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Case Report
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Anthropological Assessment of Subadult Skeletal Remains Using Bones and Teeth for Age Estimation: a Case Report

Laíse Nascimento Correia Lima[1], Ariella Cássia dos Santos Francisco de Azevedo[2], Wigínio Gabriel Lira-Bandeira[3], Raíssa Araújo Gonçalves[4], Ademir Franco[5], Ivson Bezerra da Silva[6]

[1]Adjunct professor in the Department of Clinical and Social Dentistry at Federal University of Paraíba, João Pessoa - PB, Brazil
[2]PT, Specialist in Morphology, Department of Morphology, Federal University of Paraíba, João Pessoa - PB, Brazil
[3]BSN, Master's and PhD student at Postgraduate Program in Structural and Functional Biology, Department of Morphology, Federal University of Rio Grande do Norte, Natal - RN, Brazil
[4]Specialist and master's student in Forensics Dentistry, Faculdade São Leopoldo Mandic, Campinas, SP, Brazil
[5]Full-professor in Division of Forensic Dentistry, Faculdade São Leopoldo Mandic, Campinas - SP, Brazil
[6]Adjunct professor in the Department of Morphology at Federal University of Paraíba, João Pessoa - PB, Brazil

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ABSTRACT
Introduction: age-at-death estimation is a significant challenge in forensic anthropology, with particular emphasis on subadult skeletal evaluation due to the scarcity of studies and documented osteological collections for this group. This study aimed to report a case of age estimation in the skeletal remains of a subadult, utilizing macroscopic skeletal and dental morphological methods described in the literature, and to compare the estimated age ranges with the individual's actual chronological age, evaluating their applicability in a Brazilian context. The skeletal remains of a male subadult, with a known chronological age of 16 years, were analyzed. The evaluation included long bones, the spheno-occipital synchondrosis, the pubic symphysis, the sternal end of the fourth rib, and teeth, applying traditional methods widely reported in the scientific literature. The age estimates obtained were consistent with the individual's chronological age, highlighting the applicability of the methods analyzed in a Brazilian population context. However, the absence of population-specific standards for subadults remains a significant challenge for forensic anthropology in Brazil. This study underscores the need for validation and the development of methodological approaches tailored to the characteristics of Brazilian populations, contributing to greater accuracy and reliability in forensic analysis.
Keywords: Osteology; Forensic Anthropology; Age Determination by Skeleton; Age Determination by Teeth.

Introduction

Although estimating age at death is one of the most studied topics in Forensic Anthropology, it is still controversial and full of difficulties, ranging from the investigation of age intervals after the stabilization phase (adulthood), to the conditions of the remains and resources available for analysis[1,2,3,4]. In addition, the topic is of common interest to different areas of study, such as Anthropology, Archaeology, Biology, Medicine, Forensic Sciences, among others, with the most diverse purposes[5,6].

In Forensic Anthropology, age estimation is a central procedure in obtaining the biological profile of human remains, and its correct establishment is decisive in the identification process[6,7,8]. Biological transformations in bones, which accompany advancing age — from longitudinal growth in young individuals to degenerative changes that manifest in adulthood —, allow the estimation of bone age, equivalent to an individual's biological age[9]. This concept, however, differs from chronological age[7,10,11].

Chronological age corresponds to calendar age, measured in days, months and years, while biological age refers to the physiological state of the organism, reflected by the way in which the aging process affects body tissues[7,11]. One of the essential assumptions of most methods in Forensic Anthropology is that biological age corresponds, approximately, to chronological age, and it is mentioned that the discrepancy between these increases as the individual ages[10,12,13].

Each part of the skeleton, depending on its location, structure and function, reveals different aspects of the changes associated with aging[14]. Most age assessment methods are based on the macroscopic identification of specific skeletal characteristics, which are categorized into stage or score systems, resulting in an estimate within an age range[6,12,13]. These age ranges will be more accurate in subadult individuals, in which age estimation methodologies are based on skeletal development, to the detriment of the analysis of degenerative processes, which base estimates for adult individuals[7,15]. In this context, it is

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possible to infer, from a biological point of view, that the adult phase begins when growth and development are completed[15,16].

