, and standard error of the mean 0.34 years. They added that when ancestry and sex are accounted for, the mean errors are reduced for each group (black males, white males, black females, white females). Their intra-observer error test yielded 6.5 years whereas Lamendin *et al* reported an inter-observer error of 9±1.8 and 10±2 years from two independent observers (Prince and Ubelaker, 2002).

González-Colmenarez *et al.* (2007) tested the validity of the Lamendin and Prince and Ubelaker methods. They found that the Lamendin method showed a higher mean error in estimations of the age of youngest and oldest individuals. This confirmed the need to create specific formulas for each human group in order to obtain more accurate age estimates (González-Colmenarez *et al.*, 2007).

Another study evaluated the Lamendin criteria on two historic skeletal samples from Britain (Megyesi *et al.,* 2006). Megyesi *et al.*'s results indicated that post-mortem factors affect the applicability of the Lamendin technique to archaeological and historical samples – the mean error of age estimates was higher for their sample than the original study of Lamendin *et al*.

The Lamendin method was one of four methods Martrille *et al.* (2006) evaluated and applied to their sample. They found that the Lamendin method was the most accurate method for age estimation for middle aged individuals (41-60 years). With regard to biases, they reported that all methods (Suchey-Brooks pubic symphysis method, Lovejoy auricular surface method, Lamendin method and İşcan method for fourth ribs) have the tendency to overestimate the age of young individuals and underestimate the older individuals.

Foti *et al.* (2001) confirmed the pertinence of dentin translucency as an age indicator in the Lamendin method.

Meinl *et al.* (2008) aimed at comparing the accuracy, precision and bias of two macroscopic and one histological age at death estimation methods on human teeth. These methods included the Lamendin *et al.* (1992), Bang and Ramm (1970), and the quantification of tooth cementum annulations (TCA). They found that the Lamendin *et al.* method displayed the highest precision in the young and the old age group whereas the TCA was more precise in the middle age group. The TCA was found to be the most precise method when the precision was calculated for all ages. With their study they also found that all the methods displayed a tendency to overestimate the age in the young and underestimate in the old groups (Meinl *et al.*, 2008).

Sarajlić *et al.* (2006) obtained age estimation formulae using the length of periodontosis, transparency of the root and root height in each tooth group for their male population in Bosnia and Herzegovina. They took their results and compared them to the formulae of Lamendin and Prince and Ubelaker. The highest coefficients of correlation were obtained for the maxillary canines (R=0.731) and mandibular canines (R=0.706). Within the age groups, the lowest mean error was obtained in the 40-49 years age group (ME=5.15 years). The equations obtained in their study gave statistically significant better age estimations in comparison to the original Lamendin and Prince and Ubelaker formulae (Sarajlić *et al.*, 2006).

Chapter 3: Materials and Methods

3.1. Study sample

The sample of skeletons of known sex, age and population group, used in this study, was obtained from the Pretoria Bone Collection (University of Pretoria, South Africa) and Raymond A. Dart Collection of Human Skeletons (University of Witwatersrand, Johannesburg, South Africa).

The Pretoria Bone Collection is housed in the Department of Anatomy, Faculty of Health Sciences of the University of Pretoria. It was started in 1942 and the collection has grown to such an extent that is has become a useful resource for research. It is one of the few collections in the world that is still growing through donations and unclaimed bodies, such that it can be seen to represent the current living population of South Africa. The collection comprise of full skeletons, skulls, complete and incomplete postcranial remains. Sex, population affinity/ancestry and age-at-death of all individuals are known, and they represent the spectrum of people living in South Africa. Black males are the most common, followed by white males, white females and black females. The black population group mostly comes from the lower socio-economic classes in which the sample predominantly comprises of unclaimed bodies. The black male group also includes younger individuals. The white population group consists mainly of older individuals. They are mostly of donated individuals, and represent more of the middle and higher socio-economic groups. This leads to a poor age distribution and it is difficult to obtain younger white individuals. Many of the white individuals are so old that there is extensive tooth loss that leads to difficulty in obtaining a satisfying sample size. In the white sample there are only a few individuals in the age groups 30-39 and 40-49, but the numbers increase as age increases (above 50). The black population is well represented in all the age groups (L’Abbé *et al.*, 2005).

The School of Anatomical Sciences at the University of Witwatersrand, Johannesburg houses the Raymond A. Dart Collection of Human Skeletons (Dart Collection). This collection is one of the largest documented cadaver-derived human skeletal assemblages and was started in the early 1920s by Raymond Dart. In December 2008 the Dart Collection comprised of 2605 skeletons (76% South African black, 15% South African white, 4 % Coloured/admixed and 0.3% Indian) of which the majority is male (71%). The skeletons in this collection represent a variety of indigenous and immigrant populations from South Africa, Europe and Asia. The age of the skeletons in the collection ranges from the first year to more than 100 years of age, but the most individuals died between ages 20 to 70 (Dayal *et al.*, 2009).

The upper and lower canines of 498 skulls were measured from four groups namely black males, black females, white males and white females. Table 3.1 shows the number of maxillary and mandibular canines that were measured in each of these groups. All available males and females in the younger age categories were included.

	Maxillary canine	Mandibular canine	Total
Black males	126	142	268
Black females	53	54	107
White males	43	59	102
White females	18	42	60
Total	240	297	**537**

Table 3.1. Summary of the canines measured.

Even though this study aimed to use the left canines of both jaws, all the canines that were present were measured regardless of their side. Therefore, if the left canine was absent, the right canine was used in the measurements, calculations and statistical analyses. The age of the sample ranged from 20 to 90 years (Table 3.2), which were further divided into smaller age categories of 10 years (for age estimation). Therefore it was attempted to have at least 10 to 15 canines in each of these age categories. The reason for this minimum number of canines in each group is to have a large enough sample of canines for statistical analysis.

Age categories	20-29	30-39	40-49	50-59	60-69	70-79	80+	Total
$N_{\text{Black males}}$	31	43	70	46	47	28	3	268
$N_{\text{Black females}}$	22	19	22	23	17	3	1	107
$N_{\text{White males}}$	-	7	9	23	26	28	9	102
$N_{\text{White females}}$	4	-	4	7	12	18	15	60
Total	57	69	105	99	102	77	28	**537**

Table 3.2. Number (N) of individuals in each age category.

3.2. Methods

3.2.1. Methods of measurement: general

A total of six measurements were done, and were repeated for both sides and both jaws. These measurements comprised of three measurements for sex determination (mesiodistal and buccolingual crown diameters and intercanine distance), and three for age estimation (root height, root transparency and periodontosis). These measurements were repeated three times and recorded in an Excel spreadsheet, of which the average was used. All the measurements were taken to the nearest millimetre. The following dimensions were recorded:

- Mesiodistal and buccolingual diameters, intercanine distance and periodontosis were measured first. This was done so that the remains were not handled too much due to the fragility of the remains. If this was not possible to measure *in situ*, the tooth, if loose in the socket, was taken out and these measurements were done. This occurred when there were impacted teeth and the calliper could not fit into the spaces.
- Thereafter, the tooth was taken out, if possible, washed and then measured for root height and root transparency.

The following codes were used throughout the study:

Code	**1**	**2**	**3**	**4**	**5**	**6**	**7**	**8**
Description	Upper left canine	Upper right canine	Lower left canine	Lower right canine	Black population	White population	Male	Female

3.2.2. Sex estimation

The following measurements were taken of the maxillary and/or mandibular canine(s):

- Mesiodistal (MD) crown diameter: According to Tobias (1967) this measurement is described as the distance between two parallel lines perpendicular to the mesiodistal axial plane of the tooth. Therefore this measurement is taken tangential to the most mesial and most distal points of the crown along a line parallel to the occlusal plane. In the cases where the tooth is rotated or displaced, the end points were placed where the contact should have been. For the canines, the end points are placed at the crest of curvature on the mesial and distal surfaces (Krogman and İşcan, 1986; Kieser, 1990) (Figure 3.1).

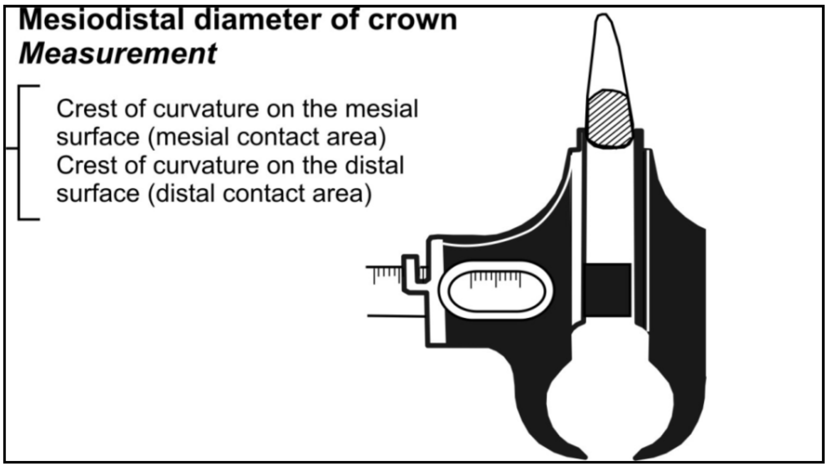


Figure 3.1: The mesiodistal diameter measured at the crest of curvature of the anterior teeth (modified from Ash and Nelson, 2003).

- Buccolingual (BL) crown diameter: This measurement is defined as the greatest distance between the buccal/labial and lingual surfaces of the crown. Therefore it is taken with the calliper held parallel to the mesiodistal axial plane of the tooth and tangential to the buccal and lingual surfaces. For canines the end points are located on the cervical third of the crown (maximum diameter) (Krogman and İşcan, 1986; Kieser, 1990) (Figure 3.2). Some authors use the term 'labiolingual' for the anterior teeth, but throughout this study the term 'buccolingual' will be used.

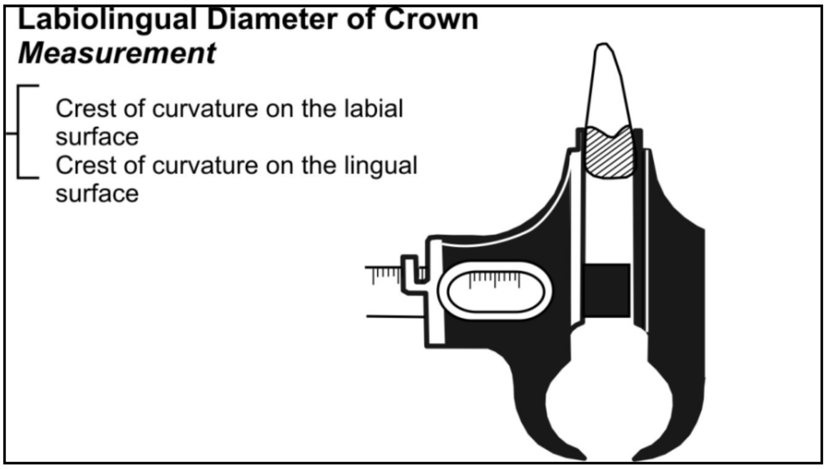


Figure 3.2: The buccolingual diameter measured at the crest of curvature of the anterior teeth (modified from Ash and Nelson, 2003).

- Intercanine distance (ID) (Maxillary/Mandibular canine arch): measured from the tip of the one maxillary/mandibular canine to the tip of the other tooth of the same jaw (Sherfudhin *et al.*, 1996; Kaushal *et al.*, 2004) (Figure 3.3).

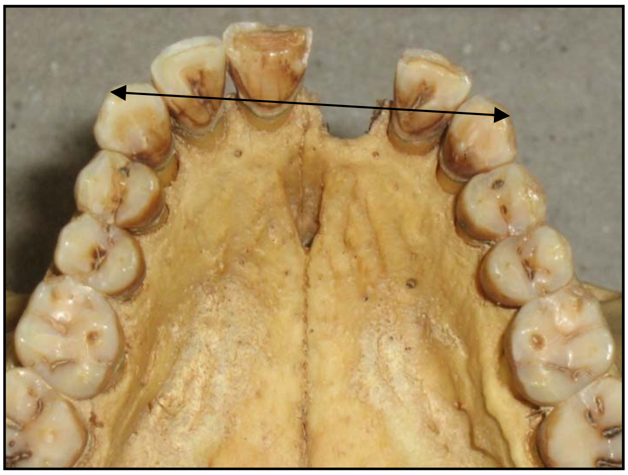


Figure 3.3: The maxillary/mandibular canine arch or intercanine distance (ID) (black arrow).

These measurements were used in further calculations and statistical analyses. In addition, a maxillary / mandibular canine index was calculated (Kaushal *et al.,* 2004):

- Maxillary/Mandibular Canine Index **(MCI) = (MD of maxillary or mandibular canine) / (maxillary or mandibular canine arch [ID]) x 100**. This feature was also used to predict the sex of the individual.

- **Standard MCI=** $(\dot{X}_{MI}-S_{MI}) + (\dot{X}_{FI}-S_{FI})/2$, where $\dot{X}_{MI}$= mean of male MCI, S_{MI}= standard deviation of male MCI, $\dot{X}_{FI}$= mean of female MCI, S_{MI}= standard deviation of female MCI, MCI (maxillary and/or mandibular canine index) = maxillary and/or mandibular canine width/maxillary and/or mandibular arch width (ID). This feature was used to compare the calculated MCI with the standard MCI of the group. If the calculated MCI is less than the standard MCI, it is classified into the female group otherwise into the male group (Sherfudhin *et al.*, 1996).

3.2.3. Age estimation

This part of the study was done following the Lamendin technique (Lamendin *et al.,* 1992) by using the maxillary and mandibular canines in the estimation of age. One of the requirements was that the teeth should be loose in the tooth socket to exclude damaging of surrounding alveolar bone. After the teeth were extracted from the alveolar bone they were rinsed with water. Much of the deposits that obscured the measurement of the teeth, specifically for the root transparency measurement, were washed off and the measurement could be done with much more certainty. The line where the root transparency begins could be seen much clearer (Figure 3.4).

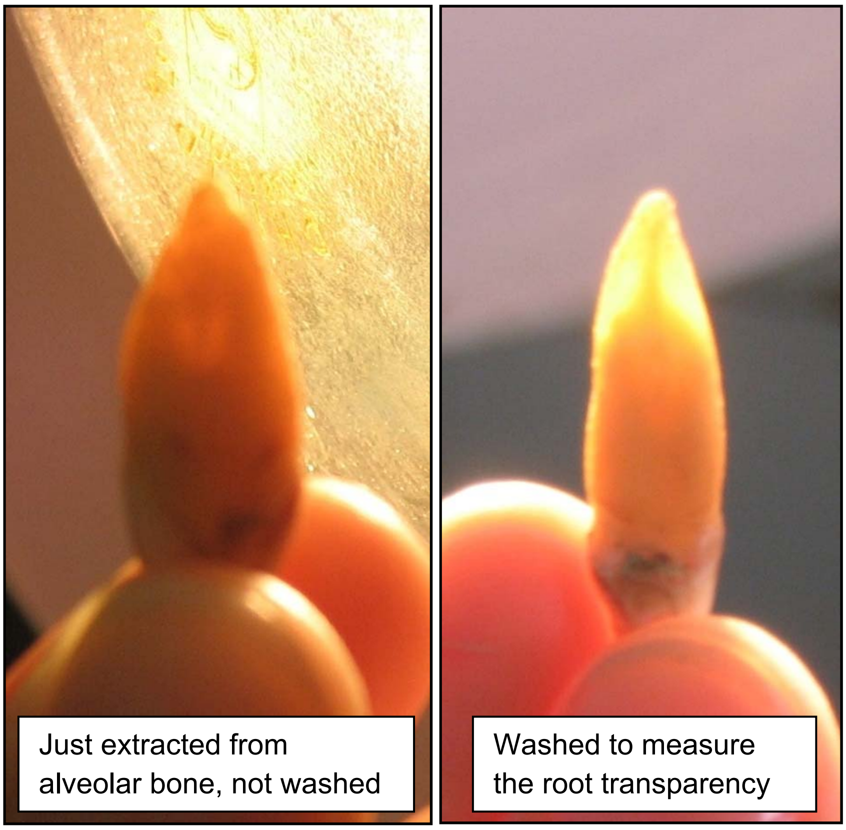


Figure 3.4. The difference between an unwashed and washed canine to measure the root transparency. The photographs were taken from a specimen from the Pretoria Bone Collection, University of Pretoria, South Africa.

The following measurements were taken on the labial surface of the teeth:

- Root height: This measurement was taken as the distance between the apex of the root and the cementoenamel junction of the canine. It is a fixed measurement and is used in multiple equations as a baseline to create standard indices for periodontal height and root transparency (Lamendin *et al.*, 1992) (Figure 3.5).

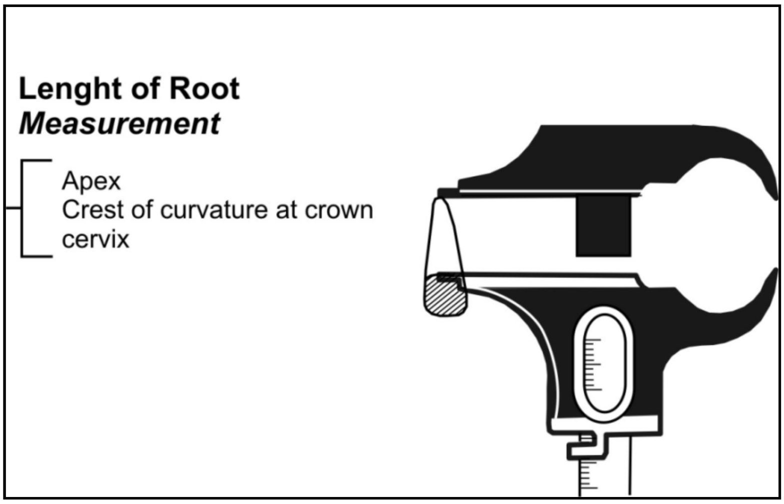


Figure 3.5: Root height of the anterior teeth (modified from Ash and Nelson, 2003).

- Root transparency: This feature is observed by viewing the tooth against a bright light source, like a light box or natural light, and measuring it from the apex of the root to the maximum height of visible transparency within the root (Lamendin *et al.*, 1992; Prince and Ubelaker, 2002) (Figure 3.6).

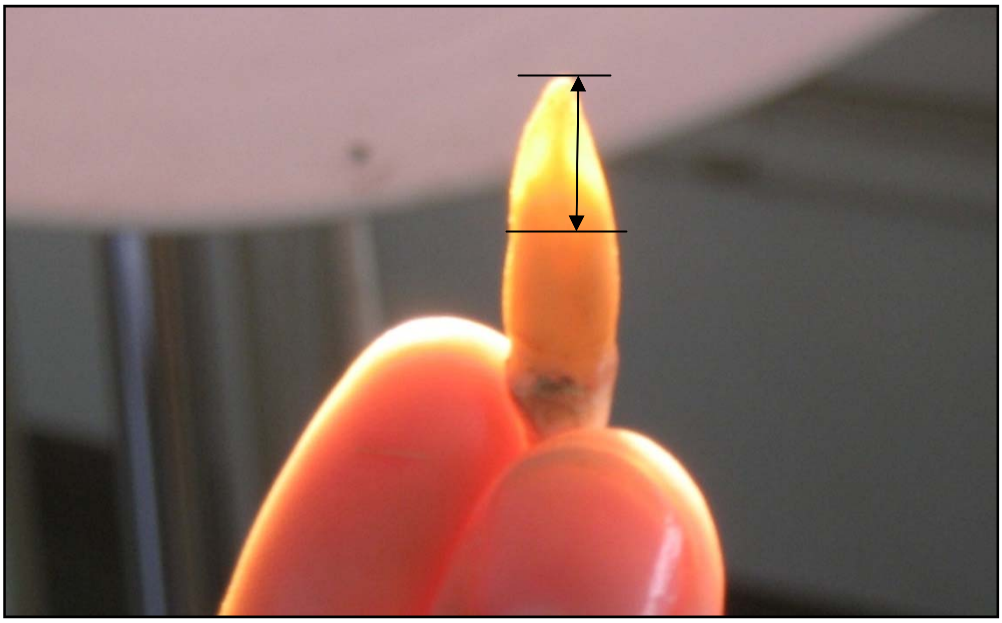


Figure 3.6: Root transparency of the canine (black arrow). The photograph was taken during this study (Pretoria Bone collection, Department of Anatomy, University of Pretoria, South Africa).

- Periodontal height or Periodontosis: This measurement is aimed at measuring the gingival tissue degeneration. It was done by recording the amount of gingival absorption as the maximum distance from the cemento-enamel junction to the line left by the soft tissue attachment on the neck and/or root of tooth (Lamendin *et al.*, 1992) (Figure 3.7).

Figure 3.7: Periodontosis (black arrows). The photograph was taken during this study (Pretoria Bone collection, Department of Anatomy, University of Pretoria, South Africa).

After all the measurements, statistical analyses were done, which include intra- and inter-observer error tests, descriptive statistics, and discriminant function analyses.

3.3. Statistical analyses

3.3.1. Intra- and Inter-observer error tests

The intra- and inter-observer error tests for repeatability of measurements were done via one-way analysis of variance (ANOVA) and the Intra Class Correlation were determined through OLS (ordinary least squares). The Intra Class Correlation (ICC) can fall in the range of greater or equal to zero or smaller or equal to one ($0 \geq ICC \leq 1$), where closer to one is the best result e.g. 100% repeatable. These tests were done by randomly selecting and remeasuring 30 sets of canines. Another researcher (inter-rater) also measured canines of 30 randomly selected crania.

3.3.2. Sex estimation

Standard descriptive statistics which include the mean, standard deviation, standard error and confidence interval for the mean of each parameter were calculated. Other statistical procedures that formed part of the study were a one-way ANOVA (analysis of variance) analysis. With one-way ANOVA one can determine whether or not two or more groups are significantly different from each other with respect to the mean of a particular variable. Therefore if the means for a variable are significantly different in different groups, then we can say that that specific variable discriminates between groups.

A study done by Rao *et al.* (1989) used the mandibular canine and stated that these measurements can be used to create an index, called the mandibular canine index (MCI). Using this index to assess the sex of an individual, they obtained accuracies of 84.3% in the male and 87.5% in the female groups respectively. This technique was implemented by this study to assess whether similar results could be obtained. However, this index did not prove to be successful with this study even when incorporated as one of the variables in discriminant function analysis. Discriminant function analyses are a more powerful statistical tool/technique to use than an index. Like other researchers (Ditch and Rose, 1972; Potter *et al.,* 2005; Acharya and Mainali, 2007; Acharya and Mainali, 2008), this study also incorporated discriminant function analyses in which four variables were used namely mesiodistal crown diameter (MD), buccolingual crown diameter (BL), intercanine distance (ID) and maxillary and/ or mandibular canine index (MCI).

The SPSS statistical package was used for the multiple discriminant function analysis. Discriminant function analysis can be used to determine which variables best discriminate between different groups (two or more) where the independent variables are the predictors and the dependent variables are the groups. As stated by Poulsen and French (2003), discriminant function analysis takes place in two steps: (1) to test the significance of a set of discriminant functions, and; (2) the classification process. In the first step a matrix of total variances and co-variances as well as a matrix of pooled within-group variances and co-variances are created. These two matrices are compared by means of multivariate F tests so as to determine whether or not there are any significant differences (with reference to all variables) between the groups. Therefore, after the multivariate test proves to be statistically significant, one can proceed to the next step to see which variables have significantly different means across the groups (classification of variables). Discriminant function analysis automatically determines optimal combinations of variables so that the first function provides the most overall discrimination between the groups, the second function the second most and etc. The contributions of the functions will not overlap, meaning the first function will pick up the most variation; the second function will pick up the greatest part of the unexplained variation, etc. The canonical correlation analysis, obtained computationally, is performed to determine the successive functions and canonical roots. Thereafter, classification from these canonical functions is possible. The subjects are classified in the groups in which they had the highest classification scores. The maximum number of discriminant functions will be equal to the degrees of freedom or the number of the variables in the analysis; whichever is smaller (Poulsen and French, 2003).

The discriminant functions are interpreted by means of standardized coefficients and the structure matrix. The standardized beta coefficients are given for each variable in each discriminant function. The larger the standardized coefficient, the greater is the contribution of the respective variable to the discrimination between the groups. In summary the means for the significant discriminant functions are examined in order to determine between which groups the respective functions seem to discriminate (Poulsen and French, 2003). One of the ways to test the significance is the Wilks' lambda. The smaller lambda is for an independent variable, the more that variable contributes to the discriminant function. Lambda varies from 0 to 1, with 0 meaning that the group means differ and 1 meaning that all the group means are the same. The F test of the Wilks' lambda will show which variables' contributions are significant (Poulsen and French, 2003).

Discriminant function analysis is done by selecting relevant features on the tooth (variables) to create a function. The variables for this part of the study were mesiodistal and buccolingual crown diameters as well as the intercanine distance. Discriminant function analysis was started by entering all the measurements into a stepwise discriminant function procedure using the Wilks' lambda to determine which variable provided the best discrimination between the sexes (with F=3.84 to enter and F=2.71 to remove). The stepwise analysis incorporates all the dental measurements that are systematically added and removed from the list. After the first variable is selected it is removed from the analysis and the remaining variables are reassessed and selected. The same analysis was done on combinations of measurements – known as functions 2 (mesiodistal and buccolingual crown diameters) and 3 (MCI). At the same time a direct discriminant function analysis was done to produce a demarking point between the sexes for each individual measurement. Discriminant function analysis assigns a sex to each case within the sample, which is based on whether the discriminant score is above or below the sectioning point (e.g. discriminant score greater than sectioning point, the individual most likely a male). This will give you some idea of the accuracies of how many individuals are correctly classified according to sex, but this will not necessarily reflect the accuracies in the general population. By incorporating a 'leave one out' classification procedure into the study, one measures the effectiveness of the functions. This classifies each individual canine by the functions derived from all the cases other than that case itself. Therefore, the accuracy of assignments to either male or female categories is cross validated.

In the calculation of the discriminant score, each dimension used in a particular function is multiplied by its unstandardized coefficient, which weights the variable according to its contribution to the sex differences. These values are then added together, which is followed by the addition of the constant. This constant value serves to calibrate the sectioning point to zero in case of unequal number of cases in the groups. In a case where the sample sizes differ, the sectioning point is calculated by averaging the two group centroids. After this the calculated discriminant score can be compared with the sectioning point (Patriquin *et al.,* 2002).

Throughout this study the black and white populations were kept separate due to a number of studies based on discriminant function analyses reporting population specificity.

3.3.3. Age estimation

The measurements for age estimation were subjected to statistical analyses which also included intra- and inter-observer error tests. For this purpose the STATA package was used.

Firstly, the root transparency and periodontal height were expressed as functions of root height (the average of each variable was substituted into the original Lamendin formula) and these were correlated against the real age and the calculated age by means of scatter plots. From these, correlation factors were obtained. Then, these variables were regressed as all four canines per individual pooled to calculate the coefficients of the three parameters. The calculated parameters were substituted into the Lamendin formula to obtain an age estimate for each individual in the sample. Lamendin's formula is as follows: **$A = (0.18 \times P) + (0.42 \times T) + 25.53$**; where A = age in years, P = (periodontal height x 100)/root height, T = (transparency x 100)/root height (Lamendin *et al.*, 1992). These estimated ages did not correlate well with the real ages of the individuals in the sample. From this it was decided that specifications were to be made to make the method more specific for the sample of this study as well as the aim to obtain better results.

Each individual were represented by 2 canines – one for the upper jaw and one for the lower jaw, irrespective of left or right. The specifications were for the first part done on the whole sample with just the periodontosis and root transparency measurements as determinants of age (scatter plots and correlation factors). This resembles what Lamendin *et* *al.* (1992) used in their study to estimate the age of the individuals in their study. Thereafter specifications were made as to see whether better results would be obtained. These include specific canine, then specific canine and sex and lastly specific canine, sex and population group (represented by means of scatter plots and correlation factors). These statistics were done for all four groups in the sample, which included correlation coefficients for each specification. Therefore, different formulae were created based on the specifications made.

Regression statistics were used to plot variables against known data to see whether there is a correlation between them as well as plotting the calculated age against the real age. If a correlation exists, it is possible to create coefficients for each variable in the process of adapting a formula to fit the purpose of the study. It was done in this study since the coefficients of the parameters in the Lamendin formula are specific for the sample used in his study. Population specific formulae were thus created.

Chapter 4: Results - Sex estimation

4.1. Introduction

The sample for this part of the study comprised of canines from 439 crania. The mesiodistal and buccolingual crown diameters as well as the intercanine distance were measured. These measurements were repeated three times. The mean of the three measurements of each variable (mesiodistal and buccolingual diameters and intercanine distance) and the maxillary and/or mandibular canine index (MCI) were calculated and these were used in statistical analyses. For the purpose of this part of the study, descriptive statistics were calculated separately for each group (black males, black females, white males, white females) and for each canine (canine 1=upper left canine, canine 2=upper right canine, canine 3=lower left canine, canine 4=lower right canine). If no clear differences were found between left and right sided canines, one canine could represent each jaw (preferably the left canine). If the left was not available the right canine could then be used instead. The two population groups were also kept separate.

4.2. Intra- and inter-observer error tests

The intra- and inter-observer error tests for repeatability of measurements were done via one-way analysis of variance (ANOVA) and the Intra Class Correlation were determined through OLS (ordinary least squares). These results can be found in Table 4.1. The Intra Class Correlation (ICC) can fall in the range of greater or equal to zero or smaller or equal to one ($0 \geq ICC \leq 1$), where closer to one is the best result e.g. 100% repeatable. These tests were done by randomly selecting and remeasuring 30 sets of canines (two canines per individual, e.g. canine 1 (upper canine) and canine 3 (lower canine)). Another researcher (inter-rater) also measured canines of 30 randomly selected crania.

In the Table 4.1, it can be seen that the process of measuring (repeatability) in the study was found reliable. The R-squared values and ICC of the principal investigator (intra-rater) were all close to 1, which means the measurement method is reproducible. Somewhat poorer results were obtained when measurements of the primary researcher were compared to that of the other researcher (inter-rater). Almost all R-squared values were above 0.8, but the ID measurements for the lower canines were somewhat problematical with ICC values of 0.675.

Intra class correlation (ICC)									
Canine	**Variable**	**N**	**Intra rater**	**95% CI**	**R^2**	**N**	**Inter rater**	**95% CI**	**R^2**
1	MD	60	0.998	0.996-0.999	0.9989	60	0.960	0.933-0.989	0.9798
	BL	60	0.998	0.998-0.999	0.9995	60	0.979	0.965-0.994	0.9895
	ID	60	0.998	0.996-0.999	0.9989	60	0.888	0.811-0.964	0.9421
2	MD	60	0.998	0.996-0.999	0.9989	60	0.977	0.961-0.994	0.9882
	BL	60	0.999	0.998-0.999	0.9995	60	0.979	0.964-0.994	0.9892
	ID	60	0.998	0.996-0.999	0.9989	60	0.888	0.811-0.964	0.9421
3	MD	58	0.998	0.996 - 0.999	0.9990	58	0.967	0.943-0.990	0.9829
	BL	58	0.997	0.996-0.999	0.9988	58	0.967	0.942-0.990	0.9828
	ID	58	0.999	0.998-0.999	0.9995	58	0.675	0.475-0.875	0.8329
4	MD	60	0.997	0.996-0.999	0.9987	60	0.967	0.944-0.991	0.9832
	BL	60	0.998	0.997-0.999	0.9993	60	0.961	0.934-0.988	0.9801
	ID	58	0.999	0.998-0.999	0.9995	58	0.675	0.475-0.875	0.8329

Table 4.1. Intra- and Inter-observer error tests. N=number of observations, R^2=R-squared.

