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ORIGINAL ARTICLE

Diagnostic accuracy of the third molar maturity index (I_{3M}) to assess the age of legal majority in Northern Brazil—population-specific cut-off values

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Abstract

International guidelines for dental age assessment have recommended the use of methods based on available population-specific data. The Third Molar Maturity Index (I_{3M}) was previously validated in several populations worldwide. This was the first study to evaluate the I_{3M} in a northern Brazilian population and to test the diagnostic accuracy of the method to distinguish between minors and adults. The sample consisted of 1.070 panoramic radiographs retrospectively collected from females ($n=595$) and males ($n=475$) with ages between 16 and 22 years. I_{3M}'s original cut-off value of 0.08 was used to classify individuals below and above the age of 18. Receiver operating characteristic (ROC) curves were plotted to assess the accuracy (ACC) of the method. In females and males separately, the overall ACC was 73.1% and 80%, respectively. The overall ACC for the combined sample was 76.1%. For northern Brazilian males, the best cut-off value remained 0.08, while for females, an adjustment to 0.12 showed optimal outcomes. The new cut-off value led to an ACC of 98.5% for females, which reflected an increase of 25.5% compared to the original cut-off value. The original cut-off value proposed by I_{3M} was applicable to the present sample of northern Brazilian individuals. Adjustments to 0.12, however, may be encouraged to enhance the performance of the method among females.

Keywords Brazil · Dental age assessment · Forensic Dentistry · Radiology · Third molar

Introduction

The decision-making process behind the selection of dental age assessment methods is guided by institutional recommendations in the form of guidelines and standards [1]. According to the Study Group of Forensic Age Diagnostics (SGFAD) of the German Society of Legal Medicine, there is a wide agreement on the suitability of dental methods for age estimation [2]. In the context of dental methods, the American Board of Forensic Odontology (ABFO) recommends those in which the expert has more familiarization [3]. Moreover, methods with population-specific data available are recommended [1]. Reference data becomes available through the publication of scientific studies for specific populations [4–7]. The so-called reference studies provide the expert with an overview of the performance of the method in a random sample from a country or region [2]. Consequently, the SGFAD explains that the method translates developmental data into a chronological scale [2]. As recommended by the International Organization of Forensic

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Odonto-Stomatology (IOFOS), in 2018, another aspect with an important part in the technical procedures is the combination of dental age estimation methods [8]. Hence, experts should be trained and be familiar with multiple methods and, ideally, data should be available for diverse populations worldwide.

The Third Molar Maturity Index (I_{3M}) was proposed in 2008 by Cameriere et al. [9] as a radio-diagnostic tool for dental age estimation. The method is based on a ratio between the measurements of tooth height and open apices of the mandibular left third molar (LL3rdM) [9]. Considering an original cut-off value of 0.08, the obtained ratio can distinguish individuals below (ratio ≥ 0.08) and above (ratio < 0.08) the age threshold of legal interest of 18 years [9]. This threshold represents the age of legal majority in most countries [10–12] and makes the I_{3M} potentially useful for age estimation of the living, such as in cases of asylum seekers [13–15] and criminal imputation of alleged minor offenders [16, 17]. The third molar is used in this method because this is the only developing tooth (position) in most of the adolescents, especially around the age of 18 [18–20].

A recent systematic review of validation studies of the I_{3M} in worldwide populations (16 eligible studies) showed an overall classification rate between 72.4 and 96% [21]. Meta-analyzed sensitivity and specificity rates were 0.88 and 0.96, respectively, and the pooled area under the curve was 0.96, revealing high discriminatory power to distinguish minors and adults based on the legal threshold of 18 years [21]. Specifically for the Brazilian population, I_{3M} was previously tested twice in the southeast region [22, 23] and twice in the northeast region [24, 25]. The southeast region is composed by four states and is considered the 2nd smallest region of Brazil; however, it has a dense population of 87 million habitants, with a predominance of mixed European ancestry. In this region, I_{3M} overall classification rates reached 87% [22]. The northeast region has nine states and over 56 million habitants with a predominance of African ancestry. In this region, I_{3M} reached accuracy rates of 79.8% [24] and 80% [25]. A recent meta-analysis of third molar dental age estimation methods in the Brazilian population ranked I_{3M} among the methods that are acceptable for application in the country [26]. However, regional analyses for the northern region are missing in the current literature.

