

FORENSIC AGE ESTIMATION USING NEW MODELS OF MATHEMATICAL REGRESSION FORMULA CONSTRUCTED WITH MOLAR INDEXES: DENTAL AGE ASSESSMENT

Cristiana Palmela Pereira^{1,2,3}, Ana Rodrigues^{1,2}, Diana Augusto^{1,2}, Adriana Santos^{1,2}, Francisco Salvado⁴, Rui Santos^{2,5}, Roberto Cameriere⁶

¹FORENSEMED research group from UICOB, Faculty of Dental Medicine, University of Lisbon, Portugal

²CEAUL – Centre of Statistics and its Applications, University of Lisbon, Portugal

³Faculty of Dental Medicine, University of Lisbon, Lisboa, Portugal

⁴Stomatology Department, Santa Maria Hospital, Faculty of Medicine, University of Lisbon, Portugal

⁵School of Technology and Management, Polytechnic Institute of Leiria, Portugal

⁶AgEstimation Project, FOR.MED.LAB, University of Macerata, Italy

ABSTRACT

INTRODUCTION: In forensic science, age estimation for identification purposes of living and deceased individuals, is of great importance. It is crucial for humanitarian reasons in both civil and criminal cases.

OBJECTIVES: The aim of this study was to contribute to the process of forensic age estimation using dental age assessment with mineralization stages of Demirjian method, and maturity indexes of permanent mandibular second (I_{2M}) and third molars (I_{3M}).

MATERIAL AND METHODS: Two samples were used from a Portuguese population; for I_{2M} , 591 orthopantomography's aged between 7 and 15 years and for I_{3M} , 350 orthopantomography's aged between 12 and 23 years. Age estimation was obtained using linear regression models, each model was estimated with all observations and without observations, in which I_{2M} or $I_{3M} = 0$.

RESULTS: The results of ICC for intra- and inter-observer validation varied between 0.608 and 0.999 for both indexes. Pearson correlation coefficient between each index and chronological age showed that all indexes were significantly negatively correlated with chronological age. The standard error of estimate and the mean absolute error were continually low without patients with null index.

CONCLUSIONS: Application of I2M is reproducible in a Portuguese population for medical-legal application in a population aged 7 to 15 years, when the corresponding index is positive. Third molar must be used only if its index is positive. The inclusion of Demirjian staging in both models significantly increases accuracy.

KEY WORDS: dental age assessment, mathematical regression formula, maturity indexes, Demirjian stages, forensic age estimation.

J Stoma 2021; 74, 2: 95-100

DOI: <https://doi.org/10.5114/jos.2021.106540>

**JOURNAL OF
STOMATOLOGY**
CZASOPISMO STOMATOLOGICZNE

OFFICIAL JOURNAL OF THE POLISH DENTAL ASSOCIATION | ORGAN POLSKIEGO TOWARZYSTWA STOMATOLOGICZNEGO



ADDRESS FOR CORRESPONDENCE: PhD Cristiana Palmela Pereira, Auxiliary Professor with aggregation Faculdade de Medicina Dentária da Universidade de Lisboa, Rua Professora Teresa Ambrósio, Cidade Universitária, 1600-003 Lisboa, Portugal, e-mail: cpereira@campus.ul.pt

RECEIVED: 15.12.2020 • ACCEPTED: 11.03.2021 • PUBLISHED: 04.06.2021

INTRODUCTION

In forensic science, age estimation for human identification of living and deceased individuals is truly remarkable [1]. It is important for humanitarian reasons in both civil and criminal cases involving young people, in the discovery of valid identification documents, for crime agents who refuse to provide their real age, and in questions related to illegal and irregular immigration as well as asylum application for foreign countries [2, 3]. Therefore, age estimation plays a significant role in the judicial system and in law enforcement worldwide [4]. Cameriere *et al.* developed a regressive formula, in which the ratio given by the sum of distances from the inner sides of the open apices and the height of the developing second and third molars (second molar maturity index – I_{2M} and third molar maturity index – I_{3M}), is a reliable method to categorize, regardless of geographic origin and socio-economic status [5, 6]. However, as third molars are often absent or extracted, they are not always available as an age indicator, and even when present, third molars cannot always be used to estimate age in children and adolescents, due to position or shape anomalies [7]. As an alternative, the development of second molar occurs earlier and in a predictable way, being a reliable tooth for age estimation [8].