Age estimation through analysis of macroscopic bone morphology tends to be more reliable in subadults, since the biological-chronological relationship is notably evident in the skeleton[6,13]. Developmental processes, because they present higher rates of change than those produced by degenerative processes, result in more noticeable modifications and, in addition, present greater genetic regulation, being less dependent on nutritional factors, habits, stresses and pathologies, such as the latter[15,17,18,19].

The scientific community disseminates that the complete fusion of the spheno-occipital synchondrosis and all long bones and the eruption of all dental elements are predictors of adulthood2[7,15]. Based on this, the morphological processes that precede these events are the characteristics used to estimate age, as well as the observation of age indicators such as: fusion of sacral vertebrae, ossification of the iliac crest[20] and the ends of the clavicle[21, 22,23] modifications in the surface of the pubic symphysis[24,25] and at the sternal end of the rib[26,27] among others.

In addition to bone analysis, the evaluation of dental elements is often used to estimate age, considering the chronology of crown-root mineralization of deciduous and permanent teeth as the most reliable method[15]. Verification of mineralization stages is even more reliable in estimating age than bone methods, and it is recommended that both ages (skeletal and dental) be estimated separately and, in the case of significant differences, the dental age closest to the individual's chronological age should be considered[15].

The scientific literature on macroscopic bone evaluation of subadults is still limited, reflecting the scarcity of documented osteological collections for this age group[28,29]. The greater prevalence of adult bones in forensic contexts is attributed to the higher mortality rates of this group, resulting in more cases found and studied. In contrast, the lower representation of subadults reflects both the lower juvenile mortality and the specificity of missing persons cases, which, although frequently involving young people, are less documented in osteological collections[30,31]. In Brazil, this issue is aggravated by the absence of national standard references, which requires the use of tables developed for populations in other countries, which are often inadequate for Brazilian biological and cultural diversity[32,33].

This study reports a case of age estimation in the bones of a subadult individual, with previously known age and sex. Based on the direct study of the bones, the aim is to compare the age ranges provided by macroscopic bone and dental morphological methods described in the literature with the actual chronological age of the individual. This approach gains scientific and practical relevance by minimizing knowledge gaps and contributing to the construction of specific standards for the national reality.

Case Report

Remains Analyzed

An anthropological, macroscopic and descriptive analysis was performed on a human skeleton belonging to a subadult individual, with known chronological age and sex, belonging to the Paraíba Laboratory of Forensic Osteology, of the Federal University of Paraíba, with approval by the Ethics Committee, under opinion 6.879.467. The work developed was structured in stages that included:

I. Carrying out a bone inventory using the form proposed by Machado *et al.*[5] in order to identify the bones present and, consequently, the possibilities of applying age estimation methods.

II. Use of age estimation methods based on the evaluation of macroscopic bone morphology in the available bones:

• Chronology of fusion of synchondroses of long bones: humerus, ulna, radius, femur, tibia and fibula;

• Fusion of the spheno-occipital synchondrosis;

• Morphology of the sternal articular surface of the clavicle (epiphyseal fusion);

• Morphology of the symphyseal surface of the pubis (pubic symphysis);

• Morphology of the costal extremity of the 4th rib;

• Chronology of dental eruption and mineralization.

III. Comparison and discussion of the results obtained with the individual's actual chronological age.

For each bone evaluated, an age range in years was obtained from estimates developed for male individuals. In paired bones, the use of the left side was recommended, with the exception of those that were absent or damaged. It is worth mentioning that in all paired bones available, the development phase between the right and left sides was congruent and that the methods chosen were based on studies carried out on dry bones, with the exception of the evaluation of dental elements, which, to a large extent, are based on studies of dental mineralization chronology.

Long bone synchondrosis

The fusion phases observed in long bone synchondroses were described in Table 1, followed by the estimated age ranges described by Schaefer[34] and Coqueugniot and Weaver[35], and are visible in Figure 1.

Spheno-occipital synchondrosis

Table 2 describes the age ranges obtained from studies developed by Sahni[37] , Scheuer and Black[38], Coqueugniot and Weaver[35] , and Shirley and Jantz[39].

