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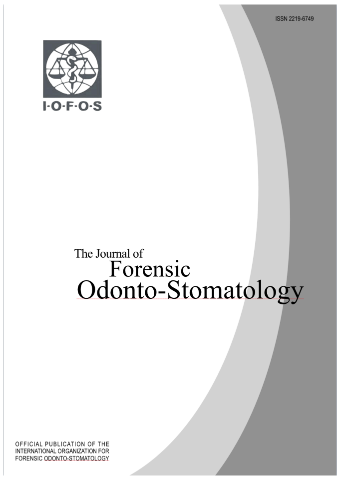

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Dental age assessment by the pulp/tooth area proportion in cone beam computed tomography: is medico-legal application for age estimation reliable?

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KEYWORDS

Forensic dentistry;
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CBCT;
Pulp/Tooth Proportion;
Kvaal's method

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ABSTRACT

Introduction: Forensic dentistry has, as one of its main goals, the identification of living and/or deceased individuals, based on the individual features of the teeth. One of the identification criteria to be established is the chronological age. Several authors, including Kvaal, have developed age estimation methods based on secondary dentine deposition. Nowadays, three-dimensional imaging tests, such as Cone Beam Computed Tomography (CBCT), are used in age estimation.
Objective: The aims of this research project were to validate Kvaal's method and its variables in age estimation and to create new linear regression formulae to better represent the study sample.
Methods: We selected 158 CBCT, with a total of 402 sound teeth (central incisors, lateral incisors and canines). The necessary measurements and ratios were calculated in both coronal and sagittal sections, with *XelisDental*®. The formulae developed by Kvaal for age estimation calculation were applied. Subsequently, the results were statistically analyzed.
Results and Discussion: The intraclass correlation coefficients from the two measurements ranged from 0.918 to 0.997. The calculated age estimation had a mean error of -21.4years (coronal section) and -26.3years (sagittal section). The *t* test revealed statistically significant differences between chronological age and estimated age. The absolute values of Pearson's correlation coefficient between age and the two Kvaal variables ranged from 0.06 to 0.38 and from 0.06 to 0.55. The coefficients of determination are lower than in the original study (between 0.03 and 0.39). In the linear regression formulae, the coefficients of determination ranged from 0.07 to 0.41.
Conclusion: This investigation concludes a non-reproducibility of Kvaal's method in the Portuguese population when applied in CBCT, with statistically significant differences between the chronological age and the dental age, estimated by the pulp/tooth proportion method, based on the teeth analyzed in this study.

INTRODUCTION

Forensic odontology is a branch of dental medicine that deals with the examination of dental evidence for further evaluation and presentation of results to the court. One aspect involves the age estimation of individuals and using it in cases related to living or dead individuals.[1]

In *post-mortem* cases, the identification is based on the characteristics present in the teeth of different individuals due to enamel's high resistance to environmental conditions.[2,3] Age estimation can also be used in *ante-mortem* criminal investigation cases, such as physical assaults or cases related to illegal immigration, due to the gradual increase of people without any identification documentation when entering European countries.[4–6] Therefore, forensic odontology plays a key role in the implementation of the United Nations' 16th Sustainable Development Objective and the Portuguese laws no. 27/2008 (updated no. 26/2014) and no. 147/99.[7–10]
In order to estimate the age of adults, several authors created methods to estimate age taking into account secondary dentine deposition and consequent reduction of pulp size.[11,12] Kvaal *et al.* developed a gender-dependent age estimation method, evaluating the pulp/tooth ratio on periapical radiographs and concluded that there are no differences between the right and left sides of the dental archs. Several authors successfully applied this method,[13,14] while others refuted it.[15,16]
Currently, complementary diagnostic tests such as CBCT can allow the visualization of oral tissues in three dimensions. Many authors used CBCT to understand whether this three-dimensional examination allows a more reliable age estimation than two-dimensional examinations [17–20], some of them applying Kvaal's method.[21,22]

AIM:

In this study, we estimated the age of a Portuguese population by dental means using the pulp/tooth ratio method initially proposed by Kvaal in the central incisors, lateral incisors and permanent canines, through CBCT.
The main aim of this investigation was to see if there were any statistically significant differences between the estimated and the chronological age of individuals.
As secondary objectives, it was intended to evaluate the correlation coefficients of the explanatory variables defined by Kvaal, when applied to this study, verifying the existence or not of statistically significant differences between the calculated and the original coefficients.
The final aim was, through CBCT analysis of tooth/pulp area proportions, to obtain a new regression formula for a more reliable calculation of age estimation with the three-dimensional method, verifying whether gender influenced age estimation.

MATERIAL AND METHODS

Experimental design
This study was conducted at a private medical dental clinic. The clinical and CBCT data of patients admitted to the Dental Medicine Clinic between 2016 and 2018 were retrospectively evaluated. The study protocol was approved by the Dental Medicine Faculty´s ethics committee.

Population sample
After the examination of medical records and CBCT data, the sample was selected based on several criteria: known gender; 21 years old or older; presence of fully visible permanent tooth of the anterior segment (central incisor, lateral incisor or canine, maxillary or mandibular), only one pulp canal, nil caries, restorations, rehabilitation, endodontic treatment, periapical/occlusal/periodontal pathology or anomalies that may cause morphological changes.
After selection, 158 CBCT where included, making a total of 402 teeth (central incisors, lateral incisors and canines, including in some cases several teeth from the same individual).

Data collection
Data from the patients referring to patient's name, gender, date of birth and date of CBCT examination was collected in *RayScan*® blindly to ensure anonymity and avoid bias.
In Kvaal's original method, a set of measurements of tooth and pulp heights and widths were calculated on periapical radiographs, in order to calculate several ratios that would allow, when applied in an equation, an estimation of the individual's age.[11]
Since three-dimensional imaging was used in the present study, measurements were recorded on coronal (C) and sagittal (S) sections, parallel to the longitudinal axis of the tooth, using the cuts in which pulp dimensions were widest. The analyzed teeth were divided into 3 different image quality groups: G0 – poor quality due to root curvature/pulp cutting; G1 – poor quality due to poor image definition; G2 – good quality.
Measurements were recorded in *XelisDental*® (as illustrated in Figure 1), using the "ruler" tool, in

order to calculate age estimation based on Kvaal's method:(11)

- Level A = tooth (At) and pulp (Ap) width at the cemento-enamel junction (CEJ) level;
- Level C = tooth (Ct) and pulp (Cp) width mid-root between CEJ and root apex;
- Level B = tooth (Bt) and pulp (Bp) width at the average distance between A and C;
- T = maximum tooth length;
- P = maximum pulp length;
- R = maximum root length.

Subsequently, the following Kvaal ratios were calculated:(11)

- Aind = Ap/At;
- Bind = Bp/Bt;
- Cind = Cp/Ct;
- RP = P/R;
- RT = T/R;
- RR = P/T;
- M = (RP+RR+Aind+Bind+Cind)/5
- W = (Bind+Cind)/2
- L = (RP+RR)/2
- Dif = W-L

Subsequently, the regression formulae defined by Kvaal were applied to the tooth groups 11/21, 12/22, 32/42 and 33/43 (dental nomenclature according to the FDI), to determine age estimation (Table 1).[11]

Figure 1. Measurements made in CBCT

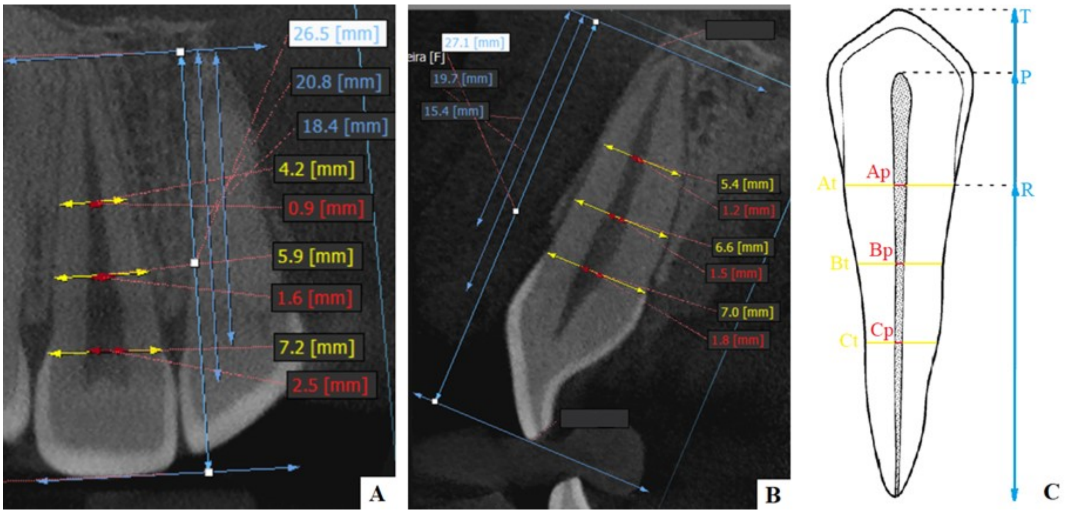

Table 1. Kvaal's regression formulae to calculate age estimation. M and Dif are the age predictors defined by Kvaal (1995). G=Gender (0 female, 1 male)

Group of teeth	Kvaal's regression formulae
11/21	Age = 110.2 – 201.4(M) – 31.3(Dif)
12/22	Age = 103.5 – 216.6(M) – 46.6(Dif)
32/42	Age = 106.6 – 251.7(M) – 61.2(Dif) – 6.0(G)
33/43	Age = 158.8 – 255.7(M)

Data validation

In order to evaluate the intra-observer variability, 10% of measurements were re-evaluated after 3 months without knowledge of the previous data. The agreement between the measurements of the observer was assessed using the Intraclass Correlation Coefficient (ICC).

Statistical data analysis

The Statistical Package for the Social Sciences (SPSS) software (version 25; IBM Corporation Armonk, NY, USA) was used for the statistical analyses. A level of significance of 5% was taken to perform all statistical tests.(23)

In the intra-observer validation, the ICC was used, based on Fleiss values.[24]
In order to confirm a normal distribution, the Kolmogorov-Smirnov test was applied (given the absolute frequency greater than 50).[23]
Student's t-test was used for paired samples to test the existence or not of statistically significant differences, between the chronological age and the dental age estimation.
Since there were different image qualities, the absolute estimation errors (AEE) were calculated for the different image quality groups: poor quality due to cutting (G0), poor quality due to poor definition (G1) and good quality (G2).
Pearson's correlation coefficient was used to measure the correlation between chronological age and the variables used by Kvaal as predictors (M and Dif) in the original study. The same predictors were evaluated based on the ANOVA linear regression model, analyzing the existence of explanatory variables and verifying whether it is sufficiently explanatory by the determination coefficient (R^2).
Finally, a multivariate linear regression was created by the ANOVA *backward method* in which only variables with *p-value*<0.1 in the Student's t test were included, using the model that best described the relationship between the dependent variable (chronological age in years) and the independent variables included.

RESULTS

Descriptive analysis

In this study, we examined 158 patients with 402 teeth between 21 and 80 years of age, where 97 (61.4%) are females and 61 (38.6%) are males. The age distribution is shown in Table 2. Regarding the analyzed teeth (N=402), 235 (58.5%) belong to female individuals and 167 (41.5%) to male. The greatest number of teeth were maxillary canines (125 teeth) and the lowest mandibular central incisors (10 teeth).

Table 2. Age distribution of the patients for each type of tooth

		Age						
		< 30	30 - 39	40 - 49	50 - 59	60 - 69	70+	Total
Tooth	11	8	5	11	7	6	0	37
	12	10	9	10	6	5	1	41
	13	14	11	22	9	8	3	67
	21	7	4	6	6	5	0	28
	22	11	5	12	4	5	1	38
	23	13	12	20	5	7	1	58
	31	2	0	2	1	1	0	6
	32	2	1	5	5	3	2	18
	33	6	3	12	12	13	3	49
	41	2	1	0	0	0	1	4
	42	3	1	3	5	0	1	13
	43	3	5	11	14	8	2	43
	Total	81	57	114	74	61	15	402

Intra-observer variability

Intra-observer variability applied to measurements recalculated from 40 teeth observed twice. The lowest ICC value was 0.918 (variable RT, coronal section) and the highest was 0.997 (both variable Cp in the coronal section and variable Bp in the sagittal section).

Age estimation by Kvaal formulae

The age estimations were made by applying the formulae deduced by Kvaal (Table 1) and subsequently verifying any significant differences between our estimation (on each section, C or S) and chronological age. Only pairs 11/21, 12/22, 32/42 and 33/43 were included in this stage, totaling 267 teeth (since Kvaal did not use the pairs 31/41 or 13/23).

When we analyze the estimation errors (EE) (Table 3), a discrepancy between the age estimations calculated by Kvaal's formulae and the chronological age is visible, with the mean of the calculated values negative, both for coronal and sagittal section. The most deviated estimations indicated a lower age than the real at about 75 years and a higher age between 24.15 (coronal) and 15.69 (sagittal) years.

Table 3. Estimation errors (EE) obtained, in years. (C=Coronal; S=Sagittal)

	EE (C)	EE (S)
Mean	-21.36	-26.28
Error Deviation	16.51	17.02
Minimum	-74.75	-75.38
Maximum	24.15	15.69

In the comparison of groups of images with different qualities, in the analysis of the good quality group, it was found that the AEE were higher than in the poor qualities (Table 4). The difference between the coronal and sagittal EE (C-S) was also evaluated for the good quality group.

It was observed, based on the means presented in Tables 3 and 4, that coronal cutting was the better in the calculation of age estimation.

Statistical test of paired sampes

In this analysis, only pairs of teeth 11/21 (N=65), 12/22 (N=79) and 33/43 (N=92) were included since the N of the group 32/42 was insufficient in number. Three analyses for each group of teeth were made: comparison 1 – relation between age estimation in coronal section and age in years; comparison 2 – relation between age estimation in sagittal section and age in years; comparison 3 – relation between age estimation in coronal section and sagittal section (Table 5).The Pearson's correlations between the variables analyzed in each pair are different from 0 (with values between 0.175 and 0.672) and all *p*-values lower than 0.05 (with only one exception occurring in teeth 33/43, coronal section and age in years).

When it comes to the *t-test*: first, it was applied to comparisons 1 and 2 to verify the existence or not of statistically significant differences between the mean age estimation (whether coronal or sagittal) and actual age; subsequently, the same test was applied to comparison 3 to assess whether there are statistical differences between the averages of the age estimations based on one section and on the other. All *p*-values were lower than 0.05, except for the comparison 3 in the 12/22 group.

Table 4. Absolute Estimation Errors (AEE), for the different groups of image quality. (G0 – poor quality due to root curvature and, consequently, pulp cutting; G1 – poor quality due to poor image definition; G2 – good quality; C=Coronal; S=Sagittal)

	AEE (C, G0)	AEE (C, G1)	AEE (C, G2)	AEE (S, G0)	AEE (S, G1)	AEE (S, G2)	Difference between EE for G2 (C-S)
N	18	83	166	32	55	180	135
Mean	17.36	21.80	23.63	24.96	23.37	27.89	2.78
Error Deviation	10.19	14.52	15.11	21.63	13.84	16.14	12.57
Minimum	2.31	0.63	0.05	0.66	0.50	0.16	-24.55
Maximum	41.42	74.75	72.32	75.38	62.88	68.67	39.12

Table 5. P-values of the Student's t test for paired samples. (comparison 1 – relation between age estimation in coronal section and real age in years; comparison 2 – relation between age estimation in sagittal section and real age in years; comparison 3 – relation between age estimation in coronal section and sagittal section).

Groups of teeth	N	Comparison 1	Comparison 2	Comparison 3
11/21	65	0.00	0.00	0.05
12/22	79	0.00	0.00	0.28
33/43	82	0.00	0.00	0.00

Pearson's correlation
As the coronal section revealed better results than the sagittal and, according to Kvaal,(11) M and Dif presented higher correlations, only the correlations of C_M and C_Dif (variables C and Dif in the coronal section) with age were analyzed with Pearson's correlation coefficient, for the entire sample and the 6 pairs of teeth separately (Table 6).
Regarding the correlation between C_M and age, all values were negative, with -0.32 (p-value < 0.05) in the entire sample and varying between -0.06 (teeth 31/41) and -0.38 (teeth 13/23). Pearson's correlation for the mandibular pairs of teeth were not statistically significant, with p-values > 0,05. When it comes to the correlation between C_Dif and age, its value for the total sample was 0.08, ranging in absolute values from 0.06 (teeth 13/23) to -0.55 (teeth 31/41). Here, only the group of teeth 32/42 showed a statistically significant value (p-value = 0.03).

Table 6. Pearson correlation coefficients, relating the chronological age with the variables M and Dif

		Age						
		Sample N=402	Teeth 11/21	Teeth 31/41	Teeth 12/22	Teeth 32/42	Teeth 13/23	Teeth 33/43
C_M	Correlation	-0.32**	-0.38**	-0.06	-0.34**	-0.24	-0.38**	-0.18
	p-value	0.00	0.00	0.87	0.00	0.20	0.00	0.09
C_Dif	Correlation	0.08	0.22	-0.55	0.19	-0.40*	0.06	0.10
	p-value	0.11	0.08	0.10	0.10	0.03	0.52	0.35

Evaluation of Kvaal's linear regression coefficients
For each group of teeth and in each section, the linear regression between age (in years) and Kvaal's explanatory variables M, Dif and G (gender) were estimated, the not significant variables were removed from the model.
As the pairs of teeth 31/41 and 32/42 have a small number of observations, only the remaining groups were evaluated (Table 7). The higher R^2 was found in the sagittal section, except for group 13/23. The highest coefficient belongs to the sagittal section of

teeth 11/21, being the lowest value in the coronal section of teeth 33/43.

Linear regression

In order to produce a formula that achieves more accurate age estimation, linear regressions between age (in years) and the remaining variables as predictors (G, RP, RT, RR, Aind, Bind and Cind) were also estimated for each group of teeth and in each section, removing all not significant variables. Table 8 presents the explanatory variables of the best model (R^2) with the respective *p-values* for the ANOVA F test and the Standard Error of the Estimate (SEE). As in Table 7, the best R^2 values are obtained in the sagittal section (except in the tooth group 13/23). The highest and lowest coefficients were observed in the sagittal section of teeth 11/21 and in the coronal section of teeth 33/43, respectively. The obtained linear regression formulae are presented in the Table 9.