Previously, Demirjian *et al.* developed a method based on several mineralization stages of a tooth formation (from A to H), which can be applied to all teeth except third molars [9].

OBJECTIVES

The aim of the present study was to contribute to the process of forensic age estimation using dental age assessment with the mineralization stages of Demirjian method, and maturity indexes for permanent mandibular second and third molar teeth, in a Portuguese population.

MATERIAL AND METHODS

ETHICAL APPROVAL AND COMPLIANCE WITH ETHICAL STANDARDS

This study was performed in accordance with ethical standards specified by health ethics committee of the Faculty of Dental Medicine, University of Lisbon, and approved with 911105 and 911106 numbers.

INFORMED CONSENT

All procedures performed in studies involving human participants are in accordance with the ethical standards of the institutional and/or national research

committee and with the 1964 Helsinki Declaration and its later amendments or comparable ethical standards. Since this was a retrospective study based on anonymized clinical records, no informed consent was obtained.

SAMPLES

To estimate age from I_{2M} , 591 panoramic radiographs (253 from females and 338 from males; age range, 7-15 years) were selected, and to estimate age from I_{3M} , we selected 350 panoramic radiographs (181 from females and 169 from males; age range, 12-23 years). The sample distribution is shown in Table 1. Samples were taken randomly from an X-ray database of stomatology department, Hospital Santa Maria, targeting patients from the Lisbon North University Hospital Center. Selection criteria included healthy subjects of Portuguese origin, presence of the second and third molars in all lower quadrants, absence of congenital dental anomalies in shape or/and position, such as root canal treatment, caries, or restorations in the left mandibular permanent teeth. Heavily rotated, impacted, and unclear panoramic radiographs were excluded from the analysis. Moreover, individual's gender, date of birth, and the date of panoramic radiograph were recorded. Chronological age (CA) for each subject was computed by subtracting the date of the X-rays from the date of birth.

MEASUREMENTS

As proposed by Cameriere *et al.* [6], the maturity index was assessed as follows: a ratio between the sum of the inner side's width of the open apices (A1 and A2) in the left and right lower second or third molars and the tooth's length (L), when more than one root were presented, or a ratio between the width of the open apex (A) and the tooth's length (L), when a single root was present. X-ray images in JPG format were analyzed by ImageJ software program. The radiographs were scaled at 400%, and the measurement was changed from pixels to millimeters. Mineralization of permanent teeth on the lower left mandible were also assessed according to Demirjian [9] classification for later comparison with a molar maturity index.

STATISTICAL ANALYSIS

The intra-class correlation coefficient (ICC) was applied to quantify intra- and inter-observer agreement in I_{3M} , and intra-agreement in I_{2M} . For this purpose, 10% of the study sample was randomly selected 3 months following the initial scoring process to determine percentage of agreement, for both intra- and inter-observer agreement analyses. Pearson correlation coefficients were

used to evaluate the correlation between the maturity indexes and chronological age. Linear and cubic regression models were investigated, in which only variables with significant coefficients (p -value lower than 0.05) in t -test were included. Estimation errors were computed. T -test to paired samples was applied in order to test the existence of significant differences between chronological age and dental age estimated by the application of indexes.

RESULTS

The results of ICC for intra- and inter-observer validation varied between 0.608 and 0.999 for the second and third molars. Therefore, the obtained values denoted, for the most part, excellent results both in precision and in reproducibility. The only values below 0.75 were observed in the maximum length of monoradicular teeth in the second molar.