Morphology of Bone Surfaces

The observation of the surface morphology of bone structures is described in Table 3. The age ranges were obtained from the analysis of the clavicle, the hip bone and the rib, whose available methods classify

Table 1. Chronology of fusion of synchondrodes of long bones: humerus, ulna, radius, femur, tibia and fibula, by Schaefer[34] and Coqueugniot e Weaver[35].

Dry bone assessment - Epiphyses - Male				
Bone	**Union Phase**	***Schaefer[34]**	***Coqueugniot and Weaver[35]**	**Estimated Age (years) Among Authors**
Humerus				
Proximal	Open	≤ 20	≤ 20	15 - 20
Distal	Complete	≥ 15	≥ 16	
* Medial Epicondyle	Open	≤ 18	≤ 19	
Radius				
Proximal	Complete	≥ 16	≥ 16	16 - 21
Distal	Open	≤ 19	≤ 21	
Ulna				
Proximal	Complete	≥ 15	≥ 16	15 - 21
Distal	Open	≤ 20	≤ 21	
Femur				
Proximal				
* Head	Open	≤ 15	≤ 20	≤ 20
* Greater trochant	Open	≤ 15	≤ 20	
* Lesser trochant	Open	≤ 15	≤ 20	
Distal	Open	≤ 16	≤ 20	
Tibia				
Proximal	Open	≤ 18	≤ 20	16 - 21
Distal	Partial	16-21	16-21	
Fibula				
Proximal	Open	≤ 19	≤ 20	15 - 21
Distal	Partial	15-20	16-21	

* Compiled by Schaefer *et al.*[36]

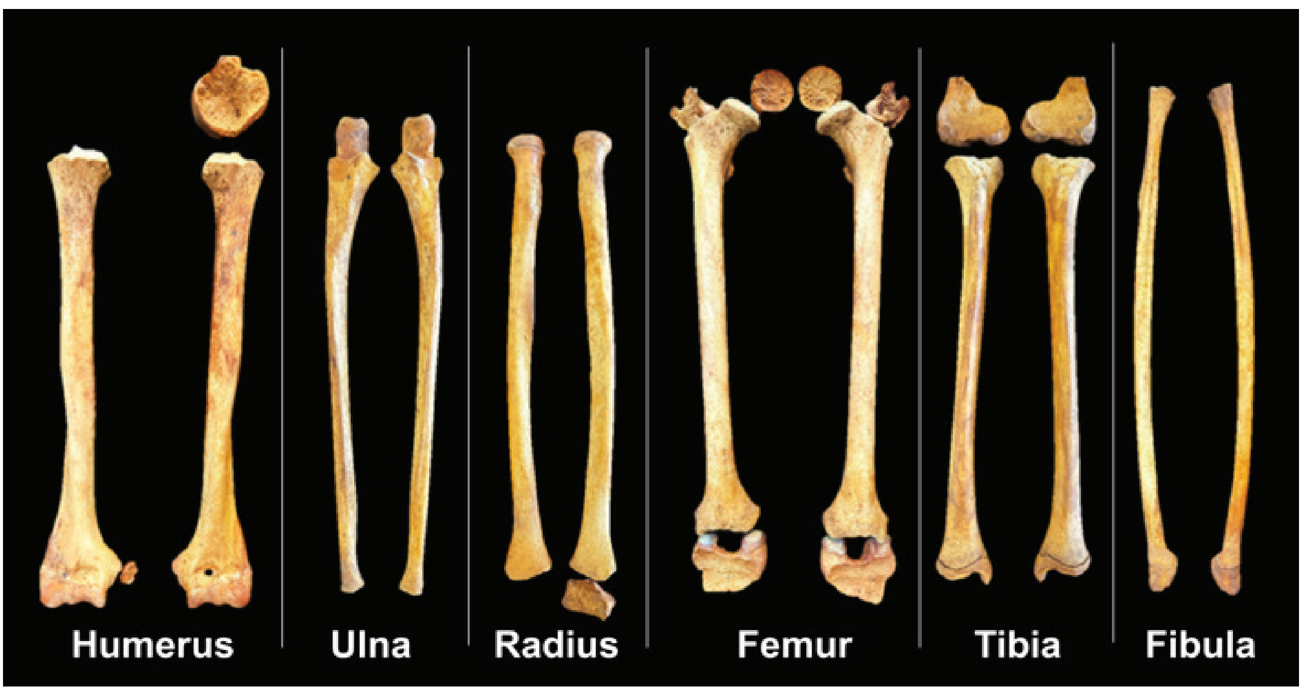

Figure 1. Epiphyses and diaphysis of long bones.