The northern region of Brazil is composed of 7 states, in which over 17 million habitants live. Amazonas is located in this region and figures as the largest state in the country (with 4 million habitants). Ancestral influence in the region is predominantly Native American. The state has borders with Venezuela, Colombia and Peru — highlighting the importance of pioneering I_{3M} validation studies to support dental age estimation cases of clandestine migration across borders. The strong miscegenation of the Brazilian population can propose a real challenge to the performance of I_{3M} (originally European), especially because of the scarce European ancestry in the northern region compared to other Brazilian regions. This study aimed to test, for the first time, the performance of I_{3M} in a population of northern Brazilian individuals sampled from an image database of the State of Amazonas.

Material and methods

Study model and ethical aspects

An observational cross-sectional study with retrospective sample collection was designed. Ethical approval was obtained from the institutional committee of ethics in research (protocol: 44,953,421.2.0000.5374). Following national [27] and international ethical standards [28] for human research, the participants involved in this study were not exposed to ionizing radiation, and there was no prospective recruitment. All the radiographs (described in the "*Settings and participants*") were obtained for clinical reasons (diagnostic, therapeutic, and follow-up purposes) that were unrelated to the present study and were collected retrospectively from an existing database. The study was reported following the "Enhancing the Quality and Transparency of Health Research" (EQUATOR Network), specifically the STROBE (The Strengthening the Reporting of Observational Studies in Epidemiology) checklist for cross-sectional studies [29].

Settings and participants

The sample consisted of panoramic radiographs collected from 595 females (mean age = 19.10 ± 1.96) and 475 males (mean age = 18.73 ± 1.95) between 16 and 22.9 years (mean age = 18.94 ± 1.97 years). The inclusion criteria were panoramic radiographs of northern Brazilians presenting the LL3rdM visible. The exclusion criteria were panoramic radiographs without records of individuals' date of birth, date of image acquisition, and sex; visible bone lesions, surgical orthopedic appliances, and deformities of the mandible; third molars in a transverse position or tilted in the buccal/lingual direction (to avoid shortened tooth images); and panoramic radiographs with bad quality [30]. The application of the exclusion criteria reduced the sample size from 1.256 originally collected images to 1.070 (Table 1). According to a recent literature review of 22 studies with the I_{3M} [31], our sample size is bigger than 19 studies (86.3%) and has an equal sex distribution.

Table 1 Sample distribution between females and males for each of the age categories addressed in this study

Age category	Females	Males	Total
16 ├ 16.99	84	76	160
17 ├ 17.99	67	88	155
18 ├ 18.99	77	61	138
19 ├ 19.99	101	72	173
20 ├ 20.99	90	69	159
21 ├ 21.99	93	61	154
22 ├ 22.99	83	48	131
Total	595	475	1.070

Age categories are expressed in years

Data sources and measurements

The panoramic radiographs used in this study were collected in 2021, retrospectively, from an existing database of a private radiology clinic in the northern region of Brazil, located in the State of Amazonas. The radiology clinic operates in Manaus, the Capital city of Amazonas, and has a predominance of patients that are (non-tribal) Amazonians. Additional patients come mainly from the other states of the northern region. Hence, the radiographs that we collected were considered as a sample of the northern region of Brazil. Image acquisition was performed with the Kodak CS8100SC unit (Eastman Kodak Company, Rochester, NY, USA). The images were stored in.tiff format (600 dpi) and exported to a personal computer equipped with a 15″ screen and GIMP software version 2.10.30 (The GIMP Development Team, International) for analysis. Image magnification of 100% and eventual adjustments of brightness and contrast were allowed prior to analysis. The analysis of the panoramic radiographs was performed in a dimmed room under standard viewing conditions [32]. No more than 25 radiographs were analyzed per day to avoid visual fatigue [33]. Following the recommendations of IOFOS [8], the method (I_{3M}) was applied as originally described in the literature [9]. The height (h) of the LL3rdM was measured in the long axis of the tooth, while the open apices (a1 and a2) were measured between root walls (separately for the mesial and distal roots). The ratio between h and a1 + a2 was calculated and considered based on the reference cut-off value of 0.08 [9]. Individuals younger than 18 years would have a ratio ≥ 0.08, while older individuals (adults) should have ratios < 0.08. LL3rdM with closed apices would automatically indicate the individual as an adult.

Variables

The age category (younger or older than 18 years) that was estimated from the ratios between *h* and a1 + a2 figured as the main variable in this study. Additionally, the chronological age (second variable of this study) of the individuals was calculated as the difference between the date of image acquisition and the date of birth. The estimated age category and the chronological age of the individuals were compared separately for females and males (sex as the third variable). It is worth pointing out that the mean age of the individuals in the younger group (below 18 years) was 16.49 ± 0.50 years, while in the older group (above 18 years), the mean age was 19.96 ± 1.36.