Pearson correlation coefficient between each maturity index and chronological age were calculated. All the values were similar, -0.781 for the tooth 37, -0.792 for the tooth 47, -0.775 for the tooth 38, and -0.751 for the tooth 48. All maturity indexes were significantly negatively correlated with chronological age (decrease when age increases, Figure 1). Furthermore, Spearman correlation coefficients were computed in order to measure monotone association with chronological age (and not only linear association), and also to compare maturity index results with the ones obtained by Demirjian stages. These coefficients were even more significant than Pearson correlation coefficients. Therefore, between the maturity index and chronological age, Spearman correlation was -0.856 for the tooth 37, -0.870 for the tooth 47, -0.862 for the tooth 38, and -0.850 for the tooth 48. Moreover, between Demirjian stages and chronological age, the coefficient was equal to 0.839 for the tooth 37 and to 0.862 for the tooth 47. Therefore, Demirjian stages were also strongly correlated with chronological age (Fig-

TABLE 1. Age distribution by sex

Age (years)	Samples for I_{2M}			Samples for I_{3M}		
	Sex		Total	Sex		Total
	Male	Female		Male	Female	
7	38	25	63	–	–	–
8	48	36	84	–	–	–
9	60	41	101	–	–	–
10	54	39	93	–	–	–
11	45	29	74	–	–	–
12	35	38	73	22	30	52
13	28	22	50	27	19	46
14	23	17	40	23	24	47
15	7	6	13	16	13	29
16	–	–	–	13	13	26
17	–	–	–	8	22	30
18	–	–	–	10	9	19
19	–	–	–	15	13	28
20	–	–	–	11	5	16
21	–	–	–	10	10	20
22	–	–	–	8	13	21
23	–	–	–	6	10	16
Total	338	253	591	169	181	350

ure 2). Hence, all these variables were strongly correlated with chronological age.

Despite genders being almost equally distributed in each age range for both I_{2M} and I_{3M} , normality was clearly rejected for the maturity index (almost null p -values) and non-parametric tests had to be applied. Thus, Mann-Whitney test revealed statistically significant differences in the I_{2M} between genders (p -values equal to 0.002 in

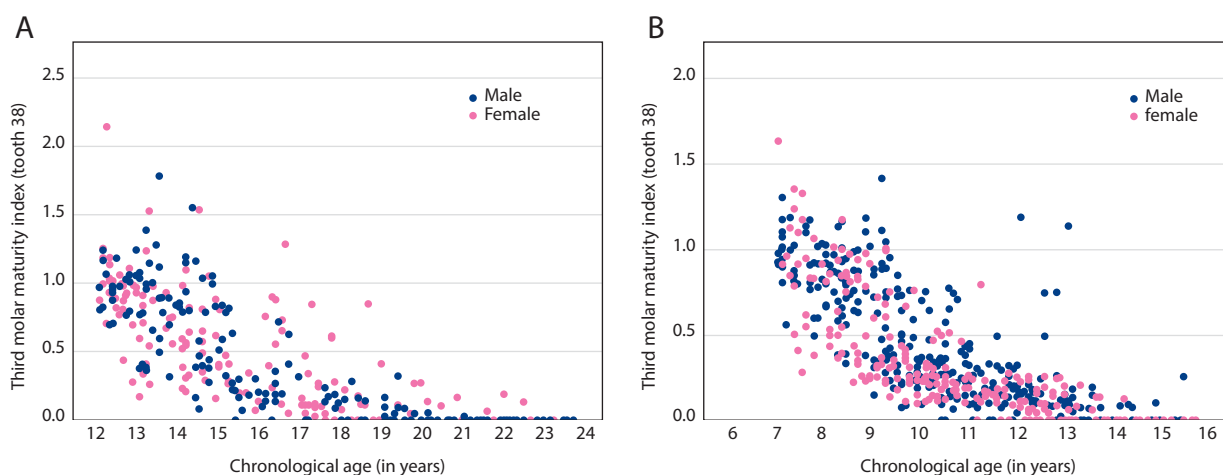


FIGURE 1. Second (A) and third (B) maturity index by chronological age (in months)

