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Forensic anthropology population data

Age estimation based on pulp chamber volume of first molars from cone-beam computed tomography images

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ABSTRACT

Aim: To establish a method that can be used for human age estimation on the basis of pulp chamber volume of first molars and to identify whether the method is good enough for age estimation in real human cases.

Materials and methods: CBCT images of 373 maxillary first molars and 372 mandibular first molars were collected to establish the mathematical model from 190 female and 213 male patients whose age between 12 and 69 years old. The inclusion criteria of the first molars were: no caries, no excessive tooth wear, no dental restorations, no artifacts due to metal restorative materials present in adjacent teeth, and no pulpal calcification. All the CBCT images were acquired with a CBCT unit NewTom VG (Quantitative Radiology, Verona, Italy) and reconstructed with a voxel-size of 0.15 mm. The images were subsequently exported as DICOM data sets and imported into an open source 3D image semi-automatic segmenting and voxel-counting software ITK-SNAP 2.4 for the calculation of pulp chamber volumes. A logarithmic regression analysis was conducted with age as dependent variable and pulp chamber volume as independent variables to establish a mathematical model for the human age estimation. To identify the precision and accuracy of the model for human age estimation, another 104 maxillary first molars and 103 mandibular first molars from 55 female and 57 male patients whose age between 12 and 67 years old were collected, too. Mean absolute error and root mean square error between the actual age and estimated age were used to determine the precision and accuracy of the mathematical model. The study was approved by the Institutional Review Board of Peking University School and Hospital of Stomatology.

Results: A mathematical model was suggested for: $\text{AGE} = 117.691 - 26.442 \times \ln$ (pulp chamber volume). The regression was statistically significant ($p = 0.000 < 0.01$). The coefficient of determination (R^2) was 0.564. There is a mean absolute error of 8.122 and root mean square error of 5.603 between the actual age and estimated age for all the tested teeth.

Conclusion: The pulp chamber volume of first molar is a useful index for the estimation of human age with reasonable precision and accuracy.

1. Introduction

Age estimation of living individuals or corpses is important in forensic sciences. Due to the fact that teeth are highly resistant to mechanical, chemical or physical impacts and time [1,2] as well as that age-related changes of tooth are minimally influenced by the nutrition, environment and living conditions that an individual is submitted to [3,4], many age estimation methods based on teeth have been established. Some of the methods were developed from the time of tooth emergence and tooth calcification in oral cavity [3,5–7]. Analysis on the stage of dentition helps in age determination in children and adolescents but is difficult in adults that the development of permanent dentition completes [3]. The methods based on biochemical characteristics of teeth such as amino acid racemization [8] and carbon-14 isotope [9] was also introduced. However, these methods are time-consuming and require sophisticated laboratory equipment and tooth extraction. Tooth extraction is unethical and impossible in living individuals. Analysis on dental wear is another most commonly used method for age estimation [10,11]. The drawback of the method is that the

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attrition level of tooth is strongly influenced by diet and personal habits [12–14].

Secondary dentine apposition is a significant morphological dental age predictor. It begins after the apical part of root has completely developed, tooth erupted into oral cavity and tooth begins to function. This apposition continues through people's whole life. Decrease in pulp cavity size is an age-associated process, and may be influenced by local factors, such as attrition, carious lesions and changes in osmotic pressure etc. [15–19]. Many two-dimensional images like panoramic [20] or periapical radiograph [21] have been used to quantify the size of pulp cavity and correlate it with age. In recent years, with the wide use of three-dimensional images in practice, three-dimensional image datasets obtained from cone beam CT, CT and Micro CT have been applied to investigating the potential relationship between age and volume ratio of pulp cavity to entire tooth [22–28], and concluded that pulp/tooth volume ratio is a useful indicator for age. In the analysis of these studies, we found that the research samples are relatively small and only included single-rooted teeth. Considering the fact that human teeth contains not only single-rooted but multi-rooted teeth, one study exclusively focusing on multi-root teeth with a relative large sample size is necessary to further disclose the relationship between secondary dentine apposition and age. Thus, the aim of the present study was to establish a method for the age estimation from multi-rooted teeth in the CBCT images and to identify whether the method is good enough for age estimation in real human cases.