Bias

Initially, a training session was established between the main observer and the supervisor (both forensic odontologists with over 10 years of experience in practice). During this period, one-hundred panoramic radiographs were measured and discussed based on I_{3M} reference ratio parameters, namely the (open) apices and the tooth height. Next, an intra-observer reproducibility test was performed by comparing the measurements of the main examiner in a sample of ten panoramic radiographs reassessed 30 days from the analysis of the main sample. The repetition of image analysis led to 30 measurements for comparison (10 h, 10 a1, and 10 a2). The number of images selected for re-analysis was based on a similar study published in 2020 [34]. Inter-observer reproducibility was assessed within the same images and considered the same number of measurements, and the same time interval between analyses. During this process, a second observer was recruited. The measurements performed by the first and second observers were compared. The intra-class correlation coefficient (ICC) was used.

Quantitative assessment and statistical analysis

The following steps were performed with R software version 4.1.2 (R Core Team, R Foundation for Statistical Computing, Vienna, Austria). All the analyses considered a significance level of 5%. Initially, the chronological age was converted into a binary variable: individuals with ages between 16 and 17.99 years (minors) vs. individuals that were 18 or older (adults). Kolmogorov–Smirnov test assessed and confirmed data normality ($p < 0.05$). A confusion matrix was produced for the total sample and females and males based on the performance of the I_{3M} to classify individuals as minors or adults. The performance of the method was quantified by means of sensitivity (SEN), specificity (SPE), positive predictive value (PV +), negative predictive value (PV −), positive likelihood ratio (LR +), negative likelihood ratio (LR −), and accuracy (ACC). A confidence interval of 95% was presented. In a deeper analysis, the sample was tested for a potential new population-specific cut-off value. In this stage, 2/3 of the samples were used for training purposes and 1/3 for (cross) validation. Logistic regression was applied considering the classification based on the chronological age as the outcome variable, and the I_{3M} ratio as the independent variable to be tested and the predictor. New cut-off values were investigated for the total sample, and separately for females and males. For each of the analyses, receiver operating characteristic (ROC) curves were produced. Using the highest SEN, SPE, and intercept obtained with the logistic regression, it was possible to detect the best cut-off values for the total sample and for females and males.

Results

ICC showed that the intra-observer reproducibility test for the height measurements of the LL3rdM was 0.997, while for the apical measurements, it was 0.996. The inter-observer ICC values were 0.972 and 0.994 for the height and apical measurements, respectively. All the measurements revealed excellent reproducibility [35].

Table 2 presents an overview of the sample and age distribution of the females and males with their mean age and standard deviation (SD) for each interval of I_{3M} ratios. It is possible to see that most of the individuals sampled in this study were allocated in the I_{3M} ratio interval of 0–0.3 (covering both the groups of minors and adults).

SD standard deviation, I_{3M} ratio interval.

The contingency table (Table 3) presents the number of correct classifications of minor/adult males and females and shows accuracy rates of 73.1% for females and 80% for males, and 76.1% for the total sample (combined sex).

F females, *M* males, *SEN* sensitivity, *SPE* specificity, *PV+* positive predictive value, *PV−* negative predictive value, *LR+* positive likelihood ratio, *LR−* negative likelihood ratio, *ACC* accuracy, *CI* confidence interval.

The logistic regression performed to investigate a potential cut-off value with a better classification of males and females (separately and combined) showed that 0.08 remained the best classifier for males and for the combined sample, while 0.12 would be the best value to classify minor/adult females. The new cut-off reached a specificity of 93%, showing the good performance of the new cut-off to avoid false positives (Table 4).

The original cut-off of 0.08 remained as the best for the combined sample and for males, while 0.12 emerged as the best for females.

F females, *M* males, *Ir.eta* metrics used to assess the best cut-off based on the highest sensitivity (SEM) and specificity (SPE) rates.

Figure 1 illustrates the ROC curves of the cut-off values plotted for the combined sample, females, and males described in Table 4. All the curves presented high specificity (related to false positives) values (99%, 93%, and 99.9% for the combined sample, females, and males, respectively) and moderate to high sensitivity (related to true positives) values (67.1%, 68.2%, and 79.8% for the combined sample, females, and males, respectively).