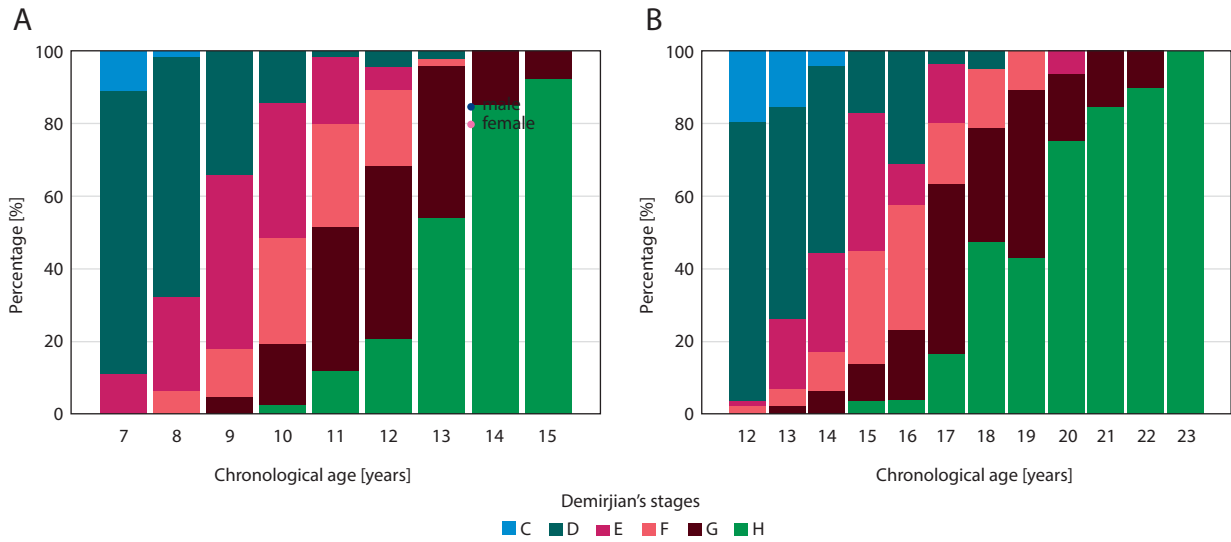


FIGURE 2. Demirjian's stages by age for the second (A) and the third (B) molars

TABLE 2. Model's estimation accuracy for maturity indexes not null (values in parentheses refer to the entire sample, including those cases with null maturity indexes)

Model	Adjust R ²	Estimated models Estimated age (in years) is equal to:	Bias (years)	SEE (years)	MAE (years)	Errors (absolute value)		
						< 1 year	Between 1 and 2 years	> 2 years
Model 1	0.639	$12.687 - 1.894 I_{2M_{37}} - 3.248 I_{2M_{47}} - 0.313 G$	0.221 (0.0)	1.195 (1.309)	0.95 (1.240)	300	152	38
Model 1 $I_{2M} \neq 0$	0.580	$12.096 - 1.412 I_{2M_{37}} - 2.822 I_{2M_{47}} - 0.391 G$	0	1.159	0.898	313	146	31
Model 2	0.748	$13.79 - 1.221 I_{2M_{37}} - 2.258 I_{2M_{47}} - 0.333 G - 2.39 E_C - 2.214 E_D - 2.631 E_E - 2.035 E_F - 1.124 E_G$	0 (0.0)	1.047 (1.086)	0.795 (0.83)	334	138	18
Model 2 $I_{2M} \neq 0$	0.653	$12.698 - 1.237 I_{2M_{37}} - 2.261 I_{2M_{47}} - 0.396 G - 1.241 E_C - 1.086 E_D - 1.506 E_E - 0.91 E_F$	0	1.055	0.804	335	136	19
Model 3	0.721	$13.217 - 13.079 I_{2M_{37}} + 12.126 I_{2M_{37}}^2 - 3.989 I_{2M_{37}}^3$	0.021 (0.0)	1.125 (1.144)	0.861 (0.887)	327	135	30
Model 3 $I_{2M} \neq 0$	0.606	$13.217 - 13.079 I_{2M_{37}} + 12.126 I_{2M_{37}}^2 - 3.989 I_{2M_{37}}^3$	0	1.126	0.858	318	140	34
Model 4	0.601	$19.341 - 6.242 I_{3M_{38}}$	0.593 (0.0)	1.934 (2.190)	1.601 (1.762)	99	73	86
Model 4 $I_{3M} \neq 0$	0.473	$17.499 - 4.096 I_{3M_{38}}$	0	1.740	1.378	105	96	57
Model 5	0.764	$21.023 - 1.355 I_{3M_{38}} - 6.372 E_C - 6.162 E_D - 5.452 E_E - 4.537 E_F - 2.718 E_G$	0 (0.0)	1.551 (1.683)	1.190 (1.307)	135	79	44
Model 5 $I_{3M} \neq 0$	0.571	$14.86 - 1.355 I_{3M_{38}} - 3.654 E_C - 3.444 E_D - 2.733 E_E - 1.819 E_F$	0	1.566	1.190	135	79	44
Model 6	0.743	$20.58 - 20.324 I_{3M_{38}} + 18.219 I_{3M_{38}}^2 - 5.061 I_{3M_{38}}^3$	0.099 (0.0)	1.638 (1.756)	1.266 (1.383)	130	80	48
Model 6 $I_{3M} \neq 0$	0.562	$19.111 - 13.377 I_{3M_{38}} + 10.158 I_{3M_{38}}^2 - 2.518 I_{3M_{38}}^3$	0	1.583	1.212	124	91	43