4.3. Descriptive statistics

The total number of teeth measured per population group can be seen in Table 4.2. The descriptive statistics shown include the mean, standard deviation and standard error (SE). The standard deviation (SD) is the combined measure of the distances of the observations from their mean. The empirical rule for a fairly typical distribution of observations is that it is usually expected to find approximately 68% of the observations within the ±1SD of the mean; approximately 95% of the observations within the ±2SDs of the mean and >99% of the observations within the ±3SDs of the mean (Samuels and Witmer, 2003).

Table 4.2 compares the dimensions of the SA Blacks and Whites. It is evident that they exhibit some differences between the similar sexes of the different population groups, with SA Blacks generally having larger teeth than SA Whites. Statistically significant differences exist between the SA Blacks and Whites with reference to all the crown diameters, intercanine distances and indices, except for the BL measurement and index of the mandibular canine (significant at p-value <0.05). Following these results, it was decided to keep the two populations separate in all subsequent analyses. All the measurements except the MCI (mandibular canine) of the SA Black group; were relatively larger than those of the SA White group.

SA Blacks					**SA Whites**					**ANOVA**
	N	**Mean**	**SD**	**SE**	**Maxilla**	**N**	**Mean**	**SD**	**SE**	**p-value**
Maxilla (C1)										
MD	93	8.01	0.486	0.050	MD	46	7.84	0.328	0.048	0.026*
BL	93	8.82	0.561	0.058	BL	51	8.58	0.507	0.071	0.011*
ID	93	36.99	3.045	0.316	ID	38	34.69	2.806	0.455	0.000*
MxCI	93	21.78	1.863	0.193	MxCI	32	22.71	1.891	0.334	0.017*
Mandible (C3)										
MD	93	7.38	0.431	0.045	MD	54	6.93	0.384	0.052	0.000*
BL	93	8.18	0.457	0.047	BL	67	8.16	0.436	0.053	0.750
ID	93	27.65	2.607	0.270	ID	54	26.78	2.404	0.327	0.047*
MaCI	93	26.87	2.589	0.268	MaCI	49	26.04	2.327	0.332	0.062

Table 4.2. The descriptive statistics for South African blacks and whites. N=number of individuals, SD=standard deviation, SE=standard error, MD=mesiodistal diameter, BL=buccolingual diameter, ID=intercanine distance, MxCI=maxillary canine index, MaCI=mandibular canine index, C1=upper canine, C3=lower canine. *significant at $p < 0.05$.

The following tables (Tables 4.3 and 4.4) show the descriptive statistics for the males and females of each group.

SA blacks	**Male**				**Female**				**ANOVA**
	N	**Mean**	**SD**	**SE**	**N**	**Mean**	**SD**	**SE**	**p-value**
Maxilla									
MD C1	88	8.03	0.476	0.051	71	7.50	0.479	0.057	0.000*
MD C2	88	7.98	0.453	0.048	76	7.41	0.447	0.051	0.000*
BL C1	88	8.84	0.542	0.058	72	8.14	0.590	0.070	0.000*
BL C2	88	8.87	0.515	0.055	77	8.12	0.541	0.062	0.000*
ID	88	37.02	3.098	0.330	67	35.19	2.898	0.354	0.000*
MxCI C1	88	21.81	1.871	0.200	65	21.29	1.801	0.223	0.09
MxCI C2	88	21.69	1.868	0.199	66	21.15	1.838	0.226	0.077
Mandible									
MD C3	88	7.36	0.432	0.046	77	6.80	0.395	0.045	0.000*
MD C4	88	7.39	0.442	0.047	72	6.79	0.395	0.047	0.000*
BL C3	88	8.16	0.454	0.048	81	7.46	0.473	0.053	0.000*
BL C4	88	8.13	0.477	0.051	79	7.42	0.424	0.048	0.000*
ID	88	27.71	2.581	0.275	77	26.92	2.666	0.304	0.056
MaCI C3	88	26.78	2.555	0.272	73	25.47	2.442	0.286	0.001*
MaCI C4	88	26.84	2.549	0.272	70	25.48	2.369	0.283	0.001*

Table 4.3. The descriptive statistics for the South African black group. N=number of individuals, SD=standard deviation, SE=standard error, MD=mesiodistal diameter, BL=buccolingual diameter, ID=intercanine distance, MxCI=maxillary canine index, MaCI=mandibular canine index, C1=upper left canine, C2=upper right canine, C3=lower left canine, C4=lower right canine. *significant at $p < 0.05$.

From Table 4.3 it can be seen that black males were larger than females in all measurements of the maxillary canines. The maxillary canine indices, however, show no significant differences. The pattern differs for the canines of the lower jaw, however, where there are significant differences between the measurements of the canines themselves as well as the indices, but not for the intercanine distance.

The males of the SA white group were also larger than the females in all measurements for upper and lower canines. This can be seen in Table 4.4. All the measurements were statistically significantly different except for the maxillary and mandibular canine indices.

SA whites	Male				Female				ANOVA
	N	Mean	SD	SE	N	Mean	SD	SE	p-value
Maxilla									
MD C1	36	7.83	0.335	0.056	32	7.53	0.379	0.067	0.001*
MD C2	42	7.80	0.324	0.050	36	7.53	0.411	0.069	0.002*
BL C1	40	8.57	0.504	0.080	33	7.98	0.502	0.087	0.000*
BL C2	44	8.58	0.604	0.091	37	8.00	0.489	0.080	0.000*
ID	36	34.75	2.870	0.478	34	32.03	3.301	0.566	0.000*
MxCI C1	28	23.72	2.178	0.345	28	23.72	2.178	0.412	0.059
MxCI C2	35	22.74	1.959	0.331	33	23.63	2.179	0.380	0.080
Mandible									
MD C3	46	6.90	0.364	0.054	51	6.51	0.384	0.054	0.000*
MD C4	49	6.88	0.318	0.046	53	6.49	0.341	0.047	0.000*
BL C3	51	8.15	0.431	0.061	54	7.39	0.450	0.061	0.000*
BL C4	51	8.10	0.412	0.058	54	7.36	0.467	0.064	0.000*
ID	48	26.65	2.165	0.313	50	25.27	2.379	0.337	0.004*
MaCI C3	45	26.00	2.296	0.342	47	25.89	2.393	0.349	0.853
MaCI C4	46	25.96	2.254	0.332	49	25.81	2.330	0.333	0.759

Table 4.4. The descriptive statistics for the South African white group. N=number of individuals, SD=standard deviation, SE=standard error, MD=mesiodistal diameter, BL=buccolingual diameter, ID=intercanine distance, MxCI=maxillary canine index, MaCI=mandibular canine index, C1=upper left canine, C2=upper right canine, C3=lower left canine, C4=lower right canine. *significant at $p<0.05$.

From Tables 4.3 and 4.4 it can be seen that the number of individuals (n) representing the white population was less than that of the black population which was well represented. This is because the sources of skeletal remains (bone collections) have many black males and the whites in the collections were from older individuals (less teeth due to dentures, for example). However, for all direct dental measurements the sample size was above 30, which is a large enough sample to use for subsequent discriminant function analysis.

From these tables it is clear that all direct measurements of the males are relatively larger (statistically significant at $p<0.05$) than those of the females, making it possible to use discriminant function analysis in an attempt to develop formula which can be used to estimate sex in unknown individuals. The indices are clearly less usable. In addition, it seems that there are no clear differences in tooth sizes between left and right sided canines, and therefore in each individual only the left sided canine was used in subsequent analysis, and if absent it was replaced by values from the right sided tooth.

4.4. Discriminant function analysis

The SPSS statistical program was used and three discriminant functions were calculated for the upper and lower jaws respectively, for each population separately. Only the left canines (or if absent, substituted by the right) were used:

- Function 1: All three diameters (Mesiodistal diameter, MD; Buccolingual diameter, BL; and Intercanine distance (ID) were entered in a stepwise analysis.
- Function 2: Only the Mesiodistal and Buccolingual diameters of a specific canine were entered using a direct approach. This can then be used if only a loose tooth is available.
- Function 3: Only the maxillary and/or mandibular canine index was used in a direct approach.

Tables 4.5 and 4.6 display the results of the discriminant function analyses in the South African black and white groups respectively, for the upper jaw. This includes the standard and structure coefficients as well as the unstandardized coefficients. For Function 1 of both populations the buccolingual diameter was selected first, followed by MD in the black group only. ID did not feature in the functions of any of two groups. For both groups, in Functions 1 and 2, values lower than the sectioning point would indicate a female individual and vice versa. However, for Function 3 in blacks a value lower than the sectioning point will also indicate a female, but the opposite is true in whites. This indicates that blacks have a relatively wider upper dental arch than whites. In all these functions the sectioning point is not equal to zero, as sample sizes of males and females differ.

Canonical Discriminant Function analysis of Canine 1 (maxilla) for SA blacks				
Functions and variables (mm)	**Standard coefficients**	**Structure matrix**	**Unstandardized coefficients**	**Centroids**
Function 1 (BL, MD, ID)				
BL	0.644	0.904	1.162	Male= 0.643
MD	0.501	0.835	1.086	Female= -0.870
Constant			-18.358	
Sectioning point			-0.1135	
Function 2 (MD, BL)				
MD	0.454	0.826	0.951	Male=0.598
BL	0.675	0.925	1.197	Female=-0.742
Constant			-17.634	
Sectioning point			-0.072	
Function 3 (MCI)				
MCI	1.000	1.000	0.543	Male=0.119
Constant			-11.722	Female=-0.161
Sectioning point			-0.021	

Table 4.5. Canonical discriminant function coefficients for maxillary canines in South African blacks. Function 1 was derived by means of a stepwise analysis, all others by a direct analysis.

Canonical Discriminant Function analysis of Canine 1 (maxilla) for SA whites				
Functions and variables (mm)	**Standard coefficients**	**Structure matrix**	**Unstandardized coefficients**	**Centroids**
Function 1 (BL, MD, ID)				
BL	1.000	1.000	1.925	Male=0.538
				Female=-0.596
Constant			-16.062	
Sectioning point			-0.029	
Function 2 (MD, BL)				
MD	0.160	0.692	0.449	Male=0.533
BL	0.897	0.992	1.741	Female=-0.619
Constant			-17.952	
Sectioning point			-0.043	
Function 3 (MCI)				
MCI	1.000	1.000	0.489	Male=-0.238
Constant			-11.325	Female=0.263
Sectioning point			-0.0125	

Table 4.6. Canonical discriminant function coefficients for maxillary canines in South African whites. Function 1 was derived by means of a stepwise analysis, all others by a direct analysis.

Tables 4.7 and 4.8 display the discriminant function analyses for the lower jaw in both SA blacks and whites. For Function 1 the buccolingual diameter was selected first, followed by MD in the black group only. ID was not selected in any of the functions. The measurement chosen first carries the most weight in the formula in determining the differences between the sexes. For both groups, all values lower than the sectioning point would indicate a female individual and vice versa.

Canonical Discriminant Function analysis of Canine 3 (mandible) for SA blacks				
Functions and variables (mm)	**Standard coefficients**	**Structure matrix**	**Unstandardized coefficients**	**Centroids**
Function 1 (BL, MD, ID - Stepwise)				
BL	0.657	0.933	1.451	Male=0.758
MD	0.454	0.853	1.103	Female=-0.914
Constant			-19.220	
Sectioning point			-0.078	
Function 2 (MD, BL - Direct)				
MD	0.440	0.852	1.058	Male=0.747
BL	0.666	0.938	1.457	Female=-0.854
Constant			-18.943	
Sectioning point			-0.0535	
Function 3 (MCI - Direct)				
MCI	1.000	1.000	0.399	Male=0.237
Constant			-10.454	Female=-0.286
Sectioning point			-0.0245	

Table 4.7. Canonical discriminant function coefficients for mandibular canines in South African blacks. Function 1 was derived by means of a stepwise analysis, all others by a direct analysis.

Canonical Discriminant Function analysis of Canine 3 (mandible) for SA whites				
Functions and variables (mm)	**Standard coefficients**	**Structure matrix**	**Unstandardized coefficients**	**Centroids**
Function 1 (BL, MD, ID)				
BL	1.000	1.000	2.290	Male=0.924 Female=-0.884
Constant			-17.730	
Sectioning point			0.02	
Function 2 (MD, BL)				
MD	-0.014	0.593	-0.038	Male=0.910
BL	1.009	1.000	2.227	Female=-0.821
Constant			-17.028	
Sectioning point			0.0445	
Function 3 (MCI)				
MCI	1.000	1.000	0.426	Male=0.020
Constant			-11.054	Female=-0.019
Sectioning point			0.0005	

Table 4.8. Canonical discriminant function coefficients for mandibular canines in South African whites. Function 1 was derived by means of a stepwise analysis, all others by a direct analysis.

Overall, the buccolingual diameter provides the most discrimination between the sexes. The bigger the value of that specific coefficient, the greater is the contribution of that measurement to the discrimination process. Therefore, statistically it was found that the buccolingual diameter was the best (first choice) discriminator between the sexes.

In order to use these formulae, the specific measurement needs to be multiplied to the unstandardized coefficient. These values are then added together, along with the constant, to obtain the discriminant score. The discriminant score is then compared to the sectioning point, if higher than the sectioning point it represents a male whereas a lower score would indicate a female. For example, using Function 2 from Table 4.7:

BL = 8.69 mm and MD = 6.59 mm (mandibular canine, black individual).

= (6.59 x 1.058) + (8.69 x 1.457) – 18.943

= (6.97222 + 12.66133) – 18.943

= + 0.69055

This value is more than the sectioning point of -0.0535, indicating a male individual. The further away the discriminant score is from the sectioning point, the more accurate the estimation.

The accuracies and cross-validation results of the blacks and whites are shown in Tables 4.9 and 4.10, respectively. In general accuracies above 80% are considered good and usable. Included in the tables are also the cross-validation results. Cross-validation or the ‘’leave one out’’ classification measures the effectiveness of a function. This will classify each canine by the functions derived from all cases other than that case itself. This process will continue for all the individual canines until all are tested, thus the accuracy of assignments to either male or female categories is cross-validated.

For the black population relatively good results were obtained (Table 4.9). For the maxillary canine, the best results were obtained using Function 1 (accuracies of 78.4% and 76.1% for males and females respectively), closely followed by Function 2. Function 3 yielded low accuracies just above chance. In the mandible, both Functions 1 and 2 yielded good results approaching 80%, followed by Function 3. Males and females were classified at about equal levels of accuracy, and no clear biases were observed. None or only one individual was lost during cross-validation.

Correct classification for blacks					
Functions	Males		Females		Average accuracy
	Count	%	Count	%	
Canine 1 (maxilla)					
Function 1					
Original	69/88	78.4	54/71	76.1	77.3
Cross-validated	68/88	77.3	54/71	76.1	76.7
Function 2					
Original	66/88	75.0	54/71	76.1	75.6
Cross-validated	65/88	73.9	54/71	76.1	75.0
Function 3					
Original	48/88	54.5	34/65	52.3	53.4
Cross-validated	48/88	54.5	34/65	52.3	53.4
Canine 3 (mandible)					
Function 1					
Original	68/88	77.3	62/77	80.5	78.9
Cross-validated	68/88	77.3	61/77	79.2	78.3
Function 2					
Original	67/88	76.1	63/77	81.8	79.0
Cross-validated	67/88	76.1	62/77	80.5	78.3
Function 3					
Original	54/88	61.4	49/73	67.1	64.3
Cross-validated	54/88	61.4	49/73	67.1	64.3

Table 4.9. Percentage accuracy and cross-validation for the derived formulae in blacks.

Correct classification for whites					
Functions	Males		Females		Average accuracy
	Count	%	Count	%	
Canine 1 (maxilla)					
Function 1					
Original	26/40	65.0	24/33	72.7	68.9
Cross-validated	25/40	62.5	24/33	72.7	67.6
Function 2					
Original	24/36	66.7	22/31	71.0	68.9
Cross-validated	24/36	66.7	21/31	67.7	67.2
Function 3					
Original	24/31	77.4	16/28	57.1	67.3
Cross-validated	24/31	77.4	16/28	57.1	67.3
Canine 3 (mandible)					
Function 1					
Original	43/51	84.3	47/54	87.0	85.7
Cross-validated	43/51	84.3	46/54	85.2	84.8
Function 2					
Original	38/46	82.6	44/51	86.3	84.5
Cross-validated	38/46	82.6	44/51	86.3	84.5
Function 3					
Original	27/45	60.0	22/47	46.8	53.4
Cross-validated	25/45	55.6	20/47	42.6	49.1

Table 4.10. Percentage accuracy and cross-validation for the derived formulae in whites.

Accuracies for the maxilla in the white population were considerably lower, while dimensions in the mandibular teeth proved to be more dimorphic (Table 4.10). Using the maxillary canine, the males obtained the highest classification percentage with Function 3 (77.4%), followed by Function 2 and then Function 1. However, the average accuracy for Function 3 was only 67.3%, indicating a clear bias. The females obtained the highest classification percentage with Function 1 (72.7%), followed by Function 2 and then Function 3. With the mandibular canine, both males and females obtained the highest classification accuracies with Function 1 (84.3% and 87.0% respectively). This was followed by Function 2 and then Function 3 which could not separate the sexes at all. In general, few individuals were misclassified during cross-validation.

Overall, mandibular teeth in white individuals gave the best results. In general Functions 1 and 2 performed equally, indicating that the addition of the measurement ID did not contribute much to distinguish males from females. It was also never selected for calculating Function 1. In black individuals the MD and BL dimensions in both upper and lower jaws thus have potential to separate the sexes. In whites, however, only dimensions of the mandible showed good results. The indices made no contribution at all.

Chapter 5: Results – Age estimation

5.1 Introduction

In this part of the study there were three measurements that were used on all four canines, if present, in the estimation of age, namely root height (RH), periodontosis (P) and root transparency (RT). These measurements were repeated three times and subjected to inter-and intra-observer error tests. The mean of each measurement was used in further analyses. These measurements were also used in the Lamendin study (Lamendin *et al.*, 1992) and were subjected to further statistical analyses using the STATA package. The preference was to measure the left canines of both jaws, but if it was not present it was substituted by the right canine of that specific jaw. The sample for this part of the study comprised of canines from 436 crania and was divided into four groups (black males, black females, white males and white females. It was attempted to have at least ten to fifteen individuals of each group in each age category (20-29, 30-39, 40-49, 50-59, 60-69, 70-79, and 80+), but this was not always possible. The sample sizes for this part of the study are shown in Table 5.1. As can be seen from this table, sample size for the black males was good, whereas especially that of the white female group was small.

Age categories	**20-29**	**30-39**	**40-49**	**50-59**	**60-69**	**70-79**	**80+**	**Total**
$N_{\text{Black males}}$	31	43	70	46	47	28	3	268
$N_{\text{Black females}}$	22	19	22	23	17	3	1	107
$N_{\text{White males}}$	-	7	9	23	26	28	9	102
$N_{\text{White females}}$	4	-	4	7	12	18	15	60
Total	57	69	105	99	102	77	28	**537**

Table 5.1. Samples in each age category.

This part of the study was divided into three steps. In the first step the measurements were correlated against age of the individuals. This was done to see whether there was any correlation between the measurements and the age of the individuals. In the second step, the three measurements namely root height, periodontosis and root transparency, were substituted into Lamendin's formula, to estimate the age of skeletal remains. These estimated ages were then plotted against the real ages in order to assess how well the Lamendin's formula predicted the ages of all individuals. Lastly, based on these results, a decision had to be made whether it was necessary to adapt the formulae to assess whether better results could be obtained.

5.2. Intra- and Inter-observer error tests

The intra- and inter-observer error tests for age estimation were done via one-way analysis of variance (ANOVA) and the interclass correlation was determined through OLS (ordinary least squares). These results can be found in Table 5.2. Intra-class correlations (ICC) range between 0 and 1, with 0 indicating no correlation between the two sets of measurements and 1 indicating complete agreement. ICC values above 0.9 usually indicate excellent repeatability. The intra-rater values indicate the reliability with which the principal investigator could measure the three parameters used in this part of the study. All the values were above 0.9, indicating high repeatability. The inter-observer repeatability indicates how well the measurements between the principal investigator and another observer could be repeated. All of these values were above 0.8, except for the periodontosis, which indicates that these measurements were less easy to record reliably.

Intra class correlation (ICC)

Canine	Variable	N	Intra rater	95% CI	R^2	N	Inter rater	95% CI	R^2
1	RH	58	0.998	0.997-0.999	0.9991	58	0.958	0.927-0.988	0.9781
	P	58	0.999	0.999-1.000	0.9999	58	0.794	0.659-0.929	0.8938
	RT	58	0.999	0.999-1.000	1.0000	58	0.822	0.704-0.941	0.9085
2	RH	58	0.999	0.999-1.000	0.9997	58	0.976	0.958-0.993	0.9875
	P	58	0.999	0.997-0.999	0.9993	58	0.720	0.543-0.897	0.8559
	RT	58	0.999	0.999-1.000	1.0000	58	0.839	0.730-0.948	0.9171
3	RH	58	0.999	0.998-0.999	0.9996	58	0.922	0.867-0.977	0.9595
	P	58	0.999	0.999-1.000	0.9998	58	0.446	0.152-0.740	0.7161
	RT	58	0.999	0.999-1.000	1.0000	58	0.820	0.699-0.940	0.9071
4	RH	60	0.999	0.999-1.000	0.9997	60	0.921	0.865-0.976	0.9589
	P	60	0.999	0.999-1.000	0.9999	60	0.716	0.541-0.892	0.8540
	RT	60	0.999	0.999-1.000	1.0000	60	0.804	0.675-0.931	0.8988

Table 5.2. Intra- and inter-observer error tests. N=number of observations, R^2=R-squared, 95% CI=95% confidence interval.

5.3. Step 1: Correlation between actual age and observed parameters

Each of the measurements was plotted against the real age of the individual to assess whether there was either a positive or negative correlation with age. The mean of the three measurements for root height, periodontosis and root transparency, of each individual in the four groups (black males, black females, white males and white females), were used and a correlation coefficient (R^2) was calculated for each. Canines 1 and 2 refer to the canine of the upper jaw (left or right) and canine 3 and 4 the canines of the lower jaw. The regressions of each measurement from the upper and lower jaws for each group can be seen in the following section. The correlation coefficient is a mathematical measure of how much one variable can be expected to be influenced by changes in another. A correlation coefficient of 1 indicates that the two variables are perfectly correlated. A correlation coefficient of zero indicates that the two variables are not related. Therefore, the closer the correlation coefficient is to zero the smaller the relationship, and low correlation coefficients mean that the relationship is not significant enough to be useful.

5.3.1. Root height

5.3.1.1. Black males

The average root height measurement was plotted against the real age of each individual to assess whether there is any correlation between them. As can be seen from Figure 5.1 which shows both the upper and lower canines, this resulted in an R-squared value of 0.003, indicating that only 0.3% of the real age could be explained by root height. This indicates that there is a very weak, positive correlation between root height and age for both the maxilla and mandible.

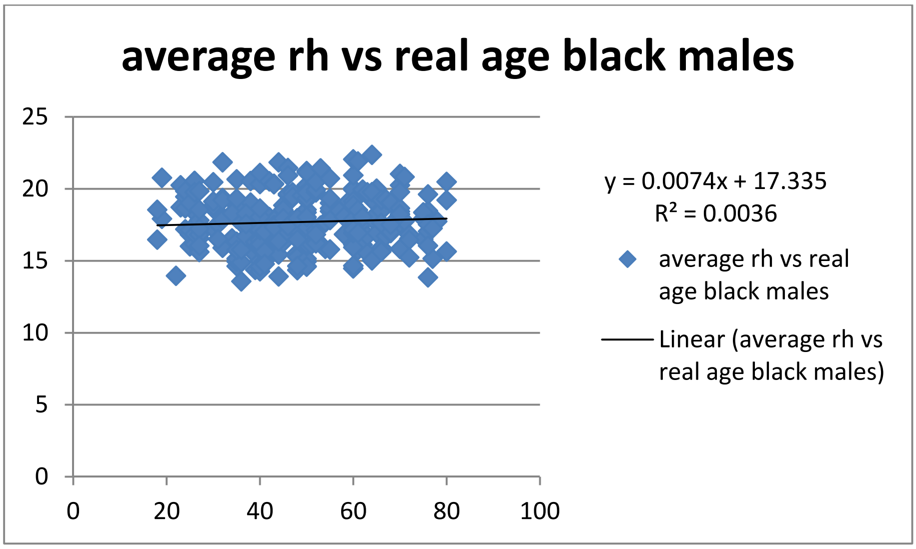


Figure 5.1. Average root height (rh) correlated against real age for black males, all canines. R^2=correlation coefficient. X-axis=real age, Y-axis=average root height (rh).

If the correlation between the mean root height of a specific canine of each jaw to the real age is assessed, the same pattern can be seen (Figures 5.2 and 5.3).

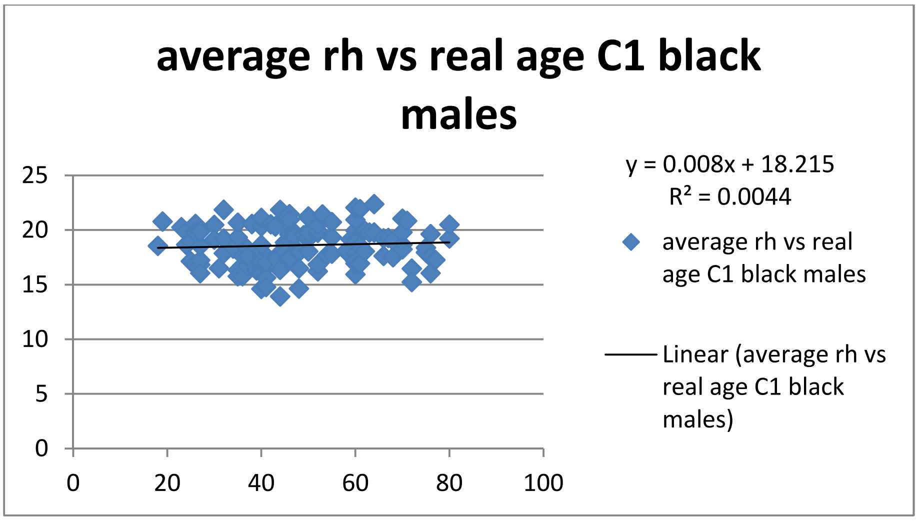


Figure 5.2. Average root height (rh) correlated against real age for canine 1 (black males). R^2=correlation coefficient. X-axis=real age, Y-axis=average root height (rh).

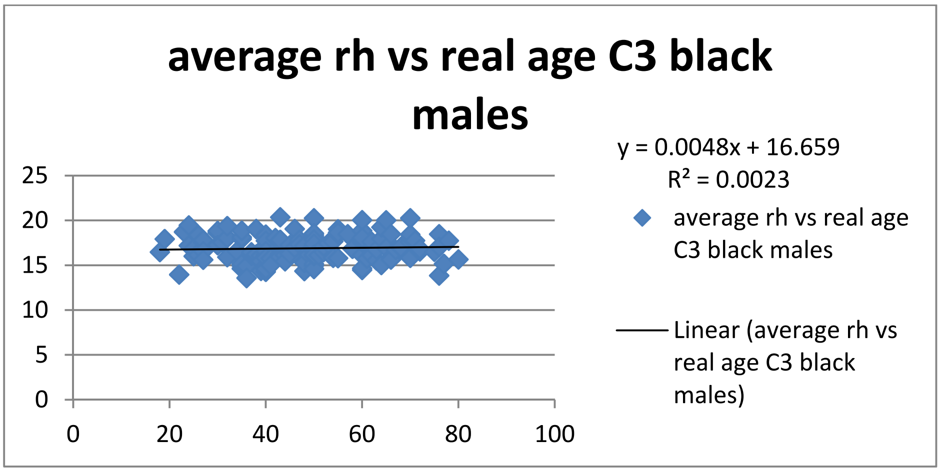


Figure 5.3. Average root height (rh) correlated against real age for canine 3 (black males). R^2=correlation coefficient. X-axis=real age, Y-axis=average root height (rh).

From this it is evident that root heights change little with age. These results are not surprising, as root height in itself has not been reported to change with age. As root height is used as a parameter against which transparency and periodontosis is judged, this implies that it is a stable characterisitc against which to judge the transparency and periodontosis.

5.3.1.2. Black females

The average root height of the black females was also plotted against the real age (Figure 5.4). A negative correlation coefficient of R^2=0.016, indicated that only 1.6% of the age could be explained by the root height.

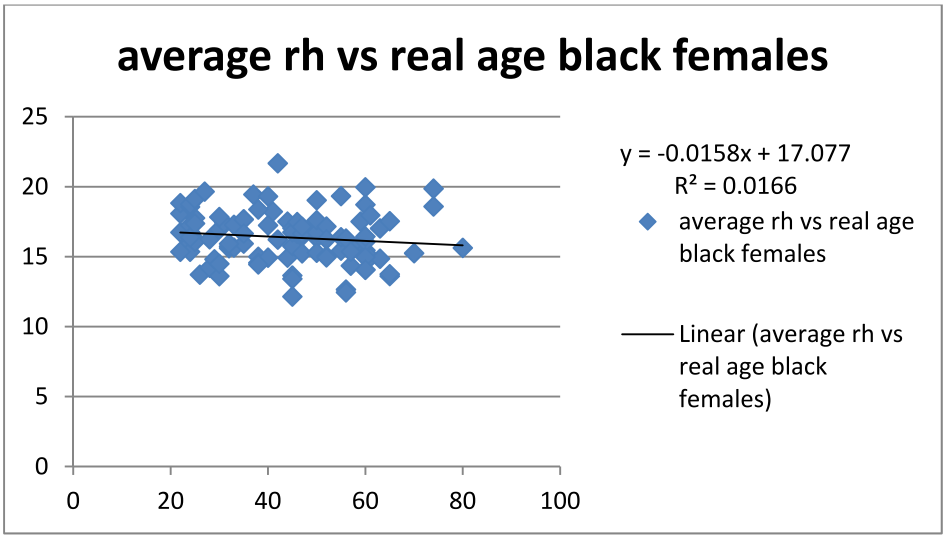


Figure 5.4. Average root height (rh) correlated against real age for black females, all canines. R^2=correlation coefficient. X-axis=real age, Y-axis=average root height (rh).

The following two figures (Figures 5.5 and 5.6) show the negative correlations separately for the upper and lower canines. Here canine 3 root height has a slightly higher correlation with age (R^2=0.024) than canine 1 (R^2=0.009), which indicates that, in especially the lower canine, some root height is lost with age.