Table 7. Evaluation of Kvaal variables for selected teeth, in both cuts, in years. (R^2=determination coefficient; Standard Error of the Estimate=SEE)

Groups of teeth	Predictive variables	N	R^2	SEE
11/21	C_M	65	0.15	12.64
	S_M	65	0.39	10.63
12/22	C_M	79	0.11	13.41
	S_M, S_Dif	79	0.24	12.47
13/23	C_M, C_Dif	125	0.17	12.60
	S_M	125	0.11	13.01
33/43	C_M	92	0.03	13.88
	G, S_M, S_Dif	92	0.15	13.14

Table 8. Predictive variables of the best model, with the p-values, R^2 and SEE. (R^2=determination coefficient; Standard Error of the Estimate=SEE)

Groups of teeth	Predictive variables	N	*p-value*	R^2	SEE
11/21C	C_RR, C_RT, C_RP	65	0.01	0.18	12.55
11/21S	S_Cind, S_RP	65	0.00	0.41	10.57
12/22C	C_Cind, C_RR, C_Aind, C_Bind	79	0.00	0.23	12.74
12/22S	S_RP, S_Aind, S_RT, S_RR	79	0.00	0.30	12.12
13/23C	C_Cind, C_RP	125	0.00	0.17	12.59
13/23S	S_Bind, S_RR	125	0.00	0.12	12.98
33/43C	G, C_Aind	92	0.04	0.07	13.65
33/43S	G, S_Bind, S_RR	92	0.00	0.16	13.06

Table 9. Linear regression formulae created, based on the best models

Group of teeth	Linear regression formulae
$11/21C_{RR,RT,RP}$	- 484.166 - 526.419(C_RP) + 363.717(C_RT) + 764.473(C_RR)
$11/21S_{Cind,RP}$	122.790 - 45.001(S_RP) - 93.512(S_Cind)
$12/22C_{Cind,RR,Aind,Bind}$	84.756 - 41.578(C_RR) - 54.754(C_Aind) + 76.288(C_Bind) - 69.786(C_Cind)
$12/22S_{RP,Aind,RT,RR}$	- 471.280 - 443.654(S_RP) + 326.338(S_RT) + 726.234(S_RR) - 75.834(S_Aind)
$13/23C_{Cind,RP}$	90.821 - 26.662(C_RP) - 85.065(C_Cind)
$13/23S_{Bind,RR}$	82.781 - 37.746(S_RR) - 43.978(S_Bind)
$33/43C_{G,Aind}$	63.380 - 5.723(G) - 45.449(C_Aind)
$33/43S_{G,Bind,RR}$	90.037 - 4.852(G) - 33.203(S_RR) - 43.764(S_Bind)

DISCUSSION

Intra-observer variability

In the intra-observer validation, no relationship was demonstrated between a specific section and a higher ICC value. Based on the presented values and on the Fleiss intervals, all ICC demonstrated an excellent level of agreement.(24) This calibration is in agreement with the existing literature: Li *et al*. (ICC between 0.837 and 0.855) and Erbudak *et al*. (highest ICC between 0.95 and 0.99).[25,26] It is important to note that this only demonstrates a high precision and not exactly a high accuracy.

Age estimation by Kvaal formulae

Since the original article, applied to periapical radiographs, argues that there are no significant differences between the right and left sides of the arches, all teeth were grouped with those of the respective teeth on the contra-lateral side .(11) Additionally, since Kvaal only developed linear regression formulae to calculate the age estimation in specific pairs of teeth, only pairs of teeth 11/21, 12/22, 32/42 and 33/43 were analyzed.

The discrepancy between age estimation and chronological age indicates that estimations are, on average, lower than actual ages, by about 21 years in the coronal section and 26 years in the sagittal section (Table 3), These findings are similar to those of Mittal *et al*. and Kanchan-Talreja.(5,27) Li *et al*. presented higher estimations.(25) The wide interval between the minimum and maximum error values shown in Table 3 (difference of 100 and 90 years, in the coronal and sagittal sections, respectively) reveal a large margin of error in calculating the age estimation.

In what extent the different image qualities (Table 4), G2 presented not only higher AEE values than the overall average, but also the highest absolute estimation errors of the 3 image quality groups, refuting the idea that the best image quality would represent a better age estimation. Also, the fact that the average difference between the EE (C-S) of G2 was low does not indicate that the values are close to the actual ones, but rather that the estimations obtained from the different sections are close. Hence, there seems to be little difference between sections to the calculation of the age estimation.

Statistical test of paired samples

The Kolmogorov-Smirnov's test confirmed that the sample followed a normal distribution. Also, for statistical purposes, the pair of teeth 32/42 was not included due to the low number available.

Pearson's correlations between the variables of each analyzed pair indicate a correlation between the variables, from weakest (0.18) to strongest

(0.67), and a *p-value* lower than 0.05 indicate that almost all correlations between estimated and chronological age were statistically significant, although far from the unit that denotes the perfect correlation.

Regarding the t test of paired samples for comparisons 1 and 2, due to the *p-value lower than* 0.05, we can affirm that there are statistically significant differences between the age estimation mean (either coronal or sagittal) and chronological age mean in years, in all groups of teeth. Thus, these results seem to reveal that this estimation methodology is biased. In comparison 3, the *p-value lower than* 0,05 obtained for teeth 11/21 and 33/43 indicates the existence of statistically significant differences. However, for the 12/22 group, the *p-value* was 0.28. Hence, no statistically significant differences were revealed between the age estimated by coronal or by sagittal section in this group of teeth (Table 5). Thus, it is unclear whether there are differences, on average, between the estimations based on the two sections.

Kvaal presented differences in age estimation and actual age between 8.6 and 11.5 years.[11] In the literature, several studies supported Kvaal's method, showing no statistically significant differences between estimated and actual age – like Paewinsky *et al.* and Bosmans *et al.*, with an EE of 6.68 and between 0.37 and 7.21 years, respectively.[13,14,27–29] Although, some studies refuted this, presenting statistically significant differences between the age estimation and the actual age – like Kanchan-Talreja *et al.* and Meinl *et al.*, with an EE of 18.1 years and between 31.4 and 47.1 years, respectively. Erbudak *et al.* showed the most closely resemble the differences observed in this study, with differences between 12.17 and 25.1 years.[5,15,25,26,30] Gopal *et al.* compared Kvaal's method to Cameriere's, concluding that the second presents better results of age estimation.[12,31]

Pearson's correlation coefficient

Pearson's correlation coefficients between C_M and age presented only negative values, indicating a negative correlation. The values ranged from -0.06 (group 31/41) to -0.38 (group 13/23), revealing despicable to weak correlations. Regarding the variable C_Dif and age, correlation values went, considering absolutes values, from 0.06 (group 13/23) to -0.55 (group 31/41), resulting in despicable to moderate correlations. Table 6 emphasizes that only 5 coefficients are statistically significant (*p-value* < 0.05). All observed coefficients were lower than in the original study based on periapical radiographs (Table 10), concluding that the variables defined as predictors by Kvaal do not show a good correlation with age in this study based on CBCT.

Table 10. Pearson correlation coefficients from this study and original study (p-value<0.05)

Variable	Teeth group	Kvaal's coefficients	Study's coefficients
C_M (M)	11/21	-0.83*	-0.38*
	12/22	-0.80*	-0.34*
	32/42	-0.71*	-0.24
	33/43	-0.75*	-0.18
C_Dif (W-L)	11/21	0.66*	0.22
	12/22	0.54*	0.19
	32/42	0.30*	-0.40*
	33/43	0.35*	0.10

When observing the minimum and maximum values described in the literature (Table 11), a similarity of the values obtained in this study is visible. Yet, studies that applied Cameriere's method in CBCT presented higher coefficients when compared to Kvaal's method in CBCT, supporting the idea that the variables analyzed by Cameriere *et al.* produced better correlations with age.(19–22)

Evaluation of Kvaal's linear regression coefficients

The groups of teeth 31/41 and 32/42 were excluded due to their small number in the sample under analysis. When applying the predictor variables of Kvaal to groups with higher N (Table 7), and considering the sections, there was an R^2 between 0.03 (group 33/43) and 0.39 (group 11/21). These values are much lower than Kvaal's original study (between 0.56 and 0.76).(11)

Several studies presented lower coefficients than those determined in this study – Sharma *et al.* and Li *et al.*, with R^2 between 0.011-0.198 and between 0.01-0.23, respectively.[25,32] Numerous articles showed higher values – Mittal *et al.* (between 0.240-0.453), Maini *et al.* (largest value of 0.517) and Akay *et al.* (values between 0.162 and 0.550 on CBCT).(22,27,29) Gopal *et al.*, once again, supported the idea that Cameriere's method presented better results, through its higher R^2 values (between 0.833 and 0.935).[31] Chandramala *et al.* presented results more similar to ours, with values between 0.014 and 0.385.[28]

As expected, the smaller SEE is found in the groups with a higher R^2 and vice versa. The SEE in this study ranged from 10.63 to 13.88 years (Table 7), which were higher than those described in the literature, including the range defined by Solheim *et al.* as acceptable for forensic application (10 years).[27,31,33] Li *et al.* presented similar SEE, between 11.4 and 12.9.[25]

Table 11. Pearson's correlation coefficients (minimum and maximum, in absolute terms) described in the literature

	Studies	**Pearson's coefficients**	
		Minimum	**Maximum**
Periapicais	Sharma *et al.* 34	-0.01	-0.44
	Gopal *et al.* 33	-0.008	0.951
Panoramic	Erbudak *et al.* 28	-0.187	0.36
	Chandramala *et al.* 30	-0.07	0.54
	Mittal *et al.* 29	0.238	0.68
	Maini *et al.* 31	-0.01	-0.493
	Roh *et al.* 32	-0.11	-0.7
	Li *et al.* 27	-0.119	-0.518
CBCT	Penaloza *et al.* 23	-0.21	-0.65
	Akay *et al.* 24	-0.117	-0.551
CBCT Applying Cameriere's method	Afify *et al.* 21	-0.829	-0.959
	Haghandifar *et al.* 22	-0.330	0.764

Linear regression

All models used can explain part of the age variation, since *p-value* < 0.05 in the ANOVA F test in all cases (Table 8). Comparing the R^2 values of the original study with the values determined in this study (both for Kvaal predictor variables and for the new variables) (Table 12), all original values were higher than in this study (not only of the variables M and Dif, but also of the new established ones). However, and considering only this study values, R^2 of the new variables (between 0.07 and 0.41) is higher

than that obtained with predictors defined by Kvaal (0.031 to 0.394), which may indicate a greater representativeness of the sample with the new variables, although the difference is always small.
The existing literature supports our values – Paewisnky *et al.* with an R^2=0.839 and Kanchan-Talreja *et al.*, Erbudak *et al.* and Roh *et al.* with R^2 between 0.11-0.44, 0.035-0.345 and 0.14-0.49, respectively.[5,13,26,30] Regarding studies in CBCT: the ones that applied Kvaal's method reveal similarities with our study, like Penaloza *et al.* (R^2 between 0.01 and 0.56); Yang *et al.*, on its pulp/tooth volume ratio, presented a lower value (R^2=0.29); the ones that applied Cameriere's method obtained not only similar values (Haghandifar *et al.* presented a R^2 between 0.109 and 0.583), but also higher values (Afify *et al.* had an interval between 0.687 and 0.919).[18–21]
When it comes to SEE (Table 12), the original study had the lowest values. Regarding this study SEE, the SEE of the new variables were slightly lower than the SEE obtained with the variables defined by Kvaal.
Despite having null mean estimation error, the estimations obtained by linear regression tend to underestimate in the lowest age groups and to overestimate in the highest age groups in all analysed models regardless of the section (coronal or sagittal) or the group of teeth used. Thus, the extreme age groups, namely the oldest, tend to have higher estimation errors. Nevertheless, there is no clear standard that distinguishes the precision of the estimations obtained by each group of teeth.
Kanchan-Talreja *et al.*, Penaloza *et al.* and Roh *et al.* stipulated SEE values from 11 to 14, 10.58 to 15.4 and 10.4 to 14.2 years, all higher than the values defined by Solheim.[5,21,30,33] The articles that applied Cameriere's method in CBCT were the only ones that presented SEE within the interval established by Solheim. Haghandifar *et al.* established an interval between 7.1 and 10.5 and Afify *et al.* presented the lowest SEE interval, between 4.76 and 8.1 years.[19,20]

Table 12. Comparative table between Kvaal's variables in the original study, Kvaal's variables in this study and the new variables

Teeth	Kvaal's R^2	Study's R^2 Kvaal's variables	Study's R^2	Kvaal's SEE	Study's SEE Kvaal's variables	Study's SEE
11/21C	0.70	0.15	0.18	9.50	12.64	12.55
11/21S		0.39	0.41		10.63	10.57
12/22C	0.67	0.11	0.23	10.0	13.41	12.74
12/22S		0.24	0.30		12.47	12.12
13/23C		0.17	0.17		12.60	12.59
13/23S		0.11	0.12		13.01	12.98
33/43C	0.56	0.03	0.07	11.5	13.88	13.65
33/43S		0.15	0.16		13.14	13.06

CONCLUSIONS

Based on the obtained results, we can conclude that there are statistically significant differences between the chronological age and the dental age of individuals, estimated by the pulp/tooth ratio method evaluated on CBCT, based on any teeth analyzed in this study. In addition, there are statistically significant differences between the correlation coefficients of the explanatory variables defined by Kvaal on periapical radiographs and the correlation coefficients of the same variables calculated in this study on CBCT, that revealed much lower values. A regression formula was created with the aim to estimate age more accurately with the three-dimensional method, through CBCT analysis of the proportions of the tooth/pulp area – however, without practical application due the reduced explanatory capacity revealed.

For forensic age estimation, it is especially important to ensure the applicability of an unbiased method with high accuracy and precision, which can achieve reliable age estimations. The age estimation according to this dental biological parameter in the usual imaging diagnosis examination CBCT has limitations to be applied. Enlargement of the database with further multi-centre studies can help to improve the model.

In conclusion, Kvaal's formulae for age estimation is not reproducible in the Portuguese population, when applied in CBCT.

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Professional liability: assessment of court sentences for lawsuits against dentists in Peru

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ABSTRACT

Introduction: The number of reported dental malpractice cases has increased in recent years. The aim of this study was to analyze the characteristics of Peruvian court sentences related to dental procedures.

Materials and methods: In the present descriptive study, 33 sentences issued by the civil court of Peru, from 2011 to 2016 were collected. Useful information from the sentences was extracted and analyzed using the SPSS 18 software.

Results: Data showed that dentists were found guilty in 84.8% of sentences due to absence of suitability in dental treatment. Male dentists (61.1%) were involved in more cases than female dentists. Prosthodontics (33.3%) was the dental specialty subject to most claims.

Conclusions: Dentists like other health professionals are regulated by legal rules in the country they practise. As part of dental practice and in order to avoid claims, having a full clinical history and informed consent should be mandatory.

INTRODUCTION

A suitable term for medical malpractice is "an act of a medical professional deviating from the set regulations and standards that result in injury or damage to a patient".[1] In recent years, claims against dental malpractice have risen. Dentistry is associated with specialist treatments and high costs due to the predominantly private relationship between the professional and the patient who has greater expectations of the results. Thereby, if the dentist fails to reach these expectations, it can result in lawsuits concerning professional liability.[2] These dental malpractice cases can be resolved in civil law involving sometimes financial implications, but in extreme cases criminal procedures may be conducted by criminal law with sanctions such as imprisonment or fine.[3] In general terms, negligent procedures include the lack of scientific qualification to perform a medical act, the violation of guidelines of therapeutic activity and the harm or injury directly caused by the dentist's negligent act for which compensation is claimed.[4] Professionals must act with expertise, prudence, and diligence, carrying out risk management before, during and after the procedure.[5]

In Peru, The General Law of Health (N° 26842) establishes that health professionals are responsible for damages caused to the patient by the negligent, imprudent and inexpert exercise of their activities (Article 36) and the health establishment is jointly liable for damages caused to patients (Article 48). For imposing a sanction, there will be taken into account: a) Damages that have occurred or may occur in patient's health; b) The seriousness of the offence; and, c) The condition of reiteration of the offender (Article 135).[6] Additionally, The Peruvian Civil Code establishes that those who do not perform their obligations due to wilful intent, inexcusable fault or slight fault are subject to compensation for damages (Article 1321). As for moral damage, it is also susceptible to compensation (Article 1322).[7] Furthermore, the practise of dentistry is ruled by the Code of Ethics and Deontology, which constitutes the set of standards that govern the disciplines that regulate and supervise the dental profession, establising the incompatibilities, limitations and prohibitions to the surgeon dentist who practises the profession in the country.[8] The purpose of this study was to assess and describe the characteristics of Peruvian sentences associated with the performance of dental procedures.

MATERIAL AND METHODS

This descriptive study was done in Lima (Peru) using 33 court sentences concerning dental treatment performed by surgeon dentists in Peru. Sentences were collected from the data base of the application vLEx (only for the Peruvian jurisdiction) using the following key words: sentence, dentist, consumer protection. All of the sentences were issued from 2011 to 2016.
From the 33 sentences, the following information was taken:

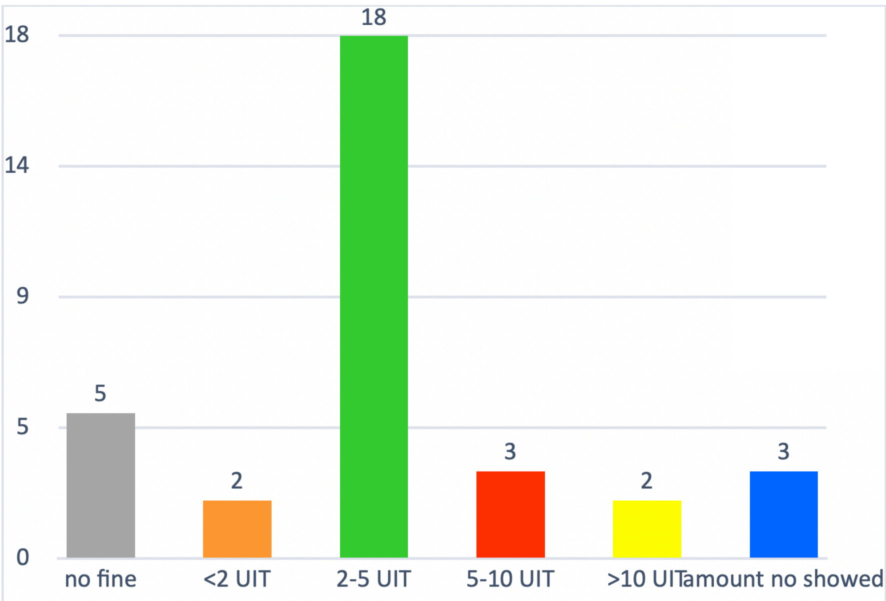
- Year of the sentence.
- Gender of the plaintiff.
- Gender of the dentist.
- Type of accused: if it was only the dentist, only the dental centre or the combination of the dentist and dental centre.
- Result of the sentence: if there was or was no suitability, status of limitation or if the court ruling is not conclusive.
- Amount of the indemnification (if any). Peruvian system of indemnifications is fixed in Levy Tax Unit (UITs in Spanish). Among 2011 and 2016, 1 UIT was (on average) approximately 1120 dollars.
- Specialty in dentistry involved.