both teeth), but not in the I_{3M} (p -values 0.293 and 0.639 on teeth 38 and 48, respectively).

The age estimation was obtained by using linear regression models. Each model was estimated with all the observations and then removing the observations, in which $I_{2M} = 0$, because from the moment $I_{2M} = 0$, there was no more evolution of the index value with age.

Hence, the first model to estimate chronological age (model 1) considered I_{2M} from the teeth 37 ($I_{2M,37}$) and 47 ($I_{2M,47}$), and gender (G ; 0 for male and 1 for female) as explanatory variables. For model 2, Demirjian stadiums were included using artificial binary variables E_C , E_D , E_E , E_P , and E_G , which were equal to one, whether the patient belonged to the corresponding stadium (equal to zero otherwise). Lastly, cubic models were also investigated (model 3). The same three models were estimated without the 101 observations, in which $I_{2M} = 0$, using a total of 490 panoramic radiographs.

The same methodology was applied for I_{3M} , first with all the observations and then without the 91 patients with $I_{3M} = 0$ (remaining 258 panoramic radiographs). Hence, the fourth model considered I_{3M} from the teeth 38 ($I_{3M,38}$) and 48 ($I_{3M,48}$), and gender (G) as explanatory variables (model 4). For model 5, Demirjian stages were included in model 4, using artificial binary variables. Nevertheless, the explanatory variables $I_{3M,48}$ and G were removed from the model since they were not significant. The obtained cubic model was classified as model 6. All the results from the six models are summarized in Table 2.

The adjusted R^2 was always higher using all observations than the same model without the cases, in which the index was null. Nevertheless, the standard error of estimate (SEE) and mean absolute error (MAE) were constantly lower without patients with null index. This difference appeared from comparing the estimates in different groups. Table 2 shows the results of both methods in the age estimation of patients with maturity index not null.

Models with greater explanatory power were the ones, which included Demirjian stages (model 2 for I_{2M} and model 5 for I_{3M}), revealing higher determination coefficients as well as lower SEE and MAE. Moreover, models 1, 3, 4, and 6 showed some bias whenever the model was estimated using the entire sample, although SEE and MAE were almost similar. In addition, age estimation errors tended to significantly increase when the models were applied to patients with null maturity index.

DISCUSSION

Over the years, age estimation methods involving the development of second and third molars have been used. For the same reason, several studies have been conducted to improve the precision of this technique. The obtained results for ICC showed excellent results both on precision and reproducibility, so much for I_{2M}