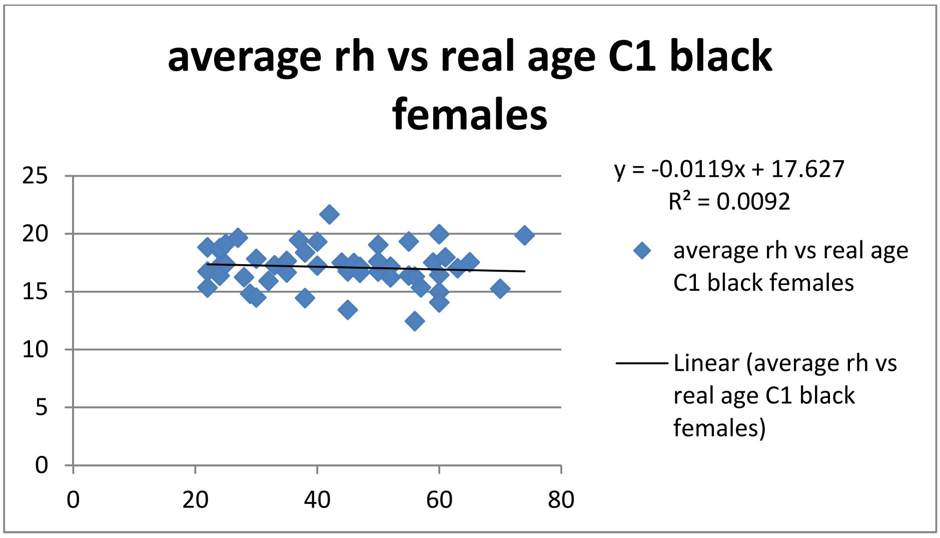


Figure 5.5. Average root height (rh) correlated against real age for canine 1 (black females). R^2=correlation coefficient. X-axis=real age, Y-axis=average root height (rh).

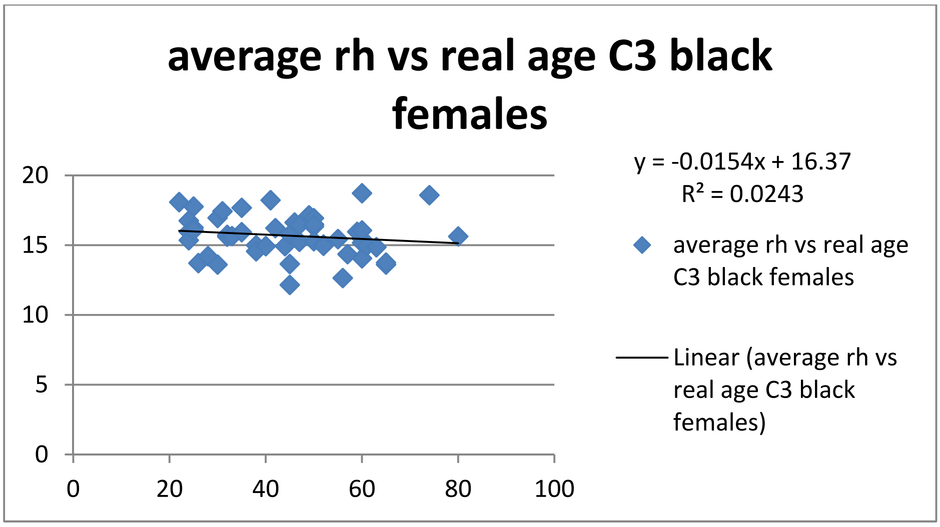


Figure 5.6. Average root height (rh) correlated against real age for canine 3 (black females). R^2=correlation coefficient. X-axis=real age, Y-axis=average root height (rh).

5.3.1.3. White males

The average root height was negatively correlated to real age in white males, with a R^2 value of 0.024 (Figure 5.7).

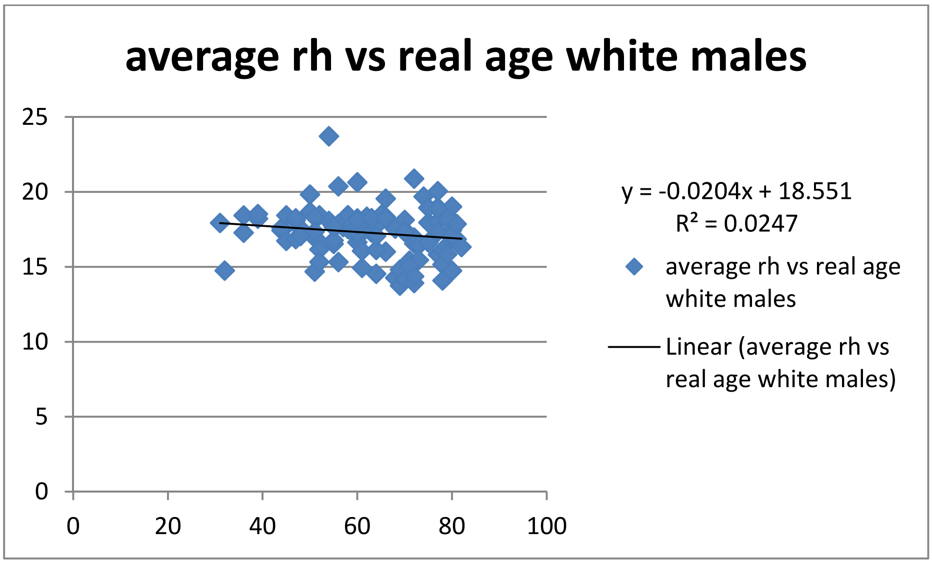


Figure 5.7. Average root height (rh) correlated against real age for white males, all canines. R^2=correlation coefficient. X-axis=real age, Y-axis=average root height (rh).

More specifically, the correlations for canines 1 and 3 can be seen in Figures 5.8 and 5.9 below. From this it is evident that rooth height in both canines 1 and 3 were negatively correlated tot age - canine 1 (R^2=0.029) and canine 3 (R^2=0.005). Once again this negative correlation indicates that the root becomes somewhat shorter with age, but this loss is relatively small.

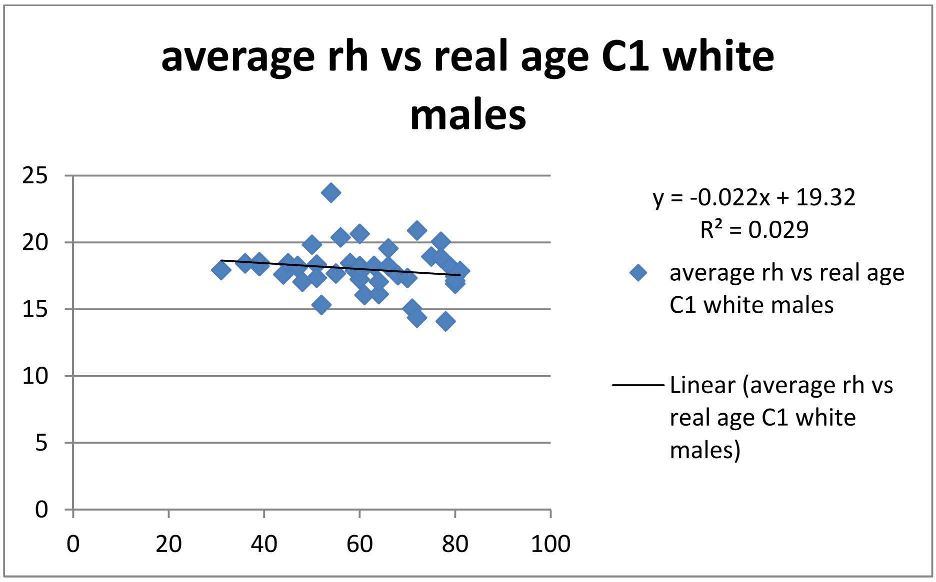


Figure 5.8. Average root height (rh) correlated against real age for canine 1 (white males). R^2=correlation coefficient. X-axis=real age, Y-axis=average root height (rh).

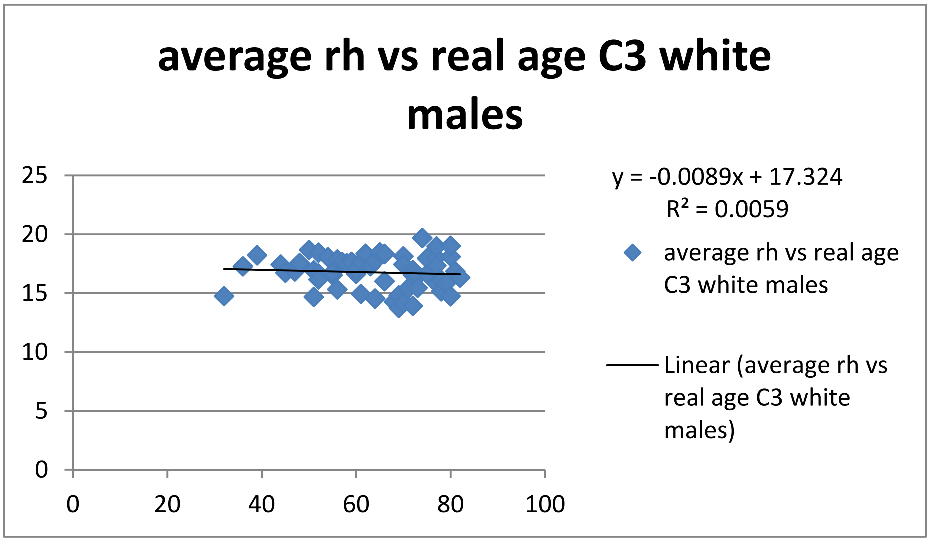


Figure 5.9. Average root height (rh) correlated against real age for canine 3 (white males). R^2=correlation coefficient. X-axis=real age, Y-axis=average root height (rh).

5.3.1.4. White females

From Figure 5.10 it can be seen that root height was also negatively correlated with real age in white females (R^2=0.012).

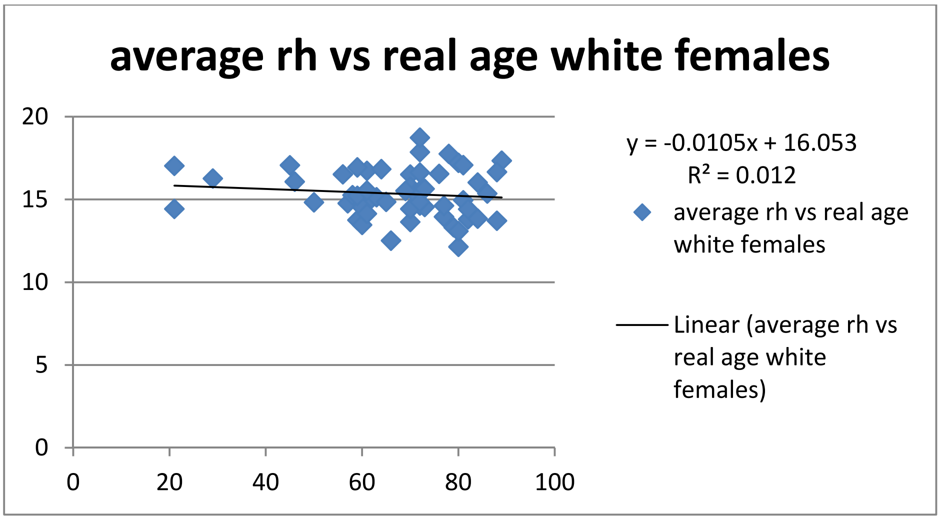


Figure 5.10. Average root height (rh) correlated against real age for white females, all canines. R^2=correlation coefficient. X-axis=real age, Y-axis=average root height (rh).

More specifically, canines 1 and 3 gave different results, but both indicate negative correlations. Canine 1 obtained a negative correlation of 0.2% with real age (R^2=0.002) (Figure 5.11) whereas canine 3 showed a 2.9% correlation with real age (R^2=0.029) (Figure 5.12). These results indicate that in females the root of the lower canine looses somewhat more height than the upper canine.

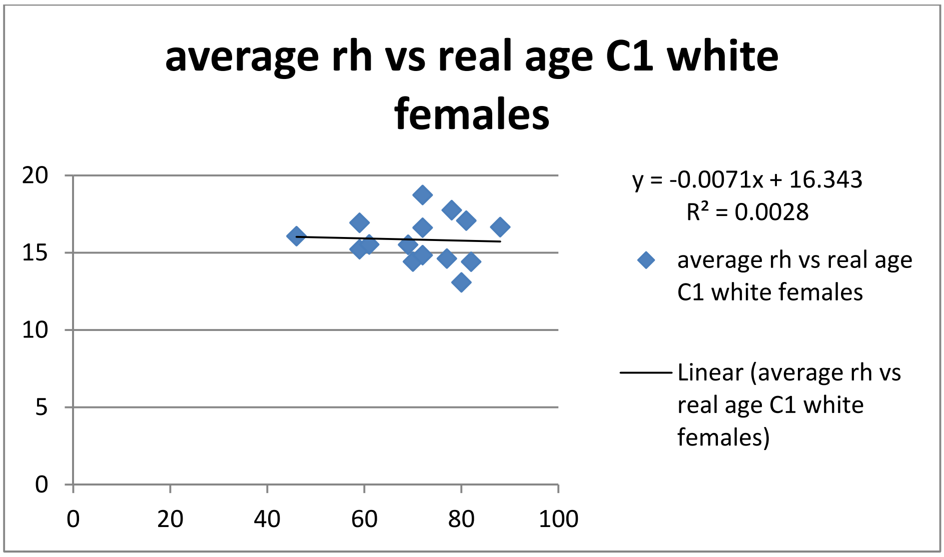


Figure 5.11. Average root height (rh) correlated against real age for canine 1 (white females). R^2=correlation coefficient. X-axis=real age, Y-axis=average root height (rh).

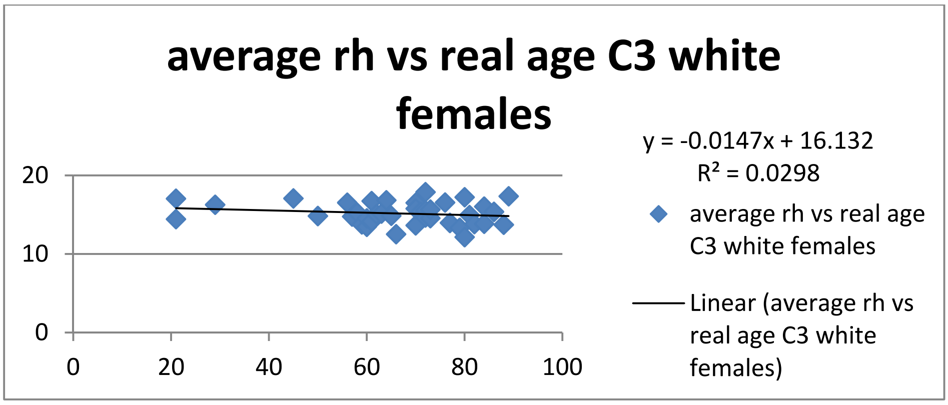


Figure 5.12. Average root height (rh) correlated against real age for canine 3 (white females). R^2=correlation coefficient. X-axis=real age, Y-axis=average root height (rh).

5.3.2. Periodontosis

5.3.2.1. Black males

The mean periodontosis height for all canines was plotted against the real age for the black male group. As can be seen from Figure 5.13, an R^2 value of 0.255 was obtained, indicating that 25.5% of age can be explained by periodontosis.

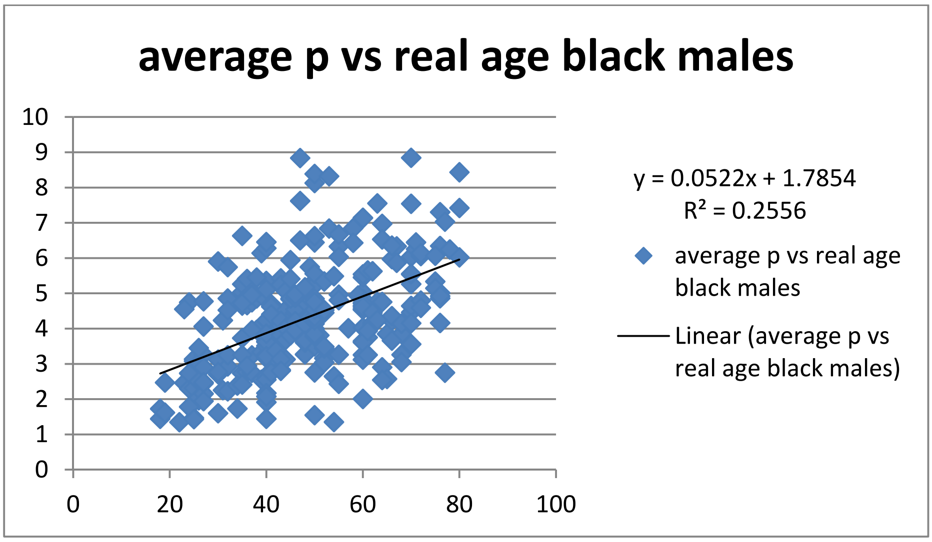


Figure 5.13. Average periodontosis (p) correlated against real age for black males, all canines. R^2=correlation coefficient. X-axis=real age, Y-axis=average periodontosis (p).

When the canines of the two jaws were plotted separately, different results were obtained for the maxillary and mandibular canines (Figures 5.14 and 5.15). It is clear from the figures that periodontosis in canines 1 (R^2=0.349) and 3 (R^2=0.181) were both weakly but positively correlated to age.

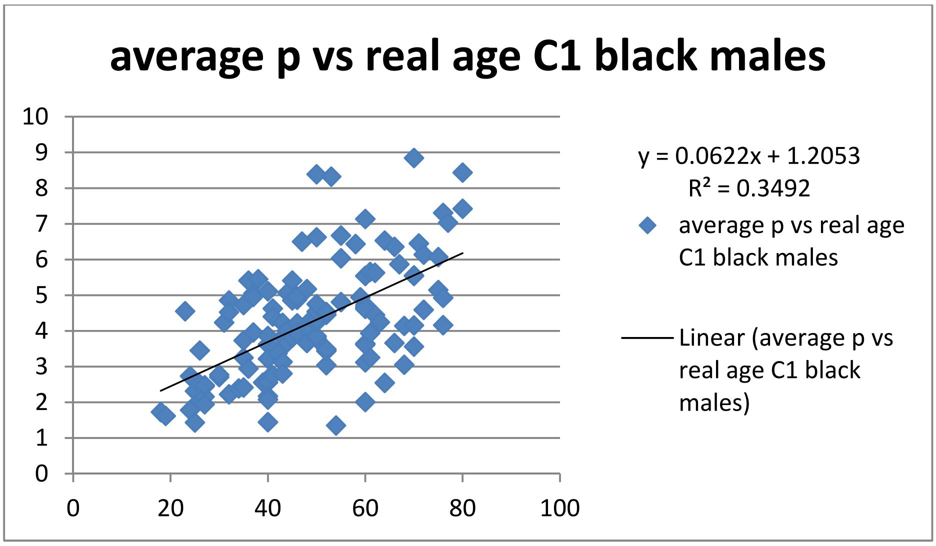


Figure 5.14. Average periodontosis (p) correlated against real age for canine 1 (black males). R^2=correlation coefficient. X-axis=real age, Y-axis=average periodontosis (p).

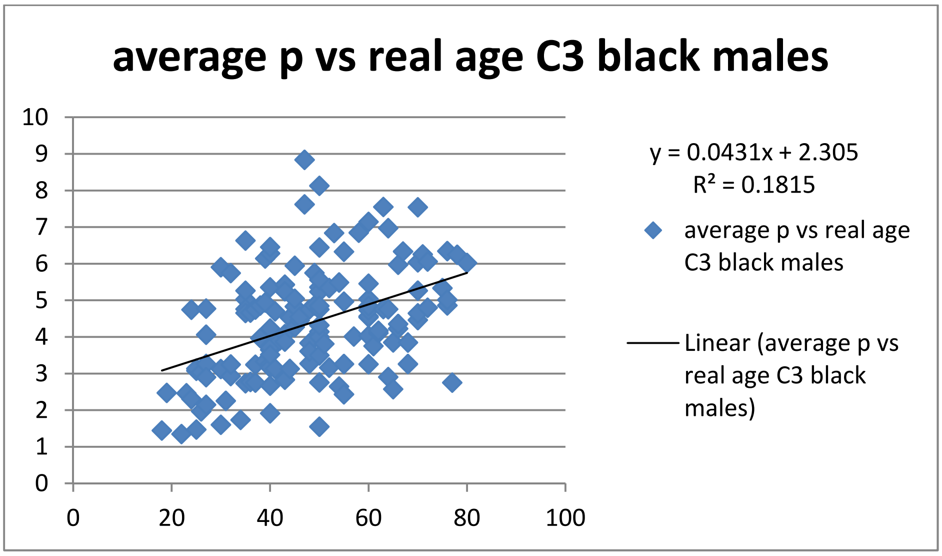


Figure 5.15. Average periodontosis (p) correlated against real age for canine 3 (black males). R^2=correlation coefficient. X-axis=real age, Y-axis=average periodontosis (p).

In Figure 5.16 the periodontosis was expressed as a percentage of root height in each individual and obtained a correlation coefficient value of 23.5% (R^2=0.235) for both upper and lower canines. When the canine was specified, canine 1 obtained a correlation coefficient value of 34.7% (R^2=0.347) whereas canine 3 obtained a value of 17.5% (R^2=0.175) (Tables 5.17 and 5.18).

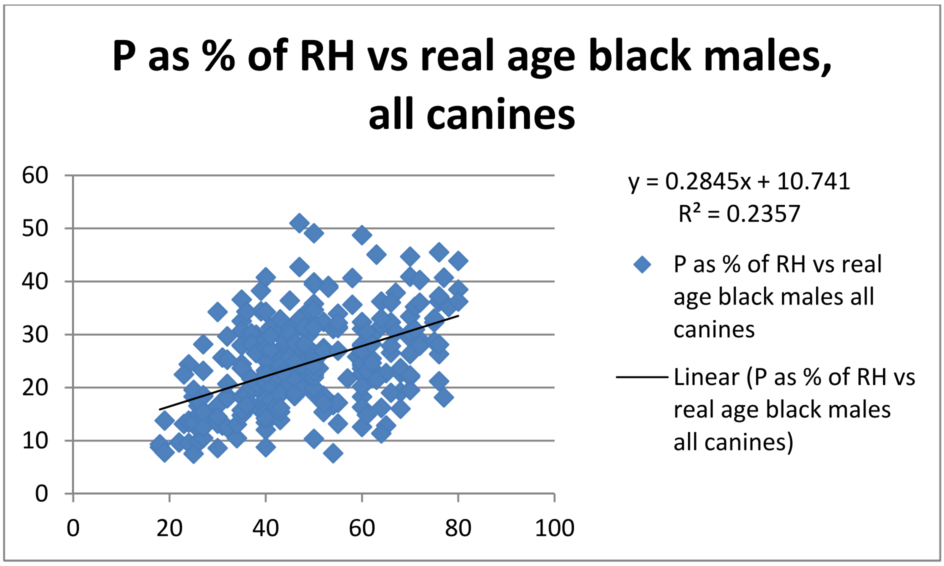


Figure 5.16. Average periodontosis (p) expressed as a function of root height (rh) against real age in black males, all canines. R^2=correlation coefficient. X-axis=real age, Y-axis=periodontosis (P) as percentage of root height (RH).

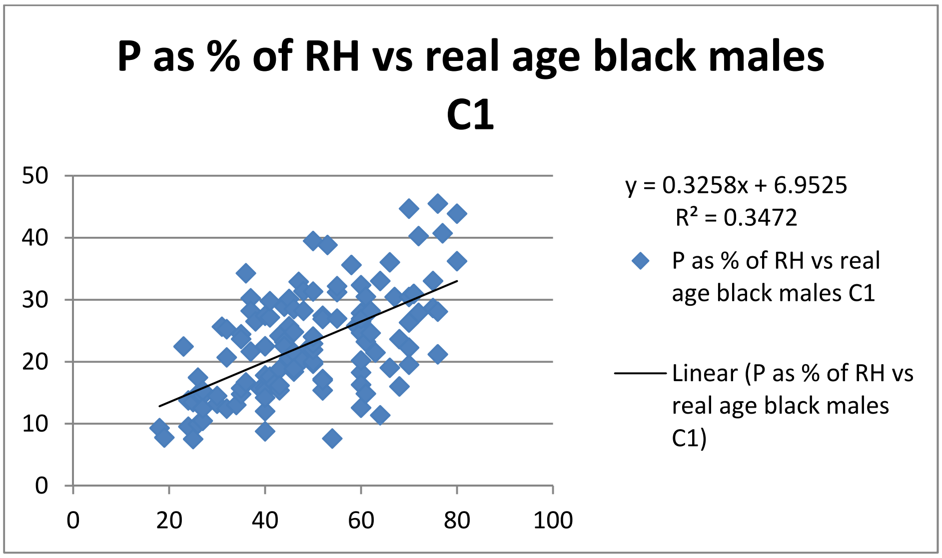


Figure 5.17. Average periodontosis (p) expressed as a function of root height (rh) against real age in black males, canine 1. R^2=correlation coefficient. X-axis=real age, Y-axis=periodontosis (P) as percentage of root height (RH).

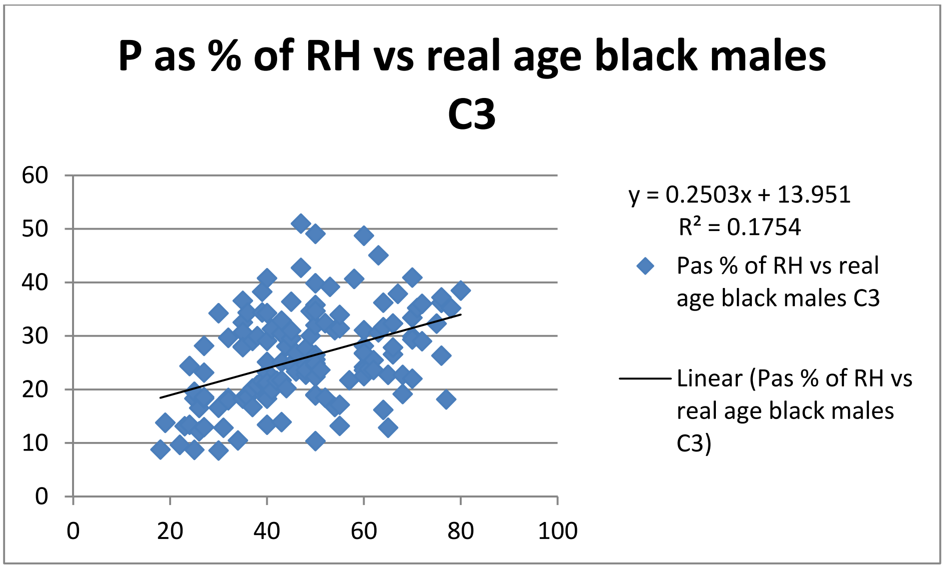


Figure 5.18. Average periodontosis (p) expressed as a function of root height (rh) against real age in black males, canine 3. R^2=correlation coefficient. X-axis=real age, Y-axis=periodontosis (P) as percentage of root height (RH).

5.3.2.2. Black females

Figure 5.19 shows the correlation of the average periodontosis against real age of each individual in the black female group for all canines. Periodontosis attributed 9.1% to the age of the black female group (R^2=0.091).

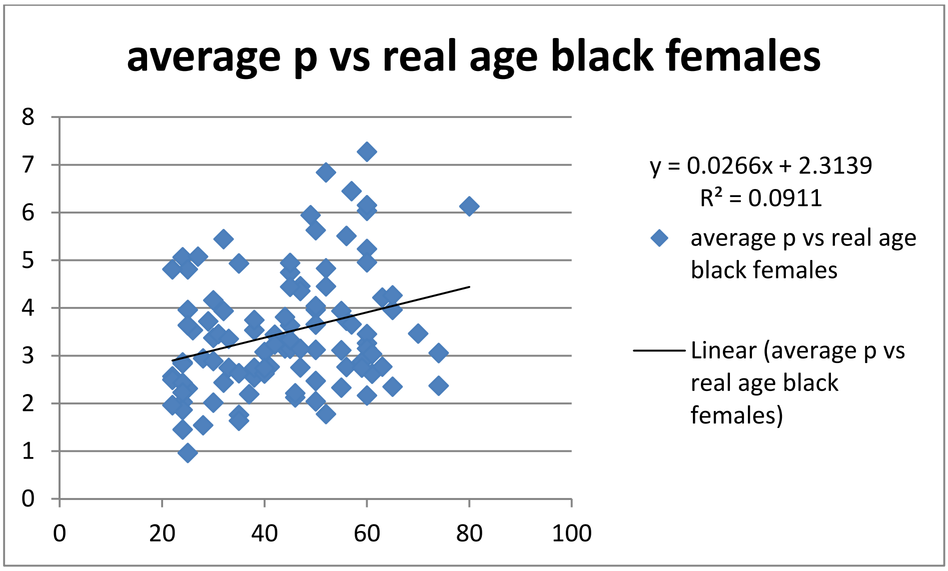


Figure 5.19. Average periodontosis (p) correlated against real age for black females, all canines. R^2=correlation coefficient. X-axis=real age, Y-axis=average periodontosis (p).

More specifically, the periodontosis measurement of canine 1 obtained a correlation coefficient of R^2=0.068 with age, which indicates that only 6.8% of the age could be explained by periodontosis (Figure 5.20). For canine 3 the corresponding correlation coefficient (R^2) was 0.113 (Figure 5.21). Therefore, 11.3% of the age could be explained by periodontosis measured from canine 3. These correlations still remain weak, but positive. It is also noticeable that the scatter is quite wide for both canines, indicating considerable variation between individuals.

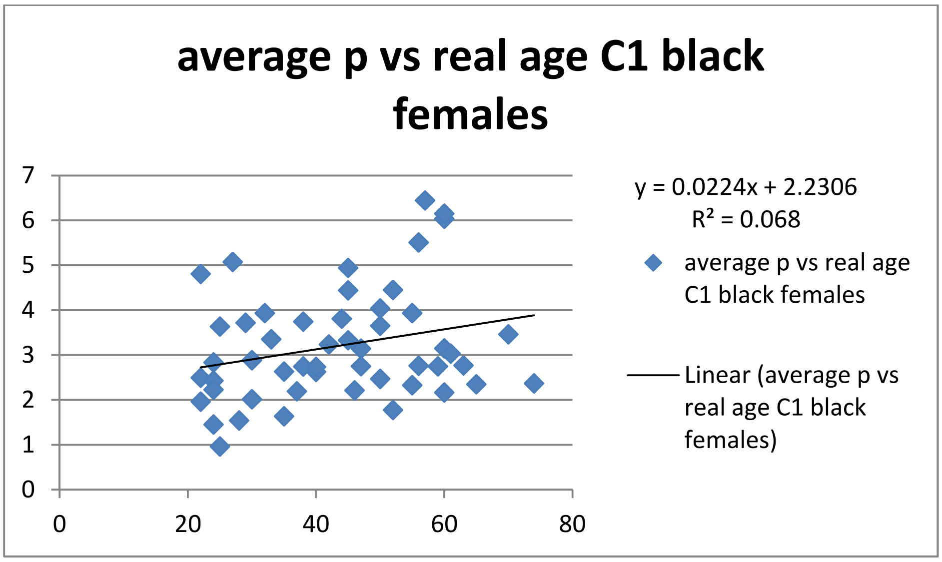


Figure 5.20. Average periodontosis (p) correlated against real age for canine 1 (black females). R^2=correlation coefficient. X-axis=real age, Y-axis=average periodontosis (p).