The statistical analysis was performed using SPSS 18 software. Data were analysed by the chi squared test and statistical significance was defined at $P < 0.05$.

RESULTS

Data obtained from the 33 sentences showed that the years with the largest number of sentences was the period 2013-2014 (54.5%) with 18 sentences. Regarding the type of accused, professionals and dental centres have the largest number of sentences (48.5% for each one).
Most of the sentences (84.8%) found no suitability in the dental treatment, meaning that the accused was found guilty. The amount of indemnification was issued in 25 sentences and most of them fluctuate between 2-5 UIT (54.5%). The largest indemnification was equal to 15 UIT for a malpractice case in implantology. Sentences which found suitability in the treatment, status of limitation or when the court ruling was not conclusive have no fine (Figure 1).

Figure 1. Amount of indemnifications awarded in sentences



The dental specialty most involved in the sentences, was prosthodontics (33.3%), followed by orthodontics (27.3%), oral and maxilofacial surgery (9.1%), implantology (9.1%), other specialties (9.1%), endodontics and more than 2 specialties (6.1% each one) (Figure 2).

Regarding the gender of the accused, the largest percentage was for males (61.1%). However, there was no significant difference between male and female dentists (p>0.05) (Table 1). On the other hand, most of the patient claimants were female (66.7%).

Figure 2. Fields of dentistry involved in sentences

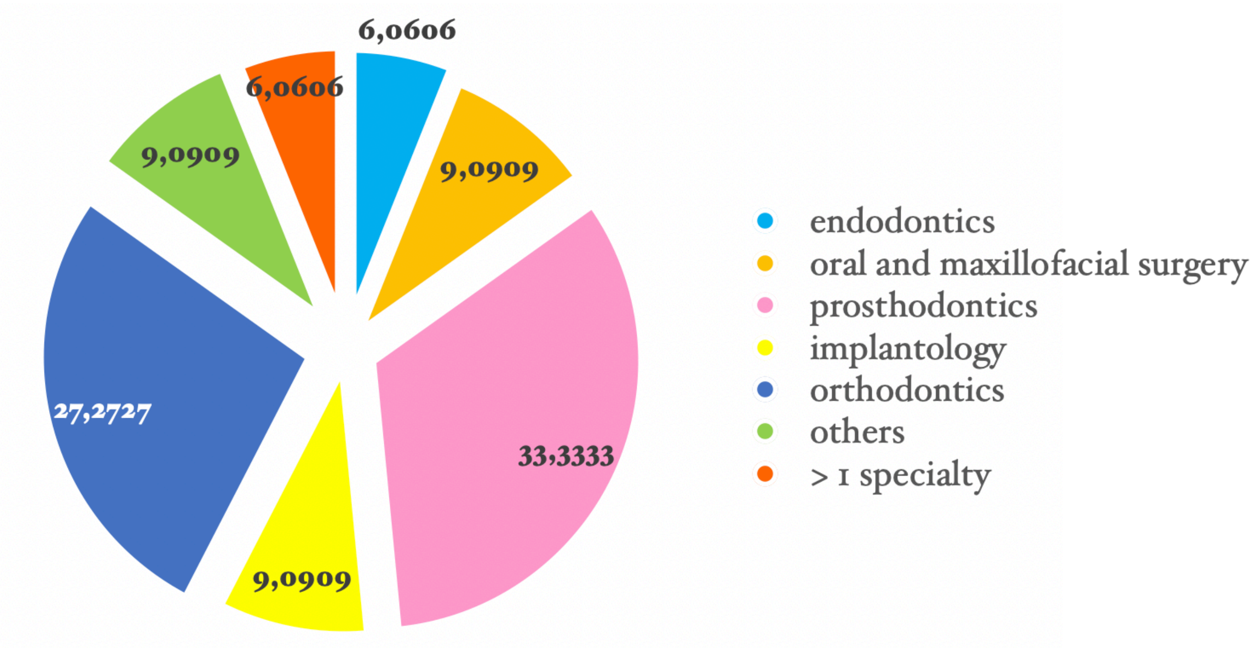


Table 1. Gender of dentist regarding type of accused

		Type of accused			
		Dentist	Dentist + DC	DC	Total
Female	Number	5	2	-	7
	%	31,3%	100%	-	38,9%
Male	Number	11	0	-	11
	%	68,8%	0%	-	61,1%

DC: Dental Centre. p=0.06

DISCUSSION

Primarily, it is very important to indicate that the literature regarding sentences involving dental treatment is limited. In addition, the access to the information was very difficult because in Peru documents of this kind are not stored in any data base; consequently, the authors had to use vLex application and even using this source the sample was restricted.

By analyzing the gender of plaintiffs, this study shows that 66.7% of all the cases were female. Other studies agree that in relation to the profile of the plaintiffs there is a predominance of women, as in the studies by Zanin et al.[9] (74%), Montagna et al.[10] (68%), Knaak et al.[11] (56%) and Marinescu Gava et al.[12] (74%). This higher number of submitted claims is explained by women's greater general interest in dental health and use of services.[13] Data from the current study show regarding the gender of the professional and the type of accused, there is no significant difference between male and female dentists (p>0.05). However, the largest percentage was for males (61.1%). Similarly, Perea-Pérez et al. found that 84.1% of all the professionals subjected to complaints or lawsuits concerning oral surgery were male.[14] Also, in the study conducted by Hashemipour et al., most complaints involved males (90.6%) and there was a significant relationship between the gender of dentists and the number of complaints (p= 0.001).[15] According to studies by Pinchi et al.[16] and Givol et al.[17] female patients are more likely to lodge complaints against treatment by male dentists, which suggest that male dentists are more likely to become involved in litigation than female dentists. The present study suggests that the relationship between male dentist/female patients is more likely to result in complaints. Results which point out that female dentists are less likely to undergo malpractice lawsuits. According to several authors this could be due to female dental practitioners tend to practise more communication that can be considered patient centred, which can minimize the probability of litigation.[18,19]

This study shows that all of the sentences (100%) involved dental treatment performed in private dental centres. In the literarure, several authors agreed with our results, for instance Perea-Pérez et al. found in 85.7% of all the cases that the care was provided in a dental clinic.[14] A study conducted by Ozdemir et al. showed that in most of the cases (81.8%) treatment was carried out in private dental practice.[20] As for the the branch of dentistry most involved in sentences, prosthodontics was claimed in 33.3% of all the cases. In practically all data the same trends are found, Kiani et al. reported that the majority of clinical complaints involved fixed prosthodontics (27.8%) followed by oral surgery (23.5%), endodontics (16.6%), periodontics (2.5%) and operative treatment (13%).[21] Also, Manca et al. found that prosthodontics was the most claimed specialty, accounting for over 44% of all cases (204 of 464).[22] This information coincides with the study published by Nassani who indicated that the available evidence suggested that prosthodontics may come at the top of dental specialties in terms of inciting patient complaints and filing of dental claims.[23] There is current evidence in the literature which point out estimations about failure in prosthodontic treatments; for instance Pjetursson et al.[24] have estimated that over one-fifth of prosthodontic cases fail after 10 years service and according to Saunders et al.[25] one-fifth of teeth prepared for full coverage crowns become non-vital within five years of placement. These results suggest taking into account other options for preserving teeth such as the use of adhesive materials in the practice of minimally invasive dentistry.[26]

As result of the sentences, in 84.8% of the cases there was no suitability in the dental treatment, consequently, the dentist was found guilty. Other studies agree that in most of the cases, the dentist is found guilty, as Hashemipour et al.[15] (56.7%) and Manca et al.[22] (74%). However, Thavarajah et al. reported a lower number of dentists pronounced guilty, 39.63% of litigations decided in favour of patients.[27]

In this study, implantology was the specialty with the highest indemnification issued (15 UIT). According to the amount of indemnifications, Perea-Pérez et al. reported that sentences involving implantology surgery had the greater amount of indemnifications.[14] Nevertheless, Thavarajah et al.[27] found that procedures involving oral surgery are often associated with high compensations and Kiani et al.[21] showed a case which involved a general dentist who worked as a prosthodontist with the most expensive compensation.

In the sentences analyzed in this study we did not find information about the operator's specilization which theoretically should reduce errors in dental treatment with a decrease in complaints from patients. Moreover, dentists should recognize their own limits and make timely referrals when they are needed. Agreeing with this statement, Hiivala et al. showed that moderate to severe harmful patient safety incidents were caused by overconfidence performing complicated treatment such as fixed prosthetics, implants or surgical procedures.[28] However, Vehkalahti et al. found no statistical differences between specialized and general dentists (p=0.963) in cases of endodontic treatment.[29]

Although the sentences assessed did not show information about dental records, research points out that a very important issue to take into account is the absence of relevant information in dental records, which should be emphasized because such a lack of information causes problems for dentists during their defence in malpractice cases.[30, 31] Hence, it is important to remember the statement of "poor records mean poor defence, no records mean no defence".[32]

Finally, we consider that data in the scientific literature should force us, as health professionals, to be mindful of having a full clinical history and before performing any dental procedure we must have the appropriate informed consent signed by the patient. This conclusion was in agreement with Marei who concluded that during the consenting process verbal and written communication are essential.[33] In addition, cases of malpractice incidents should be used for feedback to instruct dental practitioners.

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Can root pulp visibility in mandibular first molars be used as an alternative age marker at the 16 year threshold in the absence of mandibular third molars: an orthopantomographic study in a South Indian sample

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KEYWORDS

Dental age estimation;
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ABSTRACT

In many countries, the 16 years of age threshold is considered to be legally relevant according to the law. This research aims to ascertain the sensitivity and specificity of Olze et al. stages of root pulp visibility (RPV) in a sample of 760 south Indian children aged between 12 and 20 years, with an age threshold of 16 years, using receiver operating characteristic curves and area under the curve (AUC). Spearman's rho correlation showed a strong positive correlation between the RPV stages and age. No significant difference between the right and left lower first molars was seen. RPV Stage 2 showed the highest AUC in both females (0.813) and males (0.790). The performance of the RPV Stage 2 to discriminate the legal age threshold of 16 years resulted in the sensitivity, specificity and accuracy values of 0.61, 0.96 and 0.77 in males, 0.65, 0.97 and 0.80 in females. It resulted in 3.6% and 2.9% of false positives and 38.5% and 34.5% of false negatives in both sexes. Even though, RPV Stage 2 can discriminate reasonably well between two age categories, due to the high percentage of false negatives we recommend its use in conjunction with other age estimation methods.

INTRODUCTION

Juvenile crime/ delinquency is a complex problem that continues to have a toll on our society. It involves wrong doing by a child or a young person who is under an age specified by the law. The minimum age of criminal responsibility indicates the lowest age at which children are held responsible for their alleged criminal acts. It has been applied asymmetrically by different countries hugely ranging from as low as 6 up to 18 years of age.[1] The age threshold of 16 years is relevant in many countries like India, China, Maldives, Sri Lanka, Belgium, France, Netherlands, Argentina, UAE etc. It represents the age at which the criminal law is applied to juveniles (minimum of 16 years but less than 18 years) who are accused of heinous crimes and therefore treated as adults.[2, 3] Often sports organizations seek the help of medical and dental experts to estimate the age of players in the 16 year old threshold.[4] Therefore, age estimation methods that have shown reasonable accuracy should be applied to assign an age that the trial or sentencing of the accused will be conducted in the adult or juvenile justice courts.

Currently, age estimation methods are based on the projection radiography of different skeletal elements such as hand and wrist, clavicle, knee and teeth.[5-10] Depending on the question to be answered, these methods may be more or less useful and reliable.[11] Age estimation using teeth is an accurate, reliable, minimally invasive, well known and widely used historic procedure.[12, 13] For many years, prediction of the legal age thresholds in children and sub-adults was done using the mineralization and maturation of the third molars.[14-19] In 2010, Olze et al. studied regressive changes in lower third molars with completed root formation in younger age groups. They introduced a 4 stage classification based on the radiographic visibility of the root pulp. The mean age and standard deviation at each stage was used to calculate the likelihood of attaining 18 and 21 years of age.[20] Studies in different racial groups confirmed the radiographic pulp examination as a reliable method and a valuable contribution in forensic age assessment.[21-24]

The present study is a continuation of previous research[25] that tested the radiographic visibility of the root pulp in the lower first molars for prediction of the attainment of adulthood. As in the previous research, the present study aimed at determining the accuracy of Olze et al[20] classification of root pulp visibility in lower first molars for estimating another key age of medico-legal importance i.e., 16 years. The accuracy of this stage classification was evaluated by Receiver operating characteristic (ROC) analysis, which combines the sensitivity and specificity in a single accuracy measure.

MATERIAL AND METHODS

Sample

A total of 760 orthopantomograms (OPGs) from 380 male and 380 female south Indian subjects with ages ranging 12 to 20 years were collected retrospectively from the digital archives of the Department of Radiology and private dental clinics. Table 1 shows the age and sex distribution of the sample. Ethical clearance was obtained from the Institutional Ethics Committee to conduct the study. The need for obtaining informed consent was waived due to retrospective nature of the study. OPGs belonging to individuals of south Indian origin (confirmed using national identification card), with good diagnostic quality, with intact right and left mandibular first molars were included in the study. OPGs with caries, fillings, endodontically treated first molars, positional anomalies, molars with incomplete root formation or single roots were excluded from the study. Each OPG was assigned a unique identification number (UIN). Details of each individual such as sex, age, UIN, date of birth and date of radiography for each OPG were recorded separately in a Microsoft Excel file. Chronological age of each subject was calculated by subtracting the date of birth from the date of exposure of the radiograph.

Table 1. Age and sex distribution of the sample

Age groups	Males	Females	Total
12-12.9	40	40	80
13-13.9	40	40	80
14-14.9	50	50	100
15-15.9	50	50	100
16-16.9	50	50	100
17-17.9	50	50	100
18-18.9	50	50	100
19-19.9	50	50	100
Total	380	380	760

Method

The proportion of obliteration of the root pulp in lower first molars with completed root formation was assessed using the staging system described by Olze et al.[20] Figure 1 shows schematic illustrations of the stage classifications and typical radiographs.

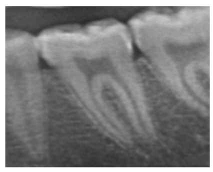
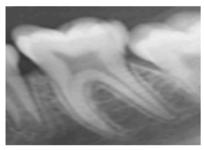
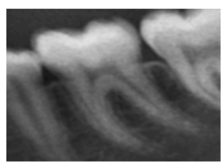
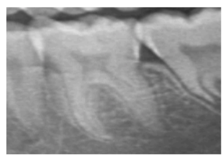
All OPGs were analyzed a by single examiner, who is a forensic odontologist with 6 years of experience in evaluating radiographic images and in age estimation. The examiner was blinded for subjects age and sex, classified the lower first molars according to the stages of root pulp visibility. To explore the intra- and inter-examiner agreement, 100 OPGs were randomly selected and re-evaluated after three months of the first evaluation.

Statistical analysis

Data was analyzed in IBM SPSS version 20.0 (SPSS Inc., Chicago, IL, USA) with a significance level set at 5% ($p<0.05$). Cohen's kappa statistics were performed to calculate the intra- and inter-examiner agreement. Descriptive statistical analysis of the data was expressed in mean, standard deviation (SD), median with lower and upper quartiles, and minimum and maximum of the root pulp visibility of lower first molars considering the 4 stage classification. Correlation between the age and root pulp visibility in lower first molars was evaluated using Spearman rank order correlation (rho). Chi- square test was used to observe the association between age as equal or above 16 years and stages of root pulp visibility. The performance of the stages of pulp grading was tested by 2×2 contingency table. It generally displays the number of participants who are true positives, true negatives, false positives and false negatives.[26] The performance of each stage was assessed using accurate classification, sensitivity, specificity, positive (LR+) and negative (LR-) likelihood ratios. In the present context, the sensitivity represents the rate of subjects correctly classified as ≥16 years old, while specificity represents the rate of subjects correctly classified as < 16 years. Likelihood ratios combine the sensitivity and specificity into a single value that indicates which cut-off is best in discriminating the age threshold. Values of LR+ > 1, i.e., LR+ between 2 to 5 present small, 5 to 10 present moderate and above 10 present large and often conclusive increases the likelihood of subject to be older than 16 years. LR- between 0.2 and 0.5 present small, 0.2 and 0.1 present moderate, while under 0.1 present large and often conclusive decreases in the likelihood of age being above 16 years.[27]

In the present study, the diagnostic accuracy of each threshold (stages of root pulp visibility) was characterized by calculating diagnostic sensitivity and specificity, which then combined in the ROC plot and by quantifying the areas under the ROC curves (AUC). ROC curve represents a graphical plot which illustrates the performance of a binary classifier system.[28] AUC represents an approximate measurement of the diagnostic accuracy of a quantity. It ranges from 0 to 1, where a value of 0 indicates a perfectly inaccurate test, while a value of 1 perfectly accurate test.[28] An AUC of 0.5 represents no discrimination, 0.7 to 0.8 considered acceptable, 0.8 to 0.9 is excellent and more than 0.9 is considered outstanding.[29]

Figure 1. Schematic representation of root pulp visualization stages proposed by Olze et al., 2010

Pulp visualization stages	Original Drawings of Olze et al.	Example X- rays for different stages in study population
STAGE 0		
STAGE 1		
STAGE 2		
STAGE 3		

RESULTS

Kappa statistics revealed the values of 0.87 for intra-examiner and 0.8 for inter-examiner, indicating substantial to almost perfect agreements (Table 2). Table 3 listed the reasons for the exclusion of OPGs that include insufficient image quality or fused roots (1.97% cases) and due to the presence of three rooted molars or multiple root canals (0.65% cases). Total analysed radiographs accounted up to 97.3%. Spearman's rho correlation showed a strong, positive and statistically significant correlation between the stages and the age for both sexes i.e., 0.823 ($p<0.05$) and 0.804 ($p<0.05$) for males and females, respectively.

