This is to certify that the
thesis entitled

AN INVESTIGATION OF STAGES OF EPIPHYSEAL UNION
IN THE CERVICAL VERTEBRAE OF YOUNG ADULT
SKELETONS

presented by

MELISSA ANN TORPEY

has been accepted towards fulfillment
of the requirements for the

M.S. degree in Forensic Science

N. Sauer
Major Professor's Signature

8-25-06
Date

PLACE IN RETURN BOX to remove this checkout from your record.
TO AVOID FINES return on or before date due.
MAY BE RECALLED with earlier due date if requested.

DATE DUE	DATE DUE	DATE DUE

AN INVESTIGATION OF STAGES OF EPIPHYSEAL UNION IN THE CERVICAL VERTEBRAE OF YOUNG ADULT SKELETONS

By

Melissa Ann Torpey

A THESIS

Submitted to
Michigan State University
in partial fulfillment of the requirements
for the degree of

MASTER OF SCIENCE

Department of Forensic Science

2006

ABSTRACT

AN INVESTIGATION OF STAGES OF EPIPHYSEAL UNION IN THE CERVICAL VERTEBRAE OF YOUNG ADULT SKELETONS

By

Melissa Ann Torpey

This research explored epiphyseal union of the cervical vertebral centra using a previously developed aging technique for the thoracic and first two lumbar vertebrae. In forensic anthropology, age estimation is an important aspect of developing a biological profile in cases of unknown identity. The timing and rate of epiphyseal union for the cervical vertebrae is studied to determine its relationship to known age of the decedents in the sample. Results from cervical vertebral ring epiphyseal union are compared to results of thoracic and first two lumbar vertebrae from a previous study to determine if the rate and pattern of epiphyses of the centra are similar or different. This study demonstrates that aging an individual through observations of cervical vertebral centra epiphyseal union is as accurate and reliable as using thoracic and first two lumbar vertebrae. Observations about the extent to which the union has progressed are important in understanding the amount of time it takes for complete fusion to occur and any pattern in how the fusion occurs. Observational and statistical analyses of the rate and pattern of the progress of epiphyseal union of the cervical vertebral centra are performed. Results from the analyses are significant and indicate an overall correlation of .781. The findings are important in skeletal age estimation both forensically and archaeologically in that they will help narrow what would otherwise be a broad age range, can be used to corroborate findings from other skeletal sites, and can be helpful in the absence of other key skeletal indicators of age.

Dedicated to:

My family who have proved to be

my sole support, comfort, and love throughout the years.

ACKNOWLEDGMENTS

I would like to thank Lyman Jellema in the Physical Anthropology section of the Cleveland Museum of Natural History for his assistance in collecting the data for this research. I would also like to thank Dr. A. Midori Albert for her constant motivation and inspiration to complete my research, follow a dream, and persevere. A special thanks to Dr. Christina DeJong for her guidance in the statistical analysis for this research. Moreover, my gratitude extends to Drs. Norman J. Sauer and Todd Fenton for enabling me to expand, as well as fine tune, my knowledge of anthropology and forensic science in the classroom, and for supervising my research.

TABLE OF CONTENTS

LIST OF TABLES

LIST OF FIGURES

Chapter 1

Introduction

"Science is, at its best, self-correcting and relentlessly self-critical" (Marks, 2002).

The field of physical anthropology is continuously changing, and for its subfield, forensic anthropology, change, correction, and improvement are of utmost importance. One of the goals of a forensic anthropologist working on a case of unknown identity is to develop, from skeletal analyses, a biological profile consisting of information on age, sex, ancestry, and stature. The biological profile is often imperative in establishing a positive identification for an unknown individual. One aspect of the biological profile that is especially important for the process of identification is age estimation. There are essentially two aspects to the methods for estimating age: (1) those based on skeletal growth and dentition and (2) those based on the deterioration of the skeleton (Byers, 2005). Methods based on skeletal growth and dentition are associated with aging children, generally 18 years and younger. Due to the rapid and regular growth of the skeleton and changes in dentition and the tighter genetic control over these growth and maturation processes throughout the subadult years, age ranges based on these methods are usually narrower in subadults, and broader in adults. The deterioration of the skeleton which begins in adulthood, or around 18 years of age produces larger estimated age ranges due to the wide range of human variation. Age ranges must be decided upon carefully because if the age range is too broad, it may be too inclusive, thereby being of little use. If the age range is too narrow, it may exclude likely candidates for a positive identification by eliminating the individual. The age estimation research in forensic anthropology is often driven by efforts to arrive at the narrowest possible age ranges within the scope of normal human variation.

Aging

Morphological changes in the skeleton are important indicators of age and are used extensively in both forensic and archaeology casework in physical anthropology. Generally, aging techniques revolve around the pelvis and the ribs for adults and long bone growth and tooth development in subadults. Although teeth form and erupt in the mouth at generally predictable ages, the third molar (Solari and Abramovitch, 2002) has become less predictable, due to its greater genetic instability, highlighting the need for an improved method to distinguish young adults and teenagers. The investigation of sex differences in age-related morphological changes has important implications in forensic anthropology, as well. Females generally develop and mature earlier than males (Pryor, 1923). Without information on skeletal development in females, criteria based solely on males are often substituted to estimate age at death for females. Not knowing whether growth or aging patterns in females and males are similar could be detrimental in the identification of an individual because ages may be over or underestimated.

In order to help narrow otherwise broader estimated age ranges, past studies have examined vertebral ring epiphyses because they fuse after long bone epiphyses and are active in the early to mid 20s. Investigated here are the stages and timing of the epiphyseal union of the cervical vertebral ring epiphyses. Examining the cervical vertebrae is unique in that no other study has previously focused solely on the timing and rate of the epiphyseal union of the cervical vertebrae and its comparability to the rest of the vertebral column. This study explored the epiphyseal rings of the cervical vertebrae and whether fusion occurs simultaneously or at different rates compared to the rest of the vertebral column.

Vertebral Aging

The scoring method of epiphyseal union for the thoracic and first two lumbar vertebrae developed by Albert and Maples (1995) was used in this study of cervical vertebral aging. Utilizing this method enabled a unique look at the applicability, consistency, and reliability of the Albert and Maples (1995) method as well as a general comparison throughout the vertebral column. The presacral vertebral column consists of seven cervical, 12 thoracic, and five lumbar vertebrae. Refer to Figure 1 on page 38 that depicts the cervical aspect of the vertebral column. Each typical cervical vertebra (C3-C7) possesses five epiphyses: one for each of the superior and inferior rings of the centrum, one for each tip of the transverse process, and one for the spinous process. This study focuses on epiphyseal rings. Due to the lack of a vertebral body for the first cervical vertebrae or C1 and the superior surface of the second cervical vertebrae or C2, the areas that are examined for this research are the inferior surface of the second and both surfaces of the last five cervical vertebrae or C3-C7. The epiphyses appear around puberty and the progression of union of the epiphyses can be used for the purpose of skeletal aging. Previous research (Albert and Maples, 1995) has concentrated on epiphyseal union of the centra of the thoracic and first two lumbar vertebrae due to the older age maturation of vertebral rings. This is important for aging individuals beyond age 18-20 where long bone epiphyses may be complete, but before skeletal maturation is reached enabling the distinction between early and mid 20s and beyond. It should be noted that a juvenile vertebral centrum is not equivalent to an adult vertebral body. The

adult vertebral body encompasses the centrum in addition to a small portion of the neural arch on either side of the centrum (Scheuer and Black, 2004). This terminology will become important in later discussions of growth and development.

Chapter 2

An Overview of Skeletal Age Estimation Methods for Young Adults

Age estimation of subadults and young adults is usually based on dental development due to the strong correlation of human tooth development and eruption with specific chronological ages (Pfau and Sciulli, 1994). When teeth are not available, other methods are used such as long bone lengths and an analysis of epiphyseal fusion. "Physiological age of the immature human skeleton must be assessed from one or more of the following systems: appearance and union of epiphyses, bone size, the loss of deciduous teeth, the eruption of teeth, and dental calcification" (Ubelaker, 1987; 1254). However, these techniques can be more environmentally sensitive than dental development causing more variation in the data and assessment of age. Methods of aging subadults can involve either an anthroposcopic analysis or a radiographic analysis as presented by Pfau and Sciulli (1994).

Third Molar Development

As an individual ages and possesses a full set of permanent teeth, tooth development obviously becomes less helpful in an age assessment. The accuracy of tooth development in determining age is best achieved when many teeth are undergoing growth and development. Solari and Abramovitch (2002) report that age estimation becomes increasingly difficult after age 14 due to most of the dentition being completely developed at this time. However, the development of the third molar can still provide evidence of chronological age. The third molars are the last teeth to erupt in human development, but they are many times the most variable tooth in regards to time of formation and eruption. As a standard, third molar eruption denotes an age of 18 years or more, but this assessment is continually becoming more unreliable (Mincer, et al., 1993).

Long Bone Epiphyses

The two most common methods of aging young adults are the development of the third molar and long bone epiphyseal union, both of which are sufficient for estimating an age of younger or older than 18 years. Although the timing of epiphyseal union varies among different individuals, the epiphyses throughout the skeleton typically follow a general sequence of union with the epiphyseal rings of the vertebral centra among the later fusing of the epiphyses (Byers, 2005). Another of the later fusing epiphyses is the medial end of the clavicle, but the clavicle is quite variable (Byers, 2005). Since, in general, long bone epiphyses complete union by the age of 20 years, the later fusing epiphyses such as the vertebral ring epiphyses can provide information on age beyond the third molar and most other epiphyses (Byers, 2005). That the vertebral ring epiphyses remain active later into the early to middle 20s extends the age range for which epiphyseal union as a method of age estimation can be useful. As a later fusing epiphysis, the vertebral ring epiphysis can provide narrower age ranges such as late teens, early 20s, and mid 20s rather than over or under 18 years of age (Byers, 2005).

Beyond the young adult years, age estimation of adult individuals is usually based on skeletal indicators representing degeneration or remodeling. These changes are most reliably observed on the pubic symphysis, fourth sternal rib end, and auricular surface. However, the auricular surface is more complicated and challenging to employ, and therefore, it is used less frequently.