Table 2 Mean and standard deviation of the age of females and males for each of the I_{3M} ratio intervals

I_{3M}	Females	Age		Males	Age	
		Mean	SD		Mean	SD
0–0.04	262	20.27	1.48	249	19.74	1.69
0.04–0.08	80	19.08	1.77	73	18.56	1.65
0.08–0.3	167	18.26	1.71	111	17.24	1.40
0.3–0.5	54	17.44	1.61	19	17.00	1.20
0.5–0.7	20	17.00	1.58	14	16.93	0.91
0.7–0.9	8	16.50	1.06	7	17.00	0.81
0.9–1.2	4	16.25	0.50	2	16.00	0.00

Table 3 Contingency table of the classification of the total sample and females and males as minors or adults based on I_{3M} ratios

Sex	(I_{3M})	True group		SEN CI 95%	SPE CI 95%	PV+ CI 95%	PV- CI 95%	LR+ CI 95%	LR- CI 95%	ACC CI 95%
		Adult	Minor							
F+M	Adult	582	82	77.0% (74.4–79.5)	74.0% (71.3–76.6)	87.6% (85.6–89.5)	57.3% (54.3–60.2)	2.9 (2.5–3.2)	0.3 (0.2–0.3)	76.1% (73.5–78.6)
	Minor	173	233							
F	Adult	313	29	70.5% (66.8–74.1)	80.8% (77.6–83.9)	91.5% (89.2–93.7)	48.2% (44.1–52.2)	3.6 (3.0–4.1)	0.3 (0.2–0.3)	73.1% (69.5–76.6)
	Minor	131	122							
M	Adult	269	53	86.5% (83.4–89.5)	67.6% (63.3–71.8)	83.5% (80.1–86.8)	72.5% (68.4–76.5)	2.6 (2.0–3.1)	0.2 (0.1–0.2)	80.0% (76.4–83.6)
	Minor	42	111							

Based on the new cut-off value of 0.12 for northern Brazilian females, a new contingency table (Table 5) was developed with descriptive data about the performance of the cut-off as a classifier of minors and adults. The discriminant power of the cut-off increased the accuracy of I_{3M} by 25.5%, as well as all the other related metrics (namely SEN, SPE, PV +, PV −, LR +, and LR −).

F females, *SEN* sensitivity, *SPE* specificity, *PV+* positive predictive value, *PV−* negative predictive value, *LR+* positive likelihood ratio, *LR−* negative likelihood ratio, *ACC* accuracy, *CI* confidence interval.

Discussion

The diagnostic accuracy of I_{3M} to classify individuals as minors or adults based on the age threshold of legal interest of 18 years has been proved worldwide [36]. The continental size of Brazil highlights the need of regional validation studies. Despite the previous studies with I_{3M} in the southeast [22, 23] and northeast [24, 25] regions, no study was performed so far with a sample from the northern region. This study was designed to test, for the first time, the performance of I_{3M}'s original cut-off value of 0.08 to classify northern Brazilian minors and adults.

Recent studies have demonstrated the importance of regional validation studies, such as in the specific southeastern region of France [37] and the south region of India [38]. In our validation study, the overall accuracy for the correct classifications of minors and adults was 76.1% for the combined sex group, and 73.1% and 80% for females and males, using the cut-off value of 0.08. These outcomes were considerably less accurate when compared to the southeastern French population, for example, in which the correct classification of females and males reached rates of 0.897 and 0.916, respectively [37]. Similarly, the overall accuracy

Table 4 Sensitivity and specificity rates obtained with the logistic regression to find the best cut-off values and test their predictive performance to classify minors/adults

Sex	SEN	SPE	lr.eta	Intercept	Coefficient	Cut-off
F + M	67.1%	99.0%	0.838	2.323	− 8.124	0.08
F	68.2%	93.0%	0.865	2.901	− 8.488	0.12
M	79.8%	99.9%	0.770	2.018	− 10.136	0.08