To test the variation between the lower first molars of right and left quadrants, analyses were performed separately for both teeth. In table 4 and 5, descriptive data of each stage of root pulp visibility is displayed for both females and males of both lower first molars. Table 6 and 7 display the proportion of the subjects under and over 16 year age threshold. Figure 2 shows the distribution of the sample according to the chronological age (in years) for different stages of the root pulp visibility in both lower first molars (FDI, 36 & 46).

Table 2. Kappa statistics of the intra- and inter-examiner agreement

	Kappa value	95% CI		Reliability
		Lower	Upper	
Intra-examiner	0.874	0.832	0.904	Substantial
Inter-examiner	0.803	0.776	0.839	Substantial

CI; Confidence Interval

Table 3. Number of evaluated teeth and the teeth that could not be reliably assessed

Tooth	Sex	Number of cases	Evaluated teeth	Insufficient quality image/ fused roots	Three rooted molars/ multiple canals
36	Male	380	368	8	4
	Female	380	372	7	1
46	Male	380	368	9	3
	Female	380	375	5	0

Table 4. Descriptive statistics of chronological age according to sex and stage of root pulp visibility in tooth 36

	Stage	N	Min	Max	LQ	Median	UQ	Mean	SD
Females	0	74	12.04	14.84	12.31	13.52	14.41	13.43	0.9
	1	162	12.06	19.93	14.53	15.67	16.65	15.56	1.5
	2	128	13.54	19.98	17.7	18.42	19.18	18.33	1.1
	3	8	16.26	19.55	17.9	18.8	19.29	18.47	1.06
Males	0	99	12	16.65	12.8	13.79	14.67	13.77	1.1
	1	140	12.07	19.62	15.21	16.08	16.98	15.96	1.4
	2	121	13.29	19.91	17.81	18.61	19.2	18.44	1.1
	3	8	18.59	19.81	18.75	19.42	19.77	19.32	0.5

N, Number; Min, Minimum; Max, Maximum; LQ, Lower quartile; UQ, Upper quartile; SD, Standard deviation.

Table 5. Descriptive statistics of chronological age according to sex and stage of root pulp visibility in tooth 46

	Stage	N	Min	Max	LQ	Median	UQ	Mean	SD
Females	0	85	12.01	17.27	12.31	13.57	14.41	13.47	1.07
	1	138	12.06	19.6	14.52	15.73	16.69	15.58	1.5
	2	142	13.54	19.98	17.3	18.35	19.17	18.08	1.3
	3	10	16.15	19.53	17.99	18.67	18.95	18.37	0.9
Males	0	87	12	15.41	12.69	13.41	14.39	13.55	0.9
	1	122	12.07	19.62	15.01	15.95	16.95	15.84	1.5
	2	154	13.29	19.91	17.25	18.44	19.1	18.03	1.3
	3	05	18.59	19.78	18.9	19.61	19.77	19.39	0.5

N, Number; Min, Minimum; Max, Maximum; LQ, Lower quartile; UQ, Upper quartile; SD, Standard deviation.

Table 6. Stage distribution according to age threshold 16 years for tooth 36

	Stage	Total (N)	<16 Years		>16 years	
			n	*Prop*	*n*	*Prop*
Female	0	74	74	1	0	0
	1	162	93	0.574	69	0.426
	2	128	5	0.390	123	0.961
	3	08	0	0	08	1
Male	0	99	93	0.939	06	0.611
	1	140	69	0.493	71	0.507
	2	121	06	0.5	115	0.95
	3	08	0	0	08	1

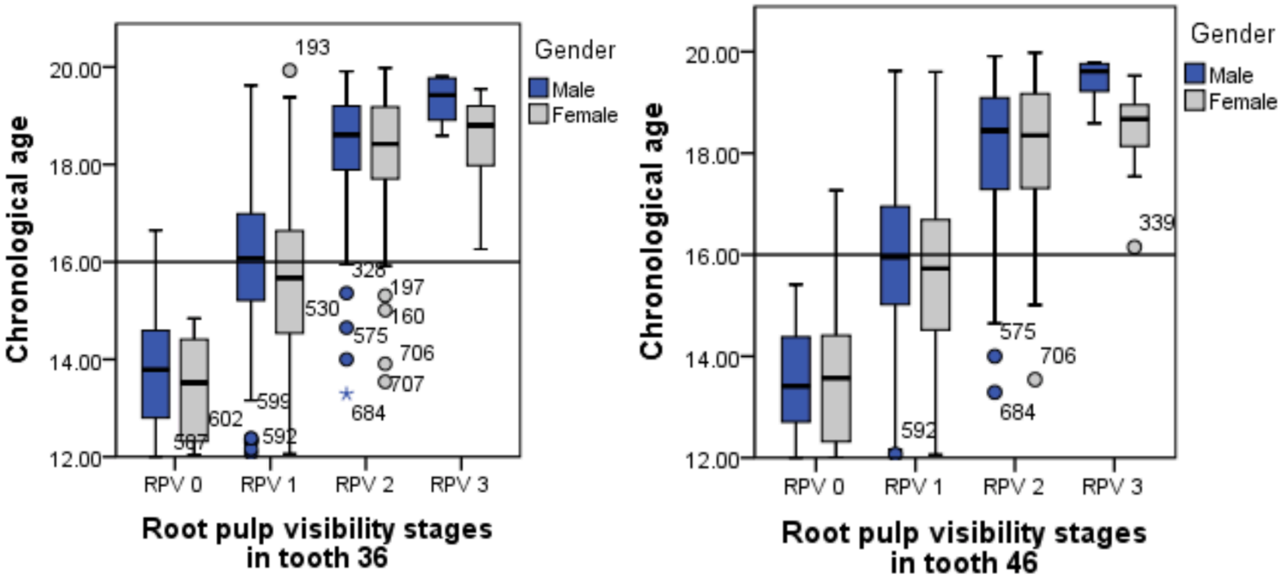
Prop; Proportion

Table 7. Stage distribution according to age threshold 16 years for tooth 46

	Stage	Total (N)	<16 Years		≥16 years	
			n	*Prop*	*n*	*Prop*
Female	0	85	84	0.988	1	0.012
	1	138	77	0.558	61	0.442
	2	142	14	0.099	128	0.901
	3	10	0	0	10	1
Male	0	87	87	1	0	0
	1	122	62	0.508	60	0.492
	2	154	19	0.123	135	0.877
	3	05	0	0	05	1

Prop; Proportion

Figure 2: Box and whisker plots of the stages of root pulp visibility for males and females for tooth 36 (left) and tooth 46 (right)



ROC curves were drawn to evaluate the discriminatory ability of the 4 stages and to determine the optimum cut-offs. Figure 3 and 4 show the graphs for females and males for both teeth with respect to the 16 year age threshold. Based on the findings, it was found that the stage 2 of root pulp visibility marked as the optimum cut-off for the 16 year old threshold in both sexes. Table 8 and 9 show the performance measures of the stage 2 root pulp visibility for both sexes in the lower first molars (FDI, 36 & 46). In females, for the lower left first molar (FDI, 36) the values of AUC, sensitivity, specificity, LR+, LR- and accuracy were 0.813, 0.65, 0.97, 22.53, 0.36 and 0.80, respectively. In males these values were 0.790, 0.61, 0.96, 17.22, 0.4 and 0.77, respectively. LR+ value of 22.53 and 17.22 in females and males. This indicates that when the stage 2 of root pulp visibility was scored then a female is 23 times and a male is 17 times more likely to be over than the under 16 years of age threshold. LR- value of 0.36 and 0.4 in females and males indicate that when the stage 2 of pulp visibility was not attained, then both the sexes are approximately 10 times more likely to be under than over 16 years of age. On the other hand, for the lower right first molar (FDI, 46), the values AUC, sensitivity, specificity, LR+, LR- and accuracy 0.805, 0.72, 0.92, 9.02, 0.3 and 0.81 in females and 0.793, 0.70, 0.89, 6.19, 0.34 and 0.79 in males, respectively.

Figure 3. ROC Curves for root pulp visibility stage 2 for 16 year old threshold in females and males for tooth 36

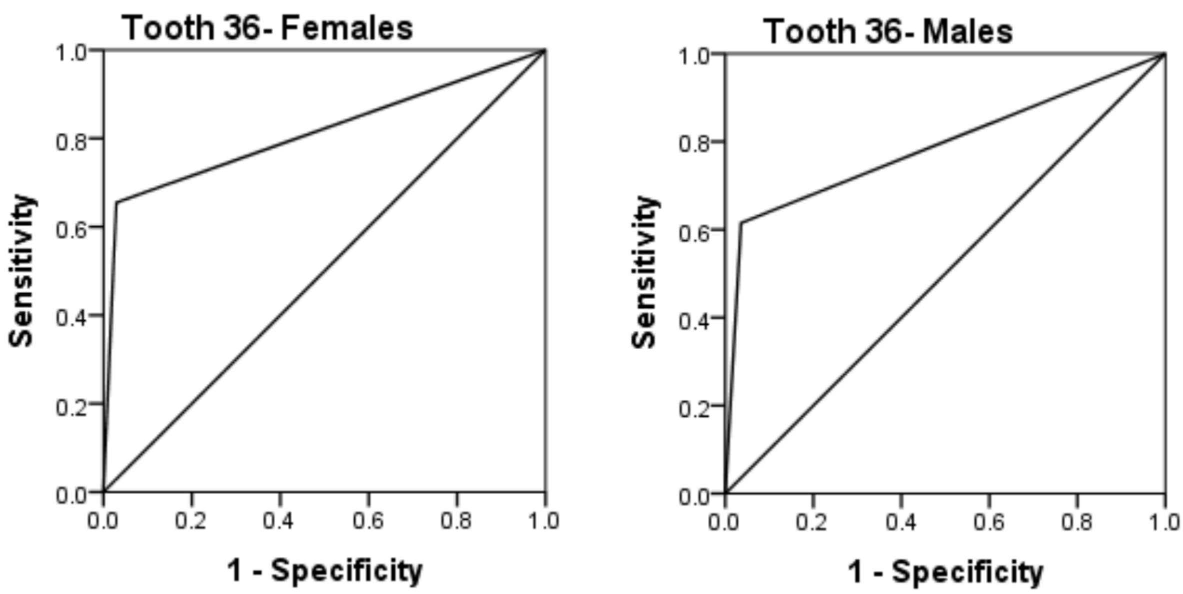
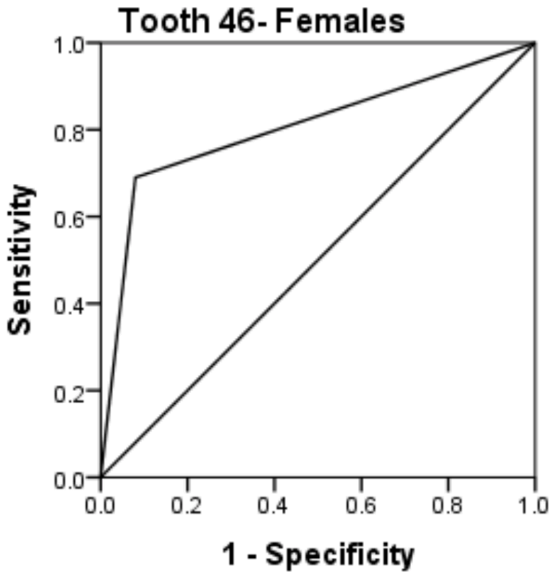


Figure 4. ROC Curves for root pulp visibility stage 2 for 16 year old threshold in females and males for tooth 46



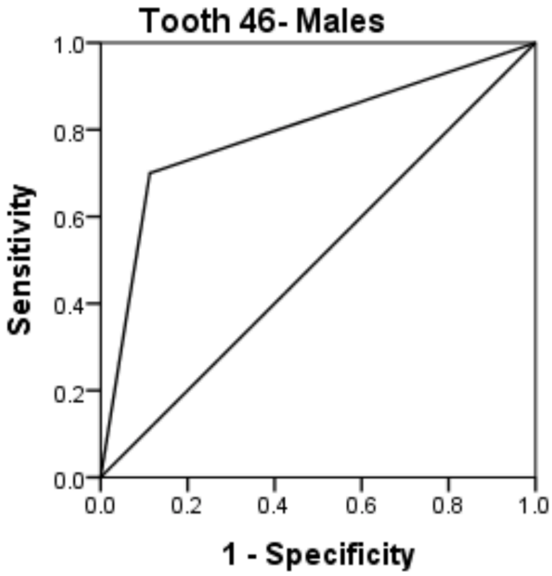


Table 8. Performance measures of pulp visibility stage 2 for legal age threshold over 16 years using tooth 36

Measures	Females	Males
Pulp visibility stage 2		
AUC	0.813 (0.768- 0.858)	0.790 (0.743- 0.837)
Sensitivity	0.65 (0.58- 0.72)	0.61 (0.54- 0.68)
Specificity	0.97 (0.93- 0.99)	0.96 (0.92- 0.99)
LR+	22.53 (9.44- 53.76)	17.22 (7.79- 38.07)
LR-	0.36 (0.29- 0.43)	0.4 (0.33- 0.48)
Accuracy	0.80 (0.76- 0.84)	0.77 (0.73- 0.82)

Table 9. Performance measures of pulp visibility stage 2 for legal age threshold over 16 years using tooth 46

Measures	Females	Males
Pulp visibility stage 2		
AUC	0.805 (0.759- 0.851)	0.793 (0.746- 0.841)
Sensitivity	0.72 (0.66- 0.78)	0.70 (0.63- 0.76)
Specificity	0.92 (0.87- 0.96)	0.89 (0.83- 0.93)
LR+	9.02 (5.42- 15.01)	6.19 (4.01- 9.54)
LR-	0.3 (0.24- 0.38)	0.34 (0.27- 0.42)
Accuracy	0.81 (0.77- 0.85)	0.79 (0.74- 0.83)

DISCUSSION

Diagnostic tests play a significant role in modern medicine and have become a critical feature of standard medical practice.[30] Sensitivity and specificity are the measures of the intrinsic diagnostic accuracy, known to be independent of the prevalence of the condition.[31] In the present investigation, we set out to find the optimum cut-off value (i.e., stage of root pulp visibility in lower first molars) that combines the highest possible specificity (important in criminal context) and the highest obtainable sensitivity (percentage of true negatives) for discriminating whether a subject is ≥16 years.

Originally, Olze et al[20] studied the root pulp visibility in the lower third molars for forensic age estimation purposes. However, in the present study we tested this method in the lower first molars. There are two important reasons why the authors in the present study chose to evaluate the lower first molars; firstly, the increased frequency of missing third molars and secondly, the presence of third molars with incomplete root formation. It is reported that the prevalence of third molar agenesis is high and is ranging up to 28%.[32] Timme et al[33] in their study reported higher number of missing third molars (46 to 60%) indicating it as a main limitation of this method. According to the original method,[20] third molars with incomplete roots (not attained Demirjian stage H) could not be included for evaluation. In the present study, when the OPGs of subjects aged 12 - 20 years were evaluated; a very high percentage of third molars (>50%) with incomplete mineralization were seen, indicating that this methodology[20] could not be applied to predict the legal age threshold of 16 years using third molars. Therefore, for the reliability of our results and based on the previous study findings,[25] we decided to study the radiographic visibility of root pulp in lower first molars.

The Study Group on Forensic Age Diagnostics (AGFAD) of the German society of Legal Medicine, recommended the use of multiple methods in combination, for optimal accuracy.[34] The ideal age estimation method is a constant quest for forensic experts. Every method exhibits variations in their accuracy, sensitivity and specificity especially when considered at different age thresholds.[35] Very few authors in the past have tested the age threshold of 16 years. Table 10 summarized the list of studies and their outcome measures. The authors have analysed the maturity of the third molars alone,[17] maturity of the second and third molars combined,[3, 16] Demirjian's staging system for second and third molars combined,[36] and for all lower seven teeth combined[37] and radiographic examination of root pulp visibility in the lower first molars (present study). Cameriere et al[16] analysed the apical maturity of second and third molars and reported sensitivity and specificity values of 0.79 and 0.81. When Wang et al[3] analysed the same parameters in a southern Chinese sample, similar sensitivity value of 0.78 was reported, however, better specificity value of 0.97 was observed. When the maturity of third molars alone was assessed, sensitivity and specificity values of 0.91 and 0.85 in males and 0.9 and 0.87 in females, were obtained in a south Indian sample.[17] Cardoso et al[36] examined the second and third molars separately, and in combination using the Demirjian staging system. They reported a sensitivity of 0.86 and specificity of 0.77 when the combination of second and third molars were assessed. In the present investigation, we have obtained optimum sensitivity values (0.61 and 0.65) and better specificity values (0.96 and 0.97) in males and females, respectively. Pinchi et al[37] tested the usefulness of Demirjian and Willem age estimation methods for the assessment of the attainment of the 14 years threshold in Italian children. Both methods have resulted in higher sensitivity and lower specificity values. These differences in sensitivity and specificity values might be related to the parameters tested and the existing developmental variations between the populations.