Table 2. Chronology of fusion of the spheno-occipital synchondrosis and the estimated age interval, in years.

Structure	Phase	Sahni [37]*	Coqueugniot and Weaver [35]*	Scheuer and Black[38]	Shirley and Jantz[39]
Spheno-occiptal synchondrosis	Partially fused	13 - 18	16 - 23	13 - 18	13 – 23

* Compiled by Schaefer *et al.*[36]

Table 3. Assessment of morphological phases of bone surfaces and estimated age range.

Bone	Phase	Author	Estimated Age (years)
Clavicle			
Sternal articular surface	0	Webb e Suchey[40]	<25
	0	Schaefer 2008[34]	<23
	2	Black e Scheuer 1996[41]	15 - 22
	Without epiphysis	Cunnigham, Scheuer, Black[23]	<18
Maximum lenght	146,4mm	Black e Scheuer 1996[41]	15 - 16
Thigh			
Pubic symphysis	1	Brooks e Suchey[42]	15 - 23
	Pre-epiphyseal	Cunnigham, Scheuer, Black[23]	<20
Rib			
Costal extremity 4th rib	0	Işcan-Loth2[7]	<17

the morphology into phases or stages (Figure 2). Although the analysis of the clavicle is based on the fusion of the epiphysis with the diaphysis, especially at the proximal end, this process is accompanied by morphological changes. The epiphysis initially appears as a protrusion, which gradually expands until it covers the entire surface of the sternal face of the clavicle. In addition, the maximum length of the clavicle between its ends was analyzed.

Dental Analysis

Although the chronology of dental mineralization, assessed radiographically, is the most reliable dental method for estimating age, the impossibility of using radiological resources led to a macroscopic dental analysis of the available third molars (upper right third molar: #18, upper left third molar: #28, and lower right third molar: #48). This approach is recommended by the international literature and predicts that the staging of radiographic dental development is similar, if not equal, to the staging performed ex vivo[43]. The third molars, visible, developing and disarticulated, allowed their classification into the stages recommended by Gleiser & Hunt[44] and modified by Köhler *et al.*[45], for use in age estimation using the method of Gunst *et al.*[46] (Figure 3; Table 4).

Figure 2. Morphology of the bone surfaces analyzed in tables 2 and 3.