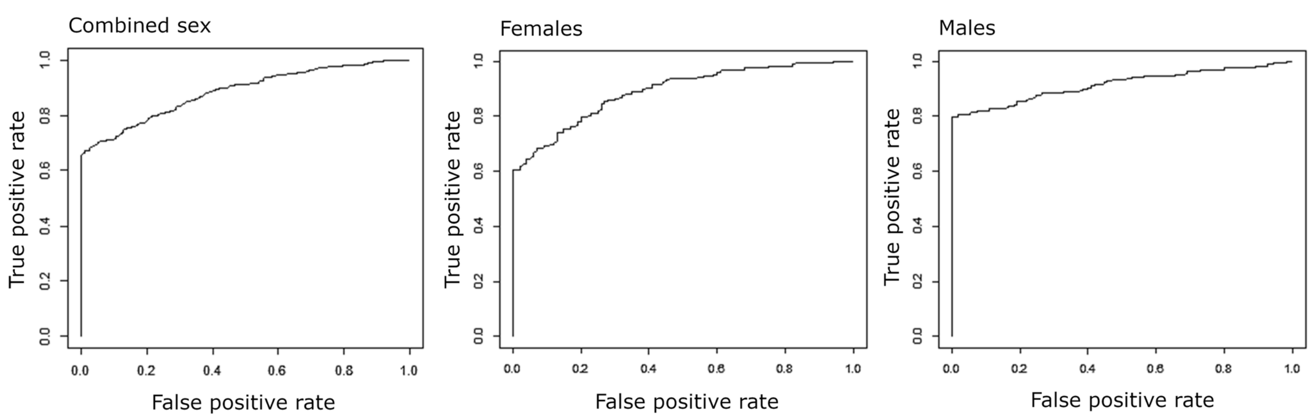

Fig. 1 Receiver operating characteristic (ROC) curves plotted for the cut-off value of 0.08 for the combined (total) sample and males, and for the new cut-off value of 0.12 plotted for the females sampled in the present study. The curves presented specificity (related to false positives) values of 99%, 93%, and 99.9% for the combined sample, females, and males, respectively, and sensitivity (related to true positives) values of 67.1%, 68.2%, and 79.8% for the combined sample, females, and males, respectively

Table 5 Female-specific contingency table for the new cut-off of 0.12 for northern Brazilian females

Sex	(I_{3M})	True group		SEN CI 95%	SPE CI 95%	PV + CI 95%	PV − CI 95%	LR + CI 95%	LR − CI 95%	ACC CI 95%
		Adults	Minors							
F	Adults	177	3	100.0%	86.3%	98.3%	100.0%	7.3	0.0	98.5%
	Minors	0	19	–	(81.5–91.0)	(96.5–99.9)	–	(6.1–8.4)	(0.0–0.0)	(96.8–99.9)

for the combined sample of females and males was below that obtained among South Indians (93.1% of correct classifications) [38]. The difference may be justified by the (more) challenging scenario proposed to test the I_{3M} in the present study. In general, studies from the past sampled individuals from 14 to 22 years [14, 37], while our sample was between 16 and 22 years.

The authors also demonstrated that less accurate classification rates are obtained by narrowing the age range from 16–21 years (90.3%) to 17–20 years (73.3%) [24]. This phenomenon can be explained by the progressive development of the LL3rdM roots, making it easier to classify someone as < 18 if the person's chronological age is 14 compared to a 16-year-old individual (time in which the root would be more developed). A second justification for the difference between our outcomes and those from other international studies is the sample size. While studies [37, 38] have sampled less than 350 panoramic radiographs, our study not only reduced the age interval but also added more radiographs to the sample ($n = 1.070$) — having a more representative number of individuals per age interval of 1 year. Specifically, between females and males, we had at least 131 individuals in each age interval from 16 to 22.9 years.

Compared to other studies previously published in Brazil, we reported accuracy rates that were slightly below those reported for the northeastern population (80%) [25]. Similarly, in our study, the accuracy rates were considerably better for males (80%) compared to females (73.1%). In the northeastern population, accuracy rates were 84% for males and 78% for females [25]. Given the good sex distribution in our sample (55.6% of females and 44.4% of males), we understood that the suboptimal predictions for females could be associated with the ancestral component (or miscegenation) of our sample. Hence, we considered a potential adjustment of the original cut-off for the female group. In fact, new cut-off values were searched for the total sample and for males as well, but 0.08 remained the best scenario in these cases. Our decision to search for adjustments came from the study of Cameriere et al. [39], in 2014, in which ethnicity/ancestry was raised as a factor that could justify the adaptation of cut-off values. Our methodological decision pointed toward the cut-off of 0.12 for northern Brazilian females — a value that reached a sensitivity of 100%, specificity of 86.3%, and an accuracy of 98.5% (increasing the previous accuracy rates by nearly 25%).

Considering the forensic expertise in the living, our outcomes may be useful to classify migrants crossing borders between the State of Amazonas and Venezuela, Colombia, and Peru. Moreover, cases of criminal imputation have been reported for alleged Venezuelan minor offenders judged in Brazil [16] — as the opposite may occur (Brazilians judged in Venezuela), the results of the present study could be useful to support dental age estimation practice abroad. When it comes to dental human identification (forensic expertise in the deceased), the State of Amazonas has seven forensic odontologists covering an area of 1.571.000 km^2, in which charred and decomposed bodies (especially retrieved from rivers) are routinely found. The I_{3M} could play an important part as an auxiliary tool to retrieve the identity of the deceased.