Table 10. Data from previously published studies with different cut-off values tested in various populations for predicting the legal age threshold of 16 years

Author (Year)	Parameters assessed	Accuracy	Sensitivity	Specificity	AUC
Pinchi et al (2012)[37]	Maturation of all mandibular teeth excluding third molars	---	D: 0.80 W: 0.95	D: 0.61 W: 0.86	---
Cameriere et al (2018)[16]	Combination of second (I2M) and third molar (I3M) maturity indices	0.80	0.79	0.81	0.890
Cardoso et al (2018)[36]	Demirjian staging system for 2nd & 3rd molars combined	0.82	0.86	0.77	0.926
SB Balla et al (2019)[17]	Third molar maturity index (I3M)	M: 0.88 F: 0.89	M: 0.91 F: 0.90	M: 0.86 F: 0.87	0.936
Wang et al (2020)[3]	Combination of second (I2M) and third molar (I3M) maturity indices	0.88	0.78	0.97	0.803 (I2M) 0.945 (I3M)
Present study (2020)	Olze et al., stages of Root pulp visibility in lower first molars	M: 0.77 F: 0.80	M: 0.61 F: 0.65	M: 0.96 F: 0.97	M: 0.790 F: 0.813

AUC, Area under the curve; I2M, Second molar maturity index; I3M, Third molar maturity index; D, Demirjian's method; W, Willems method

For each threshold, there is a combination of sensitivity and specificity which are then combined in the ROC plot, resulting in the AUC that indicates the performance of the test.[38] In the present investigation, the AUC for stage 2 root pulp visibility was 0.813 for females and 0.790 in males indicating that a randomly selected individual (either male or female) from the older age category (≥16 years) will have a greater grade of root pulp visibility (stage 2 and above) compared to a randomly chosen individual from the younger age category (<16 years) approximately 80% of the time. This indicates that diagnosing age at least 16 years from root pulp visibility stages can discriminate reasonably well between two age categories. Similarly, every threshold has the likelihood ratios of a positive (LR+) and negative (LR-) test result to express the probability of a diagnostic test result.[36] In the present study, the test result of a stage 2 root pulp visibility is more than 23 times in a female and 17 times in a male more likely to occur in an individual at least 16 years otherwise to someone younger than 16. Similarly, LR- of approximately 0.4 in both sexes indicates that an individual marked with early stages of root pulp visibility (stage 0 and 1) is one in ten more likely to be seen if age is at least 16 years than otherwise i.e., if age is in younger category.

Our study also provides the results for the error in discriminating minors (<16 years) and accountable juveniles (≥16 years). When stage 2 root pulp visibility was used as an age marker for 16 years, it resulted in ethically unacceptable (false positive) and technically unacceptable (false negative) errors. The highest error rates were false negatives i.e., 38.5% of males and 34.5% of females who were aged equal or above 16 years was classified as below 16 years. Misclassification, which is important from the medico-legal point of view (false positives) were kept to minimum i.e., 3.6% of males and 2.9% of females.

Each age estimation method must be overviewed in conjunction with its advantages and disadvantages. The advantages of this method are; firstly, it can be applied in subjects without third molars. Secondly, high specificity values were obtained in both sexes, which is extremely important in the criminal law context, which expresses the rate of false positives.[39] Even though, Wang et al[3] obtained higher specificity values using the combination of second and third molar maturity index values, they reported that this methodology could not be used in case the third molars were missing. One possible disadvantage in the present study is the lower sensitivity values in both sexes. However, it is less damaging to classify an individual as being younger than 16 years, when they are not, than classifying a minor as an accountable juvenile.[40] Although this study provides some indication of the quality age predictive model i.e., reasonably well discrimination between two age categories using stage 2 of root pulp visibility in lower first molars, we advise the combination of methods due to the high percentage of false negatives.

CONCLUSIONS

After examining a sample of south Indian children aged between 12 and 20 years using ROC curves, we found that the stage 2 of root pulp visibility can be used to predict the age of 16 years or over. In particular, this study suggests that dental age assessment can be done with reasonable accuracy, especially the prediction of the 16 year old age threshold using teeth other than the third molars. However, due to the high percentage of false negatives, we advise the use of this method in conjunction with other methods. Further research should be done to validate the use of root pulp visibility in teeth other than molars for prediction of the attainment of legal age thresholds in children and sub-adults.

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Dental age assessment based on the radiographic visibility of the periodontal ligament in lower third molars in a Thai sample

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ABSTRACT

The objective of the present study was to analyze the radiographic visibility of the periodontal ligament in completed root formation lower third molars in a sample of lower northern Thai population. Digital panoramic images from 800 patients with ages ranging from 16 to 26 years were used in this study. The visibility status of the periodontal ligament of lower third molars with completed root formation including apical closure was assessed. For each stage, the minimum age, maximum age, median, mean, and standard deviation were calculated. The minimum age found in stage 0 was 16.17 years in males and 17.00 years in females. Stage 1 was first achieved at the age of 16.17 years in males and 17.08 years in females. The earliest onset of stage 2 was 17.00 years in males and 18.17 years in females. The incidence of stage 3 was first observed at 19.17 years in males and 18.83 years in females. It may be concluded that the radiographic visibility of the periodontal ligament in lower third molars may be a useful approach in the dental age assessment in a Thai population. In case the periodontal ligament visibility is found to be in stage 2, it may be confirmed that the individual is at least 18 years of age.

INTRODUCTION

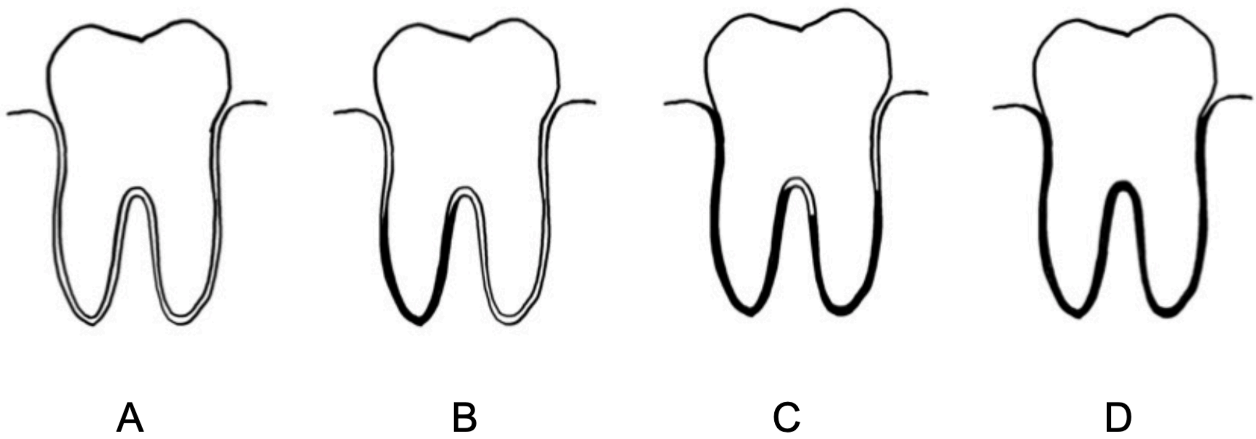
Forensic age assessment is a significant task in chronological age prediction for legal reasons, including immigration, individual identification, and sentencing in most jurisdictions.[1] A new guideline on best practice for age assessment was published as a result of multi-partner cooperation between the Ministry of Justice, the Ministry of Solidarity and Health, the Ministry of the Interior and the Ministry of Territorial Cohesion and relations with local and regional authorities.[2] In Thailand, the lawfully considered age limits are 10, 13, 15, 18, and 20 years. The international and multi-disciplinary study group on forensic age diagnostics (AGFAD) suggests chronological age evaluation processes involving three independent factors, including a physical examination, radiographic examination of the hand, and a dental examination. If skeletal development of the hand is completed, further radiography or CT examination of the clavicle's sternal extremity should be carried out.[3] The main indications for assessing chronological age based on teeth are the tooth mineralization stages.[4] In Thailand, there were studies on the root mineralization of third molars. These studies reported that mineralization of lower third molar roots could be completed over the age of 18.[5] However, it is still difficult to prove beyond reasonable doubt that a person is over 18 years of age. It would be advantageous to find a dental method after the third molar root formation was completed. Periodontal ligament (PDL) is the tooth supporting structure that is composed of strips of fibrous connective tissue and attaches tooth cementum to the alveolar bone. Periodontal ligament width could be changed because of age or tooth function. Olze et al.[6] first described the radiographic visibility of the periodontal ligament in the completed root formation of lower third molars for forensic age estimation in a German population. In addition, there are papers based on the radiographic visibility of the periodontal ligament in northern Chinese[1], Portuguese[7], and UK-Caucasian[8] populations. These previous studies described this method as a potential age estimation criterion after completed root formation. The present study was conducted to analyze the periodontal ligament visibility of completed root formed lower third molars in a sample of panoramic radiographs from Thailand and compare with the other ethnic samples.

MATERIAL AND METHODS

The institutional ethics committee approved the present study (IRB No. P10068/63). Digital panoramic images from 800 patients with ages ranging from 16 to 26 years were used in this study. The panoramic radiographs were taken between 2018 and 2020.

The visibility status of the periodontal ligament of left and right lower third molars with completed root formation including apical closure was assessed using the following four stages described by Olze et al.[6] (Figure 1):

Figure 1. Schematic drawings of the radiographic visibility of periodontal ligament in lower third molars; (A) stage 0 (B) stage 1 (C) stage 2 (D) stage 3



Stage 0: The PDL is visible along the full length of all roots.

Stage 1: The PDL is invisible in one root from the apex to more than half of the root.

Stage 2: The PDL is invisible along almost the full length of one root or part of the root in two roots or both.

Stage 3: The PDL is invisible along almost the full length of two roots.

All of the digital panoramic radiographs were evaluated by an oral radiologist, with more than 10 years of experience. The lower third molars with fused roots or narrowed furcation were not determined. The patient's age, sex, and stage of radiographic visibility of the PDL were recorded using Microsoft Excel tables. Statistical analysis was performed using SPSS Statistics version 17. Each patient's age was the difference between the date of panoramic radiographic examination and the date of birth and recorded as years and 1/100 of years. The minimum age, maximum age, median, mean, and standard deviation were calculated for each stage.

The inter-observer agreement was assessed by the random selection of 100 panoramic radiographs and examined by two examiners. To assess intra-observer agreement, the same 100 radiographs were evaluated by the first examiner after a 1-month interval. Kappa statistics were performed to calculate the intra- and inter-observer agreement

RESULTS

A total of 746 digital panoramic images (299 males and 447 females), aged 16 to 26, were recruited for the present study (Table 1).

Table 1. Age distribution of the subjects

Age (years)	Sex		Total
	Male	Female	
16	3	0	3
17	32	22	54
18	28	29	57
19	54	100	154
20	29	96	125
21	43	60	103
22	20	49	69
23	20	23	43
24	25	25	50
25	25	26	51
26	20	17	37
Total	299	447	746

There were 54 panoramic radiographs (7.24%) that could not be assessed because of fused roots or narrowed furcation. The minimum age, maximum age, median, mean, and standard deviation of radiographic visibility of the periodontal ligament for tooth 38 and 48 in both sexes were shown in Table 2. The minimum age found in stage 0 was 16.17 years in males and 17.00 years in females. Stage 1 was first achieved at the age of 16.17 years in males and 17.08 years in females. The earliest onset of stage 2 was 17.00 years in males and 18.17 years in females. The incidence of stage 3 was first observed at 19.17 years in males and 18.83 years in females.

The medians were varied in each stage. For stage 0, it ranged from 18.75 to 19.46 years. The medians of stage 1 varied between 20.33 and 20.58 years. For stage 2, the medians were between 21.42 and 22.92 years. The medians of stage 3 were between 20.58 and 23.83 years. Regarding the mean age of the stages of periodontal ligament visibility on panoramic radiographs of teeth 38 and 48, it was not statistically significant different both in males and females (Table 3). Table 4 demonstrated the distribution of periodontal ligament visibility stages on panoramic radiographs between adults and minors.

Table 2. data for the stages of periodontal ligament visibility on panoramic radiographs of teeth 38 and 48 in males and females

	Tooth	Stage	n	Min	Max	Median	Mean	SD
Males	38	0	51	16.42	25.00	18.75	18.94	1.62
		1	107	16.17	26.67	20.58	21.04	2.23
		2	90	17.00	26.92	22.92	22.83	2.28
		3	11	20.33	26.92	23.42	23.39	2.56
	48	0	45	16.17	24.00	18.83	19.09	1.59
		1	114	16.42	26.67	20.58	21.13	2.40
		2	94	18.83	26.92	21.92	22.78	2.44
		3	16	19.17	26.92	23.83	23.28	2.80
Females	38	0	70	17.00	26.92	19.46	19.72	1.91
		1	180	17.08	26.92	20.46	21.00	2.10
		2	132	18.58	26.92	21.42	21.98	2.15
		3	6	19.67	26.50	20.46	21.40	2.54
	48	0	67	17.00	26.17	19.42	19.83	1.98
		1	164	17.08	26.92	20.33	20.86	1.93
		2	145	18.17	26.92	21.67	22.26	2.43
		3	6	18.83	24.33	21.29	21.36	2.00

Table 3. Data on the mean age of the stages of periodontal ligament visibility on panoramic radiographs of teeth 38 and 48 in males and females

		Mean age		*p* Value
		Tooth 38	Tooth 48	
Males Stage	0	18.94	19.09	0.66
	1	21.04	21.13	0.77
	2	22.83	22.78	0.89
	3	23.39	23.28	0.92
Females Stage	0	19.72	19.83	0.73
	1	21.00	20.86	0.54
	2	21.98	22.26	0.30
	3	21.40	21.36	0.97

Table 4. Distribution of periodontal ligament visibility stages on panoramic radiographs between adults and minors

	Tooth	Stage	n	< 18 Years	≥ 18 Years
Males	38	0	51	16	35
		1	107	6	101
		2	90	1	89
		3	11	0	11
	48	0	45	12	33
		1	114	10	104
		2	94	0	94
		3	16	0	16
Females	38	0	70	12	58
		1	180	9	171
		2	132	0	132
		3	6	0	6
	48	0	67	15	52
		1	164	6	158
		2	145	0	145
		3	6	0	6

The intra- and inter-observer agreement were 0.874 and 0.739, respectively. These values are considered to be almost perfect for intra-observer agreement and substantial for inter-observer agreement.

DISCUSSION

According to the civil and criminal laws, 18 years of age is of legal relevance in Thailand. We analyzed the radiographic visibility of the periodontal ligament in lower third molars from a sample of the Thai population to determine if this methodology could be used to exclude subjects under 18 years of age. There are some methods to estimate the chronological age, including assessing the development of third molars on panoramic radiographs[4] and assessing clavicular ossification.[9] However, not many forensic odontologists are familiar with the clavicular assessment. Therefore, only a few studies have been published focusing on age estimation based on clavicular epiphysis.[10, 11] According to the dental age estimation study based on third molar development from panoramic radiographs in the Thai population, the root formation is completed around 22 years. However, in many cases, the root formation is completed before the age of 18 years.[4] Therefore, the alternative method is necessary to assess whether the victim or the suspect has reached 18 years of age.

Our investigation showed that from stage 2, almost all individuals were over 18 years of age, which is not in line with previous studies. Olze et al.[6] proposed the periodontal ligament visibility status in the lower third molars as a characteristic for dental age estimation after complete mineralization of the root. Their study included 1198 panoramic radiographs from Germans with ages between 15 and 40 years, the minimum age for stages 1-3 was over 18 years of age. In 2014, Sequeira et al.[7] studied 487 panoramic images of Portuguese subjects aged between 17 and 31 years. They concluded that stage 3 could be used in estimating males over 21 years of age, whereas for females, other techniques were suggested. Guo et al.[1] studied the radiographic visibility of the periodontal ligament in lower third molars of a Chinese population. They concluded that if stage 1 was determined, it was possible to prove that an individual was already 18 years of age. Timme et al.[12] assessed 2346 panoramic images in Germans and proved that stages 1 and 2 can be used to explain that individuals are older than 18 and 21 years of age, respectively.

However, our study's result was in accordance with Lucas et al.[8], who studied a UK Caucasian cohort of 2000 subjects with ages between 16 and 25 years. The authors renamed stages 0, 1, 2, and 3 to A, B, C, and D to better represent the data's categorical nature. They concluded that both in males and females, the presence of stages C and D could help accurately determine whether an individual was over 18 years.

The authors evaluated 746 radiographs of a Thai population selected from the age of 16 to 26 years, which covered the period of third molar root formation completion. The results showed that the minimum and the median ages of the individuals increased with the stages, except for stage 3 in females, which may be caused by the small number of cases. We found stage 2 first appeared at the age around 18.15 years, which is similar to that of Portuguese[7], but about 5 years younger than that of Chinese[1] and German populations.[12] The mean age of the 4 stages of periodontal ligament visibility on panoramic radiographs of teeth 38 and 48 were not statistically significant different both in males and females. Even distribution of the number of subjects in each age group may be helpful in the significance of these parameters.

The subjects in the present study were selected based on age, without the other factors such as the socio-economic status. It may be that the difference in the minimum age of different populations were caused by different origins. In 2020, Guo et al.[13] first developed a new stage classification without considering the periodontal ligament visibility status between the roots of the lower third molars, which could not be classified in many cases because of fused roots or narrowed furcation. This new criterion can be effectively used in forensic age estimation. In the present study, we found that the visibility status of 54 panoramic radiographs (7.24%) could not be classified to any stage because of fused roots or narrowed furcation, which could be considered as one of the limitations of the study. The dental age assessment using radiographs is a valid method of forensic age estimation.[14] Further study should be performed to determine whether the new classification by Guo et al is also applicable to a Thai population. According to the inter-observer error results, it can be concluded that dental age estimation should only be performed by properly trained personnel.[13]

CONCLUSIONS

Based on the results of the present study, radiographic visibility of the periodontal ligament in lower third molars may be a useful approach in dental age assessment in the Thai population. In case the periodontal ligament visibility is found to be in stage 2, it may be confirmed that the individual is at least 18 years of age. Further studies are needed to prove the relationship between ethnicity and the stage of periodontal ligament visibility on panoramic radiographs.

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Accuracy of Kvaal's radiographic and translucent dentinal root techniques of extracted teeth in Malay adults for dental age estimation

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KEYWORDS

Dental age estimation, Dentinal root translucency, Kvaal's radiographic method, Bang-Ramm method

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ABSTRACT

The use of teeth to estimate the age of unknown bodies provides much help especially in skeletal remains with no soft tissues left for identification. However, dental age estimation utilizing degenerative changes in teeth like dentinal translucency is often hampered with large margin of error. This study aims to compare the accuracy of Kvaal's radiographic method (intraoral periapical radiograph) with modified Bang-Ramm dentinal root translucency method in estimating dental age in Malay adults. One-hundred teeth of maxillary and mandibular incisors and canine were collected following dental extraction. Date of birth, date of extraction, gender and ethnicity were documented prior to extraction. All teeth were assessed using two methods of dental age estimation: 1) The equation from Kvaal's radiographic method and 2) Formula from modified Bang-Ramm dentinal root translucency method. The results from the age estimation were compared to the chronological age of the persons from whom the teeth were extracted. The average dental age estimated using both methods significantly correlated with the chronological age for both men and women. Overestimation and underestimation with mean absolute error up to 13 years and 15 years was observed in modified Bang-Ramm and Kvaal, respectively. The estimated age calculated from both methods also showed increasing standard deviation as the patient gets older. From the obtained results it is reasonable to conclude that modified Bang-Ramm method gives better accuracy for dental age estimation in Malay adults.