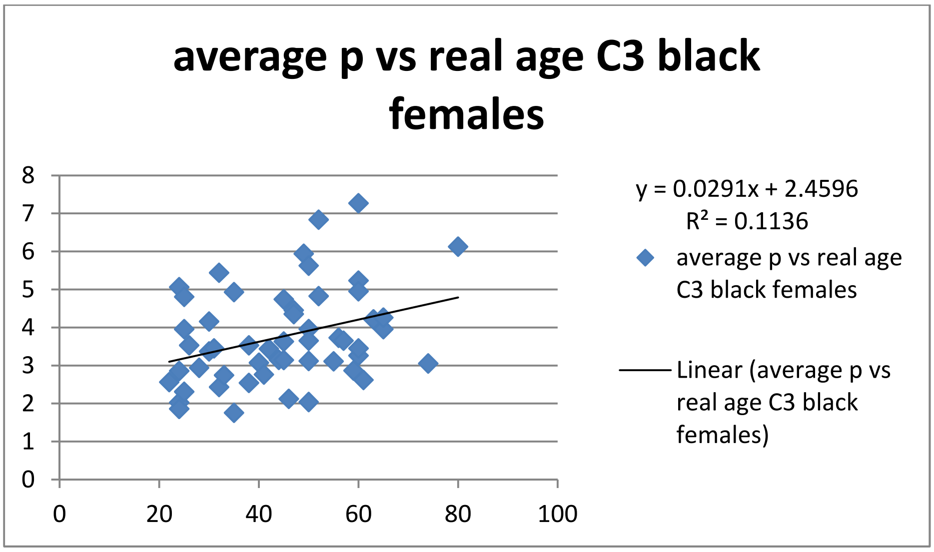


Figure 5.21. Average periodontosis (p) correlated against real age for canine 3 (black females). R^2=correlation coefficient. X-axis=real age, Y-axis=average periodontosis (p).

The periodontosis of the black female group was expressed as a percentage of root height in Figure 5.22. This measurement obtained a correlation coefficient value of 10.5% (R^2=0.105). When the specific canine was selected, canine 1 obtained a correlation coefficient of R^2=0.076 whereas canine 3 obtained an R^2 value of 0.140 (Figures 5.23 and 5.24).

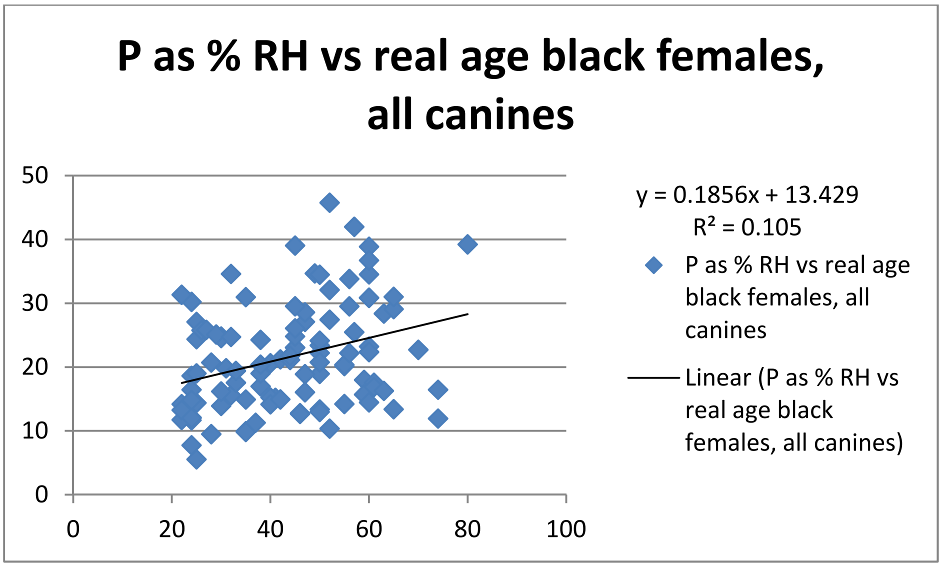


Figure 5.22. Average periodontosis (p) expressed as a function of root height (rh) against real age in black females, all canines. R^2=correlation coefficient. X-axis=real age, Y-axis=periodontosis (P) as percentage of root height (RH).

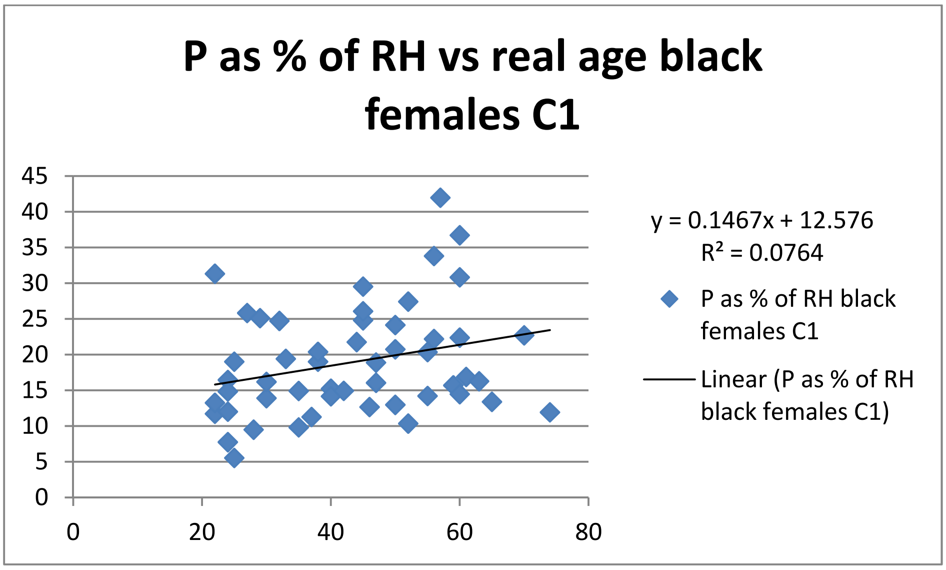


Figure 5.23. Average periodontosis (p) expressed as a function of root height (rh) against real age in black females, canine 1. R^2=correlation coefficient. X-axis=real age, Y-axis=periodontosis (P) as percentage of root height (RH).

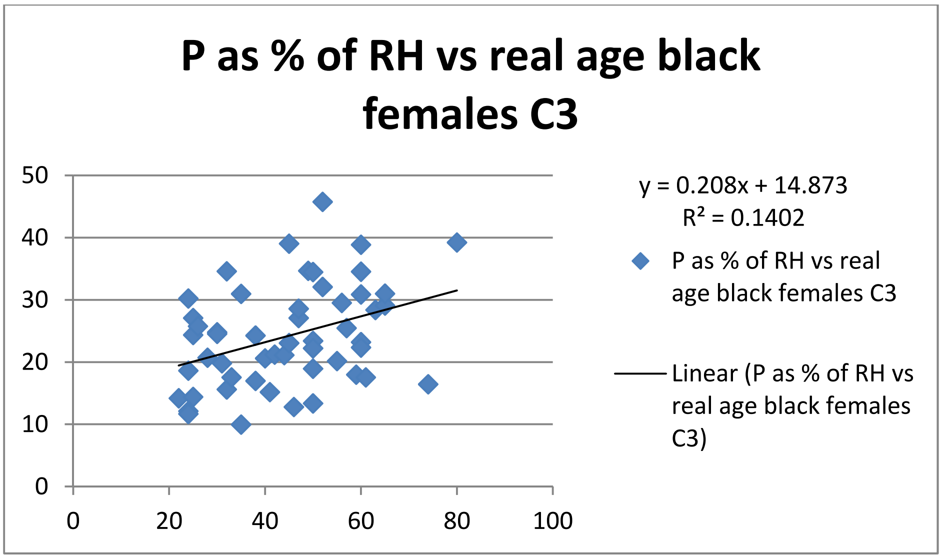


Figure 5.24. Average periodontosis (p) expressed as a function of root height (rh) against real age in black females, canine 3. R^2=correlation coefficient. X-axis=real age, Y-axis=periodontosis (P) as percentage of root height (RH).

5.3.2.3. White males

In Figure 5.25 periodontosis for the white males was plotted against real age. Here periodontosis obtained an R^2 value of 0.011, which indicates that periodontosis could explain only 1.1% of the real age. Once again the scatter indicates a high degree of variability.

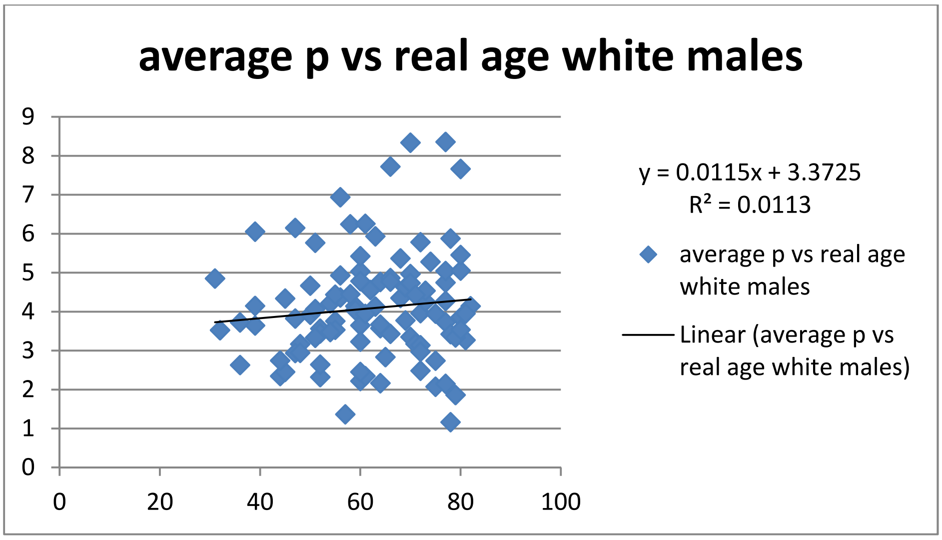


Figure 5.25. Average periodontosis (p) correlated against real age for white males, all canines. R^2=correlation coefficient. X-axis=real age, Y-axis=average periodontosis (p).

Periodontosis in canine 1 on its own (Figure 5.26) obtained a weak but positive correlation of 2.2% (R^2=0.022) with age, whereas in canine 3 (Figure 5.27) the corresponding figure was 1.3% (R^2=0.013). These were slightly better results than when the canines were pooled together, but still indicate low levels of correlation (Figure 5.25).

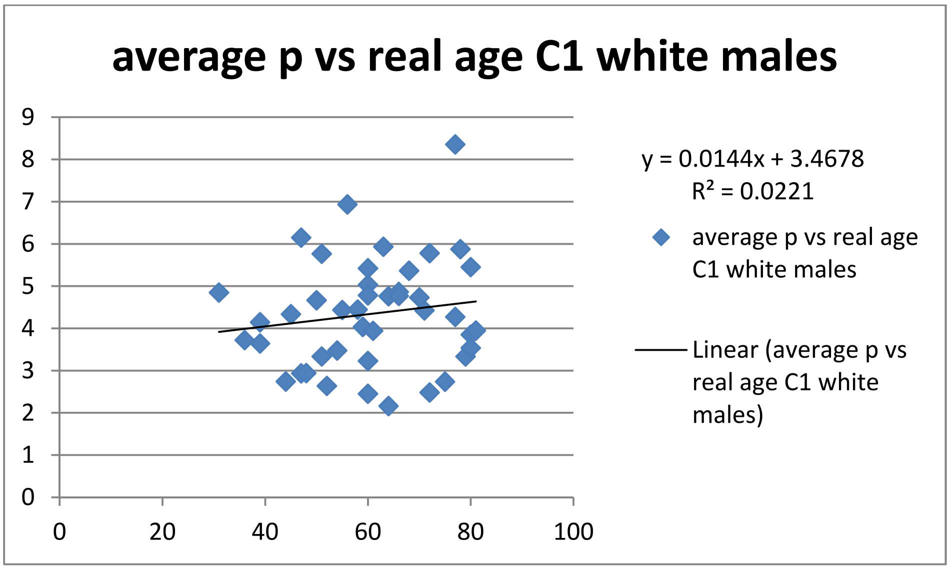


Figure 5.26. Average periodontosis (p) correlated against real age for canine 1 (white males). R^2=correlation coefficient. X-axis=real age, Y-axis=average periodontosis (p).

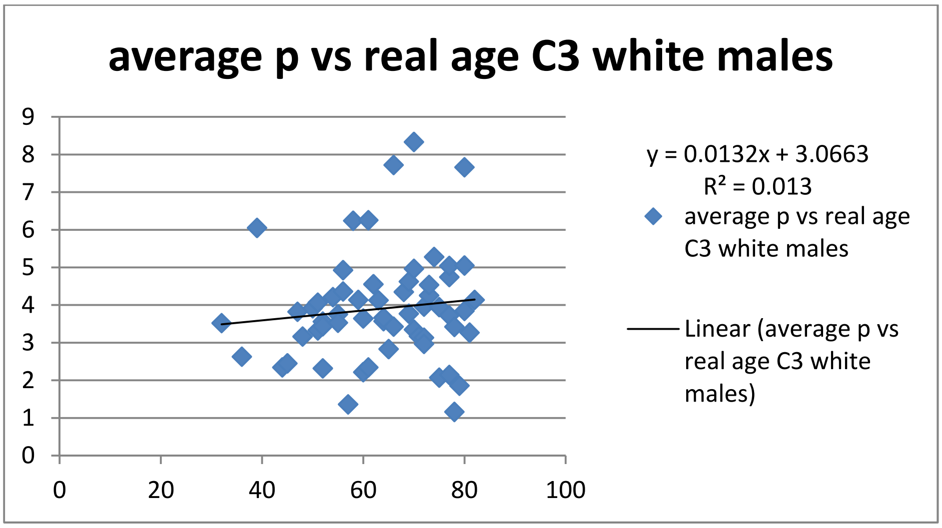


Figure 5.27. Average periodontosis (p) correlated against real age for canine 3 (white males). R^2=correlaion coefficient. X-axis=real age, Y-axis=average periodontosis (p).

The correlation of the average periodontosis expressed as a percentage of root height against real age can be seen in Figure 5.28. Here a correlation coefficient of 2.2% was obtained for all canines, whereas canine 1 obtained an R^2 value 0.045 (4.5%) and canine 3 a value of 0.015 (1.5%) (Figures 5.29 and 5.30).

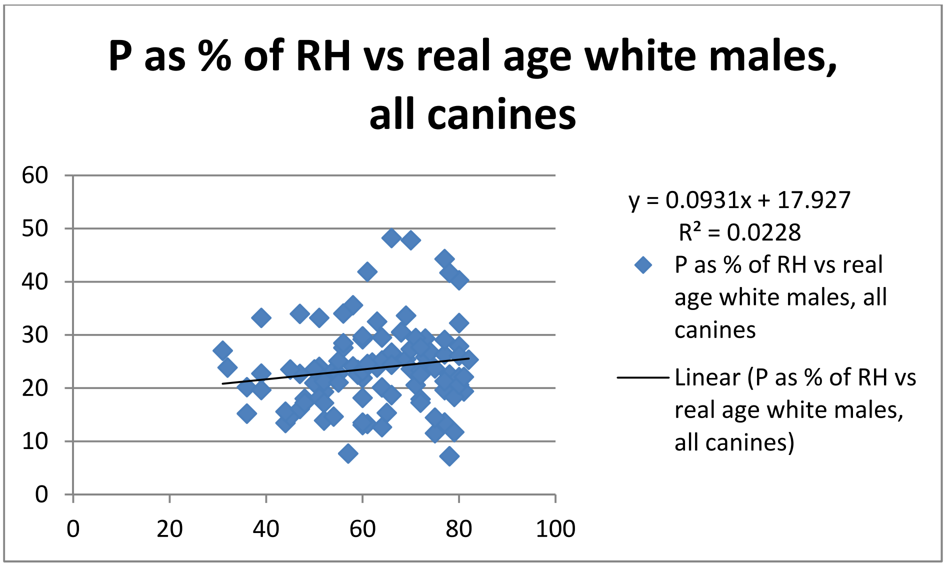


Figure 5.28. Average periodontosis (p) expressed as a function of root height (rh) against real age in white males, all canines. R^2=correlation coefficient. X-axis=real age, Y-axis=periodontosis (P) as percentage of root height (RH).

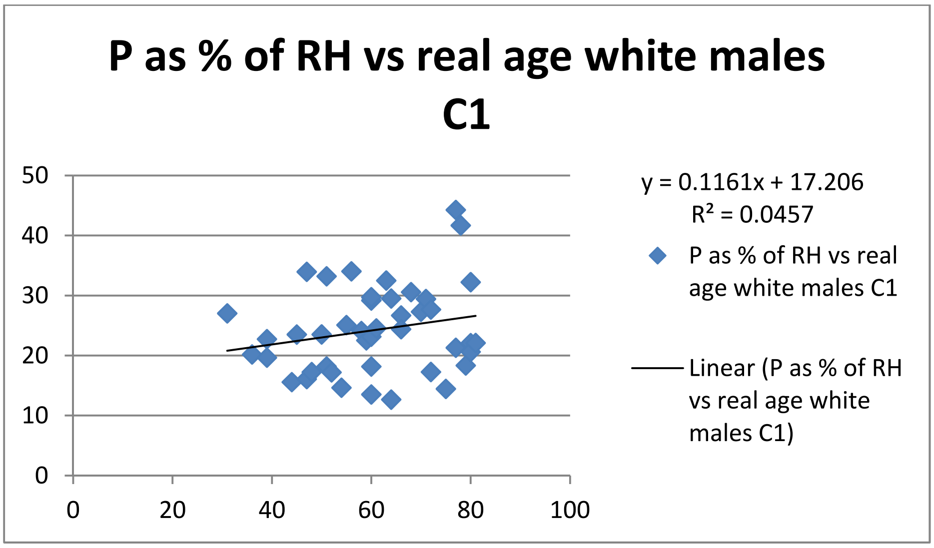


Figure 5.29. Average periodontosis (p) expressed as a function of root height (rh) against in white males, canine 1. R^2=correlation coefficient. X-axis=real age, Y-axis=periodontosis (P) as percentage of root height (RT).

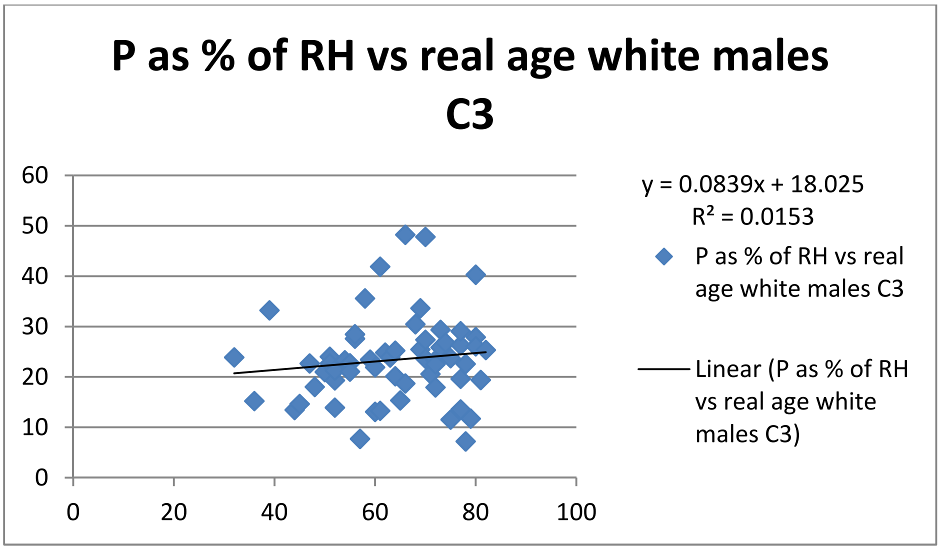


Figure 5.30. Average periodontosis (p) expressed as a function of root height (rh) against real age in white males, canine 3. R^2=correlation coefficient. X-axis=real age, Y-axis=periodontosis (P) as percentage of root height (RH).

5.3.2.4. White females

Correlation between average periodontosis (p) and real age in white females is shown in Figure 5.31, and gave a value of R^2=0.208. In this group the correlation is thus fairly high, but considerable variation between individuals is observed.

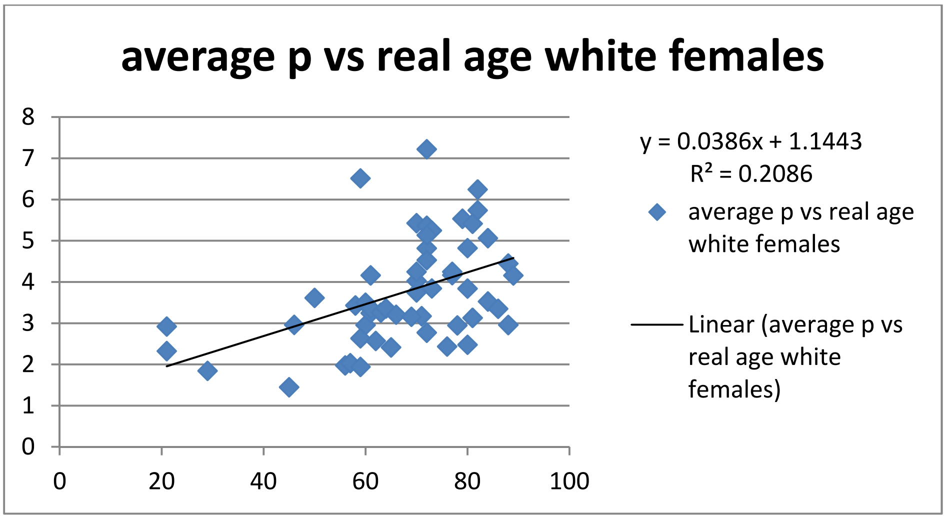


Figure 5.31. Average periodontosis (p) correlated against real age for white females, all canines. R^2=correlation coefficient. X-axis=real age, Y-axis=average periodontosis (p).

Canine 1 and real age produced a correlation coefficient (R^2) of 0.018 (Figure 5.32) whereas canine 3 gave a value of 0.307 (Figure 5.33). Periodontosis in the lower canines was thus more correlated with age than what was the case for upper canines.

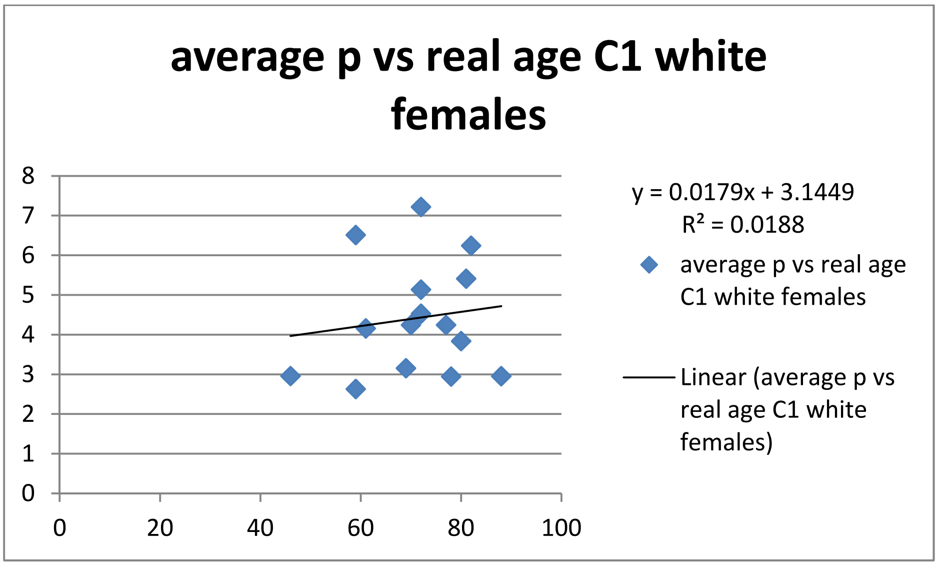


Figure 5.32. Average periodontosis (p) correlated against real age for canine 1 (white females). R^2=correlation coefficient. X-axis=real age, Y-axis=average periodontosis (p).

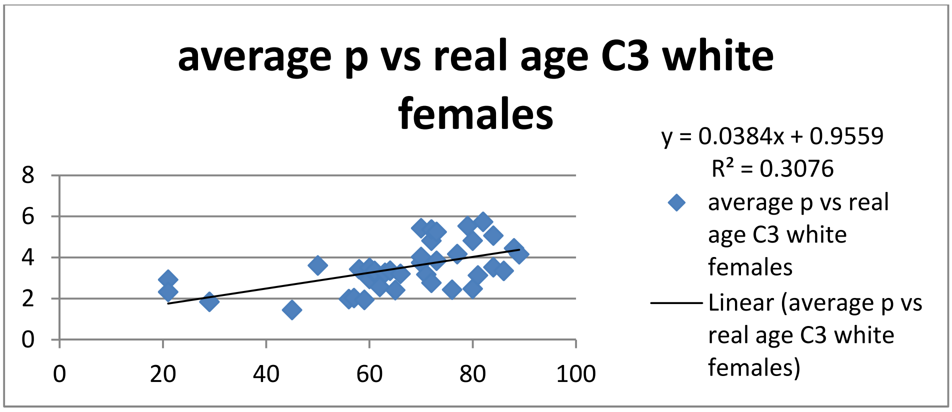


Figure 5.33. Average periodontosis (p) correlated against real age for canine 3 (white females). R^2=correlation coefficient. X-axis=real age, Y-axis=average periodontosis (p).

When the average periodontosis expressed as a percentage of root height was plotted against real age, the pooled group of canines for white females obtained a correlation of 23.2% (Figure 5.34). When the canines were specified, canine 1 obtained a correlation coefficient value of R^2=0.033 (3.3%) and canine 3 a R^2 value of 0.316 (31.6%) (Figures 5.35 and 5.36).

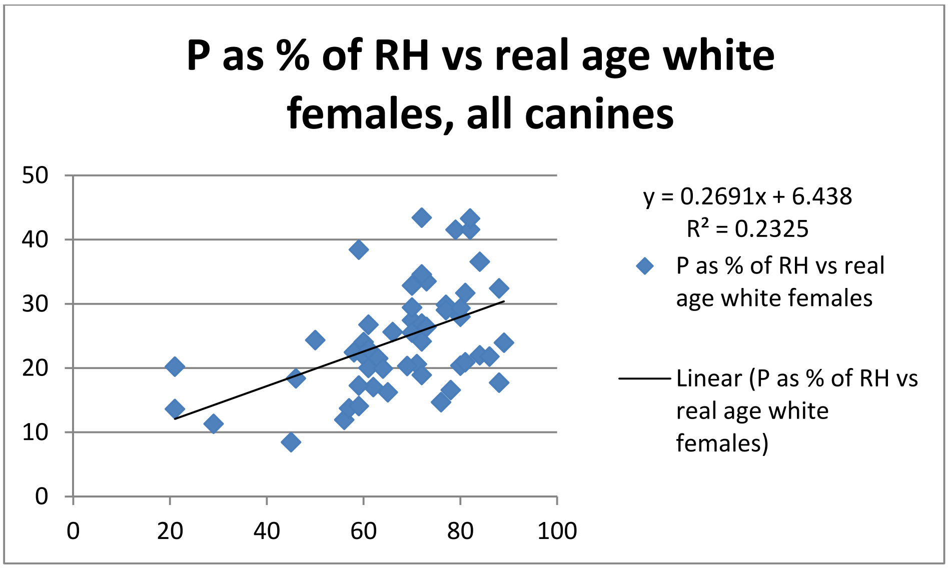


Figure 5.34. Average periodontosis (p) expressed as a function of root height (rh) against real age in white females, all canines. R^2=correlation coefficient. X-axis=real age, Y-axis=periodontosis (P) as percentage of root height (RH).

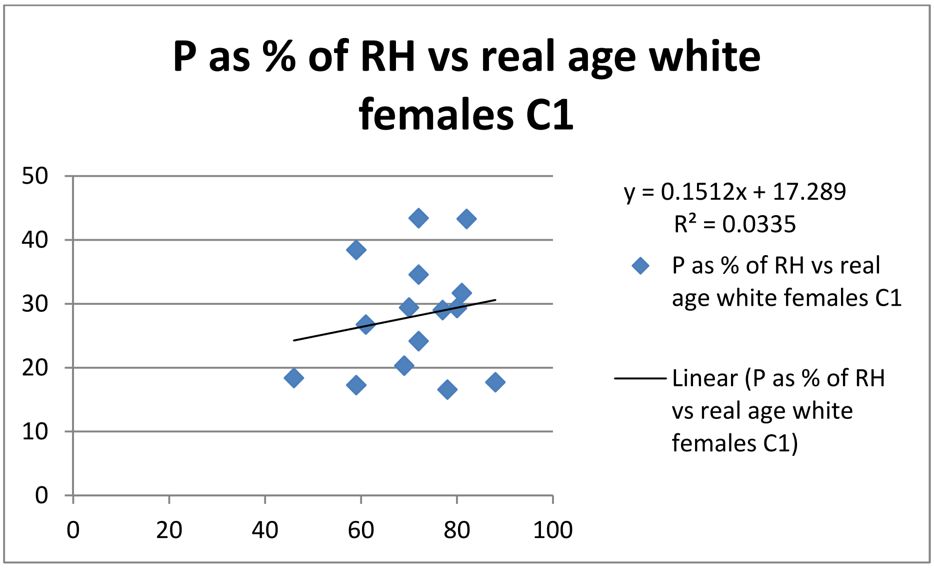


Figure 5.35. Average periodontosis (p) expressed as a function of root height (rh) against real age in white females, canine 1. R^2=correlation coefficient. X-axis=real age, Y-axis=periodontosis (P) as percentage of root height (RH).

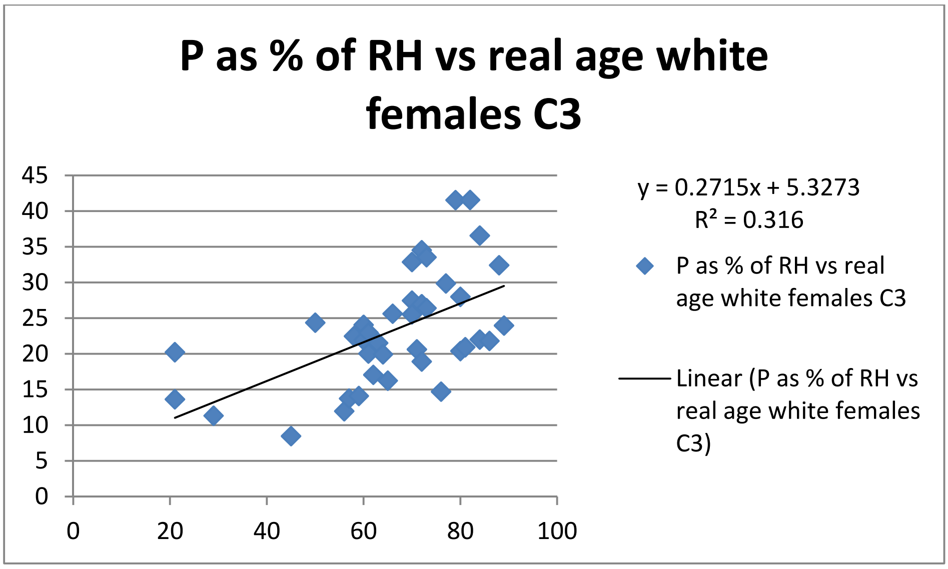


Figure 5.36. Average periodontosis (p) expressed as a function of root height (rh) against real age in white females, canine 3. R^2=correlation coefficient. X-axis=real age, Y-axis=periodontosis (P) as percentage of root height (RH).