Future studies in the field should be designed to map the different geographic regions of the country in order to test and potentially validate the I_{3M} cut-off value. Eventually, new cut-offs may emerge as the result of the country(region)-specific adaptations. In the present study, a specific cut-off proved to be more appropriate for northern Brazilian females. The authors should strive for large and balanced samples that can be more representative of the geographic regions of interest. While specific states of the southeast, northeast, and northern (current study sample) regions have been mapped so far, there are no studies with the I_{3M} with representative samples of the south and central-west regions of Brazil. Together, both regions have over 45 million habitants — highlighting the need for dedicated studies. This study recommends, however, adaptations of cut-off values only after a detailed (statistical) investigation of the performance of the original cut-off value, and the evidence of ground for improvement with the inherent adaptation.

Conclusion

The original cut-off value of 0.08 proposed with the I_{3M} showed proper diagnostic accuracy to distinguish northern Brazilians as minors and adults (considering the age threshold of legal interest of 18 years). Improvement, however, was observed for females with a new population-specific cut-off value of 0.12. With the new cut-off, the diagnostic accuracy to classify females increased by 25%, reaching an overall 98.5% rate. Among males, the diagnostic accuracy was 80% (obtained with the original cut-off of 0.08).

Declarations

Ethics approval Protocol number 44953421.2.0000.5374.

Research involving human participants and/or animals The research involved human participants. The study was observational, and the sample was collected retrospectively from an existing image database. The Declaration of Helsinki (2013) was followed, and ethical approval was obtained from the institutional committee of ethics in human research.

Informed consent N/A (the study had a retrospective sample collection of an image database).

Competing interests The authors declare no competing interests.