INTRODUCTION

According to the National Institute of Forensic Medicine Malaysia, hundreds of people die as unknown persons each year. Age assessment is an important forensic procedure in profiling the identity of these unclaimed body remains. The use of teeth to estimate the age of unknown bodies provides much help especially in skeletal remains with no soft tissues left for identification.

Dental development is complete when an individual is reaching the age of 18 to 23 years old. After this stage, dental age estimation can only be relied on degenerative changes that occur especially for early developed teeth such as permanent central incisors of maxilla and mandible. Dentin translucency in root portion is one of the major degenerative changes seen with increasing age. Root translucency develops as the dentinal tubules within a tooth root begin to mineralize from the root apex toward the crown. [1]{Kinney, 2005 #4}{Kinney, 2005 #4}{Kinney, 2005 #4}{Kinney, 2005 #4}{Kinney, 2005 #4}{Kinney, 2005 #8193}{Kinney, 2005 #8193}{Kinney, 2005 #8193}{Kinney, 2005 #8193}{Kinney, 2005 #8193}{Kinney, 2005 #8193} Many approaches have been suggested for age estimation by means of direct and indirect observation of teeth with the use of microscope or even the science of histology. [2] One of the most simple and reliable of Gustafson's criteria for age estimation is by the measurement of root dentin translucency. [3] It starts in the apical part of root and increases with age in the coronal direction. [4] Following the study by Bang and Ramm [4], modifications had been made by Acharya based on more sample size in Indian population [2]. New linear and quadratic equations to estimate age in adults had been proposed and tested. However, none of the studies utilized Malaysian adults for validation. In addition, the particular change of dentin translucency is least affected by environmental factors and the pathological process [5, 6] and therefore, may provide a concrete dental age estimation marker in estimating the age of an adult. It also shows symmetrical distribution on both sides of jaws. [7, 8] In 1995, Kvaal et al., developed a radiographic method using intraoral periapical radiograph to estimate dental age in aging individuals using several quantifiable parameters on different monoradicular teeth. [9] Among others, this method involved several simple mathematical equations that can easily be translated to dental age within a short period of time. This conventional method had been considered as the “gold standard” to evaluate the age of a human based on the radiological measurements. [10] The translucency measurements on the other hand, were sensitive to 0.1 mm. However, this method gave a wide range of age estimation corresponding to a specific length of dentin translucency. [11]

The purpose of this study is to compare the accuracy of Kvaal’s radiographic method with modified Bang-Ramm dentinal root translucency method in estimating dental age in Malay adults.

MATERIAL AND METHODS

A total of one-hundred teeth of maxillary and mandibular incisors and canine were collected following dental extraction of Malay adult population (men/women) in this study. The sample was collected from several private dental clinics in the region. The age range for the individuals was between 24 to 56 years. The age, date of birth, date of extraction, gender, and ethnicity of the patient have been recorded for each sample. Ethics approval was obtained from the Institutional Review Board (600-IRMI (5/1/6)).

The inclusion criteria for the sample selection included sound permanent maxillary and mandibular central incisors, lateral incisors and canine with well-developed root structure, absence from any pathology such as caries, cyst, and inflammation. The exclusion criteria for this study sample included endodontically treated tooth, the tooth with fractured root and the tooth with resorbed root. The extracted teeth were thoroughly cleaned, and soft tissue remnants were removed from the root surface with a scalpel. Teeth were preserved in 10% formalin.

The weighted kappa scores were used to analyze the intra- and inter-observer reliability in reproducing the measurement values based on the out-of-sample participants. The out-of-sample participants were selected based on separate data independent from the main study. Paralleling technique with complementary metal oxide semiconductor (CMOS) digital intra-oral sensor was performed to obtain periapical images in this study. The CMOS sensor (EzSensor, pixel size 35 μm, Vatech, Hwaseong, Korea) coupled to an intraoral x-ray machine (Satelec X-Mind AC/DC, Satelec ACTEON Tuusula, Finland) was used in this study. The images were stored in JPEG format. The periapical was then measured based on maximum tooth length, pulp length and root length according to Kvaal et al., 1995 protocol (Figure 1) on each sample. The radicular portion of each tooth was then divided into levels A (the root and pulp width at enamel-cementum junction), B (root and pulp width are midway between measurement levels A and C), and C (root and pulp width midway between apex and enamel-cementum junction) and the root and the pulp width were measured at each level. Chronologically, levels A and C were first determined followed with level B. The odontometric data was then used to calculate the ‘Kvaal dental ratios’: the tooth/root length; the pulp/root length; the pulp/tooth length; and the root width/pulp width ratios at the three levels (A, B and C). Age estimation was then calculated based on these ratios.

The same samples were then positioned using the L-shaped scale of the ABFO No. 2 scale on the LED light box to gauge the dentinal root translucency (Figure 2). The teeth were mounted in autopolymerizing acrylic for sectioning by a Leica SP 1600 hard-tissue microtome. The level of dentinal root translucency was measured from the apex and towards coronally for each tooth. Mounted teeth were sectioned longitudinally to 250 µm in the buccolingual plane, as close as possible to the central axis of the tooth. Tooth sections were then placed adjacent to an ABFO No. 2 reference scale on a flat-bed scanner and scanned under a resolution of 600 dpi. Scanned images were imported into Adobe Photoshop Version: 21.2.2 20200807.r.289 image-editing software program for measuring translucency length. [12] The sections were coded to ensure blind assessment of translucency length. All odontometric data was then recorded and substituted in the maximum translucency length for both linear and quadratic equations of modified Bang-Ramm method to attain the estimated dental age.[2]

The estimated age calculated from both methods was further subtracted from the chronological age to identify the error (dental age − chronological age). Based on this standardization, all positive values represented overestimation while negative values exemplified underestimation of the real age. Next, the mean errors and mean absolute errors for every age group were calculated. For calibration purposes, the error was expressed as mean error (ME) to quantify the direction of the error (overestimation or underestimation) and mean absolute error (MAE) to quantify the magnitude of the error. The results were statistically analysed using RStudio version 0.99.893 - © 2009-2016 RStudio, Inc. Boston MA, USA, and Excel 2010 version 14.0.

Figure 1. Schematic diagram for odontometric measurement adapted from Kvaal et al., 1995

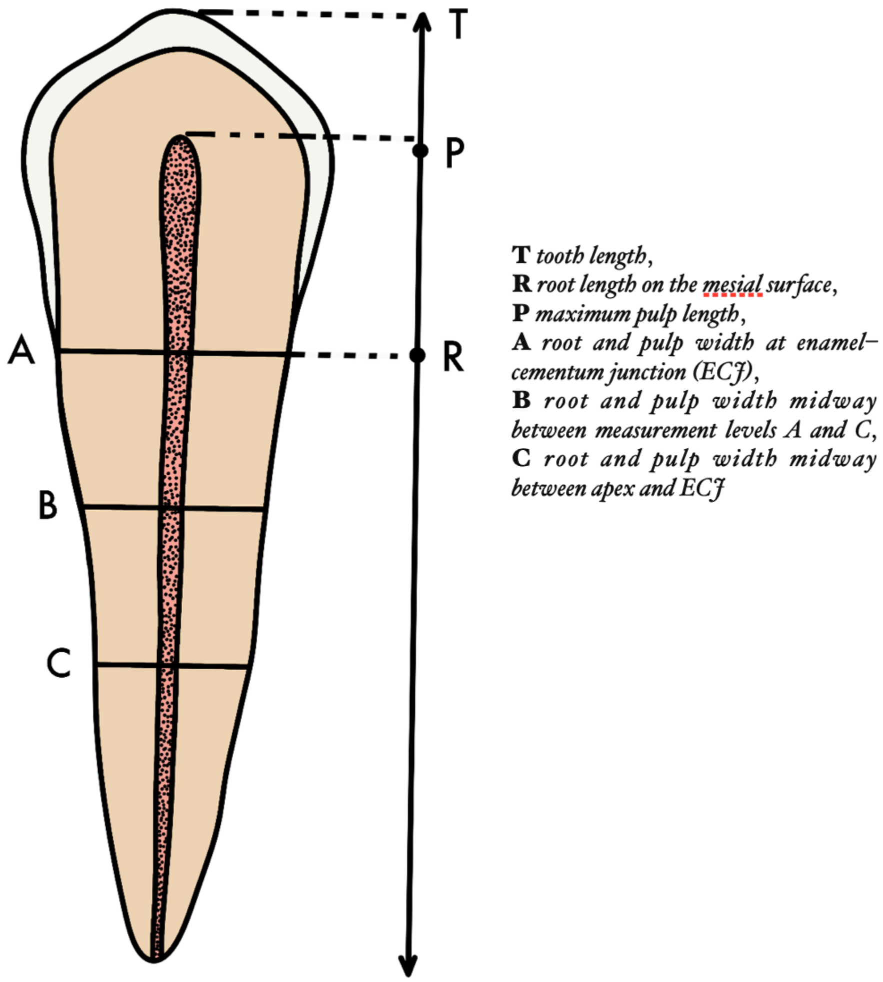
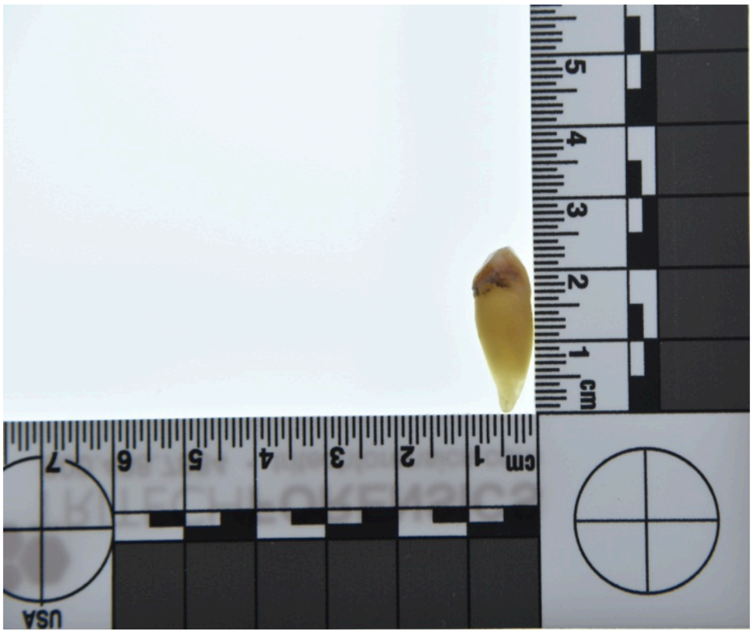


Figure 2. Enhancement of dentinal root translucency via LED light box and ABFO No. 2 scale



RESULTS

The intra- and inter-observer scores for the measurements yielded weighted kappa coefficients of 0.91 and 0.88, respectively. The mean error calculated for estimated age in relation to chronological age and tooth types (central incisor, lateral incisor, and canine) using Kvaal and modified Bang-Ramm methods are shown in Table 1. Generally, for all teeth, Kvaal method displayed positive values that indicated overestimation. In contrast, modified Bang-Ramm for both linear and quadratic equations suggested an underestimation of dental age for all monoradicular teeth. In addition, maxillary lateral incisor exhibited the lowest mean error followed with maxillary central incisor, mandibular central incisor, and maxillary canine for both equations in modified Bang-Ramm. In Kvaal, maxillary lateral incisor showed the lowest mean error followed with mandibular central incisor, maxillary canine, and maxillary central incisor.

Table 1 The mean estimated dental age in relation to chronological age by using Kvaal's and modified Bang-Ramm Linear and Quadratic equation methods

Tooth	CA (years old)	Gender	MBR_L (years old)	Error	MBR_Q (years old)	Error	Kvaal (years old)	Error
11/21	40.0	Male$_{(n=11)}$	44.7	4.7	44.3	4.3	53.4	13.4
	41.3	Female$_{(n=14)}$	47.5	6.2	47.1	5.8	54.2	12.9
12/22	42.8	Male$_{(n=12)}$	47.1	4.3	46.7	3.9	46	3.2
	43.2	Female$_{(n=13)}$	48.3	5.1	48.5	5.3	48.6	5.4
33/43	43.3	Male$_{(n=15)}$	52.1	8.8	51.4	8.1	52.3	9.0
	42.8	Female$_{(n=10)}$	53.7	10.9	52.5	9.7	54.0	11.2
31/41	34.0	Male$_{(n=12)}$	39.5	5.5	38.3	4.3	NR	NR
	36.4	Female$_{(n=13)}$	43.6	7.2	42.2	5.8	NR	NR

CA *chronological age*, **MBR_L** *modified Bang-Ramm Linear Equation*, **MBR_Q** *modified Bang-Ramm Quadratic Equation*, **M** *Male*, **F** *Female* **NR** *Not relevant*

Figure 3 exhibited the mean error for estimated age in relation to chronological age based on specific age categories. The mean error for the Kvaal technique in relation to chronological age was overestimated while the mean error for modified Bang-Ramm method for both linear and quadratic equations were underestimated. The highest mean error from the Kvaal method was at the age of 56 years (20 years) while the lowest mean error for the Kvaal method was at the age of 24 years (2.33 years). The highest mean error for linear and quadratic equations from modified Bang-Ramm method was at the age of 24 years, which were -25.33 years and -28.30 years, respectively. On the other hand, the lowest mean error was at the age of 56 years (1.3 years) calculated from the linear formula.

The mean errors generated from the modified Bang-Ramm for both linear and quadratic equation methods exhibited negative values while Kvaal method revealed a positive value (Figure 4). Furthermore, looking into the direction of the dental age estimation, the Kvaal method hinted an overestimation while the modified Bang-Ramm for both linear and quadratic equations suggested an underestimation in regard to the chronological age. Subsequently, the magnitude of the error, as calculated using the mean absolute error, showed the lowest in the modified Bang-Ramm for quadratic equation followed by linear equation and Kvaal methods. This suggests that the modified Bang-Ramm for both linear and quadratic equations method provided more accurate result in age estimation as compared to Kvaal method.

Figure 3 Mean error for estimated age in relation to chronological age based on specific age categories

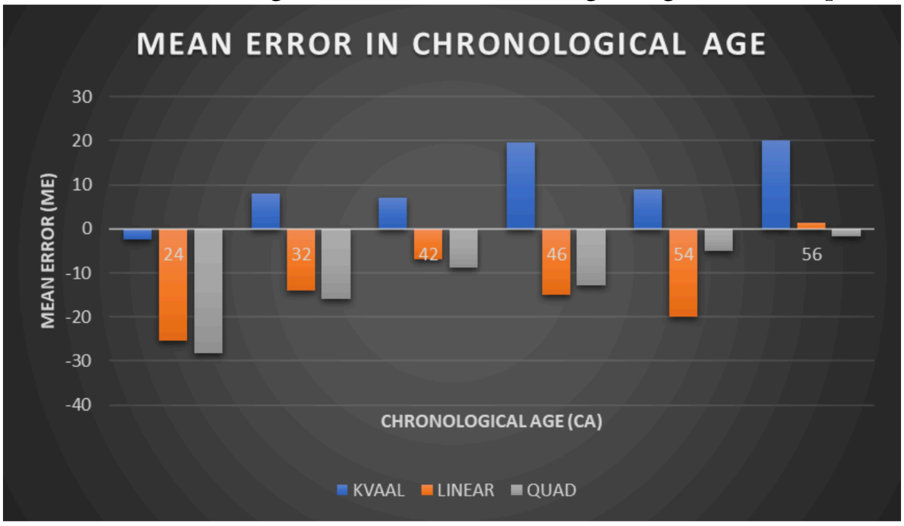
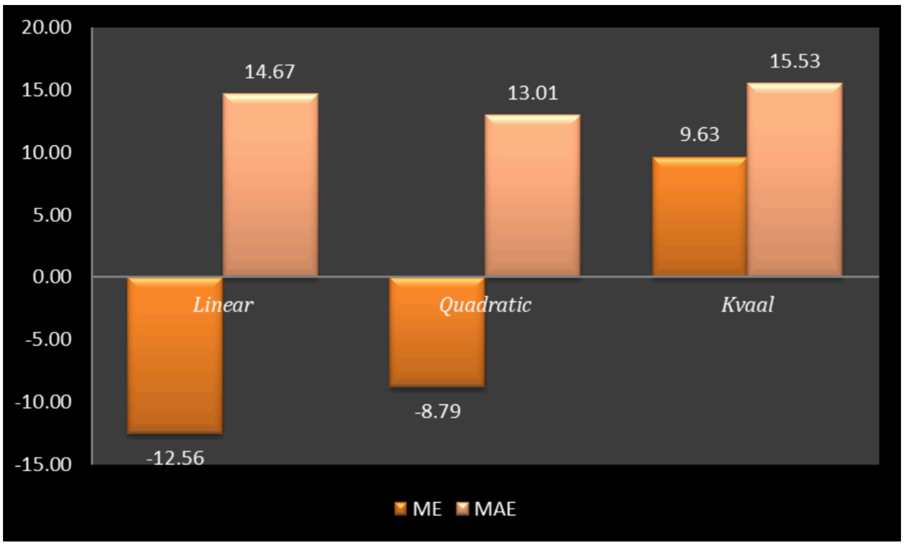


Figure 4 The mean error (ME) and mean absolute error (MAE) values from modified Bang-Ramm for Linear and Quadratic equations and Kvaal method



DISCUSSION

As part of the legal requirement and good clinical practice, the dental age estimation methods must undergo a validation to the specific population prior to being used. This is particularly pertinent to the method developed using a different set of population data. In Malaysia, there have been few studies validating dental age estimation methods using different type of predictors and age category in Malay population. [13-15] Therefore, it is imperative to continuously conduct the research and to look for the improvement on these Kvaal [9] and modified Bang-Ramm methods. [2] Hence, it is crucial to test each method in a specific population, with a large number of samples to identify method's accuracy and precision for that population.