5.3.3. Root transparency

5.3.3.1. Black males

The average root transparency of the black males for upper and lower canines was correlated against the real age of the individuals (Figure 5.37), and it resulted in an R^2 value of 0.114. This indicates that 11.4% of age could be explained by root transparency from both the maxillary and mandibular canines.

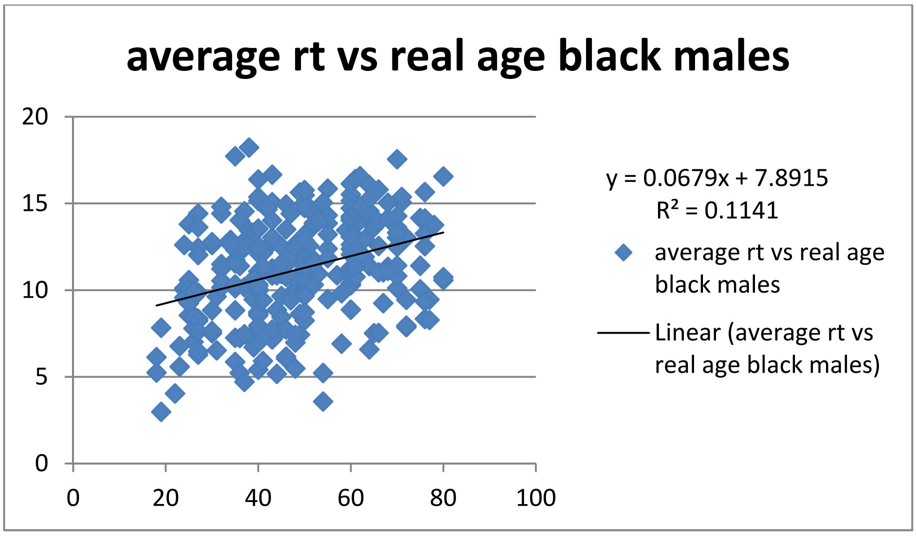


Figure 5.37. Average root transparency (rt) correlated against real age for black males, all canines. R^2=correlation coefficient. X-axis=real age, Y-axis=average root transparency (rt).

More specifically, from Figures 5.38 and 5.39 (below) it is evident that root transparency in canine 1 is somewhat better correlated with age (R^2=0.126) than canine 3 (R^2=0.104) on its own.

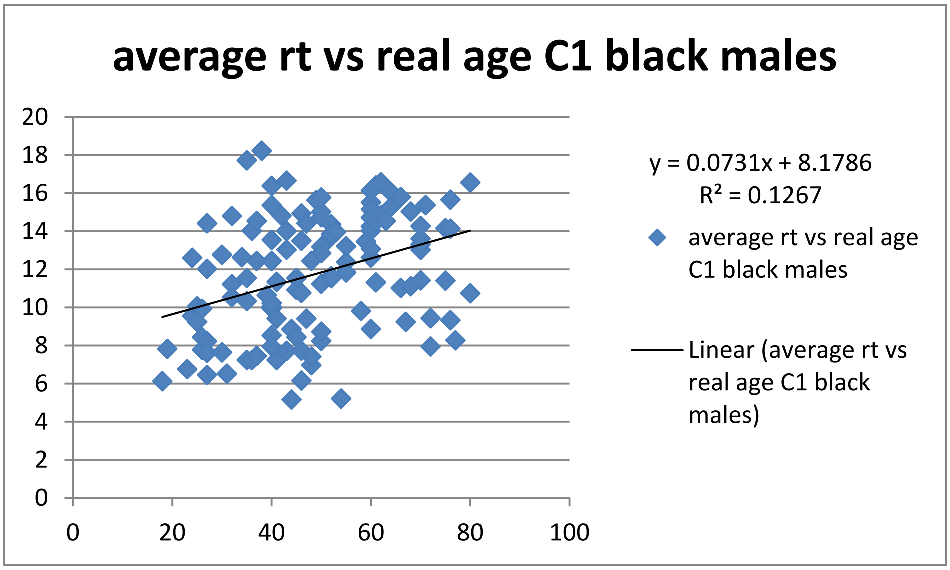


Figure 5.38. Average root transparency (rt) correlated against real age for canine 1 (black males). R^2=correlation coefficient. X-axis=real age, Y-axis=average root transparency (rt).

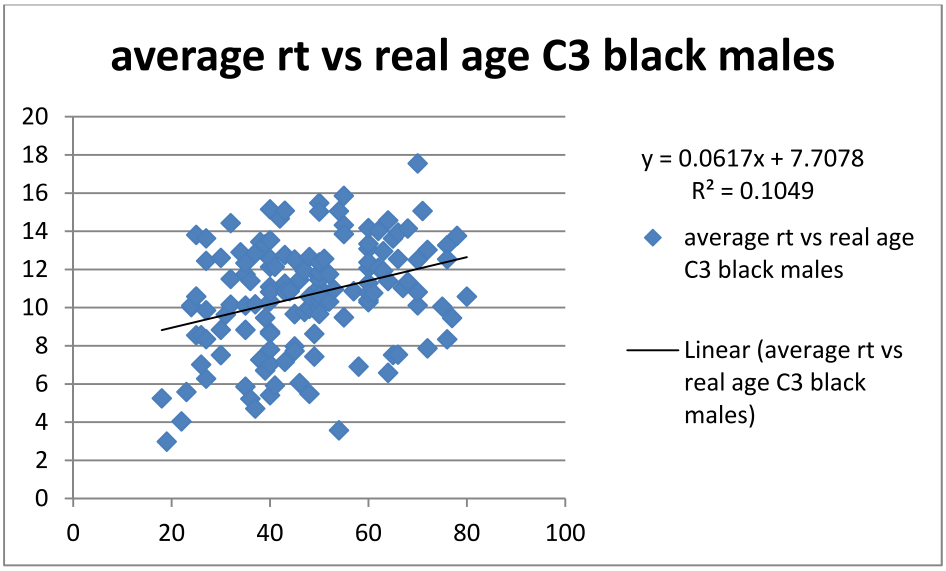


Figure 5.39. Average root transparency (rt) correlated against real age for canine 3 (black males). R^2=correlation coefficient. X-axis=real age, Y-axis=average root transparency (rt).

Root transparency was expressed as a percentage of root height (Figure 5.40) against real age and obtained a correlation coefficient value of 11.8% (R^2=0.118). Canines 1 and 3 (Figures 5.41 and 5.42) obtained correlations of 13.0% (R^2=0.13) and 10.9% (R^2=0.109), respectively.

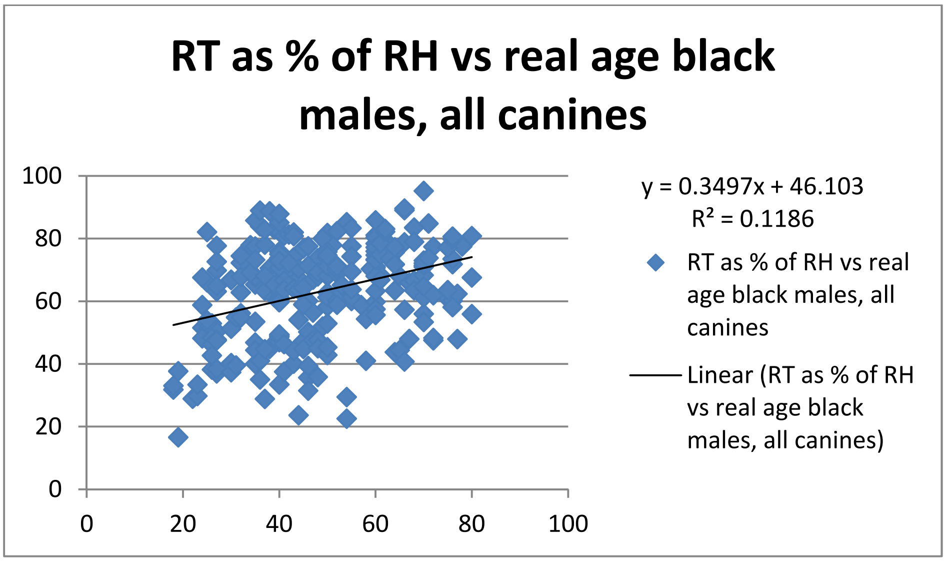


Figure 5.40. Average root transparency (rt) expressed as a function of root height (rh) against real age in black males, all canines. R^2=correlation coefficient. X-axis=real age, Y-axis=root transparency (RT) as percentage of root height (RH).

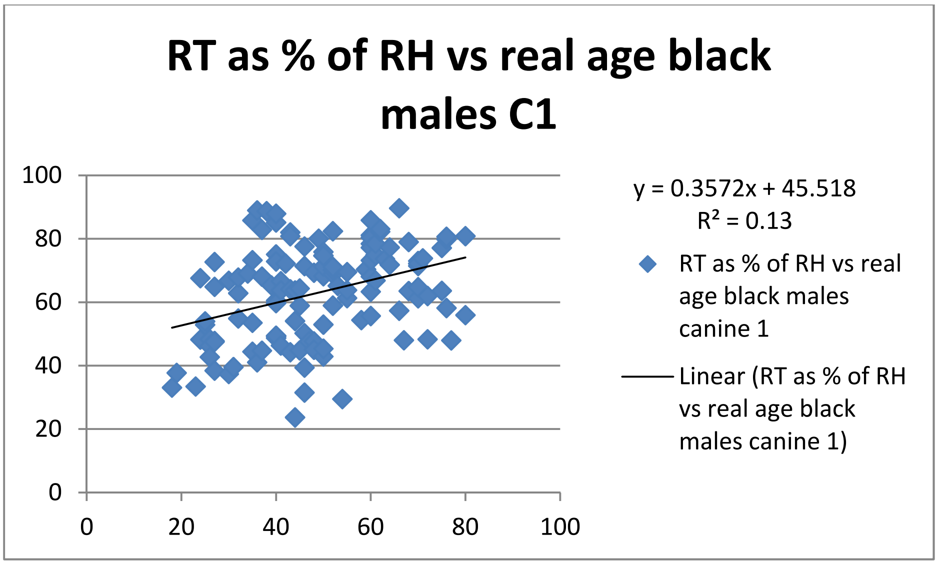


Figure 5.41. Average root transparency (rt) expressed as a function of root height (rh) against real age in black males, canine 1. R^2=correlation coefficient. X-axis=real age, Y-axis=root transparency (RT) as percentage of root height (RH).

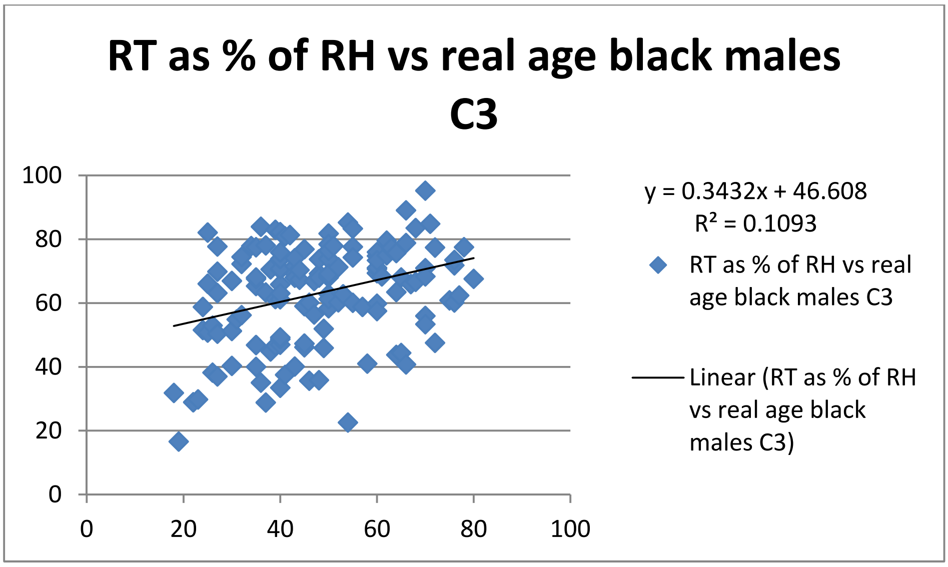


Figure 5.42. Average root transparency (rt) expressed as a function of root height (rh) against real age in black males, canine 3. R^2=correlation coefficient. X-axis=real age, Y-axis=root transparency (RT) as percentage of root height (RH).

5.3.3.2. Black females

In Figure 5.43 the average root transparency measurement for all canines was correlated against the real age of the black female group. The average root transparency obtained an R^2 value of 0.136, indicating that 13.6% of the age could be explained by root transparency.

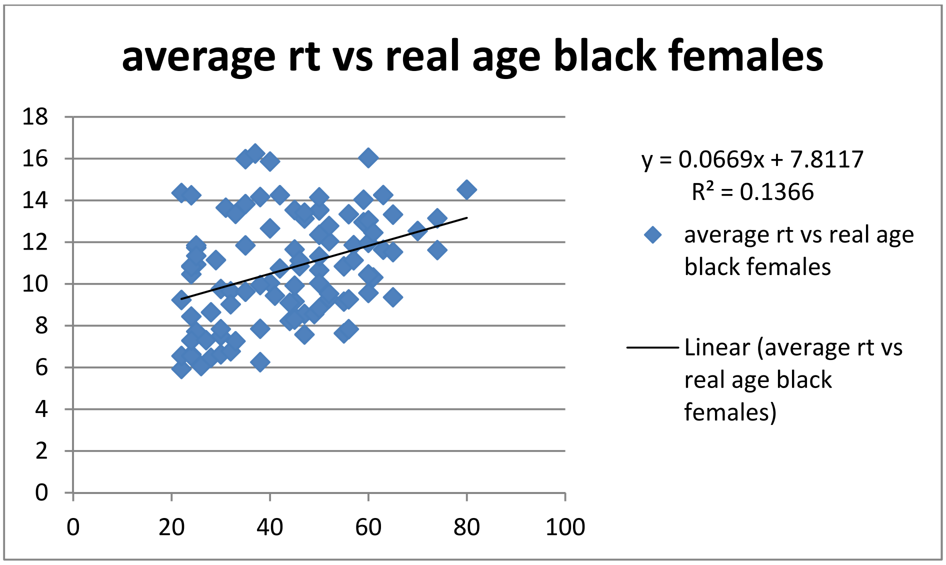


Figure 5.43. Average root transparency (rt) correlated against real age for black females, all canines. R^2=correlation coefficient. X-axis=real age, Y-axis=average root transparency (rt).

Figures 5.44 and 5.45 show that although canine 3 gave a slightly better correlation with age (R^2=0.214) than canine 1 (R^2=0.112), the correlations remain weak.

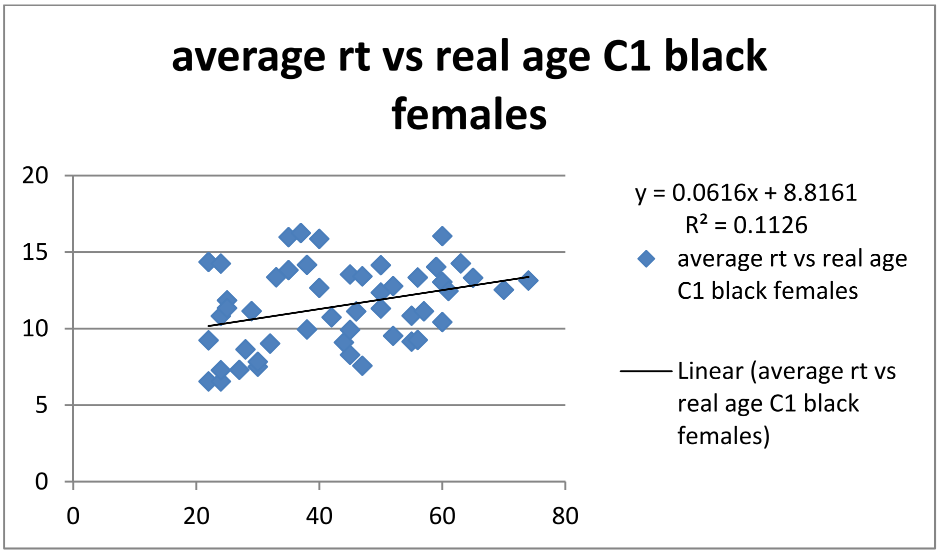


Figure 5.44. Average root transparency (rt) correlated against real age for canine 1 (black females). R^2=correlation coefficient. X-axis=real age, Y-axis=average root transparency (rt).

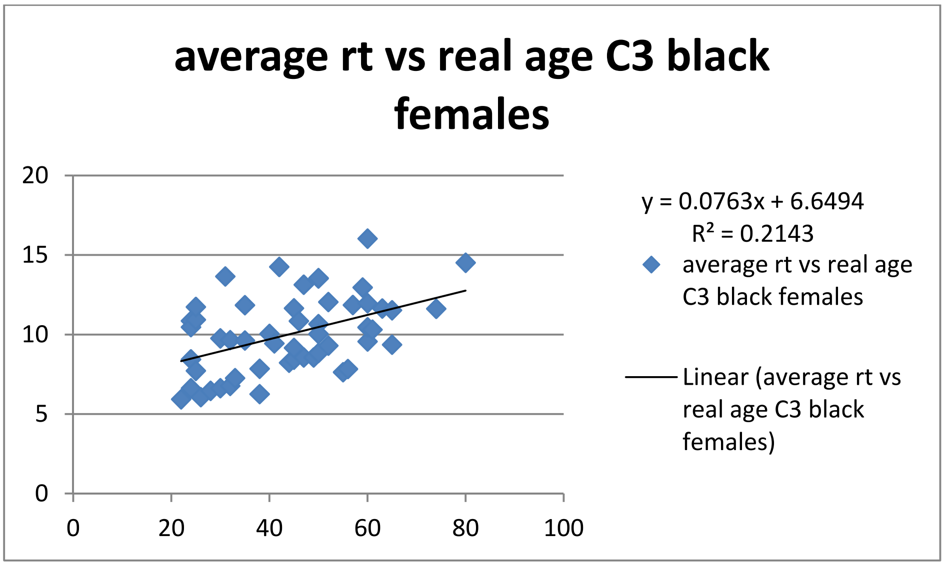


Figure 5.45. Average root transparency (rt) correlated against real age for canine 3 (black females). R^2=correlation coefficient. X-axis=real age, Y-axis=average root transparency (rt).

Root transparency was expressed as a percentage of root height against real age in Figure 5.46. It is evident that when root transparency is expressed as a percentage of root height, it obtained a correlation coefficient value of 21.9% (R^2=0.219) for all canines. If the canine was specified, canine 1 resulted in a correlation coefficient value of 16.4% (R^2=0.164) (Figure 5.47) and canine 3 a value of 29.6% (R^2=0.296) (Figure 5.48).

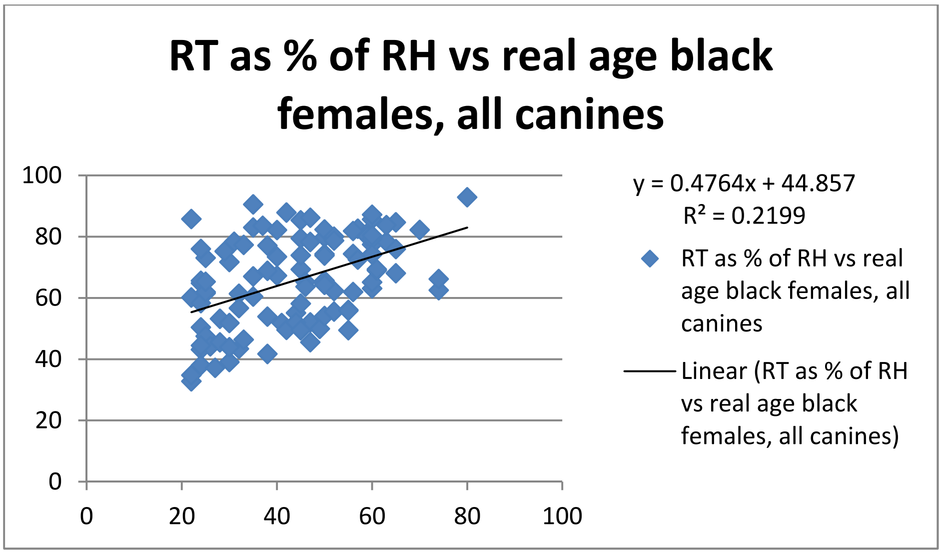


Figure 5.46. Average root transparency (rt) expressed as a function of root height (rh) against real age in black females, all canines. R^2=correlation coefficient. X-axis=real age, Y-axis=root transparency (RT) as percentage of root height (RH).

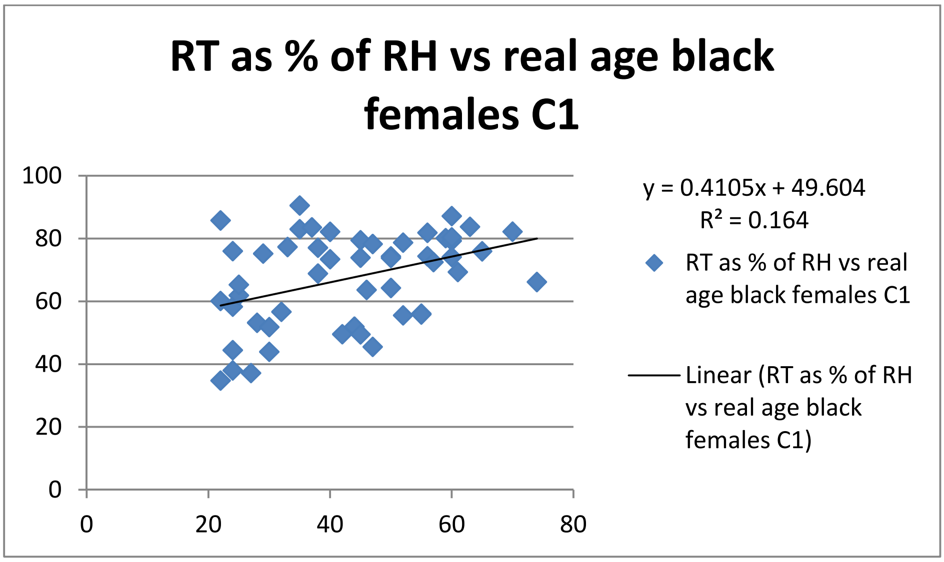


Figure 5.47. Average root transparency (rt) expressed as a function of root height (rh) against real age in black females, canine 1. R^2=correlation coefficient. X-axis=real age, Y-axis=root transparency (RT) as percentage of root height (RH).

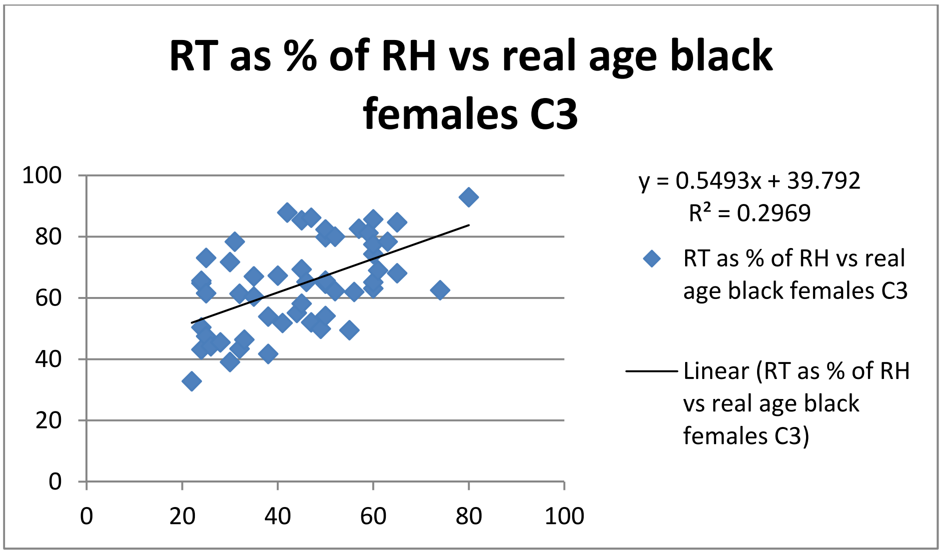


Figure 5.48. Average root transparency (rt) expressed as a function of root height (rh) against real age in black females, canine 3. R^2=correlation coefficient. X-axis=real age, Y-axis=root transparency (RT) as percentage of root height (RH).

5.3.3.3. White males

Average root transparency for all canines of the white males was correlated against the real age in Figure 5.49. The average root transparency obtained a correlation coefficient value of 4.2% (R^2=0.0425), which suggests that only 4.2% of the age could be attributed to root transparency of the pooled canines.

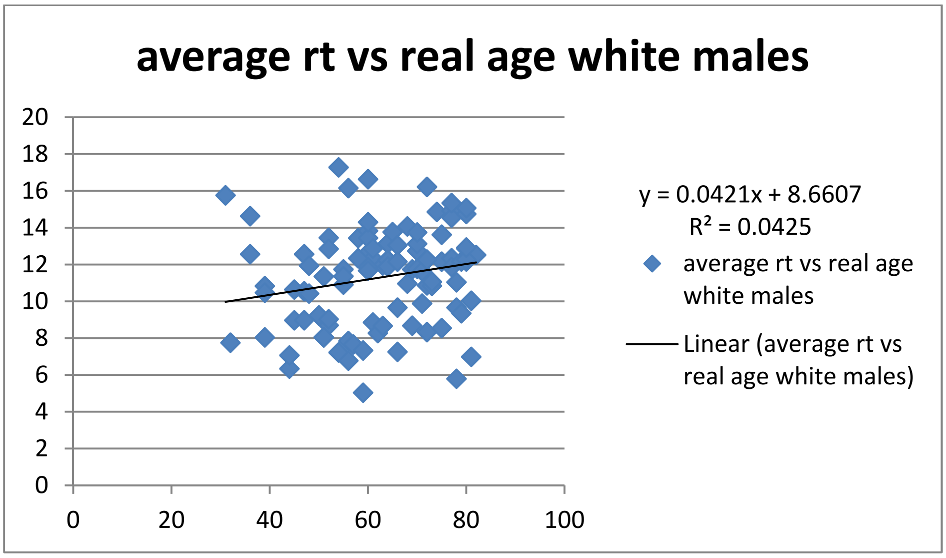


Figure 5.49. Average root transparency (rt) correlated against real age for white males, all canines. R^2=correlation coefficient. X-axis=real age, Y-axis=average root transparency (rt).

When the correlation were specified to the canine, the results obtained were that canine 3 obtained a better but weak correlation coefficient value (R^2=0.070) (Figure 5.51) than canine 1 (R^2=0.049) (Figure 5.50).

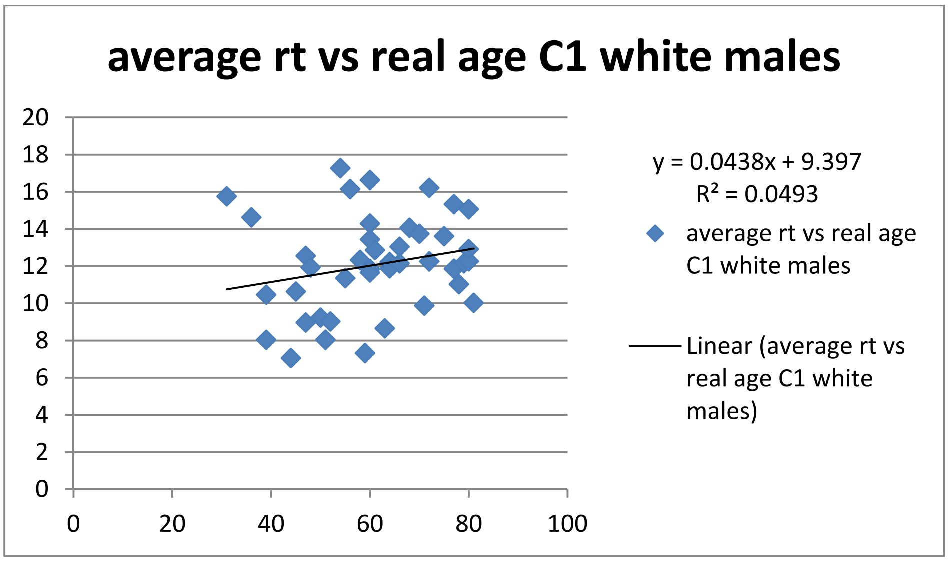


Figure 5.50. Average root transparency (rt) correlated against real age for canine 1 (white males). R^2=correlation coefficient. X-axis=real age, Y-axis=average root transparency (rt).

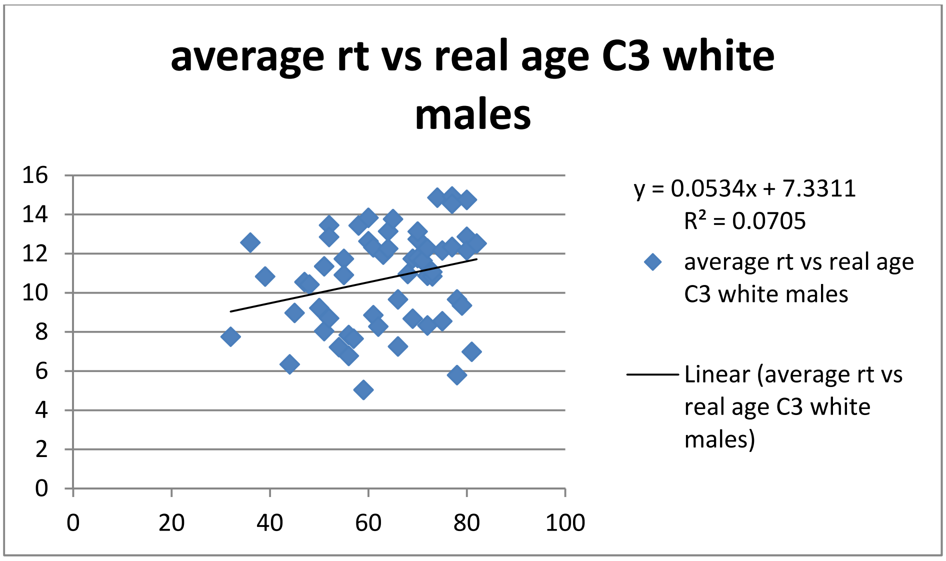


Figure 5.51. Average root transparency (rt) correlated against real age for canine 3 (white males). R^2=correlation coefficient. X-axis=real age, Y-axis=average root transparency (rt).

Root transparency was expressed as a percentage of root height against real age in Figure 5.52. Here it obtained a correlation coefficient value of 8.4% (R^2=0.084) for all canines. More specifically, canine 1 obtained a correlation coefficient value of 10.9% (R^2=0.109) (Figure 5.53) and canine 3 a value of 8.4% (R^2=0.084) (Figure 5.54).