References

1. American Board of Forensic Odontology (2020) Dental age assessment procedures. Available at: http://abfo.org/wp-content/uploads/2016/06/Dental-Age-Assessment-Procedures-Chart-2016.pdf. Accessed 19 April 2022
2. Schmeling A, Grundmann C, Fuhrmann A, Kaatsch HJ, Knell B, Ramsthaler F, Reisinger W, Riepert T, Ritz-Timme S, Rösing FS, Rötzscher K, Geserick G (2008) Criteria for age estimation in living individuals. Int J Legal Med 122:457–460. https://doi.org/10.1007/s00414-008-0254-2
3. American Board of Forensic Odontology (2020) ABFO standards and guidelines for dental age assessment. Available at: http://abfo.org/wp-content/uploads/2021/02/ADA-Technical-Report-No.-1077_July_2020.-02-2021.pdf. Accessed 19 April 2022
4. Thevissen PW, Fieuws S, Willems G (2010) Human third molars development: comparison of 9 country specific populations. Forensic Sci Int 201:102–105. https://doi.org/10.1016/j.forsciint.2010.04.054
5. Lewis JM (2010) Senn DR (2010) Dental age estimation utilizing third molar development: a review of principles, methods, and population studies used in the United States. Forensic Sci Int 201:79–83. https://doi.org/10.1016/j.forsciint.2010.04.042
6. Thevissen P, Altalie S, Brkić H, Galić I, Fieuws S, Franco A, Lee SS, Pinchi V, Yusof MYPM, Willems G (2013) Comparing 14 country-specific populations on third molars development: consequences for age predictions of individuals with different geographic and biological origin. J F Odontostomatol 31:87–88
7. Mincer HH, Harris EF, Berryman HE (1993) The A.B.F.O. study of third molar development and its use as an estimator of chronological age. J Forensic Sci 38:379–390
8. International Organization of Forensic Odonto-Stomatology (2018) Recommendations for quality assurance: dental age estimation. Available at: http://www.iofos.eu/Quality-Ass/QA_2018_final/Age_estimation_final.pdf. Accessed 19 April 2022
9. Cameriere R, Ferrante L, De Angelis D, Scarpino F, Galli F (2008) The comparison between measurement of open apices of third molars and Demirjian stages to test chronological age of over 18 year olds in living subjects. Int J Legal Med 122:493–497. https://doi.org/10.1007/s00414-008-0279-6
10. Franco RPAV, Franco A, Turkina A, Arakelyan M, Arzukanyan A, Velenko P, Bortolami PB, Makeeva I, da Silva RHA (2021) Radiographic assessment of third molar development in a Russian population to determine the age of majority. Arch Oral Biol 125:105102. https://doi.org/10.1016/j.archoralbio.2021.105102
11. AlQahtani S, Kawthar A, AlAraik A, AlShalan A (2017) Third molar cut-off value in assessing the legal age of 18 in Saudi population. Forensic Sci int 272:64–67. https://doi.org/10.1016/j.forsciint.2017.01.004
12. Gulsahi A, De Luca S, Cehreli SB, Tirali RE, Cameriere R (2016) Accuracy of the third molar index for assessing the legal majority of 18 years in Turkish population. Forensic Sci Int 266:584.e1-584. https://doi.org/10.1016/j.forsciint.2016.04.034
13. Zelic K, Galic I, Nedeljkovic N, Jakovljevic A, Milosevic O, Djuric M, Cameriere R (2016) Accuracy of Cameriere's third molar maturity index in assessing legal adulthood on Serbian population. Forensic Sci Int 259:127–132. https://doi.org/10.1016/j.forsciint.2015.12.032
14. Ribier L, Saint-Martin P, Seignier M, Paré A, Brunereau L, Rérolle C (2020) Cameriere's third molar maturity index in assessing age of majority: a study of a French sample. Int J Legal Med 134:783–792. https://doi.org/10.1007/s00414-019-02123-z
15. De Luca S, Biagi R, Begnoni G, Farronato G, Cingolani M, Merelli V, Ferrante L, Cameriere R (2014) Accuracy of Cameriere's cut-off value for third molar in assessing 18 years of age. Forensic Sci Int 235:102.e1–6. https://doi.org/10.1016/j.forsciint.2013.10.036
16. Goetten IFS, Silva RF, Franco A (2021) Skeletal and dental age estimation of the living in a criminal scenario – case report. Rom J Legal Med 29:105–108. https://doi.org/10.4323/rjlm.2021.105
17. Silva RF, Mendes SDSC, Franco A, Dias PEM, Martorell LB (2013) Documental vs biological evidence for age estimation forensic case report ROBRAC 21:6–10. https://doi.org/10.36065/robrac.v22i60.731
18. Franco A, Vetter F, Coimbra EF, Fernandes Â, Thevissen P (2020) Comparing third molar root development staging in panoramic radiography, extracted teeth, and cone beam computed tomography. Int J Legal Med 134:347–353. https://doi.org/10.1007/s00414-019-02206-x
19. Sartori V, Franco A, Linden MS, Cardoso M, de Castro D, Sartori A, Silva C, Trentin M, De Carli JP (2021) Testing international techniques for the radiographic assessment of third molar maturation. J Clin Exp Dent 13:e1182–e1188. https://doi.org/10.4317/jced.58916
20. Gaêta-Araujo H, Oliveira-Santos N, Nascimento EHL, Nogueira-Reis F, Oenning AC, Groppo FC, Oliveira-Santos C (2021) A new model of classification of third molars development and its correlation with chronological age in a Brazilian subpopulation. Int J Legal Med 135:639–648. https://doi.org/10.1007/s00414-020-02401-1
21. Santiago BM, Almeida L, Cavalcanti YW, Magno MB, Maia LC (2018) Accuracy of the third molar maturity index in assessing the legal age of 18 years: a systematic review and meta-analysis. Int J Legal Med 132:1167–1184. https://doi.org/10.1007/s00414-017-1766-4
22. Deitos AR, Costa C, Michel-Crosato E, Galić I, Cameriere R, Biazevic MGB (2015) Age estimation among Brazilians: younger or older than 18? J Forensic Legal Med 33:111–115. https://doi.org/10.1016/j.jflm.2015.04.016
23. Mazzilli LEN, Melani RFH, Lascala CA, Palacio LAV, Cameriere R (2018) Age estimation: Cameriere's open apices methodology accuracy on a southeast Brazilian sample. J Forensic Leg Med 58:164–168. https://doi.org/10.1016/j.jflm.2018.06.006
24. Correia AM, Barbosa DS, Alcantara JAS, Oliveira PMC, Silva PGB, Franco A, Bezerra TP (2020) Performance and comparison of the London Atlas technique and Cameriere's third molar maturity index (I3M) for allocating individuals below or above the threshold of 18 years. Forensic Sci Int 317:110512. https://doi.org/10.1016/j.forsciint.2020.110512
25. Nóbrega JBM, Protasio APL, Ribeiro ILA, Valença AMG, Santiago BM, Cameriere R (2019) Validation of the Third Molar Maturation Index to estimate the age of criminal responsibility in Northeastern Brazil. Forensic Sci Int 304:109917. https://doi.org/10.1016/j.forsciint.2019.109917
26. Franco A, Vidigal MTC, Oliveira MV, Nascimento CTJS, Silva RF, Paranhos LR (2020) Evidence-based mapping of third molar techniques for age estimation applied to Brazilian adolescents a systematic review. Res Soc Develop 9:e9339109395. https://doi.org/10.33448/rsd-v9i10.9395. Accessed 19 April 2022
27. BRASIL (2012) National Council of Health. Resolution n. 466 of December 12th. Available at: https://bvsms.saude.gov.br/bvs/saudelegis/cns/2013/res0466_12_12_2012.html. Accessed 19 April 2022
28. WMA – World Medical Association (2013) Declaration of Helsinki. Available at: https://www.wma.net/what-we-do/medical-ethics/declaration-of-helsinki/. Accessed 19 April 2022
29. Von Elm E, Altman DG, Egger M, Pocock SJ, Gøtzsche PC, Vandenbroucke JP, Initiative STROBE (2008) The Strengthening the reporting of observational studies in epidemiology (STROBE) statement: guidelines for reporting observational studies. J Clin