2. Materials and methods

2.1. Method establishment

2.1.1. Subjects

CBCT images of 373 maxillary first molars and 372 mandibular first molars were retrospectively collected from 190 female and 213 male patients whose age between 12 and 69 years old from the database in Peking University School and Hospital of Stomatology. The birth date of all subjects was confirmed in the hospital's patient information system. The age and sex distributions of the subjects are shown in Table 1.

The inclusion criteria of the first molars were: no caries, no excessive tooth wear, no dental restorations, no artifacts due to metal restorative materials present in adjacent teeth, and no pulpal calcification. To specify the extent of "excessive tooth wear", we borrowed the Smith and Knight's tooth wear index (TWI, Table 2) [29] and the results from the tooth wear epidemiological investigation in Chinese population [30]. The tooth with TWI ≤ 2 before 50 years and TWI ≤ 3 after 50 years was included. Only one maxillary first molar and one mandibular first molar in each person were included for analysis.

2.1.2. Image acquisition and segmentation

All the CBCT images were acquired with a CBCT unit NewTom VG (Quantitative Radiology, Verona, Italy). Exposure parameters for CBCT image were 110 kVp, 4.19–107.39 mAs in accordance with patient size and filed of view. Selection of field of view (FOV) was based on clinical need. The FOVs included 6 cm $\times$ 6 cm, 8 cm $\times$ 8 cm, 12 cm $\times$ 8 cm or 15 cm $\times$ 15 cm.

Table 1
Age and sex distribution of group samples used for method establishment.

Age (year)	Male	Female	Maxillary first molars	Mandibular first molars
12–20	43	43	86	86
21–30	47	44	91	91
31–40	51	48	91	90
41–50	36	28	56	58
51–60	23	19	33	30
61–69	13	8	14	17
Total	213	190	373	372

Table 2
Smith and Knight tooth wear index.

Score	Tooth surface	Criteria
0	B/L/O/I	No loss of enamel surface characteristics
	C	No loss of contour
1	B/L/O/I	Loss of enamel surface characteristics
	C	Minimal loss of contour
2	B/L/O	Loss of enamel exposing dentine for less than one third of surface
	I	Loss of enamel just exposing dentine
	C	Defect less than 1 mm deep
3	B/L/O	Loss of enamel exposing dentine for more than one third of surface
	I	Loss of enamel and substantial loss of dentine
	C	Defect less than 1–2 mm deep
4	B/L/O	Complete enamel loss-pulp exposure-secondary dentine exposure
	I	Pulp exposure or exposure of secondary dentine
	C	Defect more than 2 mm deep-pulp exposure-secondary dentine exposure

B = buccal; L = lingual; O = occlusal; I = incisal; C = cervical.

Acquired images were subsequently reconstructed with a voxel-size of 0.15 mm and exported as DICOM data sets. These data were then imported into a 3D image semi-automatic segmenting and voxel-counting software ITK-SNAP 2.4 (open source software, www.itksnap.org) for the calculation of pulp chamber volumes [31].

To avoid the influence of the complex root system of first molars and also to simplify the segmentation procedure, we set the pulp chamber floor as the "cut plane" to cut off the roots and calculate the volume of tooth pulp chamber. The final segmented image of tooth pulp chamber is shown in Fig. 1.

2.1.3. Mathematical model establishment

Logarithmic regression analysis was conducted with age as dependent variable and pulp chamber volume as independent variable to establish a mathematical model for the estimation of human age. To establish age estimation mathematical models suitable for unknown sex, logarithmic regression analysis was also conducted with age as dependent variable and pulp chamber volume as independent variable for maxillary first molars and mandibular first molars separately.