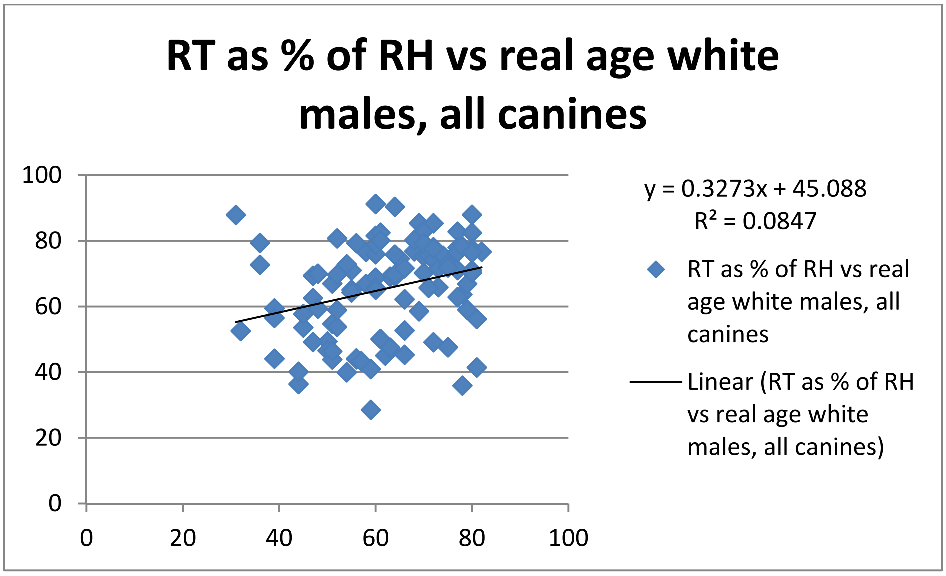


Figure 5.52. Average root transparency (rt) expressed as a function of root height (rh) against real age in white males, all canines. R^2=correlation coefficient. X-axis=real age, Y-axis=root transparency (RT) as percentage of root height (RH).

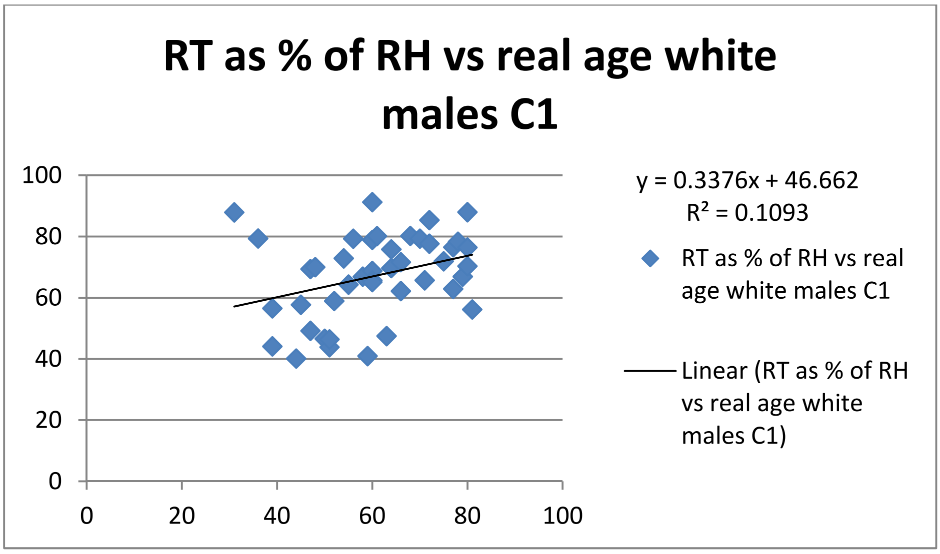


Figure 5.53. Average root transparency (rt) expressed as a function of root height (rh) against real age in white males, canine 1. R^2=correlation coefficient. X-axis=real age, Y-axis=root transparency (RT) as percentage of root height (RH).

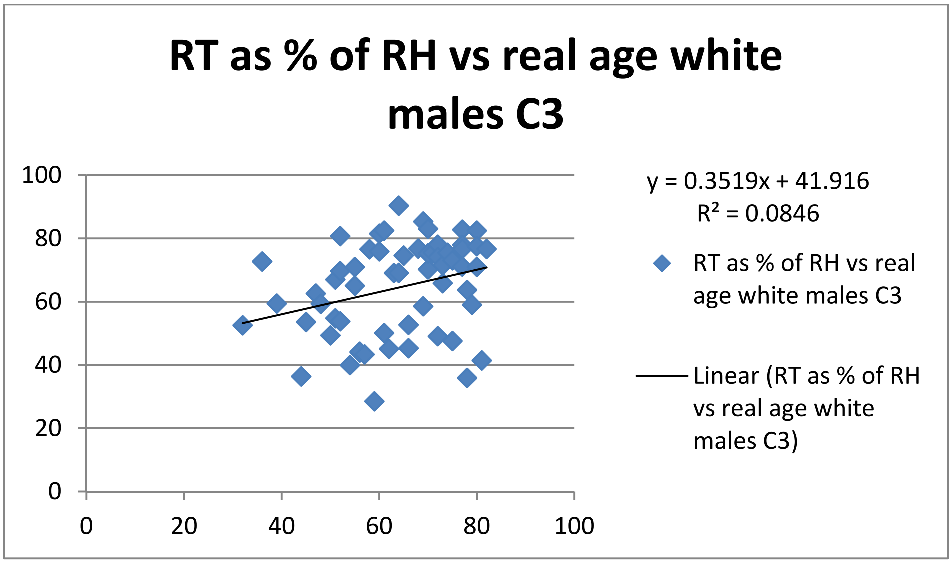


Figure 5.54. Average root transparency (rt) expressed as a function of root height (rh) against real age in white males, canine 3. R^2=correlation coefficient. X-axis=real age, Y-axis=root transparency (RT) as percentage of root height (RH).

5.3.3.4. White females

In Figure 5.55 the average root transparency (rt) for all canines was plotted against the real age of the white female group. The average root transparency obtained a correlation coefficient value of 1.2% (R^2=0.012). This means that 1.2% of age could be explained by root transparency.

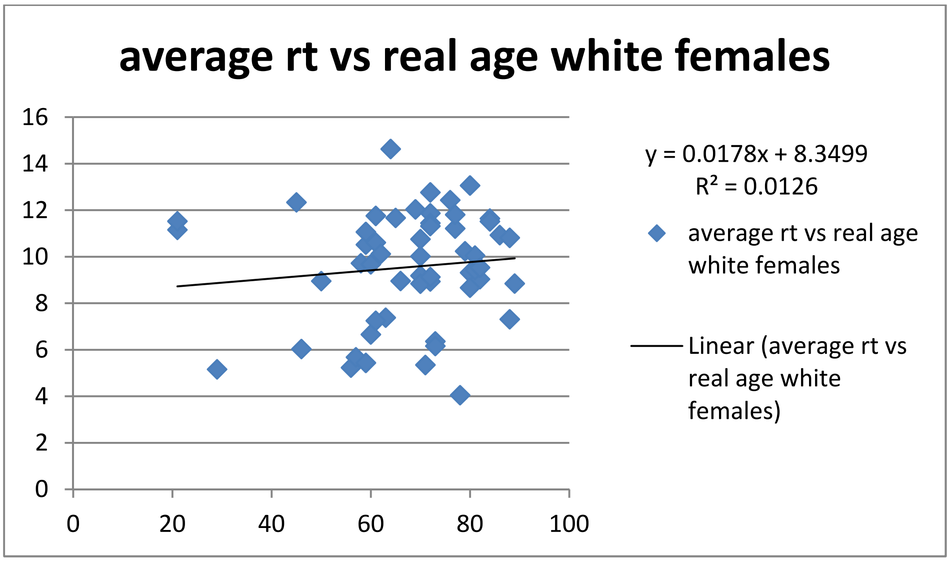


Figure 5.55. Average root transparency (rt) correlated against real age for white females, all canines. R^2=correlation coefficient. X-axis=real age, Y-axis=average root transparency (rt).

Correlations were refined to the specific canine. Canine 1 obtained a correlation coefficient value of 3.1% (R^2=0.031) (Figure 5.56) whereas canine 3 obtained a 1.6% correlation coefficient with real age (R^2=0.016) (Figure 5.57). Sample sizes were, however, small and showed a wide scatter.

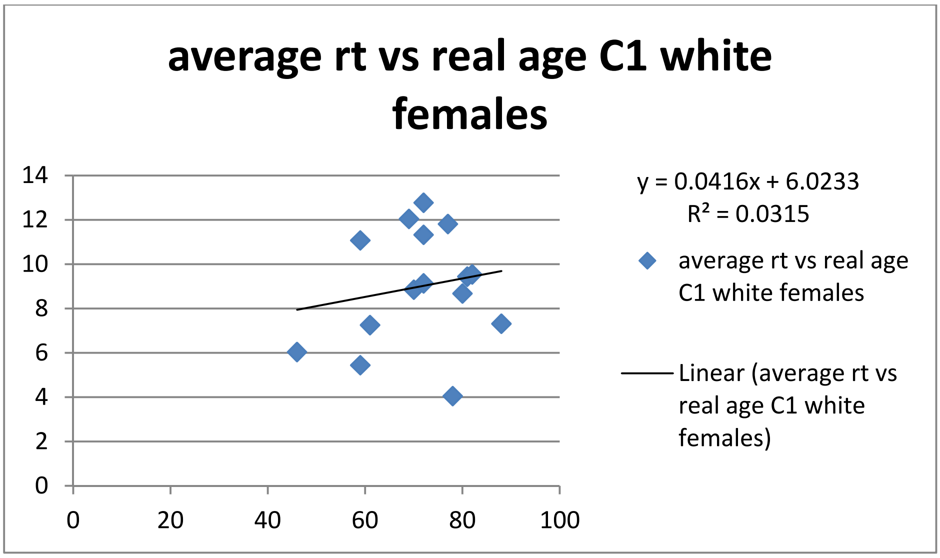


Figure 5.56. Average root transparency (rt) correlated against real age for canine 1 (white females). R^2=correlation coefficient. X-axis=real age, Y-axis=average root transparency (rt).

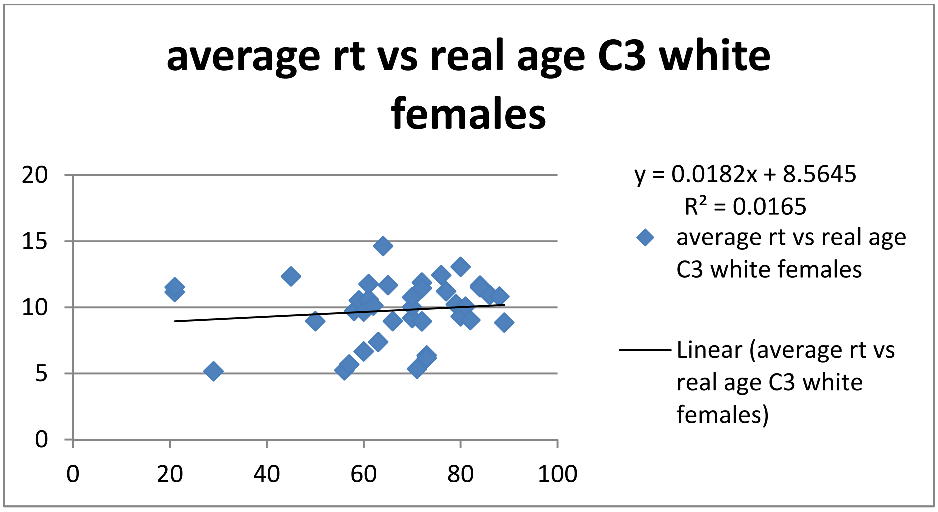


Figure 5.57. Average root transparency (rt) correlated against real age for canine 3 (white females). R^2=correlation coefficient. X-axis=real age, Y-axis=average root transparency (rt).

Lastly, root transparency was expressed as a percentage of root height against real age for all canines in Figure 5.58. Here it is evident that root transparency obtained a correlation coefficient value of 2.6% (R^2=0.026). When refined to specific canine, canine 1 obtained a correlation coefficient value of 3.9% (R^2=0.039) (Figure 5.59) and canine 3 a value of 4.2% (R^2=0.042) (Figure 5.60).

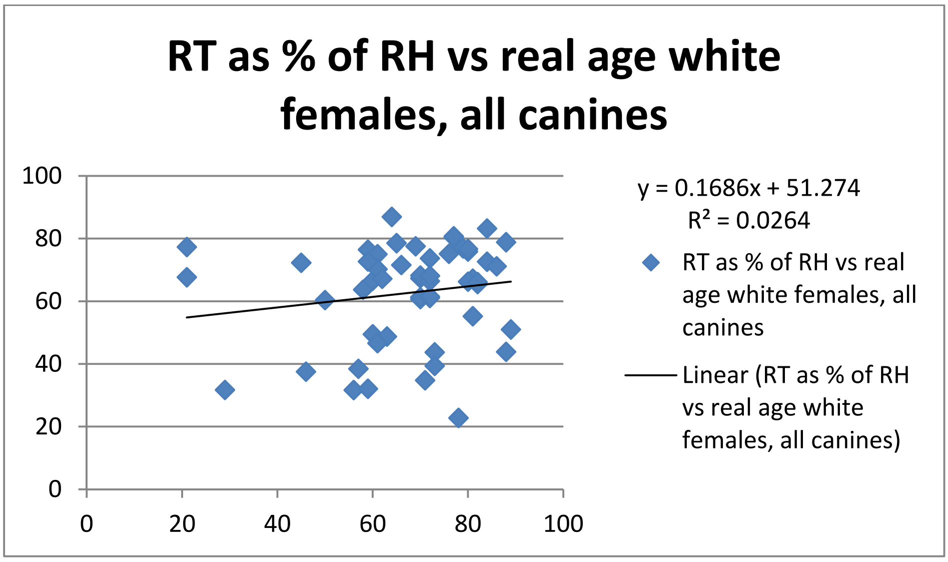


Figure 5.58. Average root transparency (rt) expressed as a function of root height (rh) against real age in white females, all canines. R^2=correlation coefficient. X-axis=real age, Y-axis=root transparency (RT) as percentage of root height (RH).

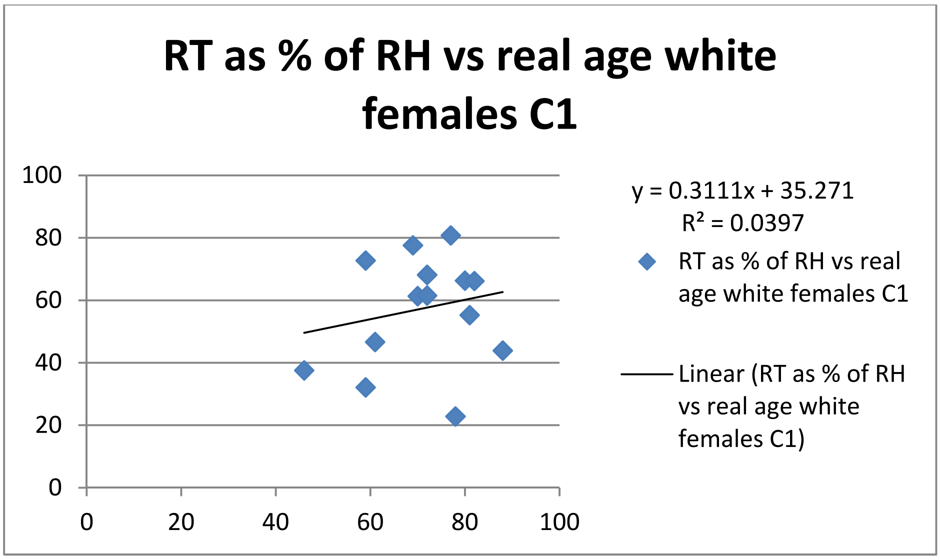


Figure 5.59. Average root transparency (rt) expressed as a function of root height (rh) against real age in white females, canine 1. R^2=correlation coefficient. X-axis=real age, Y-axis=root transparency (RT) as percentage of root height (RH).

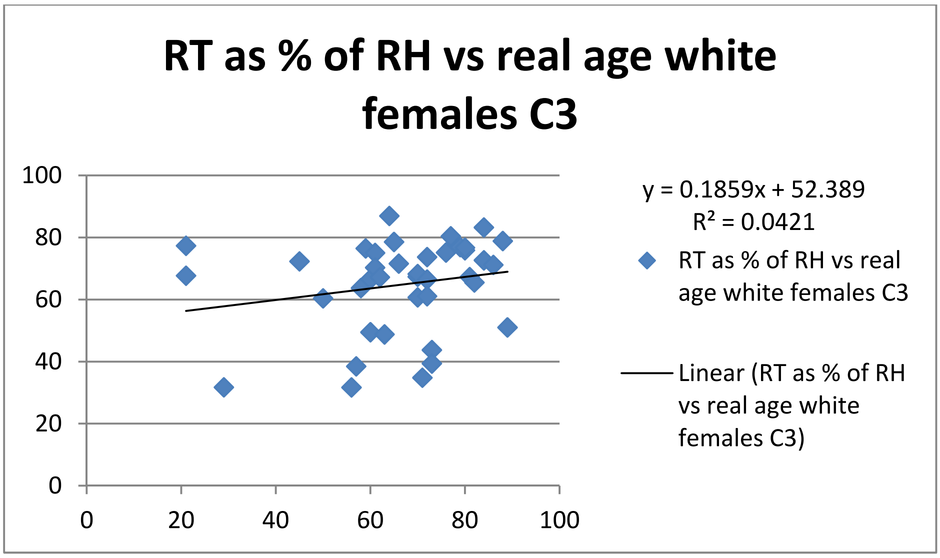


Figure 5.60. Average root transparency (rt) expressed as a function of root height (rh) against real age in white females, canine 3. R^2=correlation coefficient. X-axis=real age, Y-axis=root transparency (RT) as percentage of root height (RH).

5.3.4. Step 1: Summary

When considering Figures 5.1 to 5.60, a few relationships were noted. From all four groups (black males, black females, white males, and white females) the best correlated measurements against real age were firstly the periodontosis measurement, then root transparency and lastly root height. Root height in itself thus does not change much with age, indicating that it is a good parameter against which to standardize changes in root transparency and periodontosis. These correlations were the highest in the black male group, followed by the black females, white males and the white females. From the results of step 1 there seems to be a small but present correlation between the measurements and the real age of the individuals. Root transparency and periodontosis increase with age as expected, whereas root height slightly decreases with age. The parameter with the most correlation to age seems to be periodontosis (black males – Figures 5.13 to 5.18).

It was noted that for each group there was a specific canine that better represented the correlation between the measurement and real age. For example, canine 1 had the highest correlations with age in black and white males, whereas canine 3 worked best for black and white females. Periodontosis and root transparency were also expressed as percentages of root height against real age for all canines as well for canine 1 and 3 respectively, where periodontosis obtained the highest correlation coefficient value of 34.7% (black males, canine 1). Better correlation coefficient values were obtained for all four groups of the sample when these parameters (periodontosis and root transparency) were expressed as percentages of root height against real age. Overall, the correlations were low, but may be useful when used in combination.

5.4. Step 2: Lamendin Formula

At this stage the variables were substituted into the Lamendin formula to obtain a predicted age. Lamendin *et al*. (1992) proposed a formula, A = (0.18 x P) + (0.42 x T) + 25.53, where A is the predicted age, P is the periodontosis measurement, T is the root transparency measurement, and 25.53 is the age at which transparency of the root usually appears. This formula achieved an R squared value of 0.33. Only single rooted teeth were used in the original Lamendin study, which included the upper and lower incisors, upper and lower canines and premolars. Each individual was represented by a single tooth, but when several teeth were present for one individual the first choice was the central upper incisors, then lateral upper incisors, lower incisors, lower canines, upper canines and premolars. The periodontosis and root transparency measurements were functions of root height, as stated in the study of Lamendin *et al.* (1992). Lamendin's study showed that the males in the sample obtained a mean error of 10.1 ± 1.1 years and the females 9.4 ± 1.4 years. The different types of teeth also showed different mean errors; more specifically the upper canines obtained a mean error of 10.6 ± 3 years whereas the lower canines obtained 10.1 ± 3.8 years. In the present study the periodontosis and root transparency measurements were measured three times and the averages were used and expressed as functions of root height according to Lamendin *et al.*, 1992. These values were substituted into the Lamendin formula to get a calculated age for each of the individuals in the study. Also, the present study only used canines as the single rooted teeth.

Figures 5.61 and 5.62 show the correlations of the calculated Lamendin ages against the real ages for the black males (R^2=0.185) and females (R^2=0.266). When a specific canine was selected (not shown here as graphs), somewhat better results were obtained with canine 1 for the black males (R^2=0.217) and canine 3 for the black females (R^2=0.340).

The correlations between the calculated Lamendin ages and the real ages for all canines for the white males (R^2=0.092) and females (R^2=0.066) were also low (Figures 5.63 and 5.64). When a specific canine was selected (not shown here as graphs), canine 1 for the white males (R^2=0.129) gave better results. In the white females canine 3 provided a better but still low result, with an R^2 value of 0.096.

Figures 5.61 to 5.64 show the correlations between the calculated Lamendin ages against the real ages for the four groups in this study.

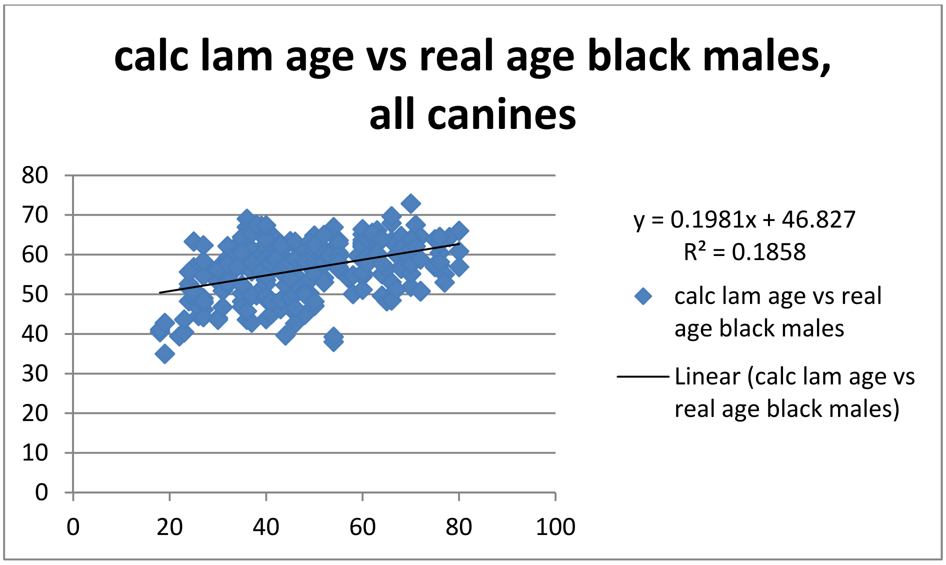


Figure 5.61. Calculated Lamendin age plotted against real age for black males, all canines. R^2=correlation coefficient. X-axis=real age, Y-axis=calculated lamendin age (calc lam age).

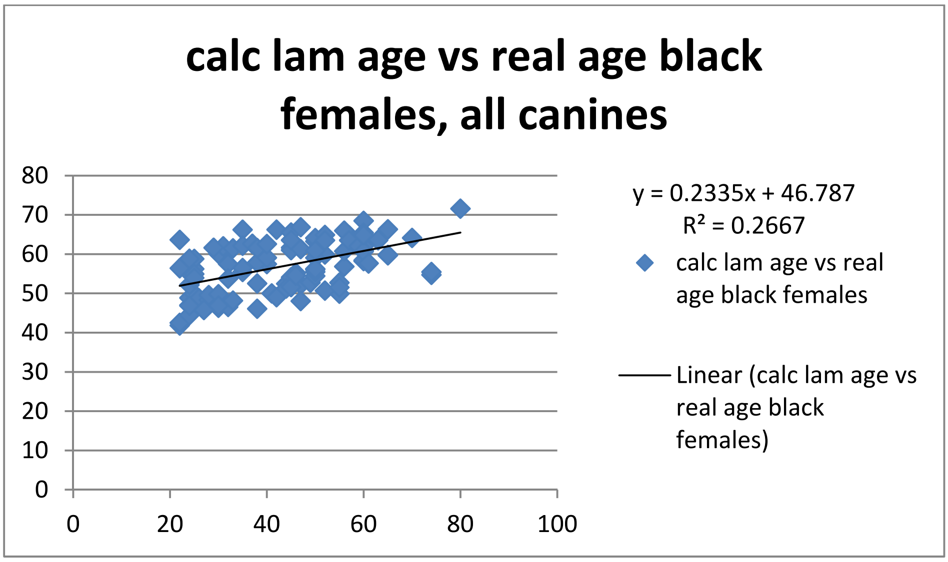


Figure 5.62. Calculated Lamendin age plotted against real age for black females, all canines. R^2=correlation coefficient. X-axis=real age, Y-axis=calculated lamendin age (calc lam age).

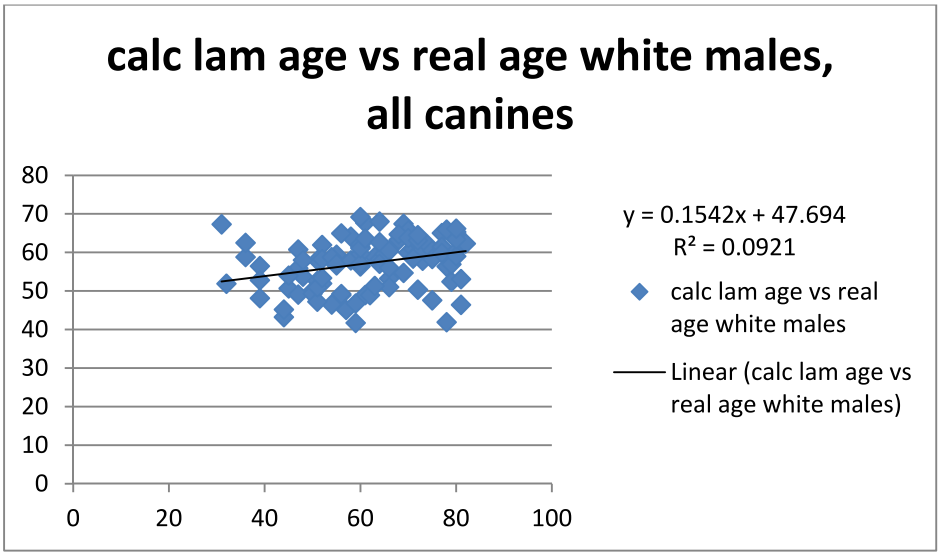


Figure 5.63. Calculated Lamendin age plotted against real age for white males, all canines. R^2=correlation coefficient. X-axis=real age, Y-axis=calculated lamendin age (calc lam age).

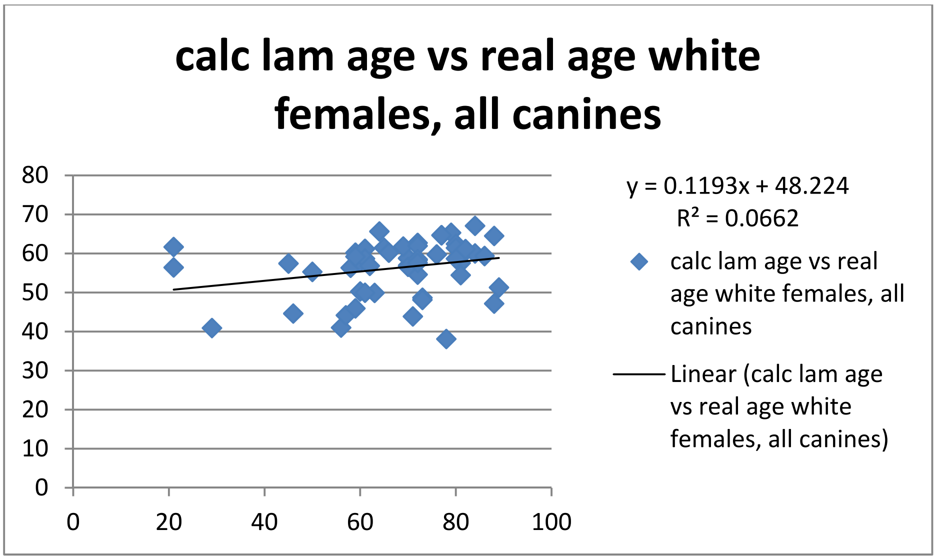


Figure 5.64. Calculated Lamendin age plotted against real age for white females, all canines. R^2=correlation coefficient. X-axis=real age, Y-axis=calculated lamendin age (calc lam age).

From these figures it can be seen that correlations between calculated Lamendin and real ages were very weak. They ranged from an R^2 value of 0.066 to 0.266, with the best results for the black group (Figure 5.61 - black males; R^2=0.185 and Figure 5.62 - black females; R^2=0.266). Correlations in the white group were particularly poor (Figures 5.63 - white males; R^2=0.092 and Figure 5.64 - white females; R^2=0.066). The small sample size for the white group may have played a role.

Lamendin *et al*. (1992) reported that the ME (mean error) of their forensic sample by decade was similar to those of their working sample and the global ME was even slightly lower in the forensic sample (8.4 versus 10 years). It was also stated that in individuals under 40 years of age, 46% of cases had an actual age included within the interval determined by the estimated age ± the ME of considered decade. A 90% accuracy was achieved for individuals over 40 years of age, which confirms that the Lamendin method is not useful in young adults. They also stated that their method had some limitations which were that large errors can be found in some individuals, mainly when they are either under 40 or over 80 years of age. However, the same positive results could not be repeated in the current study.

5.5. Step 3: Adapted Lamendin technique

Poor results were obtained with the existing Lamendin formulae. As some correlation was found to exist between actual age and periodontosis and root transparency, the existing formulae were adapted in an attempt to achieve closer estimates for practical reasons; only one canine per jaw was used.

The following sections contain the adapted formulae for the different population groups, different sex groups as well as specific canine.

5.5.1. Regression formula for all teeth for the total sample

Here Lamendin's method was applied to the whole sample, including all canines, with new coefficients applicable to the current sample. Periodontosis (P) and root transparency (RT) as percentage of root height (RH) were regressed against the age for the complete sample (n=536), and resulted in **Regression formula 1**:

Predicted age (A) = 20.68 + (0.57 x P) + (0.27 x RT), ME = 15.10, where

P = (periodontosis x100)/ root height; and

RT = (root transparency x100)/ root height.

For example, P=1.92 mm, RT=8.74 mm, RH=14.28:

P = (1.92 x 100)/14.28 = 13.44

RT = (8.74 x 100)/14.28 = 61.20

Therefore,

Predicted age (A) = 20.68 + (0.57 x 13.44) + (0.27 x 61.20)

= 44.85 ± 15.10 years.