Pubic Symphysis

The pubic symphyseal face, one of the most frequently used feature in the estimation of age in adults, is effective in that changes occurring at this site better correlate with chronological age than do other sites. It is most frequently used because the pubic symphysis is considered the most reliable due to wide variation in other methods such as dental wear or attrition and cranial suture closures. Also, the pubic symphysis is used most frequently because of the apparent clarity in its age-related changes thanks to important studies by T. Wingate Todd (1920, 1921), Brooks (1955), and McKern and Stewart (1957). Todd's studies revealed important findings and resulted in the development of four phases correlating changes in the pubic symphysis to age assessment. Phase one, denoting the earliest ages, begins with an age assessment of 25 years. After later statistical analyses, Todd's ten-phase system was merged into the Suchey-Brooks six-phase system. However useful these methods are in adult age estimation, the beginning phases only begin at the age of 25 years, and are therefore, not applicable to teenage and young adult skeletons.

Sternal Rib Ends

Another common adult aging technique was developed by İşcan and Loth (1986) using the fourth sternal rib end. İşcan and Loth studied the changes in form, shape, and texture over time to estimate an age progression. Four features of the sternal rib end change over time: surface bone, surface contour, rim edge, and rim contour. These features are used to describe the age phases established by İşcan and Loth to result in a reliable age technique for adults. Using the changes in morphological features to develop phases, İşcan, Loth, and Wright (1984, 1985) further developed these into age ranges for white females and white males. The first four phases developed for either white females or white males can be used with late teens and young adults. For white females, phase one begins at age 14, phase two encompasses 16 to 20 year olds, phase three represents 20 to 24 year olds, and phase four stretches from 24 to 40 years old. The results of this study show rapid changes in the early phases that taper off around 30 years old. In the study for white males, the findings are only slightly different and the rapid changes are still seen most often in the first four phases. For white males, phase one represents 17 to 18 year olds, phase two characterizes 18 to 25 years of age, phase three represents 19 to 33 year olds, and phase four is observed in 22 to 35 year olds. These age ranges, while not as broad as some techniques, still overlap from the late teens to the late 20s.

Radiographic vs. Anthroposcopic Techniques

Prior to extensive research on human skeletal remains, radiology was a primary source of gathering information about human bone growth. The most notable early studies on vertebral development are those performed by means of roentgenology in which living subjects of known age are studied and compared. Flecker (1942) provides an overview of the timing and sequence of ossification centers similar to that of Stevenson (1924). However, Stevenson studied dry human remains, whereas Flecker uses the roentgenographic method. Flecker's research discusses the influence to the timing of ossification centers of outside factors, such as health and endocrine factors. For each sex, Flecker provides the ages of the youngest and oldest individuals with no epiphyseal union, with at least half of the ossification centers present, and with complete union. Flecker does not differentiate between beginning union and nearly complete union; he only differentiates between fusion and non-fusion. In reference to the vertebral column, Flecker notes the lack of roentgenographic study in this area. The only mention of the fusion of the epiphyses of the vertebral bodies is noted on the lower thoracic and lumbar vertebrae. These epiphyses were observed in two individuals aged 12 years and 5 months and 13 years and 5 months.

Girdany and Golden (1952) also present a study on ossification centers based on roentgenograms. The authors report that the annular epiphyses of the vertebrae appear around puberty (defined as 16 years), but could appear by seven years of age. There is complete fusion of all of the centers of the vertebrae by 25 years of age. Girdany and Golden make special note of the sex difference in the time of appearance of ossification centers stating, "The time of appearance of centers of ossification differs considerably in the two sexes, and even in the same sex normal individual variation is great. An evaluation of the ossification age of any individual must take into account these variables" (923). Girdany and Golden do not discuss the progress of union between appearance and complete fusion because the progress of union on bones, such as the vertebrae, is not visible on radiographs.

Bick and Copel (1958) provide a radiographic and histologic study on the annular ring epiphyses of the vertebrae. The authors conclude that the epiphysis begins to fuse to the centrum around 17 years of age. By 18 years of age the fusion is apparently complete and at 20 years, the epiphyseal ring cannot be identified histologically. Hindman and Poole's (1970) research concurs with the age estimations of Girdany and Golden (1952) in that the epiphyses for the vertebral bodies appear around 16 years of age and fuse at approximately 25 years. However, the authors note several cases in which the vertebral body epiphyses are visible radiographically as early as two years of age. Although these studies are useful in observing the normal variation in time of appearance and fusion of the vertebral ring epiphyses, the radiographs cannot show the progress of union as the bone matures.

An evaluation of the studies based solely on radiographs show that x-rays are not detailed enough to show the progress of union. Radiographs are good for observing primary centers of ossification, but observing the progress of union is best accomplished by anthroposcopy. Utilizing anthroposcopic methods allows for detailed observations not obscured by features on a radiograph. Observations about how far along union has progressed is important in understanding the amount of time it takes for complete fusion to occur and any pattern in how the fusion occurs. Noting the progress of union also enables the researcher to accurately document what occurs between the appearances of the epiphyses and when the union is complete. Therefore, although radiographs could have provided a larger and more modern sample, dry bone of known individuals from a skeletal collection were used despite the limitation in sample size and ages.

Chapter 3

Vertebral Ossification

Primary Centers of Ossification

Bone development, or ossification, follows a predictable pattern in humans. There are two modes of ossification: intramembranous and endochondral (White, 2005). The vertebral column is formed through endochondral ossification, which occurs when a cartilage precursor or model is present before the bone is calcified and hardened (White, 2005). Although ossification begins before birth, it is not complete until many years afterward. The ossification centers that appear before birth are referred to as primary centers of ossification, while the ossification centers that appear after birth are known as secondary centers of ossification (Scheuer and Black, 2004). The vertebrae begin as cartilage models penetrated by a plethora of blood vessels. The marks of these blood vessels are visible on immature vertebrae as the billowed appearance on the superior and inferior surfaces of the juvenile centrum and occasionally the adult vertebral body (Scheuer and Black, 2004). The vertebrae grow both interstitially and appositionally. There are seven cervical vertebrae, the first two of which are unique, even from each other. The first cervical vertebra is formed by three primary ossification centers, the second cervical vertebra is formed by four primary ossification centers, and the last five cervical vertebrae are formed by three ossification centers (Scheuer and Black, 2004). Unlike the thoracic and lumbar regions of the vertebrae, the primary ossification centers of the neural arches appear before the ossification centers for the centra in the cervical vertebrae (Scheuer and Black, 2000). Refer to Figures 2 and 3, on pages 39 and 40, which display the general anatomical characteristics of C2 and a typical vertebra (C3-C7).

The first cervical vertebra, or the atlas, is formed by three primary ossification centers: one anterior arch and two neural arches, or lateral masses (Scheuer and Black, 2004). These neural arches later form the posterior arch. Other than an increase in size, there is relatively little morphological change in the atlas during the first year after birth. The two neural arches usually appear around the seventh fetal week. The neural arches fuse together between ages three and five (Scheuer and Black, 2004). The anterior arch is visible around one year of age. The anterior arch fuses with the neural arches between five and seven years of age (Scheuer and Black, 2004). The completion of the transverse foramina of the transverse process occurs around three to four years. By four to six years of age the atlas has grown to its final adult size (Scheuer and Black, 2004).

The second cervical vertebra, or the axis, is formed by five primary ossification centers: two neural arches, one centrum, and one for each half of the dens (Scheuer and Black, 2004). The dens, or odontoid process, a unique feature of the axis, actually forms from two separate ossification centers that appear between four and six fetal months and fuse around seven or eight fetal months or by birth (Scheuer and Black, 2004). In juveniles, the dens appears forked at the apex until the fusion of the secondary epiphyses at around twelve years of age (Scheuer and Black, 2004). The neural arches appear between seven and eight fetal weeks and fuse together between two and four years. The ossification center for the centrum appears around four or five fetal months. The centrum fuses with the dens at the dentocentral junction and with the neural arches at the neurocentral junction between the ages of four to six years (Scheuer and Black, 2004).

The dentocentral junction fuses with the paired neurocentral junctions between three and six years of age. A fusion line at this junction site may remain, but usually disappears by age nine or ten. The completion of the transverse foramina occurs between the ages of three and five years (Scheuer and Black, 2004).

Five cervical vertebrae, C3-C7, are each formed by three primary ossification sites: one centrum and two neural arches (Scheuer and Black, 2004). The appearance of the neural arches occurs first and commences at the end of the second fetal month. The ossification sites for the centra appear in the third and fourth fetal months beginning with C7 and progressing cranially until the last centra appear in C3 (Scheuer and Black, 2004). The neural arches usually fuse around two or three years of age. The centrum fuses with the neural arch at the neurocentral junction between three and four years of age. The transverse foramina are complete around three or four years of age (Scheuer and Black, 2004). Refer to Figures 4 and 5 on page 41 for a representation of the primary ossification centers of C2 and a typical cervical vertebra.

Secondary Centers of Ossification

In general, each cervical vertebra has five secondary centers of ossification, or epiphyses. Three of the secondary ossification centers are found on each tip of the two transverse processes and the tip of the spine. The last two secondary ossification centers, found in all vertebrae except C1 and the superior surface of C2, are flat, ring-like epiphyses, or growth plates, that encompass both the superior and inferior surfaces of the vertebral centrum (Scheuer and Black, 2004). Many of the research studies and anatomy textbooks state that the secondary epiphyses of the vertebrae fuse during early or young adulthood. However, the term "young adulthood" is subject to varying interpretation and can many times be defined differently or not at all depending on the source.

The secondary ossification centers for C3-C7 that appear later in childhood, including the tips of the transverse processes and spinous process, and on both the superior and inferior surfaces of the centrum (Scheuer and Black, 2004). The epiphyses of the transverse and spinous processes may persist until early in the third decade of life. The epiphyses on the superior and inferior surfaces of the centrum fuse in early adulthood. This is important because it extends the time period in which epiphyseal union can be used as an aging method. Since these are later fusing epiphyses, forensic anthropologists can utilize this method for narrowing age ranges to within early to middle 20s rather than 20 plus years. Figure 6 on page 42 displays a representation of the secondary centers of ossification of a typical cervical vertebra.