In this study, we found that the modified Bang-Ramm method gave lesser mean absolute error as compared to Kvaal method. Comparisons with other population-specific data is difficult as most studies in literatures utilized the original method of Bang and Ramm. [4] An archeological study performed by Tang et al., 2014 revealed that the average absolute difference between real age and estimated age was 10.7 years and 8.4 years for Spitalfields and All Hallows individuals, respectively, with 58% and 75% estimated within 10 years of known age, and 29% and 33% estimated within five years of known age. [16] The margin of error is comparable to our current study that shows the error ranges from 3.9 years to 10.9 years. It is apparent in our study that the dentinal root translucency is increasing with age. A reason for modified Bang-Ramm methods to have more accurate result could be explained by the fact that those methods have been developed with different regression coefficients for each tooth. In addition, modified Bang-Ramm method extracted only one value which was the length of the dentinal root translucency while Kvaal method comprised of several values extracted from the periapical radiograph which were the ratios of the tooth/root length; the pulp/root length; the pulp/tooth length; and the root width/pulp width ratios at the three levels (A, B and C) and the age estimation were then calculated based on these ratios. The result is better in terms of the mean error. Although the method by Kvaal et al., 1995 was non-destructive, the modified Bang-Ramm method were relatively simple and required less time consumption in calculating the age estimation. This could be an important factor in forensic applications especially during disaster victim identification procedures where time and resources are the uttermost necessity. [16]

One of the advantages of Bang-Ramm method is that it can still be applicable in cases where only root is present. It is well accepted that the amount of translucent dentine increases with age, with an expansion of translucency from the apical portion of tooth to the coronal. The Bang-Ramm method is used in extracted tooth and can be assessed for translucency either in whole or sectioned. From the result, the method underestimated the age, which was contrary to Kvaal method.

Kvaal on the other hand, shows more time consumptions in terms of the application of different mathematical equation for different type of tooth. [17] The equation depends on several factors such as the type of the tooth, gender, and the particular measurements. In addition, the mean error for each type of the tooth is higher compared to the Bang-Ramm method. However, should the study be conducted with larger sample size, the result for the Kvaal method could have been more accurate.

There are several limitations in this study. First, the sample size could have been larger than a hundred. The reason being, most of the sample that had been collected suffered from missing information on either the date of birth, date of extraction, gender, or ethnicity. Thus, the sample had to be excluded from the study as they were not fulfilling the criteria needed. Some of the teeth had also been excluded due to the presence of caries involving the pulp, tooth with fractured root and tooth with resorbed root. Based on the results, it is reasonable to conclude that modified Bang-Ramm method, especially the quadratic equation, gives better accuracy for dental age estimation in Malay adults as compared to Kvaal method. This can be supported by the smallest value of mean absolute error of 13.01, 14.67 and 15.53 for modified Bang-Ramm quadratic equation, linear equation and Kvaal methods, respectively.

CONCLUSIONS

The accuracy of the modified Bang-Ramm for quadratic equation is superior to linear equation and Kvaal method in estimating the Malay adults. However, in cases where the tooth is discernable, the modified Bang-Ramm for linear equation may be utilized.

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Forensic dental identification in complicated fractured skull conditions: case report with adapted algorithm for image comparison

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ABSTRACT

Objective: To analyze the perspective of using an adapted algorithm for digital images comparison while providing forensic dental identification in complicated fractured skull conditions by ante-mortem and post-mortem radiographical data sets.

Materials and Methods: Ante-mortem orthopantomogram and post-mortem peri-apical X-ray images were converted in *.jpeg format with their further import into GIMP 2.10 software (The GIMP Development Team). Segmentation of OPG-image was provided in topographical projections of jaw segments obtained directly from the victim. Comparison of analyzed image segments was provided manually within GIMP 2.10 software using functions of “Layers” and “Opacity” through the proposed algorithm.

Results: Considering the fact that 20 positive concordant dental identifiers overall were verified during comparison of AM and PM X-ray datasets, we can conclude that odontological identity was established. All above-mentioned discrepancies could be classified as explainable. Inter-agreement rate between two investigators considering correspondence between AM and PM datasets reached Cohen’s kappa level which is equal to 0,97, while positive 100% agreement was reached considering 21 out of 24 analyzed characteristics.

Conclusion: Available AM and PM radiographical datasets represent a sufficient information for effective forensic dental identification, even if such were obtained by different roentgenological techniques (orthopantomography and periapical radiography). Using of an adapted algorithm for digital images comparison with forensic dental purposes could potentially overcome cognitive bias and observer’s effect, speed up the process of analysis and increase the accuracy and inter-agreement rate while referencing AM and PM datasets.

INTRODUCTION

Three principal methodologies of person identification by dental status include evaluation and comparison of ante- and post-mortem dental records, photographs and results of radiography.[1,2,3] However, there are several causes of inability to use dental records as a reliable source of personal information in Ukraine, such as: absence of mandatory insurance program, improper filling out medical documentation in state hospitals and dental departments specifically, detention of medical documentation personally by patients during the treatment migration among different medical/dental facilities.[4-6] The latter tendency was also noted in recent systematic review demonstrated that only about 12% of dentists keeps all dental records of their patients.[7]

Based on possible condition of non-reliable person's dental record, other information also should be used for the identification purpose, which includes clinical photo and X-ray results (peri-apical, bite-wing, orthopantomograms, cone-beam computed tomographical scans).[4,6,8,9] In relevant real-life conditions orthopantomogram still considered to be one of the most significant ante-mortem source of evidences that could be effectively used for the forensic dental identification purposes.[9] Also considering problem of inconsistency of dental records, multidisciplinary approach should be provided in cases, where dental identification could not provide valued positive outcome.[5,6,8]

Nevertheless, some cases of individual identification remain complicated due to the severity of deformations, amount of pathological and iatrogenic changes, and sometimes due to the intentional actions aimed at dental status alterations. In such cases, several factors such as observer's effect or cognitive bias, could compromise the result of identification outcome while providing classic antemortem-postmortem data sets comparison especially if such held by specialists of different professional experience. Also, pronounced fragmentation of maxillo-facial region is slowing down the identification process and causing decrease of inter-agreement rate while interpreting experts' conclusions.

That is why the analyses of practical case reports of complicated identifications could be suggested as an educative measure for general dental specialists, considering legislative and procedural specifics of the forensic dental identification process in different countries. Taking into account digital conversion of dental practice in general, approbation of newly adapted methodologies using digital analysis instruments seems to be relevant and perspective within contemporary realities for solving issues of low inter-expert's agreement rate, observer's effect and cognitive bias in cases of involving dental specialists with different professional experience into forensic identification process.

OBJECTIVE

To analyze the perspective of using an adapted algorithm for digital images comparison while providing forensic dental identification in complicated fractured skull conditions by ante-mortem and post-mortem radiographical data sets.

MATERIAL AND METHODS

Present case report demonstrates possibility to compare ante-mortem (AM) orthopantomogram and post-mortem (PM) peri-apical X-ray images with obtaining of more than 12 ordinary characteristics to establish odontological identity through using for this purpose an adapted algorithm of digital images comparison.

Ante-mortem orthopantomogram was obtained during a patient's urgent visit to the Central Dental Policlinic of the Ministry of Defense in Ukraine, caused by persistent dental pain. OPG was obtained by Pax-I orthopantomography device (50-90 Kvp / 4-10 mA, focal spot – 0,5 mm; Vatech, Korea). Post-mortem peri-apical X-ray images were obtained from residual segments of maxilla and mandible manually using ProX intraoral X-ray unit (63 ±2 kV / 7 + 0.2 mA, focal spot – 0,4 mm; Planmeca, Finland).

Both OPG and peri-apical X-ray images were saved in *.jpeg format with their further import into GIMP 2.10 software (The GIMP Development Team), where images were cropped and optimized by the orientation and intensity/ contrast parameters to minimize the influence of image background on further comparison process (Fig.1).[10,11]

Segmentation of OPG-image was provided in topographical projections of segments obtained directly from the victim ("a1" – maxillary segment from 12th to 17th tooth with visualization of maxillary sinus floor; "a2" – maxillary segment from 21st to 25th tooth; "a3" – maxillary segment from 26th to 27th tooth; "a4" – mandibular segment from 37th to 38th tooth, "a5" – mandibular segment from 42nd to 43rd tooth) (Fig. 1).

Figure 1. Example of image optimization before comparison phase in the software environment: 1) periapical PM image obtained after orientational and contrast/intensity optimization; 2) periapical PM image obtained right after periapical X-ray examination before optimization; 3) cropped segment of AM orthopantomogram in projection of PM image segment.

Further comparison of analyzed segments was provided manually within GIMP 2.10 software using function of "Layer" and "Opacity" according to the next developed algorithm:

1) both segmented and previously optimized AM and PM images were imported into software as two separate layers (superimposed) of the same size;
2) operator marked unique points/lines/contours on the outer layer (PM image) with "Brush tool";
3) after all unique components were marked, operator changed the opacity of the outer layer (PM image) from 100% to 0%, while monitoring the correspondence of previously marked points/lines/contours to the inner layer (AM image);
4) if marked unique points/lines/contours from the outer layer (PM image) corresponded to the similar structures at the inner layer (AM image), operator registered results of identification according to Keiser-Nielsen principles.

Comparison of AM and PM image segments was held independently by two experts using the same algorithm described above. Inter-agreement rate between two investigators considering correspondence between AM and PM datasets was evaluated with the use of Cohen's kappa and reproducibility percentage value.[12]

Subjective expert's assurance considering concordance of dental identifiers was evaluated by experts themselves using 0-100% scale, within which 0% stands for "Completely uncertain" and 100% for "Completely certain". Specification of each scale values between 0-100% were not provided in order to not disturb the process of expert subjective grading considering the level of the personal assurance.

Results of identification were classified according to the Keiser-Nielsen principle considering ordinary concordant details: 1) odontological identity is established (12 or more unique concordant points); 2) odontological identity is probable (6-12 unique concordant points); 3) odontological identity is possible (less than 6 unique concordant points).[13]

In Ukraine forensic documentation designs do not include separate forensic dental report, so all the findings were noted in the general report of forensic expert.

Some details of the abovementioned case (date, exact territory, supposed age, gender, names and affiliations) could not be revealed considering ongoing law enforcement investigation, but details of forensic dental identification as component of general forensic examination could be represented considering ethical principles and full anonymous design of present case demonstration with agreement of all associated policies and state authorities.

Systematization, categorization and tabulation of the data was conducted in Microsoft Excel software environment (Microsoft Office 2019, Microsoft).[14]

RESULTS

Unidentified skeletonized body parts with fractured fragments of the skull and body bones were found at the territory of the Eastern Ukraine at the area of military conflict. Skull part consisted of 82 independent fractured fragments of different patterns. Primary investigation of skull fragments revealed 5 residual segments of mandible and maxilla, which potentially could be analyzed for the person's identification purpose (Fig. 2-3).

Figure 2. Initial view of all skull fractured fragments.

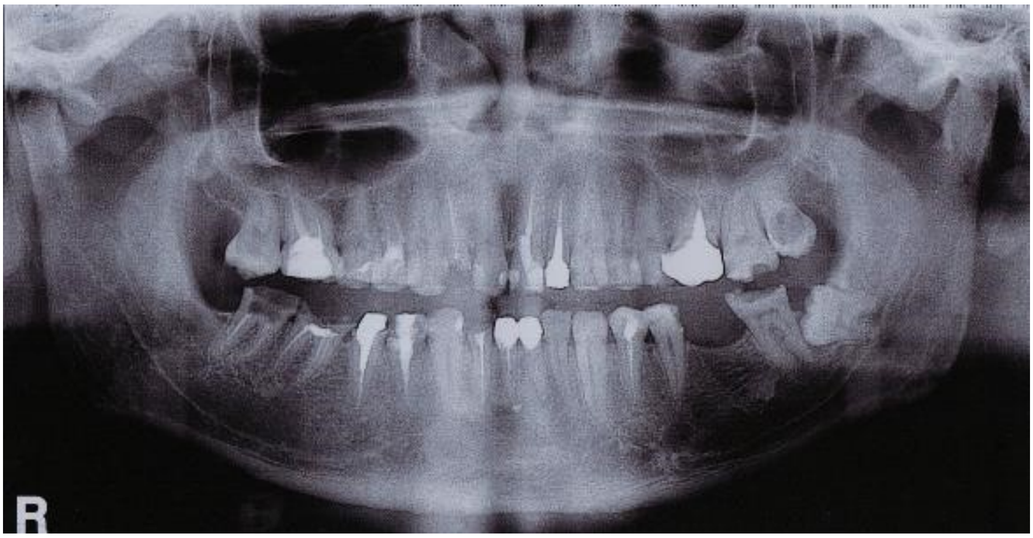
Figure 3. Initial view of the residual segments of maxilla (a1, a2, a3) and mandible (a4, a5)

Because of totally fractured conditions of the skull bones it was impossible to get a post-mortem orthopantomogram of the victim. To overcome such issue peri-apical images of fractured mandibular and maxillary fragments were obtained with the intraoral X-ray device.

Previously obtained orthopantomograms and dental X-rays of all soldiers considered to be missing during military conflict were collected from the Central Dental Polyclinic of Ministry of the Defense in Ukraine by the forensic examination bureau responsible for the identification procedures within military personnel.

Among number of analyzed orthopantomograms one demonstrated signs of previous dental interventions and unique teeth characteristics, that were similar to those, registered at the peri-apical X-ray images, obtained from residual mandibular and maxillary fragments of the victim (Fig. 4).

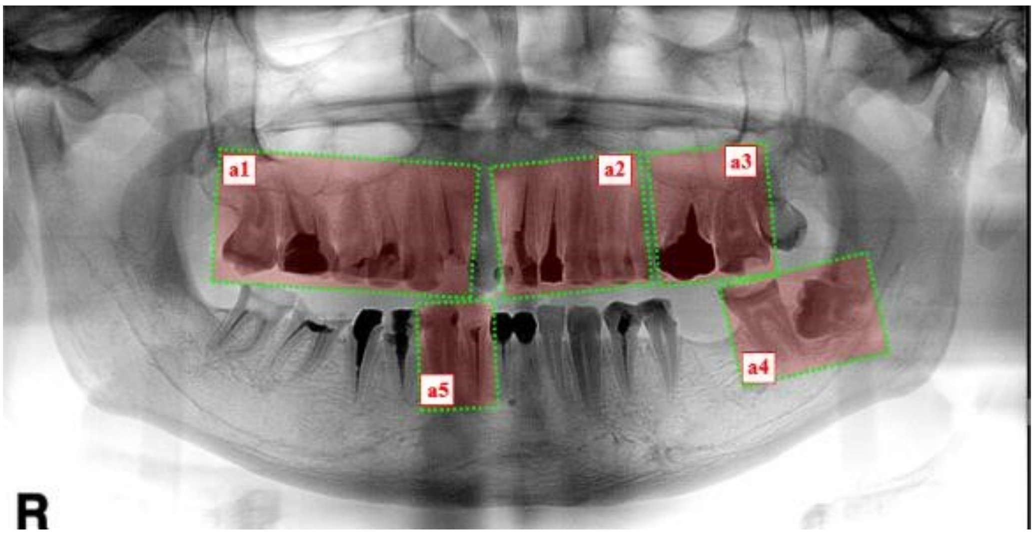
Figure 4. Orthopantomogram of suspected person



Before any superimposition of the images, they were optimized by size, orientation and contrast/intensity parameters to eliminate the influence of background on the subjective comparison process.

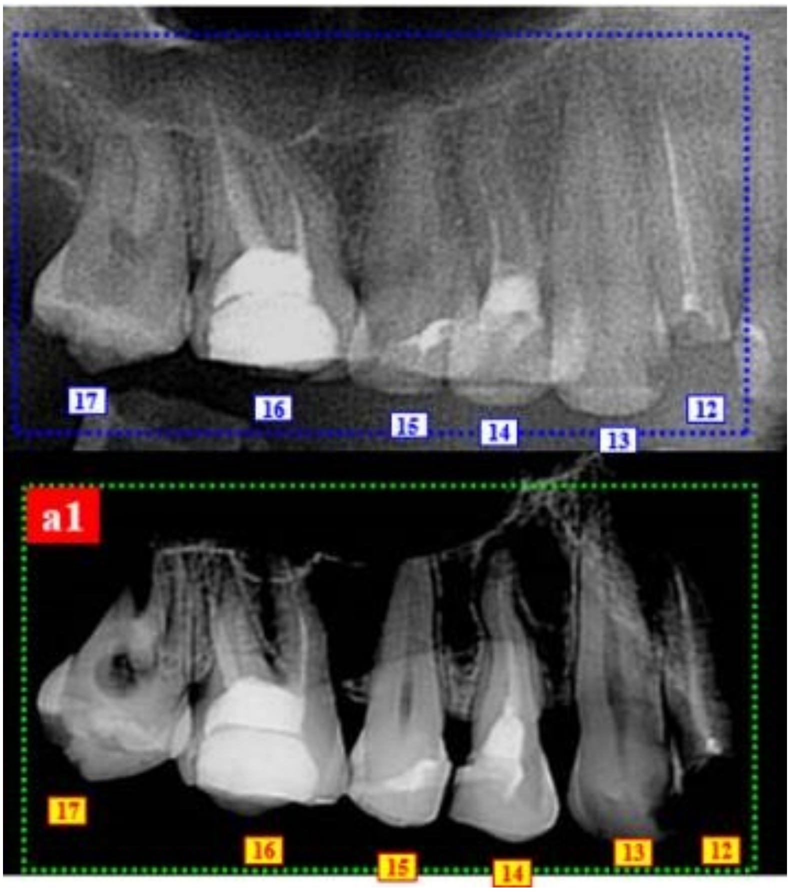
Further phase of identification included superimposition of the obtained periapical X-ray images with analogical topographical projections at selected segmented orthopantomogram according to the layer-on-layer principle and consistent change of outer layer (PM image) opacity. At the first stage the superimposition process was held subjectively by the operator's visual inspection, and after that unique components were marked and traced through the two analyzed layers of images (Fig. 5).