5.5.2. Regression formula for specific canine for the total sample

Regression formula 2 used the upper canine (canine 1) only, with both sexes and populations combined (n=239):

Predicted age (A) = 14.92 + (0.85 x P) + (0.26 x RT), ME = 13.94

Regression formula 3 was calculated by using only the lower canine (canine 3) with both sexes and populations combined (n=297):

Predicted age (A) = 24.93 + (0.35 x P) + (0.31 x RT), ME = 15.76

5.5.3. Regression Formula for specific population group and canine, but no specific sex

No specific sex was selected here. The following formulae were created for the black group for each canine:

Regression formula 4 (Black group, canine 1, n=178):

Predicted age (A) = 9.89 + (0.79 x P) + (0.31 x RT), ME = 12.39

Regression formula 5 (Black group, canine 3, n=196):

Predicted age (A) = 14.50 + (0.56 x P) + (0.28 x RT), ME = 12.85

The following formulae resulted for the white group for each canine:

Regression formula 6 (White group, canine1, n=61):

Predicted age (A) = 41.45 + (0.56 x P) + (0.10 x RT), ME = 13.79

Regression formula 7 (White group, canine 3, n=101):

Predicted age (A) = 42.28 + (0.33 x P) + (0.23 x RT), ME = 13.54

5.5.4. Regression formula for specific canine, sex and population group

A specific canine was used in this part of the study, and the upper jaw was represented by canine 1 (upper left or right) and the lower jaw by canine 3 (lower left or right).

Separate formulae for each sex are given:

Regression formula 8 (Black males, canine 1, n=125):

Predicted age (A) = 10.34 + (0.87 x P) + (0.28 x RT), ME = 12.08

Regression formula 9 (Black males, canine 3, n=142):

Predicted age (A) = 18.25 + (0.55 x P) + (0.24 x RT), ME = 13.10

Regression formula 10 (Black females, canine 1, n=53):

Predicted age (A) = 9.08 + (0.44 x P) + (0.38 x RT), ME = 12.71

Regression formula 11 (Black females, canine 3, n=54):

Predicted age (A) = 4.02 + (0.60 x P) + (0.41 x RT), ME = 12.14

The following two regression formulae were included as a summary of the results when just the sex of the specific population was specified.

Regression formula 12 (Black males, any canine, n=267):

Predicted age (A) = 15.31 + (0.66 x P) + (0.26 x RT), ME = 12.74

Regression formula 13 (Black females, any canine, n=107):

Predicted age (A) = 6.33 + (0.53 x P) + (0.40 x RT), ME = 12.26

The corresponding formulae for the white group are as follows:

Regression formula 14 (White males, canine 1, n=43):

Predicted age (A) = 32.82 + (0.31 x P) + (0.30 x RT), ME = 12.69

Regression formula 15 (White males, canine 3, n=59):

Predicted age (A) = 47.17 + (0.07 x P) + (0.22 x RT), ME = 12.02

Regression formula 16 (White females, canine 1, n=18):

Predicted age (A) = 51.32 + (0.75 x P) + (-0.08 x RT), ME = 15.54

Regression formula 17 (White females, canine 3, n=42):

Predicted age (A) = 33.42 + (0.70 x P) + (0.26 x RT), ME = 15.18

Regression formula 18 (White males, any canine, n=102):

Predicted age (A) = 43.08 + (0.14 x P) + (0.24 x RT), ME = 12.32

Regression formula 19 (White females, any canine, n=60):

Predicted age (A) = 39.50 + (0.70 x P) + (0.16 x RT), ME = 15.11

Figures 5.65 to 5.83 show the correlations between real age and estimated age, using the adapted formulae.

Figure 5.65 shows the correlation between the calculated age and real age for the new Regression formula 1. Similarly, Figure 5.66 shows the correlation for the new Regression formula 2 etc, up to Figure 5.83 which shows the same for Regression formula 19. Correlations coefficients for all these remain low, ranging from 0.02 to 0.41.

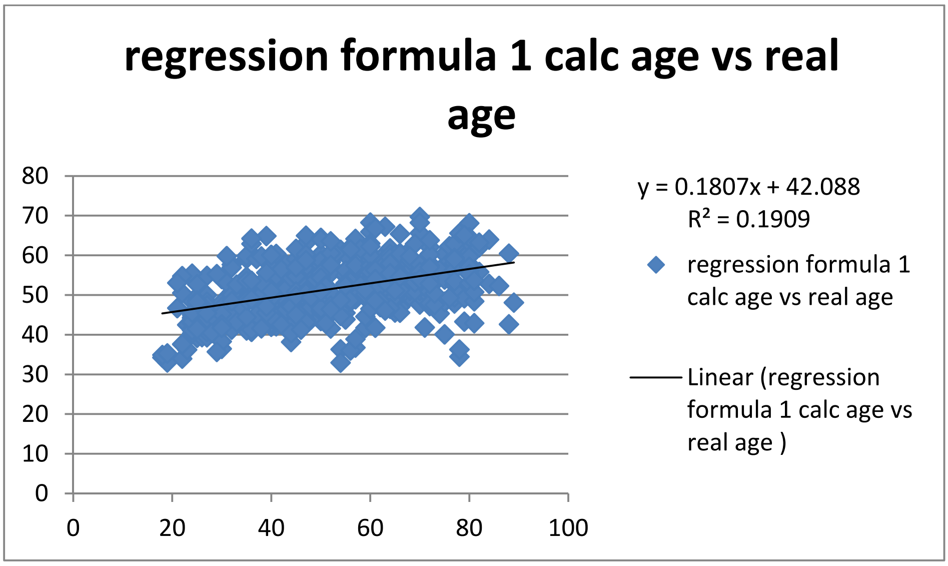


Figure 5.65. Calculated age from regression formula 1 correlated against the real age for the whole sample and all canines. R^2=correlation coefficient. X-axis=real age, Y-axis=calculated age (calc age).

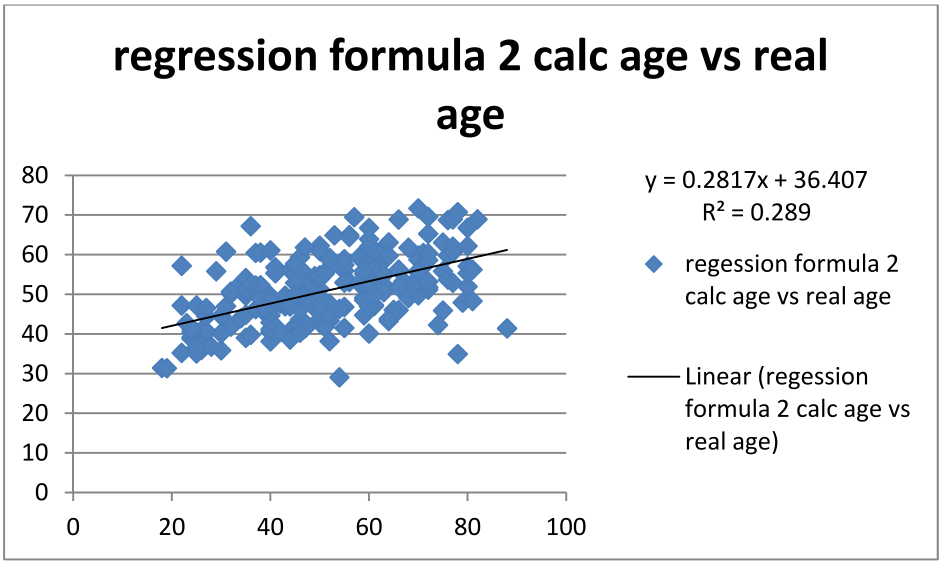


Figure 5.66. Calculated age from regression formula 2 correlated against the real age for the whole sample and canine 1. R^2=correlation coefficient. X-axis=real age, Y-axis=calculated age (calc age).

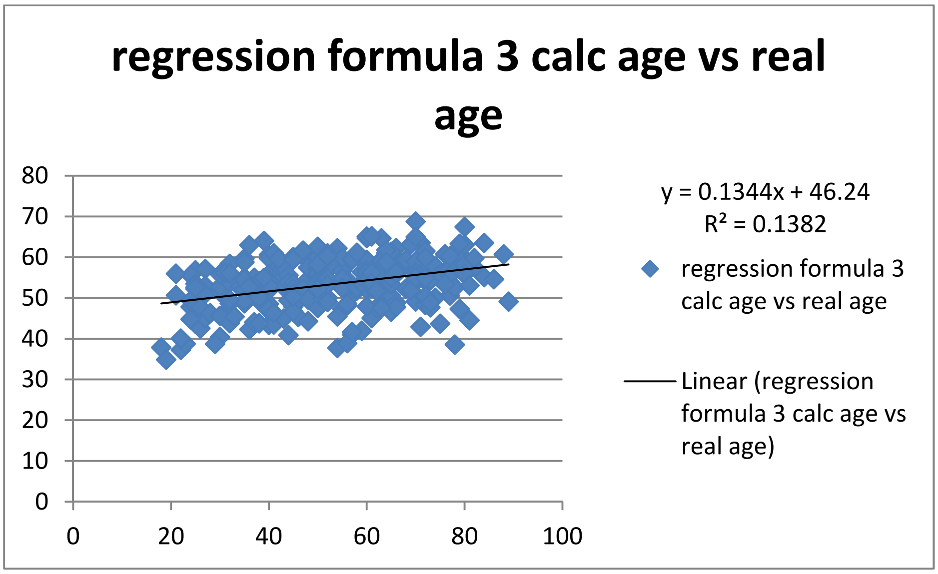


Figure 5.67. Calculated age from regression formula 3 correlated against the real age for the whole sample and canine 3. R^2=correlation coefficient. X-axis=real age, Y-axis=calculated age (calc age).

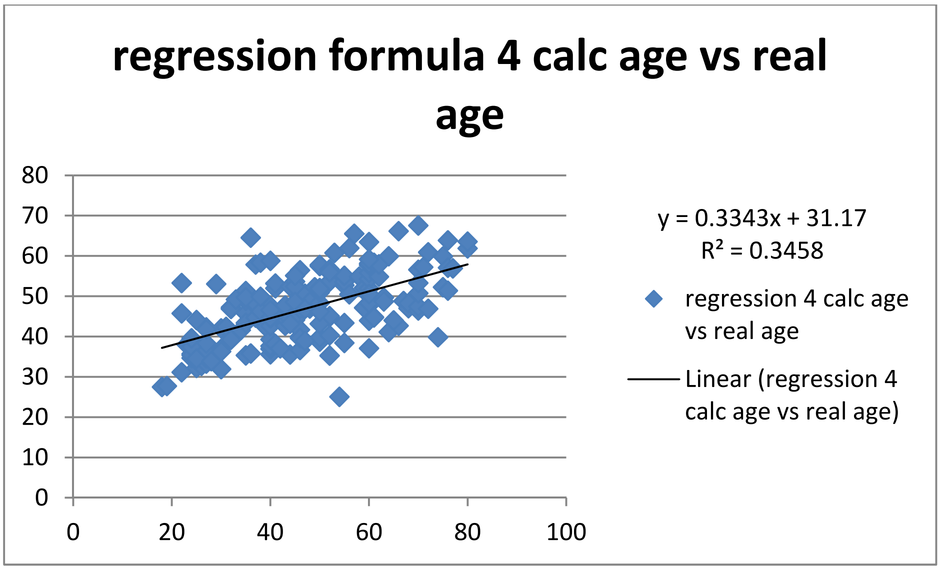


Figure 5.68. Calculated age from regression formula 4 correlated against the real age for the black population group and canine 1. R^2=correlation coefficient. X-axis=real age, Y-axis=calculated age (calc age).

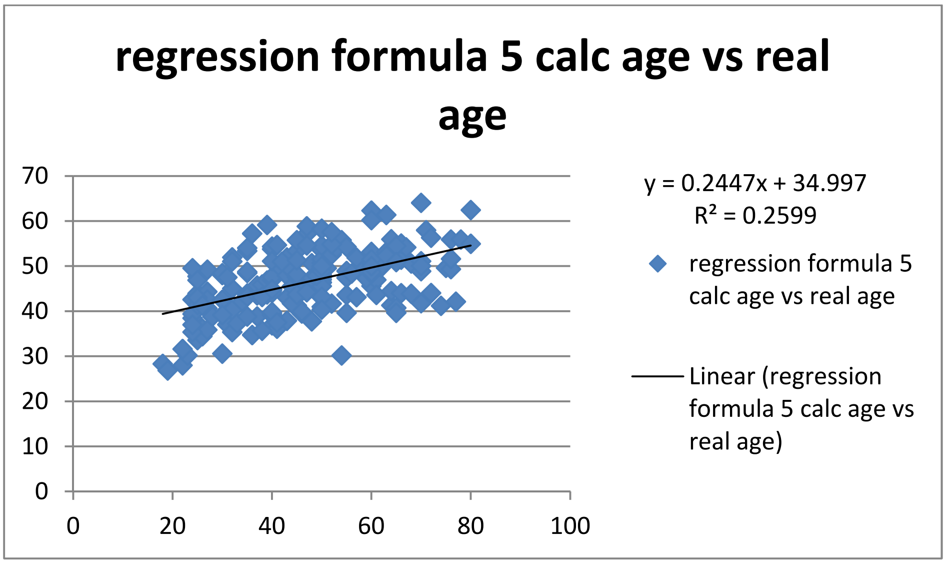


Figure 5.69. Calculated age from regression formula 5 correlated against the real age for the black population group and canine 3. R^2=correlation coefficient. X-axis=real age, Y-axis=calculated age (calc age).

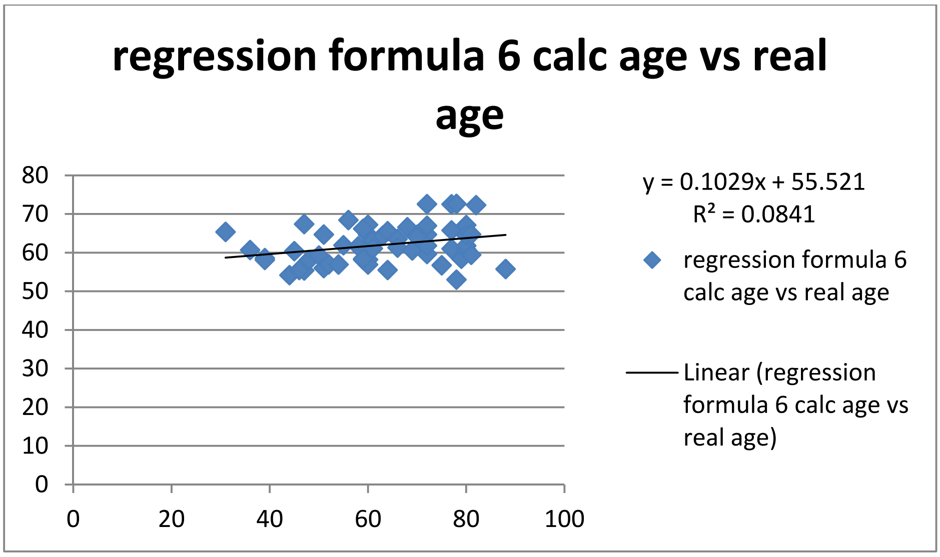


Figure 5.70. Calculated age from regression formula 6 correlated against the real age for the white population group and canine 1. R^2=correlation coefficient. X-axis=real age, Y-axis=calculated age (calc age).

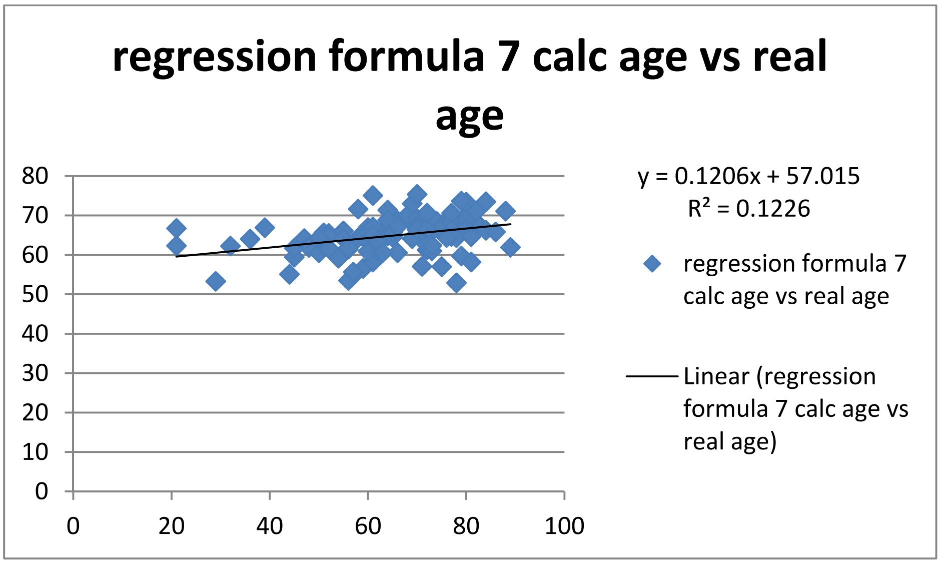


Figure 5.71. Calculated age from regression formula 7 correlated against the real age for the white population group and canine 3. R^2=correlation coefficient. X-axis=real age, Y-axis=calculated age (calc age).

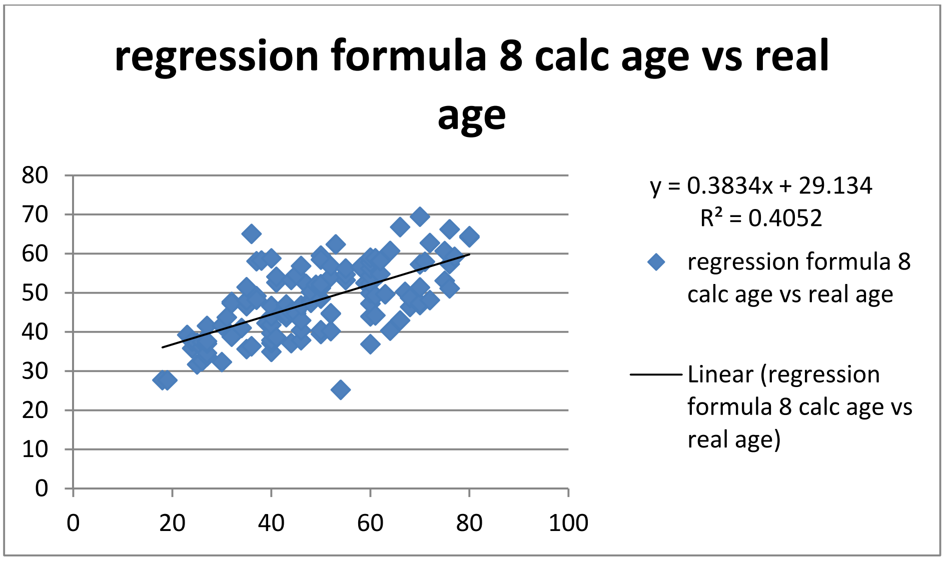


Figure 5.72. Calculated age from regression formula 8 correlated against the real age for the black male group and canine 1. R^2=correlation coefficient. X-axis=real age, Y-axis=calculated age (calc age).

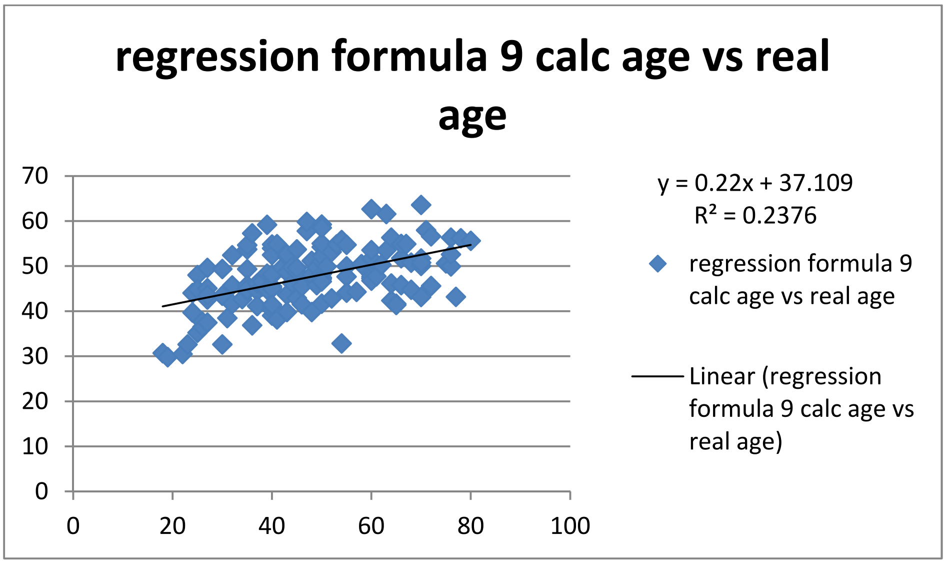


Figure 5.73. Calculated age from regression formula 9 correlated against the real age for the black male group and canine 3. R^2=correlation coefficient. X-axis=real age, Y-axis=calculated age (calc age).

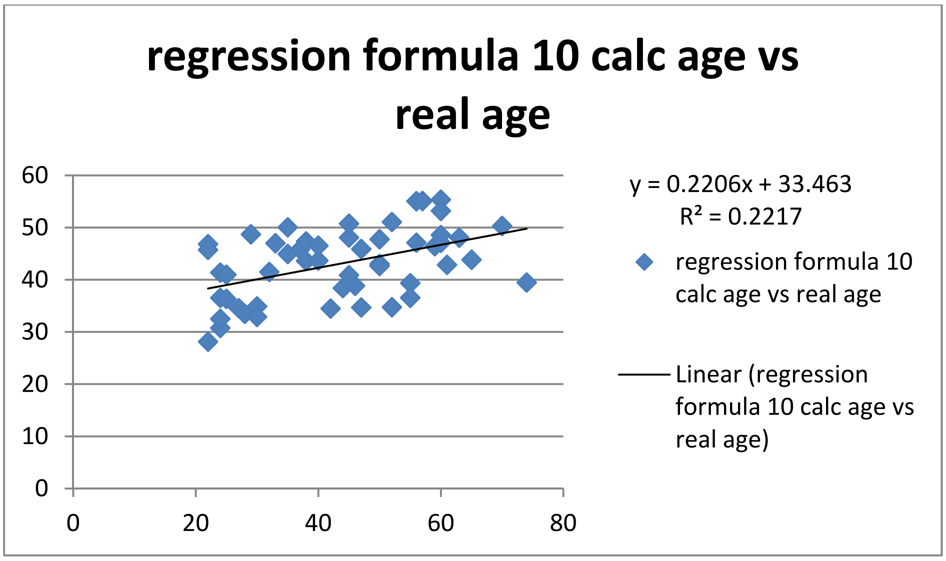


Figure 5.74. Calculated age from regression formula 10 correlated against the real age for the black female group and canine 1. R^2=correlation coefficient. X-axis=real age, Y-axis=calculated age (calc age).

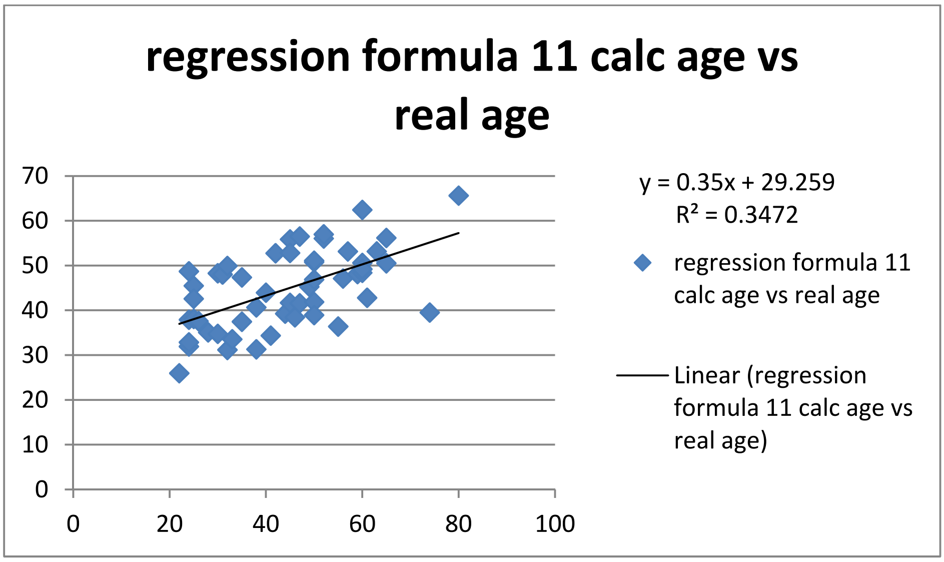


Figure 5.75. Calculated age from regression formula 11 correlated against the real age for the black female group and canine 3. R^2=correlation coefficient. X-axis=real age, Y-axis=calculated age (calc age).

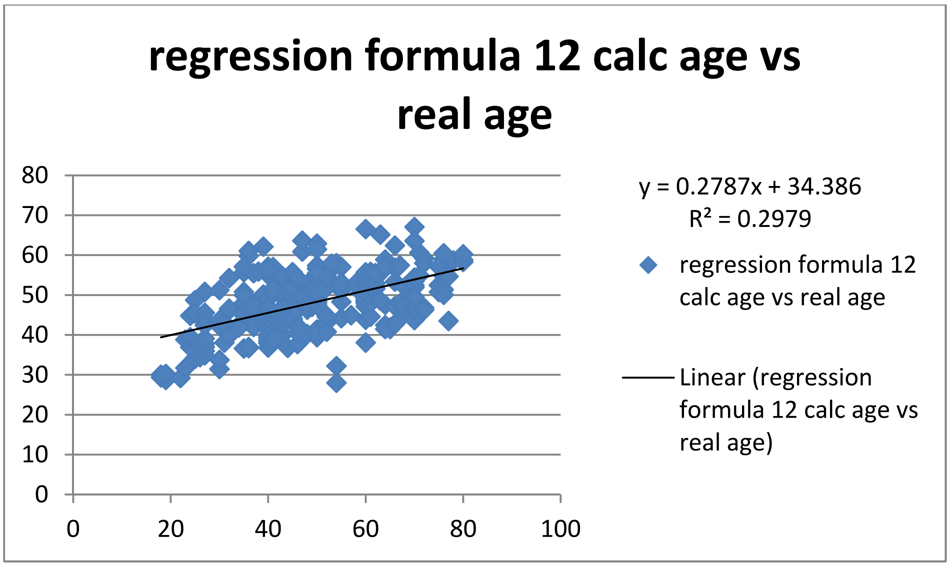


Figure 5.76. Calculated age from regression formula 12 correlated against the real age for the black male group and all canines. R^2=correlation coefficient. X-axis=real age, Y-axis=calculated age (calc age).

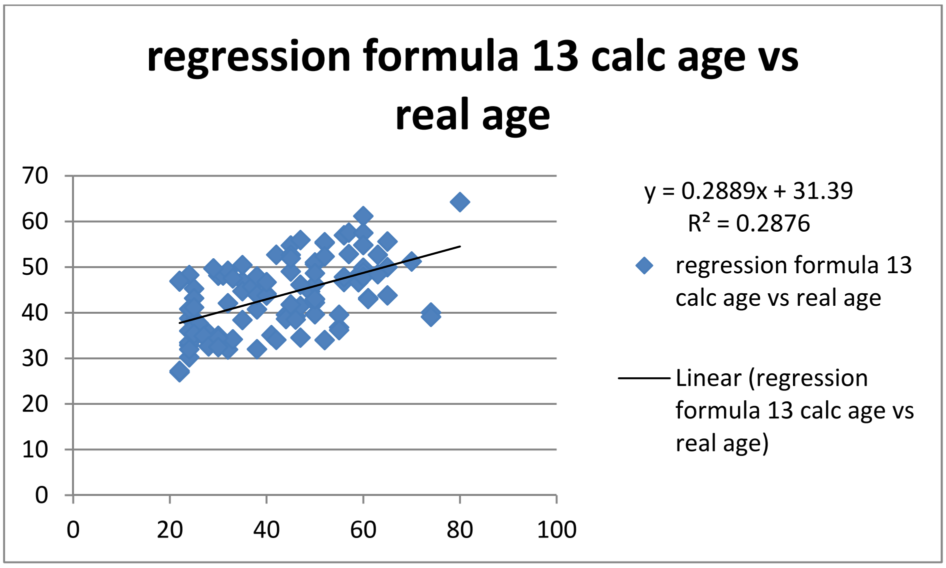


Figure 5.77. Calculated age from regression formula 13 correlated against the real age for the black female group and all canines. R^2=correlation coefficient. X-axis=real age, Y-axis=calculated age (calc age).

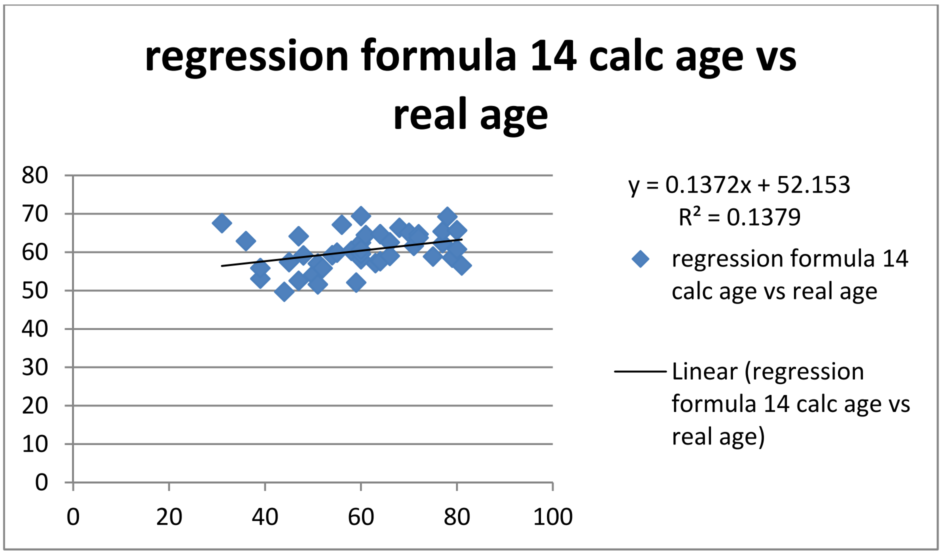


Figure 5.78. Calculated age from regression formula 14 correlated against the real age for the white male group and canine 1. R^2=correlation coefficient. X-axis=real age, Y-axis=calculated age (calc age).

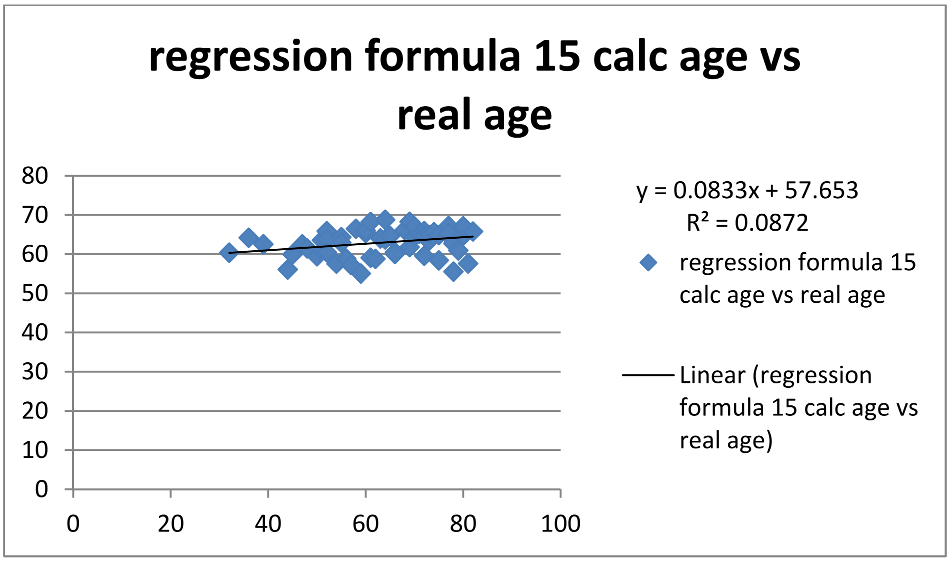


Figure 5.79. Calculated age from regression formula 15 correlated against the real age for the white male group and canine 3. R^2=correlation coefficient. X-axis=real age, Y-axis=calculated age (calc age).