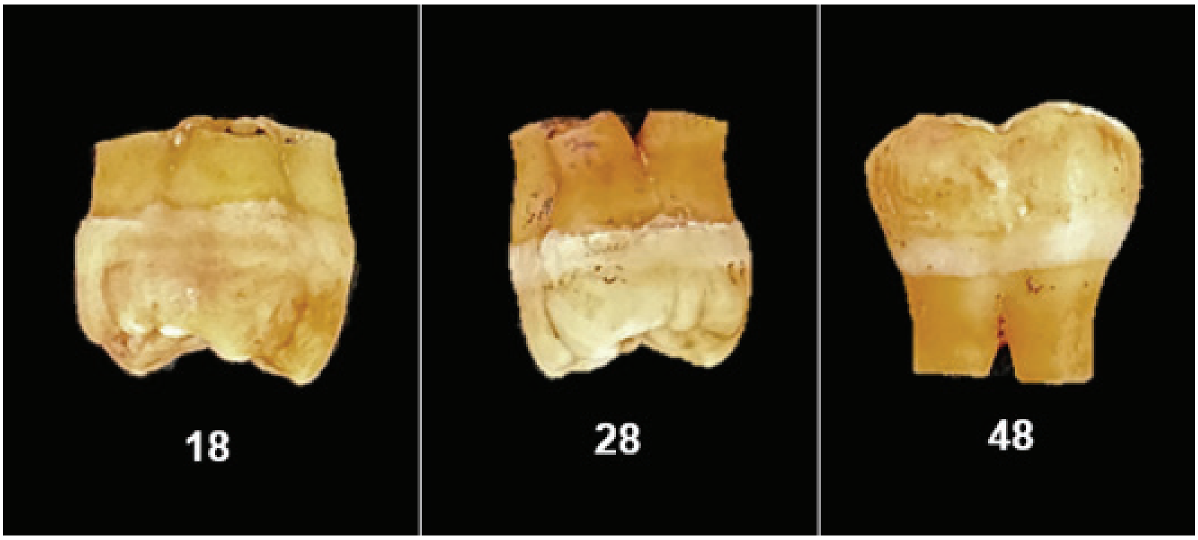

Figure 3. Development stage of available third molars

Table 4. Macroscopic evaluation of third molar mineralization and age estimated by the method of Gunst *et al.*[46]

Tooth	Stage	Formula	Estimated Age
		12.2212 + 0.6032 UR + 0.3132 LR*	17.11
18	Stage 5 ¼ root formed		
28	Stage 6 ½ root formed	11.5419 + 0.4426 UR + 0.4651 LR**	16.54
48	Stage 6 ½ root formed		

Formulas considering the presence of *03 third molars and **04 third molars

Discussion

The significant increase in the number of studies focused on age estimation highlights the relevance of the topic in the current context. Unpretentiously, bone indicators, which aimed to understand atypical delays or advances in human growth, proved to be fundamental in estimating an individual's age for forensic purposes[47]. Age is an essential normative reference that regulates a person's interaction with the legal system and their rights and duties throughout life, and its estimation is essential both for identification processes and for the fulfillment of these legal prerogatives[2,3,4].

Macroscopic evaluation of bone morphology in the context of anthropological studies of unidentified bones is the initial step and, in some circumstances, the only tool available to provide information about the biological profile of an individual. This approach stands out for the wide range of information that can be obtained, combined with its simplicity of application, effectiveness and low cost. In subadult individuals, the process of bone maturation significantly expands the possibilities of analysis, since morphological changes occur at predetermined times. However, it is essential to consider sexual differences, especially those resulting from the hormonal influence specific to each sex, as well as population variations.

Analysis of the chronology of fusion of the synchondroses of the long bones revealed that only the epiphyses of the bones involved in the elbow joint (proximal of the radius and ulna, in addition to the distal of the humerus) presented complete fusion, although the medial epicondyle of the humerus remained separate. This pattern contrasted with that observed in the knee joint, where the epiphyses – distal of the femur and proximal of the tibia – were not fused, as was the proximal epiphysis of the fibula, which is adjacent to this joint. The other distal epiphyses, such as those of the tibia and fibula, were partially fused, while those of the radius and ulna, as well as that of the femur, remained without signs of fusion.

The evaluation of the knee region, observed by Alzyoud *et al.*[48], although radiographic, demonstrated the non-fusion of the distal femoral epiphysis and proximal fibula and tibia around 14.5 years, with an average deviation of 1.50, which corroborates the score of Schaefer *et al.*[36], but underestimates the case in question. The age estimated through the chronology of synchondrosis fusion by the authors encompassed the age of the individual reported in the evaluation of all epiphyses, with the exception of the proximal femur, by Schaefer *et al.*[36], as mentioned. Although the studies were carried out in other populations, there was correspondence with the real age.

Partial fusion of the spheno-occipital synchondrosis is a biological indicator of failure to reach adulthood. In the case presented, the spheno-occipital synchondrosis was classified as partially fused, which is consistent with the estimated age of 16 years, since complete fusion of this joint usually occurs between 18 and 25 years. The presence of the synchondrosis at an intermediate stage reinforces its usefulness as a reliable marker for age estimation in subadult and young adult individuals. However, the variability associated with population and individual factors highlights the importance of interpreting this data in conjunction with other skeletal and contextual characteristics.