Chapter 4

Vertebral Ring Epiphyseal Union

Past studies of vertebral ring epiphyseal union from skeletal remains include Stevenson (1924), McKern and Stewart (1957), Buikstra (1980, 1984), Steele and Bramblett (1988), and Albert and Maples (1995). What is known from these studies concerning vertebral ring epiphyseal union is presented below.

In 1924, Stevenson pioneered an in-depth study into the order of epiphyseal union and the correlation of age to this order. Stevenson's sample consisted of 128 individuals of known age between 15 and 28 years. Stevenson established four distinct phases of epiphyseal union. The first stage was that of no union, characterized by a centrum with rough edges and a billowy surface. The second stage is that of beginning union, characterized by the change in the space between the epiphyses and the centra being replaced by a fine line, an increase in new bone deposition on the once billowed surface, and a smoothing out of the rough edges. The third stage is that of recent union and is characterized solely by the retention of a line of demarcation that can persist into the fourth stage as an epiphyseal scar. The fourth stage is that of complete union with the sole remaining characteristic at this point being the epiphyseal scar. Stevenson applies these stages of union to 28 epiphyses on 10 bones in order to draw a conclusion of a sequence of the union of epiphyses in the human body. Although Stevenson did not include any vertebrae in his study, the general results and conclusions regarding epiphyseal unions are significant. Stevenson notes that the period between 15 and 20 years is distinguished as the period of epiphyseal union. Also, Stevenson's results depict a fusion curve in which relatively few epiphyses begin the process of uniting prior to the age of 18 years. Throughout his study, Stevenson stresses that "At the outset it may be stated that the determination of age depends not so much upon the condition of any one or two epiphyses as upon the general condition of all epiphyses of the body" (1924; 82).

Anatomy textbooks have generally stated that the epiphyseal ring of the vertebral body fuses by the age of 25 years. McKern and Stewart were the first to examine the progression of union from epiphyseal early attachment to ages of final fusion. McKern and Stewart's 1957 publication encompassed a series of aging methods for young American males in response to the need for establishing positive identifications during the Korean War. McKern and Stewart studied skeletal indicators of age in 259 racially mixed males between the ages of 17 and 25 years. McKern and Stewart emphasized that the progression of union of all of the epiphyses of the vertebrae are important in skeletal aging. For the superior and inferior surfaces of the thoracic vertebrae, McKern and Stewart developed a scoring method using five stages (0-4) representative of the progress of union. McKern and Stewart reported that stages 2 and 3 appear most commonly between 19 and 21 years of age. The last stage, stage 4, is reportedly always present by 24 years. McKern and Stewart found no significant difference in the rate of union between superior or inferior rings. Regarding the sequence of epiphyseal union across various thoracic vertebrae, a consistent delay in union was found on the centrum surfaces between T2 and T7, especially T4 and T5, until the age of 24 years when compared to other thoracic vertebrae. By the age of 20 years, complete union was seen exclusively in T1 and T8-T12. Complete union in T2-T7 was achieved during the ages of 20-24 years. McKern and Stewart described the appearance of the superior and inferior surfaces of the vertebral centrum and developed a scoring method to estimate the amount of epiphyseal union. Five stages (0-4) were developed to score the superior and inferior surface of each vertebral centrum in the sample. These stages represented no union, beginning, active, recent, and complete union respectively. One of the observations McKern and Stewart noted was the billowing or striations that appeared on the immature surfaces of the centrum. According to the study, over time, the striations disappear and the surfaces become pitted and irregular. However, McKern and Stewart emphasize the persistence of the striations with age and ultimately state that human variation hinders the results of this study as assisting in age identification.

Whereas McKern and Stewart studied thoracic vertebrae, Buikstra, et al. (1980, 1984) focused on cervical vertebrae. During a case that called into question whether or not two vertebrae, a third and fourth cervical vertebra, were of the same individual, a black female. To answer this question, a study was conducted using a small sample of black females between 17 and 25 years from the Terry Collection housed at the Smithsonian Institute in Washington D.C. Epiphyseal ring union was observed for the second, third, and fourth cervical vertebrae. Epiphyses of each superior and inferior surface, as well as the posterior (dorsal) and anterior (ventral) halves, were scored based on McKern and Stewart's (1957) five stage scoring method where stage 0 represented no union and stage 4 represented complete fusion. Of several results, Buikstra found that the epiphyseal rings are generally fused to the centra by the age of 25 years. Buikstra stated that "By age 23 epiphyseal rings for most upper cervical vertebrae should show at least Stage 3 fusion" (1984, 131). The results also suggest that an advanced stage of maturation is found at any given age on the cervical vertebrae that are closer to the skull rather than that of the lower cervical vertebrae. Also, in general, the ventral half appears more advanced in maturation than the dorsal aspect.

In addition to McKern and Stewart and Buikstra, et al., Steele and Bramblett, in their textbook The Anatomy and Biology of the Human Skeleton, the growth and development of the vertebral column is described. The authors reported that the epiphyses of the cervical vertebral centra of males appear to begin fusion around the age of 17 years with fusion being complete around 25 years of age. Steele and Bramblett state that the sequence of vertebral ring fusion of epiphyses generally proceeds from the two ends (cranial and caudal) toward the middle (thoracic) (Steele and Bramblett, 1988). The cervical vertebrae are further broken down into the atlas (C1), axis (C2), and the true cervical vertebrae (C3-C7). The axis is described as the strongest of the cervical vertebrae because of the combination of the dens and the body, and it carries a unique statement by the authors denoting that the inferior surface of the axis unites by young adulthood, qualified as 19-20 years. There are two spans of time differentiated by Steele and Bramblett (1988) that are important in the fusion of the vertebral ring epiphyses. The first is adolescence, spanning ages 13 to 24 years, and second is young adulthood, from 25 to 49 years. In adolescence, the vertebral ring epiphyses appear and begin to fuse to the primary centers of ossification. By the end of adolescence, the vertebral ring epiphyses are completely fused to the vertebral bodies. Young adulthood is characterized by the complete fusion of the epiphyseal rings of the vertebral bodies, typically occurring around the age of 25 years. Steele and Bramblett report that during early adulthood, the vertebral bodies exhibit radiating striae, but as adulthood progresses and bone is deposited over the surface, the striae are gradually obliterated. In the later stages of adulthood and senescence, the striae become completely obliterated. Also, the distinction between the epiphyseal rings and the surface of the vertebral bodies will disappear (Steele and Bramblett, 1988). That the ages correlating with these stages of young, later, and old adulthood are not defined specifically prohibits any informative comparison to the conclusions of other studies.

One study that focused on the stages of progress of union of the vertebral ring epiphyses was Albert and Maples (1995), who studied the stages of epiphyseal union of the vertebral centra of the thoracic and first two lumbar vertebrae. This study took McKern and Stewart's stages one step further to explore sex and race differences in epiphyseal union, and to see if any secular changes might have occurred since it was about 40 years since McKern and Stewart (1957) published their results. While McKern and Stewart's sample consisted only of males, Albert and Maples included females into their sample. Also, Albert and Maples were able to expand the investigated ages to earlier than 17 and later than 25 to explore how early the epiphyseal ring union begins and if it extends beyond 25 year olds. Albert and Maples collected vertebrae from 55 individuals during autopsy for a sample that included American males and females, of African American and European ancestry. McKern and Stewart's original five stages were modified into four stages to better account for the last signs of union. This change enabled the stages to have early and late phases to better represent the progression of the epiphyseal union. Albert and Maples found that McKern and Stewart's stages did not account for the time frame in which the skeletal changes seem to occur. For example, beginning union and progressing union were placed in the same stage (Stage 1), but defined as an early and late phase because this skeletal modification occurs over the period of only a few months. Progression slows toward the completion of union and can continue for several years. The results showed that for the earliest or youngest age complete union was 24 to 25 years old, although the youngest to show complete union of all epiphyses was 18 years old. Also, Albert and Maples found no statistically significant sex or race differences in the timing or rate of the epiphyseal union. The first two lumbar vertebrae also followed a rate of union comparable to the thoracic vertebrae. In addition, the superior and inferior surfaces of the vertebrae showed no significant differences in the sequence of union. Although occasionally one surface of a vertebra either the superior or inferior epiphyses would show a more advanced phase of union than the other, the differences were not systematic. The superior and inferior epiphyses generally demonstrated the same stage of union.

As is evident from the above literature review, there have been no rigorous efforts to explore skeletal maturation of the cervical vertebral centra. Buikstra, et al.'s (1980, 1984) severed skull study was an attempt to apply to the cervical vertebrae McKern and Stewart's thoracic vertebral ring epiphyseal union aging method. However, this was a case that required aging only two cervical vertebrae, and was not an in-depth research study. Therefore, Buikstra's case did not involve comparisons of maturation among all cervical vertebral ring epiphyses or between cervical and thoracic and lumbar vertebral ring epiphyses. Data on cervical vertebral ring epiphyseal union can lead to a better understanding of the pattern of vertebral union in general. This in turn would aid skeletal age estimation efforts in that (1) it would help narrow what would otherwise be a broad age range, (2) could be used to corroborate findings from other skeletal sites, and (3) can be helpful in the absence of other key skeletal indicators of age.

Chapter 5

Methods

Sample

The cervical vertebrae of 77 individuals of known age, sex, and ancestry between the ages of 12 and 27 years were studied. The sample consisted of 11 white females, 27 black females, 15 white males, and 24 black males aged 12 to 27 years at death from the Hamann-Todd Osteological Collection housed in the Physical Anthropology section of the Cleveland Museum of Natural History in Cleveland, Ohio. Epiphyseal union data were collected from the inferior surface of the centrum of C2, and from the superior and inferior surfaces of the centrum of C3 through C7. The first cervical vertebra (C1) has no centrum, and the superior surface of C2 does not have a ring epiphysis, so they were excluded from this study.

Table 1 – Sample Demographics.

	Blacks	Whites
Males	24	15
Females	27	11

Scoring the Progress of Vertebral Ring Union

Epiphyseal union is a continuous process; however, the points at which key changes occur can be represented by using a basic scoring system. The scoring method utilized in this study was based on the method Albert and Maples (1995) modified for their study of the thoracic and first two lumbar vertebrae. The stages are described below.