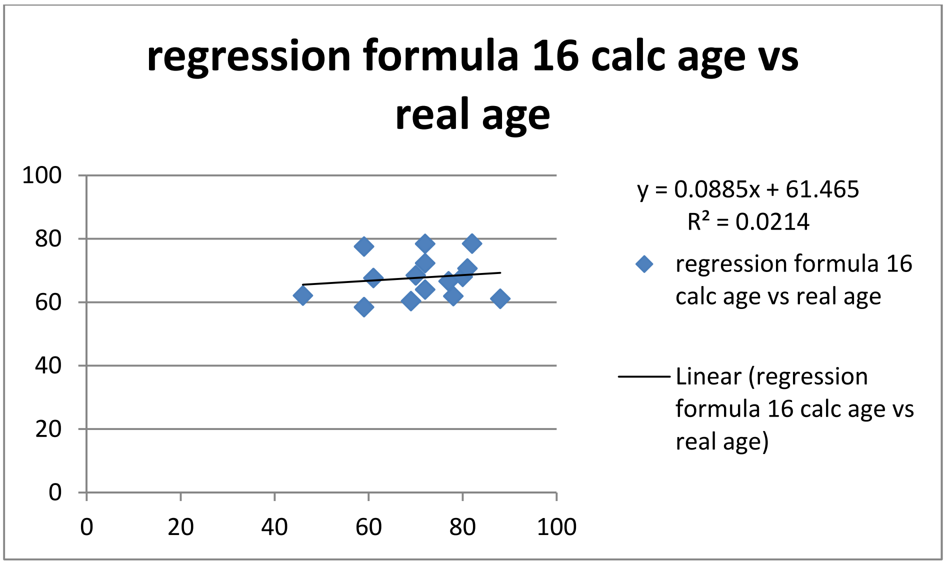


Figure 5.80. Calculated age from regression formula 16 correlated against the real age for the white female group and canine 1. R^2=correlation coefficient. X-axis=real age, Y-axis=calculated age (calc age).

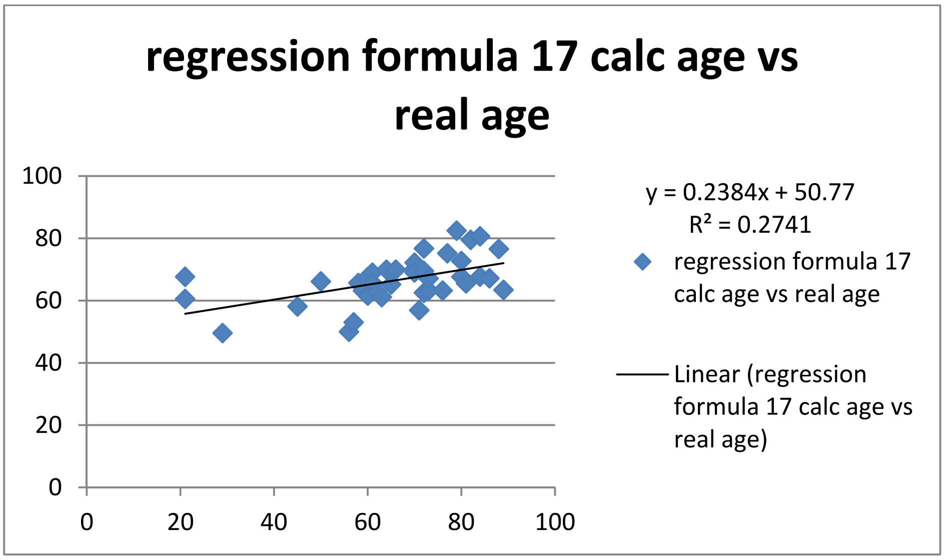


Figure 5.81. Calculated age from regression formula 17 correlated against the real age for the white female group and canine 3. R^2=correlation coefficient. X-axis=real age, Y-axis=calculated age (calc age).

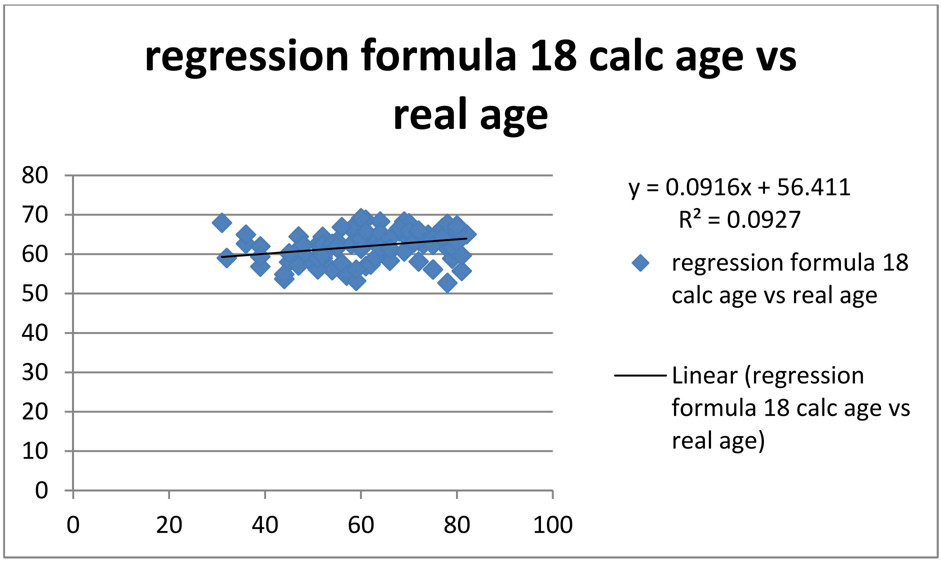


Figure 5.82. Calculated age from regression formula 18 correlated against the real age for the white male group and all canines. R^2=correlation coefficient. X-axis=real age, Y-axis=calculated age (calc age).

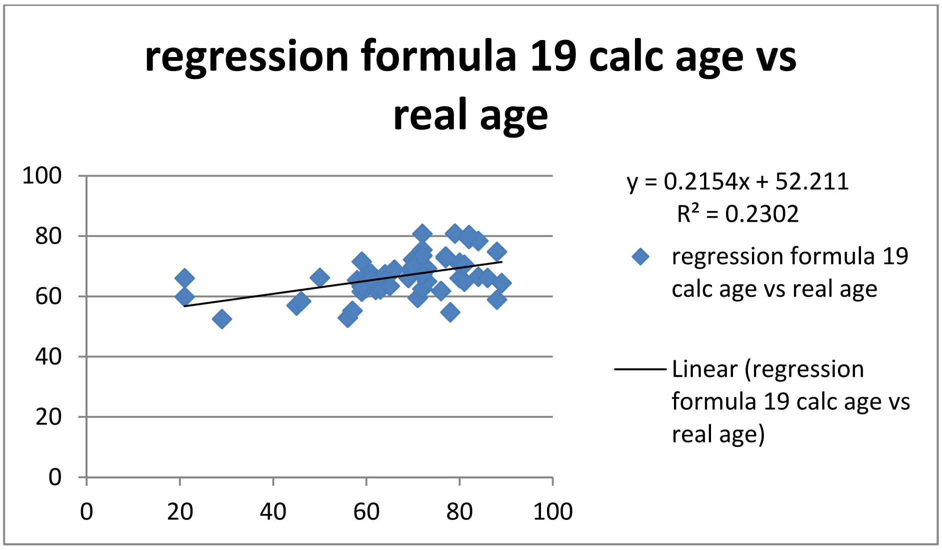


Figure 5.83. Calculated age from regression formula 19 correlated against the real age for the white female group and all canines. R^2=correlation coefficient. X-axis=real age, Y-axis=calculated age (calc age).

From the correlations of the newly calculated ages with real age of the individuals, it is clear that the predictive values are still very low. The black group gave better results with R squared values as high as 0.405, in comparison to the white population group with the highest R-squared value of 0.274. When the correlations were done for a specific canine (graphs not shown here), canine 1 (upper canine) gave better results for the males of both the black and white groups, whereas canine 3 gave better results for the females of the black and white groups. The correlations are weak, but better results were obtained with the newly calculated ages than with the calculated Lamendin ages in step 2. Another observation made was that the constant values of the newly adapted regression formulae varied throughout step 3. The value closest to Lamendin *et al*.'s (25.53) was obtained by regression formula 3 (whole sample, canine 3), of 24.93. This value, according to Lamendin *et al*. (1992) indicates the age at which root transparency usually appears. It was also stated that this constant of the equation makes the latter useless for individuals under 25 years of age. Also noted was that the ME (mean error) of each newly adapted formula varied from those obtained by Lamendin et al. (1992). The ME of the newly adapted formulae ranged from 12.02 to 15.76. From these formulae it was not possible to get close to the reported ME of Lamendin *et al*. (1992).

Chapter 6: Discussion

6.1 Introduction

The aim of this study was to estimate the sex and age of a South African sample by using the permanent maxillary and mandibular canines. This chapter will include the discussion on the measurements taken, sample size and the sex and age estimation parts of the study. Throughout the discussion the successes and challenges of this study will be mentioned.

6.2. Accuracy of measurements

The theoretical basis of metric analyses of teeth lies in the precision and repeatability of measurements, the conservative nature of the continuous variation, the direct link with the past as well as the demonstration of a heritability component for this category of biological variation. When looking from a statistical and mathematical standpoint, the continuous and correlated nature of measurements makes them highly suited for the application of multivariate statistical procedures (Katzenberg and Saunders, 2000). By using standardized measurement techniques, it allows accurate comparisons of individual teeth as well as those of populations and various subgroups. Caution should still be taken since there is a wide range of variability in tooth size both inter-populationally and intra-populationally.

Various authors have reported difficulties with measuring teeth (Goose, 1963; Tobias, 1967; Kieser, 1990; and Hillson, 1996), and reassessed the way in which the mesiodistal and buccolingual crown diameters are measured. Hillson *et al.* (2005) stated that the definitions given by Moorrees and Reed (1964) are the most widely followed. Although the system of measurements centres on the axis of the mesiodistal crown diameter, the line of the axis itself is still not clearly defined. In 1963 Goose suggested that the mesiodistal diameter axis should be between the contact points (Figure 2.9) of the tooth with its neighbours in normal occlusion. When malocclusion is present, it is said that the positions on the crown at which the contact points would have been in normal occlusion are used instead. In the case of unworn incisors and canines the definitions of the mesiodistal crown diameter are the same (Hillson *et al.*, 2005). As Hillson *et al.* (2005) stated, any change theoretically, in the axis of the mesiodistal crown diameter would lead to a change in the measurement axis of the buccolingual crown diameter. This buccolingual axis should be perpendicular to the mesiodistal crown diameter (Figure 2.10), but in no way could the angle of it be checked in practice. The easiest way of taking this measurement (buccolingual) on the incisors, canines and premolars is to find the maximum diameter from the buccal/labial to the lingual crown area. This is done by slightly rotating the tooth crown to get the maximum and by repeating the calliper readings. This will give an average diameter which may not be the actual perpendicular reading to the mesiodistal diameter or to the occlusal surface. Therefore, most observers consciously or unconsciously ''bend'' the rules of measurement to some extent (Hillson *et al.*, 2005). In this study, the maximum measurements for both the mesiodistal and buccolingual diameters were taken and good intra- and inter-observer repeatabilities (Intra-rater R^2= 0.996-0.999; inter-rater R^2= 0.833-0.989) were found, suggesting that they could be measured with reasonable accuracy, discussed in more detail below. The intercanine distance formed part of the set of measurements taken for sex estimation and no problems were noted. Acceptable accuracy was obtained, as discussed in more detail below.

The next set of measurements was used to test their usability in age estimation. Lamendin *et al.* (1992) defined three measurements on the tooth, namely periodontosis, root transparency and root length. Periodontosis (P) (gingival regression) occurs due to the soft tissue degeneration which surrounds the tooth and it progresses from the neck to the apex of the root. It is stated that it appears as a smooth yellowish area below the enamel which is darker but still lighter than the rest of the root. Tartric deposits are often found at this level. Therefore, this feature is measured on the labial surface as the maximum distance between the cemento-enamel junction and the line of soft tissue attachment. At the beginning of the measuring process, this feature (periodontosis) was difficult to measure since that “smooth yellowish area below the enamel”, was not that clearly visible but tartric deposits were found at this level most of the time. As the measuring process continued, it became easier to measure periodontosis since the eye became more adapted at looking at the feature with much more accuracy (more experience).

Root transparency is a physiologic feature and according to Lamendin *et al*. (1992), never appears before the age of 20 years, but it appears to become more common with advancing age. Root transparency can be present in the entire root of the tooth and can be seen through a light source. In this study a lamp was used as the light source. This feature is measured as the maximum height from the apex of the root to the visible transparency seen within the root. This measuring of the maximum height of transparency posed some difficulties. Some canines, when viewed in front of the light source, showed transparency along the margins of the root of the tooth and had no transparency in the central axis. Thus the maximum measurement was taken from the apex of the root up to the furthest point of transparency along the margins. Similar to what was found by other researchers (Miles, 1963; Johnson, 1968; Bang and Ramm, 1970; Vasiliadis *et al*., 1983; Bang, 1989; Lorentson and Solheim, 1989; Drusini *et al*., 1991; Thomas *et al*., 1994; Whittaker and Bahri, 1996; Sengupta *et al*., 1999; Katzenberg and Saunders, 2000), measuring root transparency proved to be difficult. It was found that it can be taken with much more precision when the tooth is cleaned with water. The line, at which this measurement should be taken, is much clearer and definite (see Figure 3.6 - Chapter 3) when viewing it in front of the light source, since all the dust and/or build-up are no longer present on the surface of the tooth. Therefore this measurement could be taken with much more accuracy after washing and would be recommended for future use. With this said, it should be noted that there was a sequence in which measurements were taken. Firstly, periodontosis was measured since the tooth needs to be *in situ.* Once the tooth is extracted, the root height and root transparency could be measured. Root height was the easiest to measure since the points where the measurement started and ended were very clearly stated in the definition provided by Lamendin *et al*. (1992). Root height was used as a reference tool in which periodontosis and root transparency were expressed as percentages of root height.

In this study, very good repeatability was found both within and between observers. These assessments of repeatability were done by means of one-way analysis of variance (ANOVA) and the Intra Class Correlation was determined through OLS (ordinary least squares). The intra-rater values (R-squared values) for measurements pertaining to both sex and age estimation were above 0.9, which indicates that the method of measuring is reproducible. The Intra Class Correlation (ICC) can fall in the range of greater or equal to zero or smaller or equal to one ($0 \geq ICC \leq 1$), where the correlation closer to one is the best result e.g. 100% repeatable. The R-squared values and ICC of the principal investigator (intra-rater) were all close to 1, which means the measurement method is reproducible. The inter-observer repeatability indicates how well the measurements between the principal investigator and another observer could be repeated. All of these values were above 0.8, except for the periodontosis of canines, which indicates that these measurements could not be repeated with a high degree of accuracy. This may be due to the difficulty in establishing and applying the definitions of the measurements to measuring the teeth and the fact that the teeth are not in the sockets by the time it has to be measured again (e.g. it might be difficult to measure it if the teeth are already taken out of the jaw). This in turn can affect the repeatability of the measuring method as well as the results of the study.

6.3. Sample size

The upper and lower canines of 498 skulls formed the sample of this study. Difficulties were found in the availability of enough specimens in each sex group of each population. Therefore two bone collections were used in an attempt to obtain an adequate sample. However, despite the best efforts, it was only the black males that were fully represented in number; the other groups were a bit more difficult especially when further categorizing them in age groups. Especially in the case of whites, particularly the females, it was difficult to obtain the same number of individuals as with the Black group. Reason being that there were not many of a specific group in the collection, some individuals were edentulous and some presented with tooth pathology, which made the use of specific teeth not possible. This may be one of the factors that can influence results, since a relatively good number of specimens are needed for statistical tests to be done to their full potential. This was especially true in the case of age estimation, where number of individuals per age cohort ranged between 0 and 70. Especially in the white sample, this sample is probably inadequate. Future research should attempt to enlarge the sample size of particularly the white population.

6.4. Sex estimation

Numerous studies, including those of Otuyemi and Noar (1996) and Brooke *et al.* (2009) have shown that significant differences exist between ancestral groups with regard to tooth size, particularly in the mesiodistal and buccolingual crown diameters. Apart from population differences, there are other factors contributing to tooth size variability such as sex, hereditary factors, bilateral differences, environmental and secular changes (Otuyemi and Noar, 1996; Kaushal *et al.*, 2003; Hemanth *et al.*, 2008; Brooke *et al.,* 2009). Therefore, tooth size, particularly the mesiodistal and buccolingual crown diameters, can be used in estimation of sex.

The sex determination was done by measuring the mesiodistal and buccolingual crown diameters of all four canines. It was decided to use only one canine per jaw, since no significant differences between left and right sided teeth were found. In addition, the intercanine distances for the upper and lower jaws were measured. These measurements would give an idea of the tooth size and general jaw size (intercanine distance). For both black and white populations of South Africa, the maxillary and mandibular canine indices where included. The black and white groups differed from one another with regard to tooth size, therefore it was decided to keep them apart. In general, the teeth of black South Africans were found to be considerable larger than that of white South Africans (Otuyemi and Noar (1996), Brooke *et al.* (2009), Ackermann and Steyn, 2010).

Statistically significant differences exist in the tooth sizes between the males and females, thus indicating their potential to be used in sex estimation. The index however, did not show any difference between males and females and was not used in subsequent analyses. This index was used in the past (Anderson and Thompson, 1973; Rao *et al*., 1986 and 1989) but presented unsatisfactory results. Modern statistics give a much more powerful way or method to analyze sex differences. With the stepwise discriminant function analysis, the maxillary and/or mandibular canine index was never selected. Descriptive statistics and discriminant function analyses were done. One-way ANOVA was used to obtain the significances for the measurements and indices. From the results obtained it was evident that there were statistically significant differences in the crown diameters and intercanine distances of the maxillary canines in both black and white males and females.

In general, it is clear that all measurements (MD and BL) of the males are relatively larger (statistically significant at $p<0.05$) than those of the females, making it possible to use discriminant function analysis in an attempt to develop formula which can be used to estimate sex in unknown individuals. The indices are clearly less usable and the intercanine distance did not contribute to the estimation of sexes. The results of the present study concur with the numerous other studies done, which include Seipel (1946), Moorrees (1959), Garn *et al.* (1964), Scott and Turner (1997), that reported considerably larger dental dimensions in the males than in females (where male teeth are 2-6% larger than female teeth, $p<0.001$) and that the largest differences were found in the canines in spite of the reduction in the tooth size of humans (Kieser, 1990; İşcan and Kedici, 2003).

It was also found that for all canines, the BL measurement of both populations were greater than their MD measurements. Therefore, the canines of both populations seem to be more cube-like (approximately equal diameters), which was also found by Hillson (1996). With the discriminant function analysis it was found that the BL measurement in general, was the best indicator for sex determination in both populations. As stated before, this may be due to the fact that although dental wear is one of the difficulties found in measuring tooth crowns, the only effect that wear has on the buccolingual diameter is when most of the crown has been lost through occlusal attrition. The mesiodistal diameter, however, is strongly affected by approximal (interproximal/ interstitial) attrition, even at the earliest stages (Jacobson, 1982; Hillson, 1996; Hillson *et al*., 2005). The present study sample was relatively smaller for some groups (white males and white females) due to the presence of tooth wear and/or alterations, therefore these teeth could not be included in the study.

Classification accuracies of discriminant functions were good. The highest accuracies for the blacks range from 76.1% to 81.8%, whereas the highest for the whites range from 72.7% to 87.0%. Overall, Function 1 (MD, BL, ID) gave the best percentages in sex classification with the maximum percentage of 87%, but only the mesiodistal and buccolingual crown diameters contributed to the estimation of sex. The intercanine distance did not have an effect. The mandibular canines presented with the highest classification accuracies for both populations, therefore it should be considered first in estimating the sex. Accuracies did not reduce significantly during cross-validation.

The accuracies found in this study correlate well with those obtained by numerous other researchers, which range from 70.9% to 93.3% (Kieser, 1990; Scott and Turner, 1997; İşcan and Kedici, 2003). Acharya and Mainali (2008) found that in their Nepalese sample the mesiodistal diameter showed an accuracy ranging from 77.4% to 83% whereas the buccolingual diameter resulted in 62.3% to 64.2% accuracy. This differs from the results obtained by the present study. To obtain the best possible results, both measurements should preferably be used. There are numerous studies that focused on discriminant function analyses using the pelvis, skull, femur, humerus etc (e.g. Steyn and İşcan, 1997; Bidmos and Asala, 2003; Bidmos and Dayal, 2003; Bidmos and Asala, 2004; Dayal and Bidmos, 2005; Patriquin *et al*., 2005; Franklin *et al.*, 2006; Barrier and L'Abbé, 2008; Dayal *et al.*, 2008). The accuracy of using teeth versus the for example the pelvis, femur or humerus; does not compare well since one of the studies done using discriminant function analyses on the pelvis of blacks and whites, resulted in accuracies reaching up to 95.5% (Patriquin *et al.*, 2005). The results of the present study does however show a relative comparison to other less dimorphic areas of the skeleton such as for example the talus (Bidmos and Dayal, 2003) or radius and ulna (Barrier and L'Abbé, 2008), which resulted in accuracies ranging from 76% to 88%. These studies were all based on South African samples.

The third function, MCI as sex indicator, seems to have difficulty in separating the sexes, producing accuracies not higher than 77.4%. The same accuracy for the MCI as sex indicator was reported by Anderson and Thompson (1973), whereas Rao *et al.* (1986 and 1989) found accuracies of 88%, 84.7% and 87.5% in their studies. The results from this study do not support those from Rao *et al*.'s studies and indicate that the indices did not contribute in separating the sexes successfully.

In conclusion, the canines, in particular the direct measurements of the canines, performed well in separating the sexes. Although not as accurate as, for example the pelvis, it can be useful particularly in juvenile individuals where the secondary sexual characteristics have not developed yet, or where preservation is poor.

6.5. Age estimation

For the purposes of age estimation, a different set of measurements is used. Lamendin *et al.* (1992) defined three measurements, namely periodontosis, root transparency and root length. They created a method based on Gustafson's technique and focused their study on two dental features: root transparency (RT) and periodontosis (P), on single rooted teeth (incisors, canines and premolars) which are both measured and expressed as an index value by relating these measurements to a fixed tooth measurement (root height (RH)). They applied multiple regression analysis to the variables and it resulted in the formula: A (age) = (0.18 x P) + (0.42 x RT) + 25.53; where P = periodontosis height x 100/root height, RT = root transparency x 100/root height.

Age estimation in this study was based on the same three measurements. One canine was taken as representative of each jaw, preferably the left canine – meaning canine 1 for upper jaw and canine 3 for lower jaw. The age estimation part of this study was done in three steps. The first step was to find correlations between the measurements (RH, P and RT) and the real age of the individuals. Root height in itself was not expected to change with age as it is used in the Lamendin formulae as a standard against which to judge increasing root transparency and periodontosis, although it was found to shorten somewhat with age. Positive but weak correlations were obtained for the other two features, where periodontosis obtained the highest correlation with age (R-squared = 0.35). It was also noted that canine 1 (maxillary canine) obtained the highest correlation for the males and canine 3 (mandibular canine) for the females. P and RT were also expressed as percentages of root height against real age and it was found that there is a slight increase in R-squared values in most of the cases. Positive but again weak correlations were obtained, where the highest correlation against real age was obtained by periodontosis (R-squared = 0.35). Although there was a slight increase noted in the R-squared values when these measurements were expressed as percentages of root height, the increase was not of such magnitude and thus it would be the same if these measurements are used on their own versus real age. Again canine 1 provided better correlations for the males and canine 3 for the females.

Root transparency did not obtain the highest results as was found in previous works (Miles, 1963; Johnson, 1968; Bang and Ramm, 1970; Vasiliadis *et al.*, 1983; Bang, 1989; Lorentsen and Solheim, 1989; Drusini *et al.*, 1991; Thomas *et al.*, 1994; Whittaker and Bakri, 1996; Sengupta *et al.*, 1999; Katzenberg and Saunders, 2000; Foti *et al.*, 2001). Periodontosis surprisingly gave better results than root transparency in this study. Hillson (1996) also reported that there are large clinical studies that showed a clear relationship between periodontosis and age. Dental hygiene and care play a big role in studies based on dentition, since it can influence the periodontosis measurement. For both measurements, the SA blacks obtained better correlations whereas the SA whites had very low correlations. The smaller sample sizes of the South African whites may have influenced these results. It was also clear from the results that canine 1 gave the best results overall for the males of both populations whereas canine 3 was best for the female groups.

Megyesi *et al*. (2006) used the Lamendin criteria on two historic skeletal samples and found that the post-mortem factors affect the applicability of the Lamendin technique to archaeological and historical samples. They reported that the root translucency disappears with time or is obscured by unknown post-mortem taphonomic effects related to the length of interment or post-mortem environment. They stated that caution should be taken when applying this technique to these types of samples or remains.

Step 2 was the application of the Lamendin technique to this study's sample, in which the measurements where substituted into the Lamendin formula. Overall very weak correlations between the calculated Lamendin ages and the real ages of the individuals were obtained - R^2 values of 0.092 to 0.266. The highest R^2 value was obtained by the black group. With this part of the study it was not even possible to report an R-squared value of 0.33 (Lamendin *et al.*'s study, 1992), e.g. where the age could not even be correctly estimated with an accuracy of 33%. Again the sample size of the whites could have influenced the results. The age of the sample could not be estimated satisfactorily when measurements were substituted in the Lamendin formula. The Lamendin technique was found to overestimate the younger individuals and underestimate the older ones. These results are in agreement with findings of other researchers (Foti *et al.*, 2001; Prince and Ubelaker ,2002; Megyesi *et al.*, 2006; Sarajlic *et al.*, 2006; González-Colmenares *et al.*, 2007; Martrille *et al.*, 2007; Meinl *et al.*, 2008)

Step 3 was the adaptation of the Lamendin technique to this sample. New coefficient and constant values were calculated throughout step 3, resulting in different formulae. Step 3 was started off by just incorporating the measurements into the adapted formula without any specifications as to sex, population group and specific canine. This part resembles the Lamendin technique in which the Lamendin formula was adapted and used on the whole sample. The differences came in that this study used canines, which also fall under the anterior teeth, and population specific coefficients and constant values. When only the two variables (P and RT) were regressed against the age of the whole sample without any specifications, it resulted in an R-squared value of 0.19. The constant value equalled 20.68, indicating that root transparency starts being visible from the age of approximately 20 years. A mean standard error of 15.10 years was obtained. When specifications were made with reference to the specific canine, sex and population group in differing combinations, the R-squared values ranged from 0.02 to 0.41 and the mean errors from 12.02 to 15.76. From these formulae the constant values also differed with each group and canine. The constant values are low for the black population (10.34, 18.25, 9.08, and 4.02), whereas for the white population this value became greater (32.82, 47.17, 51.32, and 33.42). According to Lamendin *et al*. (1992), this value represents the age at which root transparency sets in. This may be accepted for the rest of the groups but for canine 1 of the black males and canines 1 and 3 of the black females, it cannot be accepted that root transparency appears at the age of approximately 4 to 9 years of age if the rest of the permanent dentition should still erupt at these ages.

The sample size for the whites may have had an influence on the values calculated for the groups and thus the onset of root transparency may be at an earlier age. It differs from the standard Lamendin gave (25.53) and it also differs greatly from population to population, which was already mentioned by previous studies that population specific data should be collected. From this it is evident that population affinity as well as dental hygiene play a big role and can influence results greatly.

With these results it was found that canine 1 worked best for the males and canine 3 for the females. Thus, the overall result of this method in the estimation of age for a South African sample seems to be of relatively poor precision and accuracy. One objective that this study reached was that different formulae for each group in this sample could be created based on specifications made, seeing that there are differences between sexes as well as between populations. Although the results are unsatisfactorily, this part of the study not only created different formulae for each specification but also improved on the R-squared value from the original study on which this was based.

The mean error of the present study ranged from 12.02 to 15.76 years. Although a mean error of 15 years is high, Keough (2007) also reported a mean error of 13.31 to 14.04 years for age estimation with bone histology. When methods based on e.g. the pubic symphysis (Hanihara and Suzuki, 1978; Meindl *et al.*, 1985; Katz and Suchey, 1986; Brooks and Suchey, 1990; Sinha and Gupta, 1995; Berg, 2008) and auricular surface (Lovejoy *et al.*, 1985; Buckberry and Chamberlain, 2002; Osborne *et al.*, 2004; Mulhern and Jones, 2005; Hens and Belcastro, 2012), are considered, it is clear that their mean errors are also high. It seems that age estimation in adults still remains challenging, and that the Lamendin and modified Lamendin techniques are probably on par with other methods used in adult age estimation.

Chapter 7: Conclusion

In this study it was attempted to establish the sex and age of unknown individuals in a South African sample by using the permanent canine. It was concluded that:

1. Statistically significant sex differences exist in the dimensions of the canines of SA blacks and whites, especially in the crown diameters (MD and BL).

2. All direct measurements of the males were relatively larger (statistically significant, $p<0.05$) than those of the females. It was therefore possible to use discriminant function analysis to develop formula which can be used to estimate sex in unknown individuals.

3. It was possible to differentiate between the sexes with a relatively good accuracy of up to 87% by using the mesiodistal (MD) and buccolingual (BL) crown diameters as the basis of differentiation.

4. The mandibular canine presented with the highest classification percentages for both populations; therefore it should be considered first in estimating the sex.

5. Age estimation was based on applying the Lamendin technique on the South African sample. Positive but weak correlations were obtained, where periodontosis obtained the highest correlation with age (R-squared = 0.349).

6. Canine 1 (maxillary canine) obtained the highest correlation with age for the males and canine 3 (mandibular canine) for the females.

7. The Lamendin technique was found to work well in the mid-30's and -40's, but tend to overestimate the younger individuals and underestimate the older ones.

8. The Lamendin technique was adapted and modifications were made to develop formulae for each group. Although the R-squared values were still low, it provided better results than the original formula (highest R-squared value of 0.41). The mean errors for the different formulae ranged from 12.02 to 15.76 years.

9. With the Lamendin technique, it was found that canine 1 (upper jaw) worked best for the males and canine 3 (lower jaw) for the females. Overall this method produced results of bad precision and accuracy, although it is on a par with many other methods used in adult age estimation.

10. The canines remain the tooth of choice when considering techniques used to establish a profile of an unknown individual.

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