as for I_{3M} , which was confirmed in literature [6, 10-14]. Regarding Pearson correlation of I_{2M} , it was possible to verify that I_M of the 47 tooth revealed a slightly more significant correlation with chronological age than I_M of the 37 tooth. In fact, the use of I_{2M} makes this study relevant because there were quite few studies in literature regarding the application of I_{2M} in age estimation. Mann-Whitney test revealed statistically significant differences between genders in I_{2M} , which was experienced in some published literature, since girls usually tend to have a faster development of second molars than boys [15]. For I_{3M} , it was evaluated both 38 and 48 teeth, contrasting with most studies using the same method [5, 10-12, 14, 16, 17], including only the lower left third molar (38th tooth). This was a significant evaluation since we concluded that the information of both teeth did not allow to improve the estimates since the information of the index of one of the teeth was considered irrelevant, when the index of the other was already in a model. Regarding gender, the obtained results revealed that there were no statistically significant differences, which was sustained with most of the published studies [5, 10, 12]. However, there are some studies with opposite evidence. According to Balla *et al.* [16], the correlation between I_{3M} and real age shows better results in girls, while Kelmendi *et al.* [18] proved that boys presented better results.

The association between Demirjian stages and chronological age for both methods revealed no statistically significant differences between the two genders, which was also in accordance with the existing literature [6, 19].

To the best of our knowledge, there are no other studies regarding age estimation by regression models with I_{2M} as an explanatory variable. This highlights the results of this investigation, which are quite satisfactory, although future confirmation of the application of different samples is necessary. We can confirm that the best models included Demirjian stages and maturity index. Hence, the use of both methods simultaneously provides a more accurate age estimate. Regarding the inclusion of $I_{2M} = 0$ (as it is typical in this kind of studies), despite the improved determination coefficient, the estimation errors increased, and the estimates were less accurate. Thus, we cannot advise the use of $I_{2M} = 0$.

In case of the I_{3M} , the same occurs. Thus, the inclusion of Demirjian stage classification improves the model, achieving more accurate estimates. Additionally, with the inclusion of all observations, the SEE was much higher than the one obtained in the original study [6].

In relation to the cubic regression model, it has an explanatory power, which is clearly superior to the linear regression model as a function of the I_{3M} of tooth 38, but lower than the regression including Demirjian stages as explanatory variables. The R^2 values were similar to those obtained by Balla *et al.* [20] in a sample with 1,283 OPG, age range, 7-22 years (0.72 for girls and 0.74 for boys) as well as the mean absolute error (18.36 meses for girls and 19.08 for boys).

Therefore, we advise the use of maturity index and Demirjian stages simultaneously in a regression model, restricted to non-zero indexes. Moreover, until I_{2M} equals zero, the age estimate based on the second molar is more accurate. Then, when $I_{2M} = 0$, the third molar shall be applied. However, it should only be applied until $I_{3M} = 0$, since from there, the age estimation errors significantly increase.

CONCLUSIONS

Based on the obtained results, it is possible to state that the application of the maturity index based on the second molar is reproducible in a Portuguese population (age range, 7-15 years) for medical-legal application as the corresponding maturity index is positive. Subsequently, the third molar must be only used as long as its maturity index is positive. Moreover, the inclusion of Demirjian staging in both models significantly increases the accuracy of the estimates. Hence, the provided estimates have the required precision to be used in medico-legal applications.

HIGHLIGHTS

Age estimation based on the second molar maturity index I_{2M} is accurate in a Portuguese population aged 7 to 15 years, only when this maturity index is positive.

Age estimation based on the third molar maturity index I_{3M} is accurate in a Portuguese population aged 12 to 23 years, only when this maturity index is positive.

The use of maturity index and Demirjian stages simultaneously with a regression model is more accurate.

ACKNOWLEDGMENTS

This research was supported by the Centro de Estatística e Aplicações da Universidade de Lisboa, CEAUL, FCT – Fundação para a Ciência e a Tecnologia Project reference UIDB/00006/2020.

CONFLICT OF INTEREST

The authors declare no potential conflicts of interest with respect to the research, authorship, and/or publication of this article.