Stage 0 – This stage is characterized by no union, where there is no attachment of the ring epiphysis of the centrum. The superior and inferior surfaces of the centrum may be billowed or striated due to the radiating vascular channels stemming from the anterior and posterior central perforating arteries. The edges of the centrum are dull and rounded until they become sharper right before union begins. The surface of the centrum is rough and usually has a bare appearance. There is clearly no evidence of epiphyseal ring adhesion on the centrum (Albert and Maples, 1995). (See Figures 9, 10, and 11)

Stage 1 – This stage indicates beginning union of the epiphysis and the vertebral centrum. In the early phase of this stage, the epiphyseal ring is attached in only a small area to the vertebral centrum. The epiphyseal ring is described as thin and fragile. The late phase is represented by progressing union. At this point, the epiphysis is more solidly attached to the centrum and has less open spaces, but in less than half of the centrum. Essentially, more than half of the area of union is open and unfused (Albert and Maples, 1995). (See Figures 12, 13, and 14)

Stage 2 – This stage feature almost complete union in the early phase and recently complete union in the late phase. In the early phase, where there is almost complete union, the epiphysis has united with more than 50% of the vertebral centrum surface. Therefore, there is a smaller space between the epiphysis and vertebral centrum. In some areas where union has occurred, a distinct, indented line or groove appears between the epiphysis and the centrum. In the late phase, where there is recent union, there are no spaces left and there is a distinct and uniform line or groove between the epiphysis and the centrum (Albert and Maples, 1995). (See Figures 15, 16, and 17)

Stage 3 – This stage represents a completely mature vertebral body, where the epiphysis is fully fused to the centrum and the vertebra and appears as one piece. Also, the line or groove seen in previous stages that marks the epiphyses from the centrum is completely obliterated. However, a scar may persist where the line demarcating the epiphysis from the centrum has remained. A scar is described as a smooth, shallow depression, whereas the line seen in the recent union phase is wider, deeper, rougher, and coarser (Albert and Maples, 1995). (See Figures 18, 19, and 20)

Analyses

Both statistical and observational analyses were conducted. Statistical analyses were performed to calculate the mean value stage for each individual to correlate the known age and assessed stage. Statistical analyses were also carried out to calculate the mean stage for each individual in the sample and correlating this number with the known age to determine the reliability of the method.

Observations beyond classifying each vertebra to an epiphyseal stage were described. The youngest and oldest individuals to exhibit each stage of epiphyseal union were noted between the sexes to show progression of growth and variation. Patterns in the sequencing of the epiphyseal union along the cervical vertebrae were observed, as well as any differences in the stages of the superior or inferior surface of any given vertebra. The sequence of epiphyseal union throughout the cervical vertebrae and other significant patterns are crucial to understanding the growth and development of not only the cervical vertebrae, but the entire vertebral column.

Chapter 6

Results

Statistical Analyses

Statistical analyses were performed on data collected on 77 individuals. Eleven surfaces, superior and inferior aspects of five centra (C3-C7), as well as inferior C2, were observed. Of the 77 individuals, five individuals had vertebral epiphyses omitted because a particular vertebra was missing, too damaged, or fused to a neighboring vertebra. These individuals were not removed from the study, but less than 11 surfaces were appropriately scored. Because the study focused on the maturation of the cervical vertebra, the stages of each surface are not necessarily the same for a given individual since growth is in progress. A way to simplify each individual represented by 11 numbers was needed and therefore statistical analyses were performed.

Since there were as many as 11 variables (stages of union) recorded for each individual in the sample, mean values representing the overall progress of union were calculated. The mean represents the average stage number for each individual in the sample. Mean epiphyseal union values for individuals in the overall sample were correlated with their known age at death. Further, mean values of females and males were correlated with known age separately to test for sex differences. Therefore, the correlation of means and the age of 38 females were calculated and subsequently for the 39 males, and for 51 blacks and 26 whites.

Also worthy of consideration were calculations of the *maximum number* and *mode* of epiphyseal union for each individual in the sample. The *maximum number* is the most advanced stage recorded on any cervical vertebrae for each individual in the sample representing the most advanced progress in each individual. The *mode* is the stage most frequently observed on the cervical vertebrae of each individual in the sample representing the overall progress of the cervical vertebrae as a whole. These calculations are encompassed within the results of the observational analyses.

The maximum number, while it may be a sign of stage progression or development, and the mode, while it shows the stage of greatest frequency, fails to represent the individual's stage as a whole. The average, or mean, for each individual allows all stages to be accounted for equally. Also, the mean is used in this analysis due partly to its usage in the original Albert and Maples (1995) thoracic and first two lumbar vertebrae study.

An overall correlation of .781 was found between mean values computed from the stages of epiphyseal ring union of all vertebrae for each individual in the study, and known age at death, for the overall pooled sample. This was calculated using the mean value stage for each individual in the study. The calculations indicate a positive correlation meaning that as the mean stage increases, age increases. Correlations were also calculated separately for males and females. The correlation between mean stage of vertebral ring epiphyseal union and age for females was higher at .854 than males who showed a correlation of .688. The correlation suggests that this scoring method for vertebral ring epiphyseal union is stronger and more accurate for females than males. Refer to Table 2 below for a summary of the correlation between the mean stage value and the known age of an individual. Scatter plot graphs depicting the correlations between mean stage of vertebral ring epiphyseal union and age listed in Table 2 can be referenced on pages 43 and 44 (Figures 7 and 8).

Table 2 – Statistical Analyses: Correlation Results.

Correlations	R	Significance
Overall (n=77)	.781	<.000
Female (n=38)	.854	<.000
Male (n=39)	.688	<.000

Observational Analyses

While results of the correlation analyses were informative, observational results of the progression of union – from the time of onset to the time of complete union – provided information beyond what statistical analyses was able to reveal. Results are presented for females and males, by stage of union. The sample was not broken down into ancestry because the diversity in the sample was too small and there were no differences found. Observations for the early and late phases of each stage are discussed below, as well as any results based on sequencing and patterns.

Upon tabulation of the collected data, several key observations were made and are described below. Refer to Table 3 for a summary of the observations for stages of vertebral ring epiphyseal union. Also, refer to Tables 4 and 5 on pages 54 – 57 for complete scoring data for each individual in the sample.

<u>Stages</u>

Stage 0

The early phase of stage 0 is characterized by no attachment of epiphyseal rings in any vertebrae. The youngest female to show this phase was 12 years old. The youngest male to exhibit this early phase was 17 years old.

The late phase of stage 0 is characterized by the oldest individual to show no union in any vertebrae. The oldest individual to show non union on any vertebrae is 18 years in females (C5, superior; C6, superior; C7, superior) and 19 years in males (in all vertebrae).

Stage 1

The early phase of stage 1 is beginning union. There are two females who are the youngest to show beginning union on any vertebrae at the age of 12 years (the first female: C3, superior and inferior; C4, superior and inferior; the second female: C3, inferior; C4, superior and inferior). The youngest male to show beginning union is 16 years of age (C2, inferior; C7, inferior).

The oldest female to show beginning union on any vertebrae is 23 years of age (C5, superior; C6, superior; C7, superior). There are two males who are the oldest to exhibit Stage 1 on any vertebrae at the age of 21 years (the first male: C2, inferior; C3, superior and inferior; C4, inferior; C6, superior and inferior; C7, superior and inferior; the second male: C4, inferior; C5, inferior; C7, inferior).

Stage 2

The early phase of stage 2 is almost complete union. There are two individuals who are the youngest females to exhibit almost complete union at the age of 16 years (the first female: C3, superior; the second female: C2, inferior; C3, inferior). There are three males at the age of 18 years who are the youngest to show almost complete union on any of the vertebrae (the first male: C2, inferior; the second male: C2, inferior; C3, superior and inferior; C4, superior; C5, inferior; the third male: C3, inferior; C4, inferior; C6, inferior; C7, inferior).

The late phase of stage 2 is recent union. The oldest individuals to exhibit recent union on any vertebrae are 27 years of age for both males and females, in which there are three males (all three males: in all vertebrae) and three females (the first female: C7, inferior; the second female: in all vertebrae; the third female: in all vertebrae except C2, inferior).

Stage 3

The early phase of stage 3 is complete union in any vertebrae. The youngest female to show complete union on any vertebrae is 19 years of age (C4, superior). The youngest male to show Stage 3 on any vertebrae is 18 years of age (C2, inferior; C3, superior; C4, superior; C5, superior and inferior; C6, superior; C7, superior).

This phase of Stage 3 is usually described as the youngest individuals to show complete union of the epiphyseal rings in all of the vertebrae. However, none of the individuals in the study exhibit complete union in all of the vertebrae.

Table 3 – Summary Observations of Vertebral Ring Epiphyseal Union Stages.

	Females	Males
Earliest Stage 0 Latest Stage 0	12 years 18 years	17 years 19 years
Earliest Stage 1 Latest Stage 1	12 years 23 years	16 years 21 years
Earliest Stage 2 Latest Stage 2	16 years 27 years	18 years 27 years
Earliest Stage 3 Latest Stage 3	19 years Unavailable	18 years Unavailable

Patterns

Observational analyses shows no significant differences regarding the sequence of union between the superior and inferior ring epiphyses for either a particular vertebra or among any vertebrae for a given individual. Out of 385 vertebrae observed, only 71 vertebrae or 18% revealed different stages for either the superior or inferior surfaces. The second cervical vertebrae were excluded from this analysis due to the lack of a superior surface. Among C3-C7 there is a relatively even distribution of the number of vertebrae (between 12 and 17) that exhibit different stages on either the superior or inferior surface of one vertebra. Of the 71 vertebrae that showed different stages, 46 vertebrae exhibited a more advanced stage on the superior surface than on the inferior surface while only 25 showed a more advanced stage on the inferior surface than on the superior surface. In general, the superior and inferior ring epiphyses stage only differs within one stage. No particular sequences of stages (stage 0 – stage 1; stage 1 – stage 2;

stage 2 – stage 3) were over-represented to suggest a significant delay between any specific stage. However, there was one individual, 16 years of age, which exhibited a stage 2 on the superior surface and stage 0 on the inferior surface. This is the only anomalous vertebra found in this study.