2.1.4. Segmentation accuracy

To validate the measurement accuracy of image segmentation and volume calculation, images of ten extracted molars were acquired with the CBCT unit NewTom VG and a high-resolution Micro CT unit (Inveon, Siemens, Germany). Projecting parameter of the Micro CT was 80 kV, 500 mA, and 8.82 μm-effective pixel size. The images were then imported into the software ITK-SNAP 2.4 to calculate the pulp chamber volume. With the volume calculated from Micro CT images as the reference standard, the volume calculated from the CBCT images was quantified for the accuracy of the volume calculation.

2.1.5. Inter-and intra-observer variability

All measurements were carried out by the same examiner. To test intra-examiner reproducibility, slice data of a random sample of 20 maxillary first molars and 20 mandibular first molars were re-examined after an interval of 3 weeks. At the same time, the same slice data of 20 maxillary first molars and 20 mandibular first

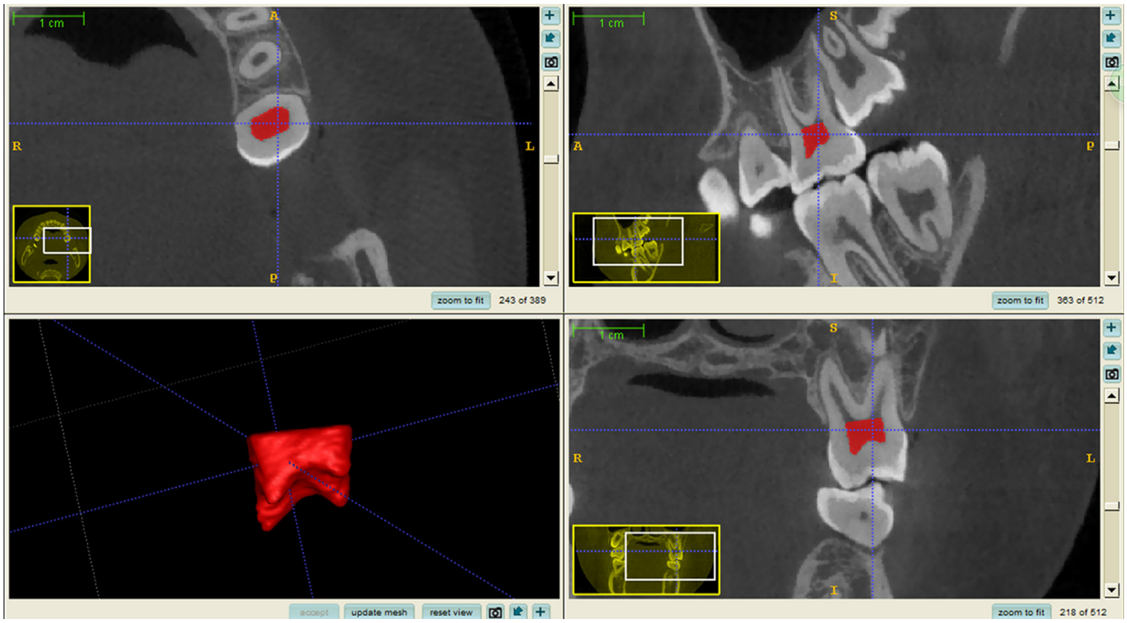

Fig. 1. The final segmented image of tooth pulp chamber.

molars were examined by another calibrated examiner to test the inter-examiner reproducibility.

3. Method validation

Another group of CBCT images which was not used for the establishment of the mathematical model was collected to validate the precision and accuracy of the model for age estimation. 104 maxillary first molars and 103 mandibular first molars from 55 female and 57 male patients whose age between 12 and 67 years old were collected from the database in Peking University School and Hospital of Stomatology with the same inclusion criteria as aforementioned in the Method Establishment. The age and sex distributions of the subjects are shown in Table 3.

After the procedures of image acquisition and segmentation described above, the volume of tooth pulp chamber were calculated. Take the numerical value of the volume of pulp chamber into the mathematical models, we get the estimated age of these subjects. The actual age and estimated age of the subjects were then compared to determine the precision and accuracy of the established model.

4. Statistical analysis

Statistical analysis was performed using SPSS® Statistics 19.0 (SPSS, Inc., Chicago, IL).