References

- Kumagai A, Takahashi N, Andrea L, et al. Accuracy of the third molar index cut-off value for estimating 18 years of age: validation in a Japanese samples. *Leg Med [Internet]* 2019; 38: 5-9. Available from: <https://doi.org/10.1016/j.legalmed.2019.03.001>.
- Alves N, Azevedo A, Michel-Crosato E, Rocha M, Cameriere R, Haye Biazevic MG. Dental age estimation in a Brazilian adult population using Cameriere's method. *Braz Oral Res* 2015; 29: S1806-83242015000100215.
- Santiago B, Almeida L, Cavalcanti YW, Baraúna Magno M, Cople Maia L. 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* 2018; 132: 1167-1184.
- Apaydin BK, Yasar F. Accuracy of the Demirjian, Willems and Cameriere methods of estimating dental age on Turkish children. *Niger J Clin Pract* 2018; 21: 257-263.
- Balla S, Galic I, Karunakar P, Vanin S, De Luca S, Cameriere R. Validation of third molar maturity index (I_{3M}) for discrimination of juvenile/adult status in South Indian population. *J Forensic Leg Med* 2017; 49: 2-7.
- Cameriere R, Ferrante L, De Angelis D, Scarpino F, Galli F. The comparison between measurement of open apices of third molars and Demirjian stages to test chronological age of over 18 years old in living subjects. *Int J Legal Med* 2008; 122: 493-497.
- Guo Y, Chu G, Olze A, et al. Age estimation of Chinese children based on second molar maturity. *Int J Legal Med* 2018; 132: 807-813.
- Fins P, Pereira ML, Afonso A, Pérez-Mongiovi D, Caldas IM. Chronology of mineralization of the permanent mandibular second molar teeth and forensic age estimation. *Forensic Sci Med Pathol* 2017; 13: 272-277.
- Demirjian A, Goldstein H, Tanner JM. A new system of dental age assessment. *Hum Biol* 1973; 45: 211-227.
- Balla SB, Reddy Banda T, Galic I, Madhuri Naga N, Naishadham PP. Validation of Cameriere's third molar maturity index alone and in combination with apical maturity of permanent mandibular second molar for indicating legal age of 14 years in a sample of South Indian children. *Forensic Sci Int* 2019; 297: 243-248.
- Ranasinghe S, Perera J, Taylor J, Tennakoon A, Pallewatte A, Jayasinghe R. Dental age estimation using radiographs: towards the best method for Sri Lanka children. *Forensic Sci Int* 2019; 298: 64-70.
- Dogru H, Gulsahi A, Burçak Çehreli S, Galic I, van der Stelt P, Cameriere R, et al. Age of majority assessment in Dutch individuals based on Cameriere's third molar maturity index. *Forensic Sci Int* 2018; 282: 231.e1-231.e6.
- Mazzilli L, Melani R, Lascala CA, Velandia Palacio LA, Cameriere R, et al. Age estimation: Cameriere's open apices methodology on southeast Brazilian sample. *J Forensic Leg Med* 2018; 58: 164-168.
- Spinas E, De Luca S, Lampis L, Palacio LA, Cameriere R. Is the third molar maturity (I_{3M}) useful for a genetic isolate population? Study of a Sardinian sample of children and young adults. *Int J Legal Med* 2018; 132: 1787-1794.
- Guo Y, Chu G, Olze A, et al. Age estimation of Chinese children based on second molar maturity. *Int J Legal Med* 2018; 132: 807-813.
- Balla S, Chinni S, Galic I, et al. A cut-off value of third molar maturity index for indicating a minimum age of criminal responsibility: older or younger than 16 years? *J Forensic Leg Med* 2019; 65: 108-112.
- Tafrount C, Galic I, Franchi A, et al. Third molar maturity index for indicating the legal adult age in southeastern France. *Forensic Sci Int* 2019; 294: 218.e1-218.e6.
- Kelmendi J, Cameriere R, Koçani F, Galic I, Mehmeti B, Vodanović M, et al. The third molar maturity index in indicating the legal adult age in Kosovar population. *Int J Legal Med* 2018; 132: 1151-1159.
- Koshy S, Tandon S. Dental age assessment: the applicability of Demirjian's method in South Indian children. *Forensic Sci Int* 1998; 94: 73-85.
- Balla SB, Lingamb S, Kotrac S, et al. New regression models for dental age estimation in children using third molar maturity index: a preliminary analysis testing its usefulness as reliable age marker. *Leg Med (Tokyo)* 2019; 39: 35-40.