Chapter 7

Discussion

Results of this study of the vertebral ring epiphyseal union of cervical vertebrae are significant, especially when compared with what is also known about the thoracic and first two lumbar vertebral ring epiphyseal union. The observations and stages recorded in this study show that although McKern and Stewart (1957) only look at the thoracic vertebrae of males from age 17 to 25, when the sample age is expanded in either direction, more information can be gleaned about the maturation of the vertebrae in regards to the vertebral ring epiphyseal union. Not only was the age of the sample expanded, but sex and ancestry were also evaluated. This variation was unavailable in McKern and Stewart's sample.

The results found in this study and the original Albert and Maples (1995) results are remarkably close. Albert and Maples (1995) report an overall correlation of 52 individuals at .78 (.7754). Likewise an overall correlation of .78 (.781) was found for the overall combined sample between mean values from the stages of epiphyseal ring union of all vertebrae for each individual in this study, and known age at death. The correlation results of this study were consistent with findings from the thoracic and first two lumbar vertebrae, which demonstrates the efficacy of this aging method as well as its applicability to the cervical vertebrae (McKern and Stewart, 1957; Albert and Maples, 1995).

Although the demographics of this study did not allow for sufficient analysis of ancestral differences, sex differences were evaluated. Sex differences were found based on the calculated correlations between the mean stage of vertebral ring epiphyseal union and age. There are 38 females and 39 males in the sample allowing for an equal comparison between sexes. Females were found to have a higher correlation at .854, while males in this sample show a correlation of .688. The higher correlation for females suggests a higher reliability and effectiveness in the method when used to evaluate females. However, both correlations are significant and suggest an overall efficacy for both sexes.

While Albert and Maples (1995) reported the youngest age to show beginning union on any vertebrae as 14 years in females and 16 years, 4 months in males, beginning epiphyseal union was found in the cervical vertebrae of this sample as early as 12 years of age. This supports the notion that epiphyseal union initiates at either end of the vertebral column and progresses toward the middle (Steele and Bramblett, 1988). However, at this time, only the cervical, thoracic, and first two lumbar vertebrae have been investigated. Further research would be necessary on the lower lumbar vertebrae or the entire vertebral column to better corroborate these findings.

Secular changes in skeletal growth and maturation were reviewed recently by Schaefer and Black (2005). The authors compared the rate of epiphyseal union on ten skeletal sites between a Bosnian population from the 1990s and the rates previously reported by McKern and Stewart from an American population in the 1940s and 1950s. Schaefer and Black (2005) reported some populational differences at the time of complete union of some of the epiphyses. Although McKern and Stewart's (1957) sample, Albert and Maples' (1995) sample, and the sample from the Hamann-Todd Osteological Collection used for this study are all from the American population, the sample for each study originate in different time periods. The Hamann-Todd

Osteological Collection was acquired over the course of many years between the 1900s and 1920s, McKern and Stewart's sample was acquired from the 1940s and 1950s, and Albert and Maples collected their samples in the 1990s. Due to the consistency in the results that can be compared in this study, it can be noted that although there were secular changes between populations, there were no secular changes observed within the American population spanning through the 20^{th} century with regards to vertebral ring epiphyseal union.

Chapter 8

Conclusion

Aging techniques in forensic anthropology inherently possess several limitations. The initial downfall is the challenge presented by human variation and its effect on the progression of age on the bone. Human variation produces age ranges that can be broad, spanning several decades at times and decreasing the utility of the method. So the aim is to continue research efforts into revealing, which, if any, skeletal sites show less variation and therefore, lead to narrower estimated ranges which result in greater utility, especially in forensic cases. Studies in the area of vertebral aging are important for the field of forensic anthropology because it can provide information that can help distinguish late teens to early 20s and from early 20s to mid 20s. Current methods of aging provide a wide age range such as the methods developed for the pubic symphysis and the ribs, but by using the vertebral aging method in combination with other methods of aging, the age range can be narrowed down. Essentially, vertebral ring epiphyseal union aids skeletal age estimation by narrowing otherwise broad age ranges, corroborating findings from other skeletal sites, and using the method in the absence of other key skeletal indicators of age.

The results of this study indicate that the same staging methods developed by Albert and Maples (1995) for the thoracic and first two lumbar vertebrae can be applied to the cervical vertebrae. The similarity between the correlations found in the original Albert and Maples (1995) study and those found in this study show a consistency in the scoring method throughout the vertebral column, its reliability in correlation with age, and its repeatability among different observers. As the results have shown a consistency in age and vertebral ring epiphyseal union, it is important to note that the data collected can be used in both a forensic and archaeological setting.

Future research could focus on scoring every surface of the centrum on the entire vertebral column of an individual and comparing that correlation with the ones found here and for the thoracic and first two lumbar vertebrae. Also, although the sample size in this study was sufficient for statistical analyses, there were a low number of whites, especially white females, available for this study. Further research utilizing a comparable sample size of whites and blacks, and males and females could yield important results regarding sex and race differences. As is evident by the overview of previous research in the area of vertebral ring epiphyseal union, there is a need to further explore the maturation of the vertebral column more thoroughly, to not only continue to utilize this information for age estimation, but to better understand the general maturation of the vertebral column and its significance to the overall maturation of the entire skeleton.

It is the responsibility of a forensic anthropologist to utilize as many parts of the human skeleton as possible to appropriately acquire an estimation of age. The vertebral column as a whole has largely been ignored. While many other aspects of the skeleton can assist in developing an age estimation, it is important that all aspects of the skeleton be explored for its potential in not only age, but sex, ancestry, stature, pathology, and any other information it could potentially yield. Exploring the potential of the entire skeleton can be especially important in a few specific situations. We have the opportunity as a global society to be prepared for mass disaster situations like we have never been able to prepare before. Part of that preparation includes adjusting every method necessary to, when the time comes, be prepared to handle, analyze, and identify human remains. In cases where bodies are commingled, the ability to associate the remains with each other is paramount in ultimately making a positive identification. While the field of forensic anthropology, skeletal analysis, and human identification is not perfect and never will be, the world's tragedies continue and the best methods and techniques we can provide is the least we can do as human beings and responsible forensic science professionals.

APPENDIX A

Figure 1 – Cervical Aspect of the Vertebral Column

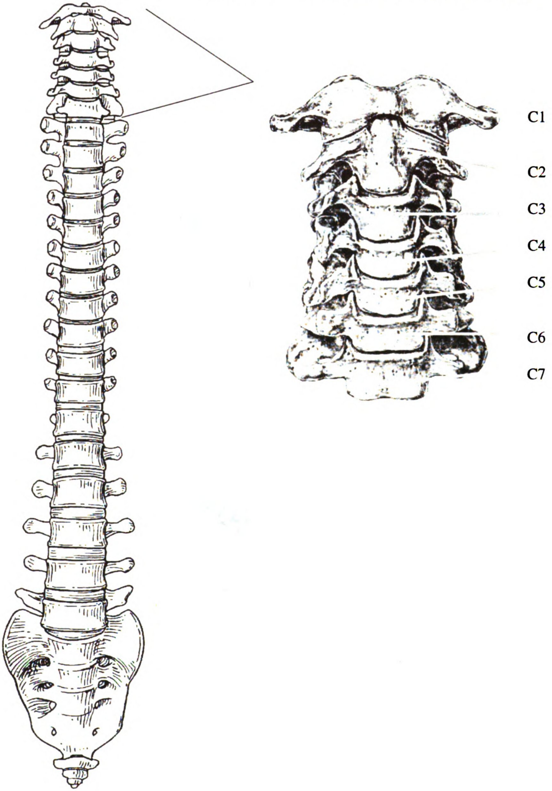
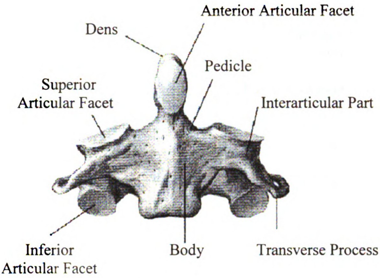
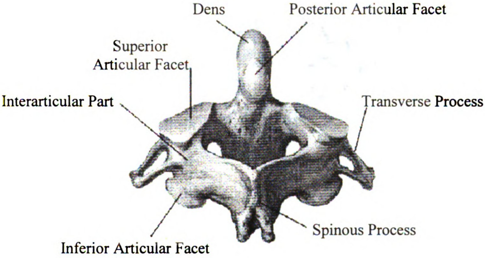


Figure 2 – Characteristics of the Second Cervical Vertebra



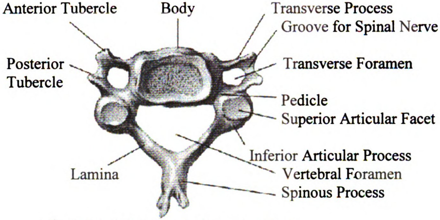
Axis (C2): Anterior View



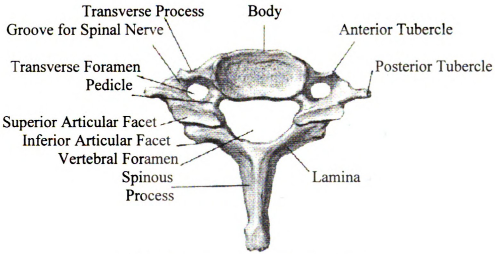
Axis (C2): Posterosuperior View

(Netter, 2003)

Figure 3 – Characteristics of a Typical Cervical Vertebra (C3-C7)



4th Cervical Vertebra: Superior View



7th Cervical Vertebra: Superior View

(Netter, 2003)

Figure 4 – Primary Centers of Ossification of the Second Cervical Vertebra

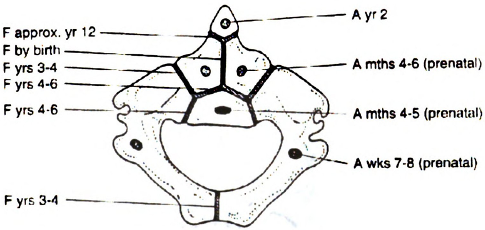
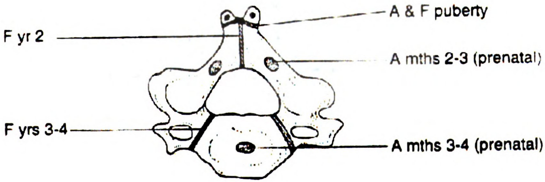