Figure 5. Primary topographical projection of segments obtained from victim on OPG of suspected person



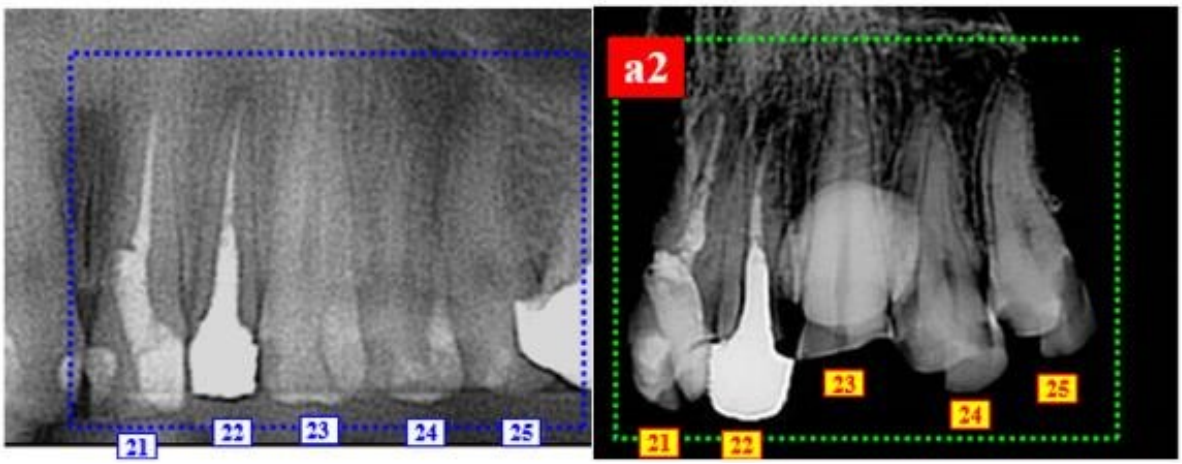
“a1” maxillary segment of PM periapical image demonstrated identical unique characteristics with AM OPG-image segment by means of: 1) root canal treatment of 12th tooth; 2) lost coronal part of 12th tooth; 3) caries at mesial surface of 13th tooth; 4) specific form of apical third of 14th tooth (curved); 5) root canal treatment of 16th tooth; 6) filling of the 16th tooth; 7) contour of maxillary sinus floor (Fig. 6).

Figure 6. Comparison of OPG-image segment topographically identical with periapical X-ray image, obtained from the a1 maxillary segment



“a2” maxillary segment of PM periapical image demonstrated identical unique characteristics with AM OPG-image segment by means of: 1) post and core build-up within 22nd tooth; 2) root canal treatment of 22nd tooth; 3) periapical lesion at 22nd tooth; 4) root canal treatment of 21st tooth; 5) filling of the 21st tooth (Fig. 7).

Figure 7. Comparison of OPG-image segment topographically identical with periapical X-ray image, obtained from the a2 maxillary segment



“a3” ... stics

with AM OPG-image segment by means of: 1) post and core build-up within 26th tooth; 2) occlusal caries at 27th tooth (Fig. 8).

"a4" mandibular segment of PM periapical image demonstrated identical unique characteristics with AM OPG-image segment by means of: 1) position of 38th tooth in the relation to 37th tooth (impaction of 38th tooth); 2) developmental stage of 38th tooth; 3) coronal defect of 37th tooth (Fig. 9).

"a5" mandibular segment of PM periapical image demonstrated identical unique characteristics with AM OPG-image segment by means of: 1) root canal treatment of 42nd tooth; 2) lost coronal part of 42nd tooth; 3) restoration on the mesial surface of 43rd tooth (Fig. 10).

Figure 8. Comparison of OPG-image segment topographically identical with periapical X-ray image, obtained from the a3 maxillary segment

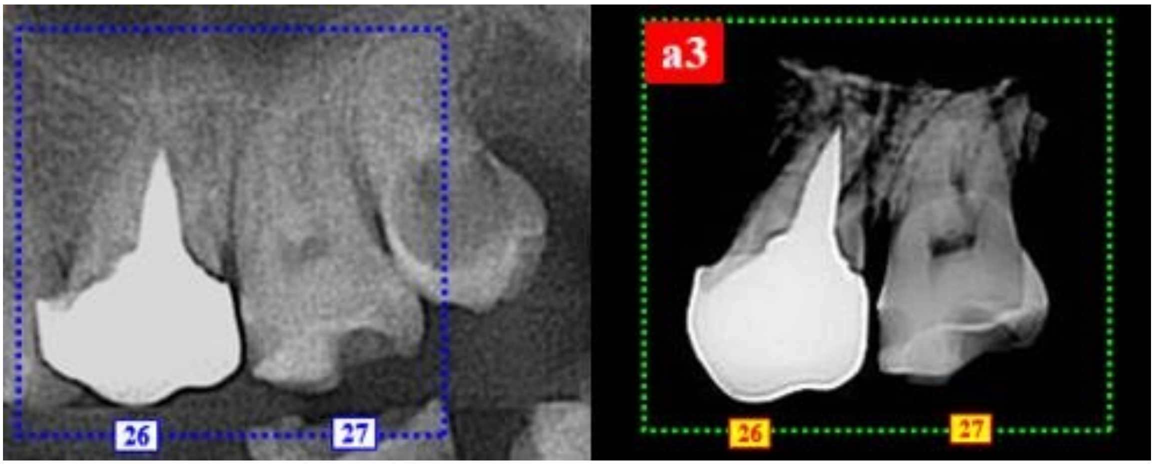


Figure 9. Comparison of OPG-image segment topographically identical with periapical X-ray image, obtained from the a4 mandibular segment

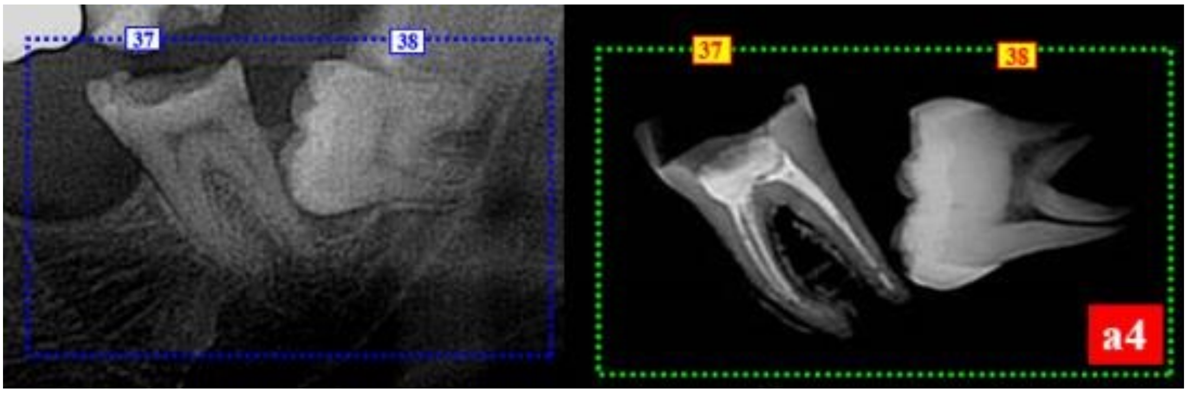
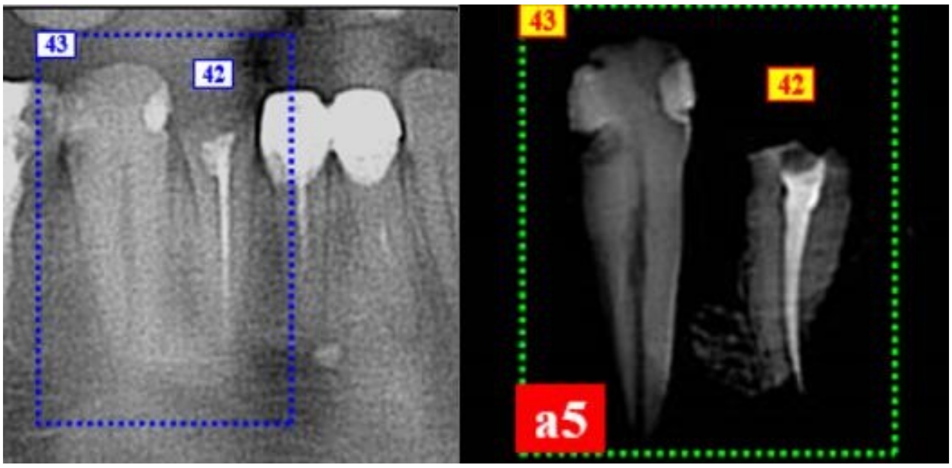


Figure 10. Comparison of OPG-image segment topographically identical with periapical X-ray image, obtained from the a5 mandibular segment



Considering that overall 20 positive concordant dental identifiers were verified during comparison of AM and PM X-ray datasets, we can conclude that odontological identity was established. Some inconsistencies also were verified during comparative images analysis: 1) "a1" postmortem periapical X-ray image demonstrated highly contrasted results of 14th tooth's root canal treatment, which could not be recognized at AM orthopantomogram; 2) "a2" postmortem image demonstrated broken crown of 23rd tooth, which was not altered at AM image; 3) also "a4" PM image demonstrated signs of 37th tooth's root canal treatment, which were absent at AM orthopantomogram (Table 1).

Table 1. Consistency of identification points between AM and PM image datasets, established by two independent experts

№	Segment	Identification point	AM image (OPG)	PM images (periapical X-rays)	Consistency of the identification point concordance between AM and PM images Expert 1/Expert 2	Subjective Expert 1 assurance	Subjective Expert 2 assurance
1	a1	Root canal treatment of 12th tooth	Present	Present	+/+	100%	100%
2	a1	Caries at mesial surface of 13th tooth	Present	Present	+/+	100%	100%
3	a1	Specific form of apical third of 14th tooth (curve)	Present	Present	+/+	100%	100%
4	a1	Root canal treatment of 14th tooth	Questionable	Present	-/-	70%	60%
5	a1	Root canal treatment of 16th tooth	Present	Present	+/+	100%	100%
6	a1	Lost coronal part of 12th tooth	Present	Present	+/+	100%	100%
7	a1	Filling of the 16th tooth	Present	Present	+/+	100%	100%
8	a1	Contour of maxillary sinus floor	Present	Present	+/+	100%	80%
9	a2	Post and core build-up within 22nd tooth	Present	Present	+/+	100%	100%
10	a2	Root canal treatment of 22nd tooth	Present	Present	+/+	100%	100%
11	a2	Periapical lesion at 22nd tooth	Present	Present	+/+	100%	100%

12	a2	Filling of the 21st tooth	Present	Present	+/+	100%	100%
13	a2	Root canal treatment of 21st tooth.	Present	Present	+/+	100%	100%
14	a2	Broken crown of 23rd tooth	Absent	Present	-/-	100%	100%
15	a3	Post and core build-up within 26th tooth	Present	Present	+/+	100%	100%
16	a3	Occlusal caries at 27th tooth	Present	Present	+/+	100%	100%
17	a4	Position of 38th tooth in the relation to 37th tooth	Present	Present	+/+	100%	100%
18	a4	Developmental defect of 38th tooth	Present	Present	+/+	100%	100%
19	a4	Coronal defect of 37th tooth	Present	Present	+/+	100%	100%
20	a4	Root canal treatment of 37th tooth	Absent	Present	-/-	100%	100%
21	a5	Root canal treatment of 42nd tooth	Present	Present	+/+	100%	100%
22	a5	Lost coronal part of 42nd tooth	Present	Present	+/+	100%	100%
23	a5	Restoration on the mesial surface of 43rd tooth	Present	Present	+/+	100%	100%
24	a5	Caries lesion at distal surface of 43rd tooth	Questionable	Present	-/+	60%	70%

All the abovementioned discrepancies could be classified as explainable, since endodontic treatment and crown's trauma could be provided/ occurred after initial OPG was obtained. The issue of caries at distal surface of 43rd tooth at "a5" segment remained controversial, considering points of two experts' view (Fig. 11)

Inter-agreement rate between two investigators considering correspondence between AM and PM datasets reached Cohen's kappa level which was equal of 0,97, while positive 100% agreement was reached considering 21 out of 24 analyzed characteristics.

Figure 11. Comparison of OPG-image and actual residual fragments of the maxilla and mandible

DISCUSSION

There are several components of the dental status that potentially could be used as important identification criterions during ante- and post-mortem data comparison. Latter includes teeth themselves and their specific characteristics (impaction, rotation, displacement, hypo- or hyperodontia), signs of caries or periodontal pathologies, coronal and hidden restorations, bone patterns and landmarks as well as its individual and pathological changes, air-borne spaces.[9, 15]

Morphological features of teeth and jaws have been categorized as the most validated dental identifiers used for person's identification.[16] At presented case the curved shape of 14th tooth's apex and position of 38th tooth (impaction) with its specific developmental stage could be interpreted as so-called morphological identifiers with high level of uniqueness. Potentially AM OPG could represent a greater amount of unique dental characteristics, but due to the lack of PM jaws segments such could not be used during AM-PM comparison in the present case.

Differentiation of dental patterns based on the OPG-image could be provided by 11 prespecified grouped parameters (virgin, missing, filling, defect, crown, residual root, bridge pontic, dental implant, endodontic treatment, impacted and dental anomaly), which consist of strictly defined derivates.[17] Within present case 6 parameters could be verified, including missing, virgin, crown, filling, impacted and defect, which is optimal for reliable identification of dental pattern based on OPG-image. Previously, authors mentioned that "6 parameters-pattern" stands for 99,95% specific pattern among dental status diversities, and in this means is analogical to "11 parameters-pattern".[17]

Brkic et al. reported that prosthetic appliance were the most frequently registered environmental dental findings with sufficient identification potential.[18] Also, disturbance of tooth eruption as identification criterion was noted in 22% of analyzed cases.[18] In present case AM and PM images of the person demonstrated the presence of post-and-core build-up constructions and specific impacted position of 38th tooth, which due to the previous Brkic's findings could be interpreted as characteristics with sufficient potential for positive identification outcome. Silva et al. highlighted the importance of periapical radiographs with registered signs of endodontic treatment for forensic identification purposes.[19,20] In present case 7 out of 20 positive concordant points (35,0%) were related to root canal treatment outcomes registered both at PM and AM X-ray images.

Effectiveness of forensic dental identification based on the available AM and PM periapical radiographs demonstrated direct statistical association with investigators' level of experience, while even unexperienced undergraduates showed acceptable results, but postgraduates obtained the results which were characterized with 89,3%, 92,3% and 90,5% of sensitivity, specificity and accuracy respectively.[21] Similar results were also reported by Pinchi et al., due to which experienced forensic odontologists demonstrated high levels of accuracy and repeatability (0,97-1) during comparative analysis of available datasets.[22] In analyzed case report reproducibility level between two investigators regarding uniqueness of evidences and level of their full correspondences between X-ray datasets reached high Cohen's kappa level and more than 90% of inter-concordance.

Such outcome could be directly related to the fact that both dental specialists were previously trained within the framework of post-graduate specialized dental courses provided under the IOFOS umbrella. Also, use of adapted algorithm for digital image comparison optimized the identification process both by means of high-level inter-agreement rate and by means of unambiguousness of obtained outcomes interpretation. Such perspectives of proposed algorithm for digital image comparison is highly relevant, especially in the context of young dental specialists involving in complicated dental identification cases.

Previously, the influence of cognitive bias and observer's effect on the outcome of identification efficiency, especially during comparison examinations, were highlighted.[22] Considering these facts, in present case report we have used not only subjective comparison of AM and PM images, but also we have developed algorithm for its phased comparison with use of layer-to-layer superimposition principle and tracing of unique characteristics through the images layers. In such way, we tried to compensate cognitive bias influence, mentioned in a previous study: proposed objective contour-to-contour comparison through images layering excluded any ambiguity regarding dental identifiers in the process of analyzing complicated fractured case with the absence of post-mortem OPG.

Relevant improvements in OPG analysis with forensic identification aim include methods of speeded up robust features and convolutional neural network.[23,24,25] In previous research it was shown that semi-automated approach could provide high accuracy of mandibular structure analysis in the manner of finger-print based on matching protocol consisted of region of interest verification, voxel-based registration and further results classification.[26] Also Lin P.-L. demonstrated the high efficiency of using weighted Hausdorff distance during comparisons of bitewing dental radiographs.[27] Proposed method was based on contouring of specific dental features with contour's further realigning among comparator images.[27]

In present case we implemented somewhat similar, but simpler algorithm of 2D images comparative analysis, which consisted of the image segmentation, optimization, layer-by-layer superimposition and further tracing of unique characteristics by opacity changes. Even though presented case was characterized with high level of inter-agreement rate between two investigators regarding correspondence between AM and PM datasets, in future research we will analyze changes of such rate while providing image optimization with layer-to-layer comparison and within conditions of ad oculus analysis of AM and PM datasets.

Use of segmentation process described in proposed protocol of OPG and periapical X-ray image superimposition argumented by the results of systematic review, which demonstrated that similar approach supports the noise reduction and enhances the accuracy of the image for the comparative analysis.[28] Image preprocessing with reasonable purpose represent the useful tool for forensic dental practice during comparative and reconstructive identification, age estimation and bite mark analysis. Nowadays, new software packages and mobile application aimed at X-ray image improvement could be effectively implemented in forensic dental practice after precise evaluation of their validness, sensitivity and specificity parameters.[28]

Future trends in forensic dental identification are closely related to the digitalization shift in the field, which are associated with possibility to gain larger amount of unique criterions during individual and mass cases of ante-mortem and post-mortem dental datasets comparison.[29] Digitalization itself expands the number of comparative elements due to analog-to-digital conversion and supports their processing with use of adaptive algorithms and software. But on the other side, the progressive increase of identification points associated with the diversity of their identification value and level of their uniqueness, so we need to assure an adequate attention to the process of their phased stratification and differentiation with further categorization according to actual identification impact and importance. Also, there is still a gap in translation of modern achievements within digital dentistry into forensic dental practice, which could be compensated by the cooperation of corresponding societies and associations from both sides. For instance, modern approaches of image and 3D-objects superimposition, previously used for surgical and prosthetic treatment planning, and consisted of several techniques as nurb-to-nurb, mesh-to-mesh and point-to-point overlapping, potentially could improve the algorithm of comparative forensic dental post- and ante-mortem analysis.[30,31]

CONCLUSIONS

Available AM and PM radiographical data sets represent sufficient information for effective forensic dental identification, even if such were obtained by different roentgenological techniques (orthopantomography and peri-apical radiography). The accessibility of adequate AM data volume (provided by orthopantomogram), possibility of X-ray assessment for mandibular and maxillary residual segments and registration of previously provided iatrogenic interventions and associated changes (being assessed as important identification criterions) were the key components of successful identification outcome, despite complicated fractured conditions of the skull. Nevertheless, modern approaches of graphical superimposition expand the possibilities for person's identification by dental status and could be effectively implemented in forensic dental practice especially under the condition of deficiency or total absence of dental and medical records. Moreover, use of an adapted algorithm of digital images comparison with forensic dental purposes could potentially overcome cognitive bias and observer's effect, speed up the process of analysis and increase accuracy and inter-agreement rate while referencing AM and PM datasets.

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