The medial epiphysis of the clavicle, with its slow maturation process, begins its formation as a rudimentary nodule, with complete fusion usually occurring after 22 years of age and ending around 30 years of age[21,23,41]. In the case of this 16-year-old individual, the epiphysis was not fused, which is expected for this age group, since fusion between the epiphysis and the diaphysis begins to occur between 16 and 21 years of age. The variation in the age of fusion, which can be influenced by genetic and environmental factors, should be considered, but the non-fused condition observed in this case corroborates the estimate of the individual's biological age. Furthermore, the maximum length of the clavicle (146.4 mm) was considered, and, among the various age estimates, this parameter proved to be the most accurate, with an age range of 15 to 16 years, according to data from Black and Scheuer[41].

Although the pubic symphysis methodology is widely described for age estimation in adults, it is important to note that the age ranges predicted by the morphology of the symphysial surface of the pubis also cover ages below 18 years , which justifies its application in younger individuals. The evaluation was performed using the Brooks and Suchey method[42], which classifies the pubic symphysis in phase 1 (figure 1). In addition, a more recent approach for the analysis of the pubic symphysis was considered[23], with partially distinct descriptions of the phases, but with a shorter age range compared to the original method.

The analysis of the sternal end of the fourth rib was performed based on the method of Iscan, Loth and Wright[26] which classifies the phase of the extremity into nine stages (0 to 8), evaluating characteristics such as the depth and shape of the fossa, in addition to the configurations of the edge and wall. In phase 0, the classification in which the analyzed rib was found, the individual is considered to be under 17 years of age. In the present case, the rib had a shallow fossa, rounded and firm edges, characteristics that, according to Hartnett[49], are typical of a young rib.

The dental analysis took into account the presence of 3 or 4 third molars. Although tooth 38 was not visible, the possibility of it being intraosseous was considered in the age estimate. The classification of third molar development was included in the male-specific formulas of Gunst *et al.*[46] that are based on their stages of mineralization. The analysis, without the use of radiography, was feasible as evidenced by Franco *et al.*[43], which reinforces the reliability of the macroscopic classification of the developing tooth and expands the possibility of using the method.

In this scenario, it is worth highlighting that an individual's biological age can be influenced by several factors, such as environmental, genetic and individual conditions, which can result in variations in bone development and fusion patterns. Furthermore, these variations can occur between different populations, making it even more important to consider these factors when applying age estimation methods[47,50]. However, in this case, the methods used for skeletal and dental analysis, based on anatomical macroscopic characteristics, have proven to be reliable, with the results obtained showing a good correlation between biological age and actual chronological age.

This case highlights the pressing need to establish more robust population standards for subadults in Brazilian anthropological forensics. The lack of clear and consistent references for this age group hinders the accuracy of biological age estimation in forensic cases involving young individuals. The analysis of bones and anatomical features in this study contributes to filling this gap, demonstrating the applicability of estimation methods in subadults and highlighting the importance of developing and validating specific standards for this population. This will allow anthropological forensics in Brazil to have a more solid basis for the interpretation of forensic cases involving individuals under 18 years of age, improving the accuracy and reliability of the results.

This study did not aim to estimate the age of the analyzed individual, but rather to verify the applicability of the methods in a Brazilian population context, highlighting the development phases observed in the bones of a 16-year-old individual. According to Machado *et al.*[15], despite being widely used, the multifactorial morphoscopic approach does not, by itself, resolve the difficulties in estimating age at death. When poorly executed, it can generate redundancy, multicollinearity, and analytical complexity, compromising the interpretability and practical value of the results.

Conclusion

This study reinforces the importance of comparing the age ranges provided by macroscopic bone and dental morphological methods described in the literature with the actual chronological age of subadult individuals. In the case analyzed, the estimates were consistent with the actual age, demonstrating the applicability of traditional methods of skeletal and dental analysis. However, the lack of specific population patterns for subadults in Brazil still represents a significant challenge. The validation of new approaches, combined with the integration of different methods and the consideration of individual variations, is essential to improve the accuracy and reliability of anthropological expertise in the country, contributing to advances in the forensic analysis of cases involving subadults.

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Corresponding author
Laíse Nascimento Correia Lima
E-mail: laise.lima@academico.ufpb.br