Figure 5 – Primary Centers of Ossification of a Typical Cervical Vertebra



A – Time of Appearance
F – Time of Fusion

Figure 6 – Secondary Centers of Ossification of a Typical Cervical Vertebra

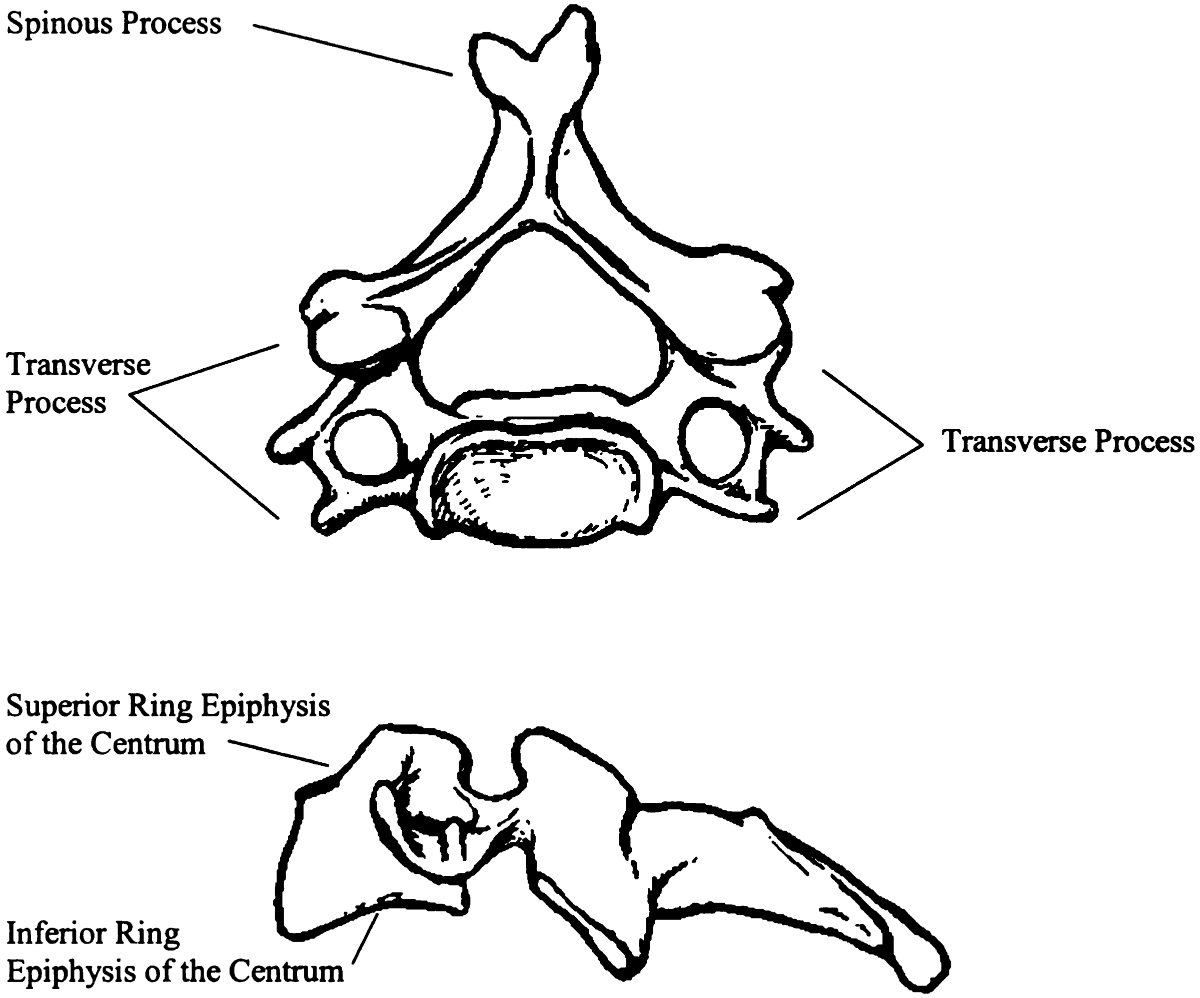


Figure 7 – Correlation between Mean Stage of Vertebral Ring

Epiphyseal Union and Age for Females and Males

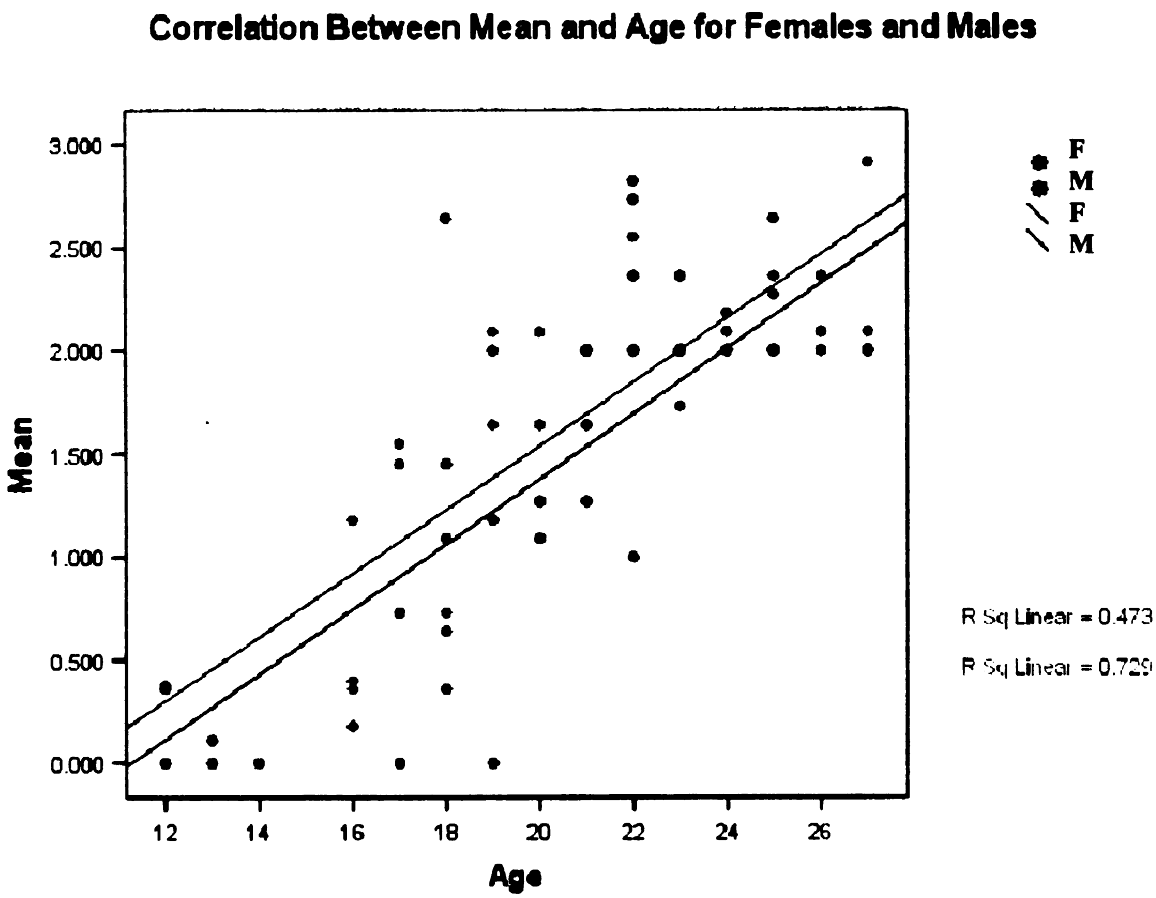
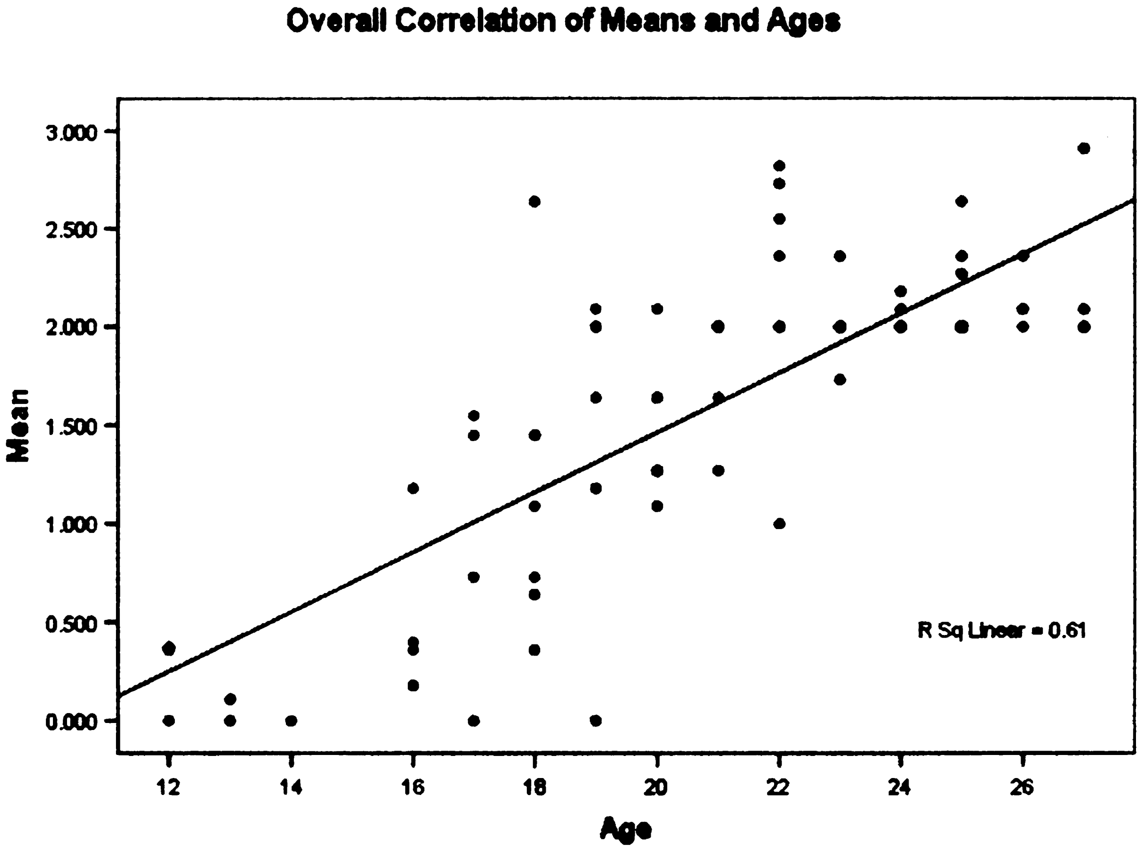


Figure 8 – Overall Correlation of Mean Stage Vertebral Ring

Epiphyseal Union and Age



APPENDIX B

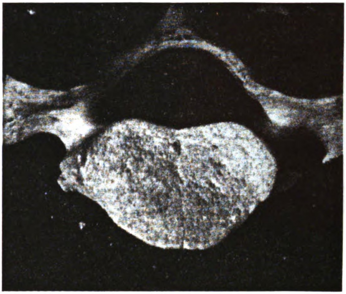


Figure 9 – Example of Stage 0 Vertebral Ring Epiphyseal Union (C7-S)

Images in this thesis are presented in color.