Epidemiol 61:344–349. https://doi.org/10.1016/j.jclinepi.2007.11.008

30. Moukarzel M Angelakopoulos N, De Luca S, Palacio LAV, Aquilanti L, Coccia E, Pigolkin YI, Cameriere R (2020) Validity assessment of the third molar maturity index (I3M) in a Lebanese sample of adolescents and young adults Aust J Forensic Sci Epub:1–12 https://doi.org/10.1080/00450618.2020.1805008
31. De Micco F, Martino F, Velandia Palacio LA, Cingolani M, Campobasso CP (2021) Third molar maturity index and legal age in different ethnic populations: accuracy of Cameriere's method. Med Sci Law 6:105–112. https://doi.org/10.1177/0025802419900686
32. Vasconcelos KF, Nicolielo LFP, Nascimento MC, Haiter-Neto F, Bóscolo FN, Van Dessel J, EzEldeen M, Lambrichts I, Jacobs R (2015) Artefact expression associated with several cone-beam computed tomographic machines when imaging root filled teeth. Int Endod J 48:994–1000. https://doi.org/10.1111/iej.12395
33. Nascimento EHL, Gaêta-Araujo H, Vasconcelos KF, Freire BB, Oliveira-Santos C, Haiter-Neto F, Freitas DQ (2018) Influence of brightness and contrast adjustments on the diagnosis of proximal caries lesions. Dentomaxillofac Radiol 47:20180100. https://doi.org/10.1259/dmfr.20180100
34. Neves JA, Antunes-Ferreira N, Machado V, Botelho J, Proença L, Quintas A, Delgado AS, Mendes JJ, Cameriere R (2020) Validation of the Third Molar Maturation Index (I3M) to assess the legal adult age in the Portuguese population. Sci Rep 10:18466. https://doi.org/10.1038/s41598-020-75324-x
35. Koo TK, Li MY (2016) A guideline of selecting and reporting intraclass correlation coefficients for reliability research. J Chiropr Med 15:155–163. https://doi.org/10.1016/j.jcm.2016.02.012
36. Angelakopoulos N, Galić I, Balla SB, Kiş HC, Gómez Jiménez L, Zolotenkova G (2021) Comparison of the third molar maturity index (I 3M) between left and right lower third molars to assess the age of majority: a multi-ethnic study sample. Int J Legal Med 135:2423–2436. https://doi.org/10.1007/s00414-021-02656-2
37. Tafrount C, Galic I, Franchi A, Fanton L, Cameriere R (2019) Third molar maturity index for indicating the legal adult age in Southeastern France. Forensic Sci Int 294:218.e1-218.e6. https://doi.org/10.1016/j.forsciint.2018.10.013
38. Balla SB, Galic I, Karunakar P, Vanin S, De Luca S, Cameriere R (2017) Validation of third molar maturity index (i3m) for discrimination of juvenile/adult status in South Indian population. J Forensic Legal Med 49:2–7. https://doi.org/10.1016/j.jflm.2017.05.003
39. Cameriere R, Santoro V, Roca R, Lozito P, Introna F, Cingolani M, Galic I, Ferrante L (2014) Assessment of legal adult age of 18 by measurement of open apices of the third molars: study on the Albanian sample. Forensic Sci Int 245:205.e1–5. https://doi.org/10.1016/j.forsciint.2014.10.013