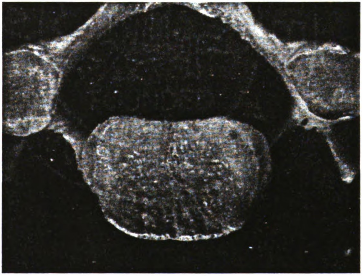


Figure 10 – Stage 0 Vertebral Ring Epiphyseal Union (C4-I)

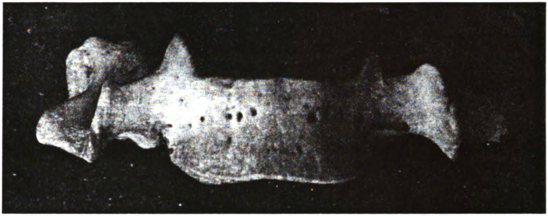


Figure 11 – Stage 0 Vertebral Ring Epiphyseal Union (C5)

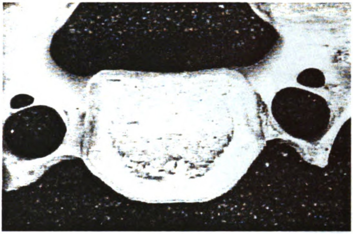


Figure 12 – Example of Stage 1 Vertebral Ring Epiphyseal Union (C6-I)

Figure 13 – Stage 1 Vertebral Ring Epiphyseal Union (C7-I)

Figure 14 – Stage 1 Vertebral Ring Epiphyseal Union (C6)

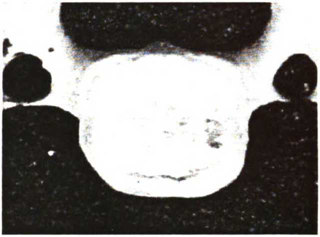


Figure 15 – Example of Stage 2 Vertebral Ring Epiphyseal Union (C5-I)

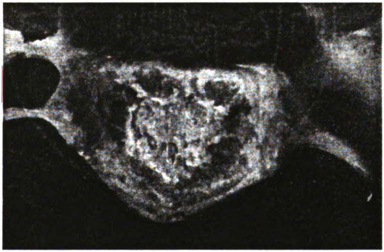


Figure 16 – Stage 2 Vertebral Ring Epiphyseal Union (C3-S)

Figure 17 – Stage 2 Vertebral Ring Epiphyseal Union with Persistent Line between Epiphyses and Centrum (C7)

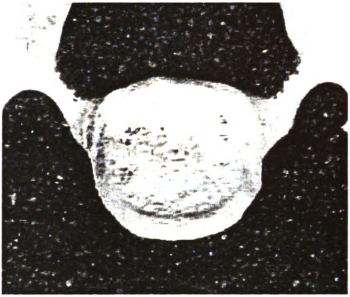


Figure 18 – Example of Stage 3 Vertebral Ring Epiphyseal Union (C3-I)

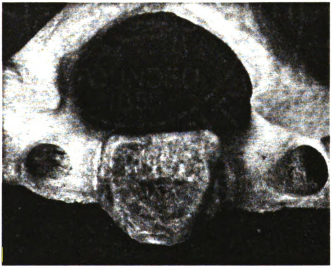


Figure 19 – Stage 3 Vertebral Ring Epiphyseal Union (C2-I)

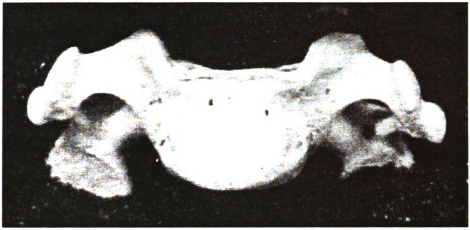


Figure 20 – Stage 3 Vertebral Ring Epiphyseal Union; Full Adult Vertebra (C3)

Table 4a – Vertebral Ring Epiphyseal Union Data for Females

Age	Ancestry	C2-I	C3-S	C3-I	C4-S	C4-I	C5-S	C5-I	C6-S	C6-I	C7-S	C7-I
12	W	0	1	1	1	1	0	0	0	0	0	0
12	W		0	1	1	1	0	0	0	0		
12	W	0	0	0	0	0	0	0	0	0	0	0
13	B	0			0	0	0	0	0	0	0	1
13	B	0	0	0	0	0	0	0	0	0		0
14	B	0	0	0	0	0	0	0	0	0	0	0
16	B	0	0	1	1	0	0	0	1	0	1	0
16	B		2	0	0	0	1	0	1	0	0	0
16	B	2	1	2	1	1	1	1	1	1	1	1
17	B	2	2	2	2	2	2	1	1	1	1	1
17	B	2	2	2	1	2	2	1	2	1	1	0
18	B	2	2	2	1	2	1	2	1	1	1	1
18	W	1	1	1	1	1	0	1	0	1	0	1
19	B	2	2	2	2	2	2	2	2	2	2	2
19	W	2	2	2	3	2	2	2	2	2	2	2
19	B	2	2	2	2	2	2	2	2	2	2	2
19	W	2	2	2	2	1	2	2	1	1	1	2
20	B			2	2	2	2	2	2	2	2	2
20	B	3	2	2	2	2	2	2	2	2	2	2

Table 4b – Vertebral Ring Epiphyseal Union Data for Females

Age	Ancestry	C2-I	C3-S	C3-I	C4-S	C4-I	C5-S	C5-I	C6-S	C6-I	C7-S	C7-I
21	B	2	2	2	2	2	2	2	2	2	2	2
22	B	3	3	3	3	3	3	2	2	3	3	2
22	B	1	1	1	1	1	1	1	1	1	1	1
22	B	3	3	3	3	2	3	2	3	2	2	2
23	B	2	2	2	2	2	1	2	1	2	1	2
23	B	2	2	2	2	2	2	2	2	2	2	2
23	B	2	2	2	2	2	2	2	2	2	2	2
23	W	2	2	2	2	2	2	2	2	2	2	2
23	W	2	2	2	2	2	2	2	2	2	2	2
24	B	2	2	2	3	2	2	2	2	2	2	2
24	B	2	2	2	2	2	2	2	2	2	2	2
25	B	2	2	2	2	2	2	2	2	2	2	2
25	B	3	3	3	3	2	2	2	2	2	2	2
25	W	3	2	2	2	3	2	2	2	3	2	2
25	W	2	2	2	2	2	2	2	2	2	2	2
26	B	3	3	2	2	2	2	2	2	2	3	3
27	B	3	3	3	3	3	3	3	3	3	3	2
27	W	2	2	2	2	2	2	2	2	2	2	2
27	B	3	2	2	2	2	2	2	2	2	2	2

Table 5a – Vertebral Ring Epiphyseal Union Data for Males

Age	Ancestry	C2-I	C3-S	C3-I	C4-S	C4-I	C5-S	C5-I	C6-S	C6-I	C7-S	C7-I
16	B	1	0	0	0	0	0	0	0	0	0	1
17	B	0	0	1	1	1	1	1	1	1	0	1
17	B	0	0	0	0	0	0	0	0	0	0	0
18	B	2	1	1	1	1	1	1	1	1	1	1
18	B	1	0	1	1	0	0	0	1	0	0	0
18	B	2	2	2	2	1	1	2	1	1	1	1
18	W	3	3	2	3	2	3	3	3	2	3	2
18	W	1	1	1	1	1	1	1	0	0	0	0
19	B	0	0	0	0	0	0	0	0	0	0	0
19	W	2	2	1	1	1	1	1	1	1	1	1
20	B	2	1	1	1	1	1	1	1	1	1	1
20	B	2	2	1	2	1	1	1	1	1	1	1
20	W	2	2	2	1	1	1	1	1	1	1	1
21	W	2	2	2	2	2	2	2	2	2	2	2
21	B	2	2	2	2	2	2	2	2	2	2	2
21	B	2	2	2	2	1	2	1	2	1	2	1
21	B	2	2	2	2	2	2	2	2	2	2	2
21	W	1	1	1	2	1	2	2	1	1	1	1
22	W	3	2	3	3	3	3	3	3	3	3	2
22	B	2	3	2	3	2	3	2	3	2	2	2
22	B	2	2	2	2	2	2	2	2	2	2	2
22	W	2	2	2	2	2	2	2	2	2	2	2

Table 5b – Vertebral Ring Epiphyseal Union for Males

Age	Ancestry	C2-I	C3-S	C3-I	C4-S	C4-I	C5-S	C5-I	C6-S	C6-I	C7-S	C7-I
23	B	2	2	2	2	2	2	2	2	2	2	2
23	B	2	2	2	2	2	2	2	2	2	2	2
23	W	2	2	3	3	3	2	3	2	2	2	2
23	W	2	2	2	2	2	2	2	2	2	2	2
24	W	2	2	2	3	2	2	2	2	2	3	2
24	B	2	2	2	2	2	2	2	2	2	2	2
24	W	2	2	2	2	2	2	2	2	2	2	2
25	B	2	2	2	2	2	2	2	2	2	2	2
25	W	2	2	2	2	2	2	2	2	2	2	2
25	B	3	3	3	3	3	2	2	3	2	3	2
25	B	2	2	2	2	2	2	2	2	2	2	2
25	W	2	2	2	2	2	2	2	2	2	2	2
26	B	2	2	2	2	2	2	2	3	2	2	2
26	B	2	2	2	2	2	2	2	2	2	2	2
27	B	2	2	2	2	2	2	2	2	2	2	2
27	B	2	2	2	2	2	2	2	2	2	2	2
27	W	2	2	2	2	2	2	2	2	2	2	2

REFERENCES

Acheson RM. Maturation of the skeleton. In: Falkner F, editor. Human development. Philadelphia: W.B. Saunders, 1966; 465-502.

Albert AM. The use of vertebral ring epiphyseal union for age estimation in two cases of unknown identity. *Forensic Science International* 1998; 97: 11-20.

Albert AM, Maples WR. Stages of epiphyseal union for thoracic and lumbar vertebral centra as a method of age determination for teenage and young adult skeletons. *Journal of Forensic Sciences* 1995; 40(4): 623-633.

Bagnall KM, Harris PF, Jones PRM. A radiographic study of the human fetal spine 2. The sequence of development of ossification centres in the vertebral column. *Journal of Anatomy* 1977; 124: 791-802.

Bass WM. Human osteology: a laboratory and field manual. Columbia MO: Missouri Archaeological Society, 1987.

Berzlanovich AM, Sim E, Muhm, MA. Technique for dissection the cervical vertebral column. *Journal of Forensic Sciences* 1998; 43(1): 190-193.

Bick EM, Copel JW. The ring apophysis of the human vertebra. *Journal of Bone and Joint Surgery* 1958; 33A: 783-787.

Brooks ST. Skeletal age at death: the reliability of cranial and pubic age indicators. *American Journal of Physical Anthropology*, 1955; 13: 367-597.

Buikstra JE, Gordon CC, St. Hoyme L. The case of the severed skull. In: Rathburn TA, Buikstra JE, editors. Human identification. Springfield Ill: C.C. Thomas, 1984; 121-135.

Buikstra JE, Gordon CC. Individuation in forensic science study: decapitation. *Journal of Forensic Sciences* 1980; 25(1): 246-259.

Burns KR. Forensic anthropology training manual. Upper Saddle River, New Jersey: Prentice-Hall, Inc, 1999.

Byers SM. Introduction to forensic anthropology: a textbook, 2^{nd} ed. New York: Pearson, 2005.

Clark CR. The cervical spine, 4^{th} ed. New York: Lippincott, Williams, and Wilkins, 2005; 40-43.

Dickson RA, Deacon P. Spinal growth. *Journal of Bone and Joint Surgery*. British volume 1987; 69(5): 690-692.

Dormans JP. Evaluation of children with suspected cervical spine injury. *The Journal of Bone and Joint Surgery* 2002; 84A(1): 124-132.

Flecker HM. Time of appearance and fusion of ossification centers as observed by roentgenographic methods. *Journal of Roentgenology and Radium Therapy* 1942; 47: 97-159.

Gilbert BM, McKern TW. A method for aging the female os pubis. *American Journal of Physical Anthropology* 1973; 38: 31-38.

Girdany BR, Golden R. Centers of ossification of the skeleton. *American Journal of Roentgenology, Radium Therapy, and Nuclear Medicine* 1952; 68(6): 922-924.

Hindman BW, Poole CA. Early appearance of the secondary vertebral ossification centres. *Radiology* 1970; 95: 359-361.

Hoffman JM. Age estimations from diaphyseal lengths: two months to twelve years. *Journal of Forensic Sciences* 1979; 24(2): 461-469.

İşcan MY, Loth SR. Osteological manifestations of age in the adult. In: İşcan MY, Kennedy KAR, editors. Reconstruction of life from the skeleton. New York: Wiley Liss, 1989; 23-40.

İşcan MY, Loth SR, Wright RK. Age estimation from the rib by phase analysis: white females. *Journal of Forensic Sciences* 1985; 30(3): 853-863.

İşcan MY, Loth SR, Wright RK. Age estimation from the rib by phase analysis: white males. *Journal of Forensic Sciences* 1984; 29(4): 1094-1104.

Klonowski EE, Sarajilc N, Drukier P, Klonowski AM. The impact of age related changes in vertebral column on age determination for identification purposes. American Academy of Forensic Science meetings, abstract, 2005.

Kreitner KF, Schweden FJ, Riepert T, Nafe B, Thelen M. Bone age determination based on the study of the medial extremity of the clavicle. *European Radiology* 1998; 8: 1116-1122.

Krogman WM, İşcan MY. Skeletal age: cranium and post-cranium. In: The human skeleton in forensic medicine. Springfield IL: Charles C. Thomas 1986.

Krogman WM. A problem in the ageing of human skeletal remains. *Journal of Forensic Sciences* 1962; 7(3): 255-264.

Lustrin ES, Karakas SP, Ortiz AO, Cinnamon F, Castillo M, Vaheesan K, et al. Pediatric cervical spine: normal anatomy, variants, and trauma. *RadioGraphics* 2003; 23: 539-560.

Macalister A. Notes on the development and variations of the atlas. *Journal of Anatomy and Physiology* 1893; 27: 519-542.

Marks J. What it means to be 98% chimpanzee: apes, people, and their genes. London: University of California Press, 2002.

Martínez-Lage JF, Poza M, Arcas P. Avulsed lumbar vertebral rim plate in an adolescent: trauma or malformation? *Child's Nervous System* 1998; 14(3): 131-134.

McKern TW. Estimation of skeletal age from combined maturational activity. *American Journal of Physical Anthropology* 1957; 15(3): 399-408.

McKern TW, Stewart TD. Skeletal age changes in young American males. Technical Report EP-45. Headquarters, Quartermaster Research and Development Command, Quartermaster Research and Development Center Environmental Protection Research Division, Natick, Massachusetts 1957.

Meindl RS, Russell KF, Lovejoy CO. Reliability of age at death in the Hamann-Todd collection: validity of subselection procedures used in blind tests of the summary age technique. *American Journal of Physical Anthropology* 1990; 83: 349-357.

Meindl RS, Lovejoy CO. Ectocrannial suture closure: a revised method for the determination of skeletal age at death based on the lateral-anterior sutures. *American Journal of Physical Anthropology* 1985; 68: 57-66.

Mincer HH, Harris EH, Berryman HE. The ABFO study of third molar development and its use as an estimator of chronological age. *Journal of Forensic Sciences* 1993; 38(2): 379-390.

Muller-Bolla M, Lupi-Pegurier L, Quatrehomme G, Velly AM, Bolla M. Age estimation from teeth in children and adolescents. *Journal of Forensic Sciences* 2003; 48(1): 140-148.

Nakai M. Vertebral age changes in Japanese macaques. *Journal of Physical Anthropology* 2001; 116: 59-65.

Netter FH. Atlas of human anatomy. Teterboro, NJ: Icon Learning Systems, 2003.

Pfau RO, Sciulli PW. A method for establishing the age of subadults. *Journal of Forensic Sciences* 1994; 39(1): 165-176.

Pryor JW. Differences in the time of development of the centers of ossification in the male and female skeleton. *Anatomical Record* 1923; 25: 252-273.

Roche AF. Bone growth and maturation. In: Tanner JM, editor. Human growth: a comprehensive treatise. New York: Plenum Press 1986; 25-60.

Schaefer M, Black S. Comparison of ages of epiphyseal union in North American and Bosnian skeletal material. *Journal of Forensic Sciences* 2005; 50(4): 1-8.

Scheuer L, Black S. The juvenile skeleton. London: Elsevier Academic Press, 2004.

Scheuer L, Black S. Developmental juvenile osteology. London: Academic Press, 2000.

Schmitt A, Murail P, Cunha E, Rougé D. Variability of the pattern of aging on the human skeleton: evidence from bone indicators and implications on age at death estimation. *Journal of Forensic Sciences* 2002; 47(6): 1203-1209.

Snodgrass JJ. Sex differences and aging of the vertebral column. *Journal of Forensic Sciences* 2004; 49(3).

Solari AC, Abramovitch K. The accuracy and precision of third molar development as an indicator of chronological age in Hispanics. *Journal of Forensic Sciences* 2002; 47(3): 531-535.

Steele DG, Bramblett CA. The anatomy and biology of the human skeleton. College Station: Texas A&M University Press, 1988.

Stevenson PH. Age order of epiphyseal union in man. *American Journal of Physical Anthropology* 1924; 7(1): 53-93.

Stewart TD. Sequence of epiphyseal union, third molar eruption, and suture closure in Eskimos and American Indians. *American Journal of Physical Anthropology* 1934; 19(3): 433-452.

Suchey JM, Katz D. Applications of pubic age determination in a forensic setting. In Reichs K, editor. Forensic osteology: advances in the identification of human remains. Springfield, IL: Charles C. Thomas 1998; 204-236.

Todd TW. The anatomical feature of epiphyseal union. *Child Development* 1930; 1(3): 186-194.

Todd TW. Age changes in the pubic bone, II-IV. *American Journal of Physical Anthropology* 1921; 4: 1-70.

Todd TW. Age changes in the pubic bone, I: the white male pubis. *American Journal of Physical Anthropology* 1920; 3: 285-334.

Ubelaker DH. Estimating age at death from immature human skeletons: an overview. *Journal of Forensic Sciences* 1987; 32(5): 1254-1263.

Whitaker JM, Rousseau L, Williams T, Rowan RA, Hartwig WC. Scoring system for estimating age in the foot skeleton. *American Journal of Physical Anthropology* 2002; 118: 385-392.

White TD. The human bone manual. San Diego, CA: Elsevier Academic Press, 2005.

White TD. Human osteology. San Diego, CA: Academic Press, 1991.