Table 3
Age and sex distribution of group samples used for method validation.

Age (year)	Male	Female	Maxillary first molars	Mandibular first molars
12–20	9	9	18	18
21–30	10	10	20	20
31–40	10	11	20	20
41–50	10	10	20	19
51–60	11	11	17	19
61–67	7	4	9	7
Total	57	55	104	103

Paired-*t* test was used to determine the statistical significance of measurement accuracy and inter- and intra-observer variability. A p value of 0.05 or less was considered significant.

Independent-sample t test was applied to comparing the difference of the pulp chamber volume among male and female, maxillary first molars and mandibular first molars. A p value of 0.05 or less was considered significant.

Mean absolute error (MAE) and root mean square error (RMSE) between the actual and estimated ages were used to determine the precision and accuracy of the mathematical models.

5. Results

The calculated volume of pulp chamber in the group samples used for the method establishment ranged from 7.5465 mm^3 to 69.2447 mm^3 with a mean value of 26.2547 mm^3 (SD = 10.0869). The distribution of volume in every age section is shown in Fig. 2 for both group samples used for method establishment and method validation.

An independent-sample t test showed that the difference in volume between genders was statistically significant ($p = 0.013$), so was the difference between the volumes of maxillary molars and mandibular molars ($p = 0.028$).

The equation from the logarithmic regression analysis for all teeth was:

$$\text{AGE} = 117.691 - 26.442 \times \ln(\text{pulp chamber volume})$$

For unknown sex, the equations obtained from logarithmic regression analysis and separately used for maxillary first molars and mandibular first molars were:

$$\text{Maxillary first molars}: \text{AGE} = 134.837 - 31.451 \times \ln(\text{pulp chamber volume})$$

$$\text{Mandibular first molars}: \text{AGE} = 122.927 - 28.989 \times \ln(\text{pulp chamber volume})$$

Since the difference between genders and tooth positions are significant, the equations obtained from logarithmic regression

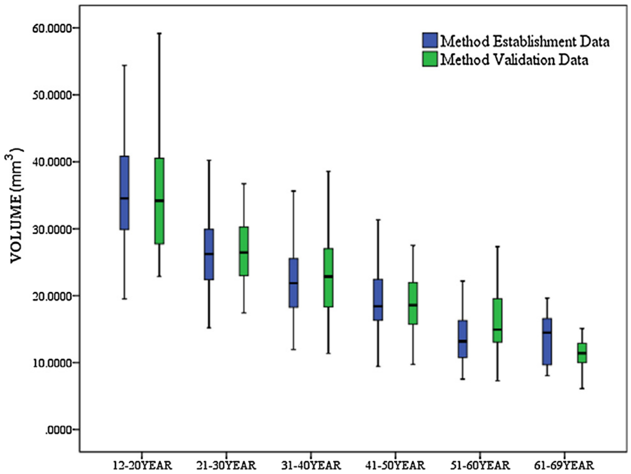

Fig. 2. The distribution of pulp chamber volume in different age groups used for method establishment and method validation.

analysis and separately used for male maxillary first molars, male mandibular first molars, female maxillary first molars and female mandibular first molars were:

$$\text{Male maxillary first molars}: \text{AGE} = 118.456 - 25.67 \times \ln(\text{pulp chamber volume})$$

$$\text{Male mandibular first molars}: \text{AGE} = 118.398 - 26.756 \times \ln(\text{pulp chamber volume})$$

$$\text{Female maxillary first molars}: \text{AGE} = 131.455 - 30.685 \times \ln(\text{pulp chamber volume})$$

$$\text{Female mandibular first molars}: \text{AGE} = 119.519 - 28.182 \times \ln(\text{pulp chamber volume})$$

The regressions were statistically significant ($p = 0.000$).

Scatter diagram shows the relationship between the volumes of pulp chamber and ages for all teeth is shown in Fig. 3.

No significant differences were found for inter-observer ($p = 0.347$) and intra-observer ($p = 0.202$) variances.

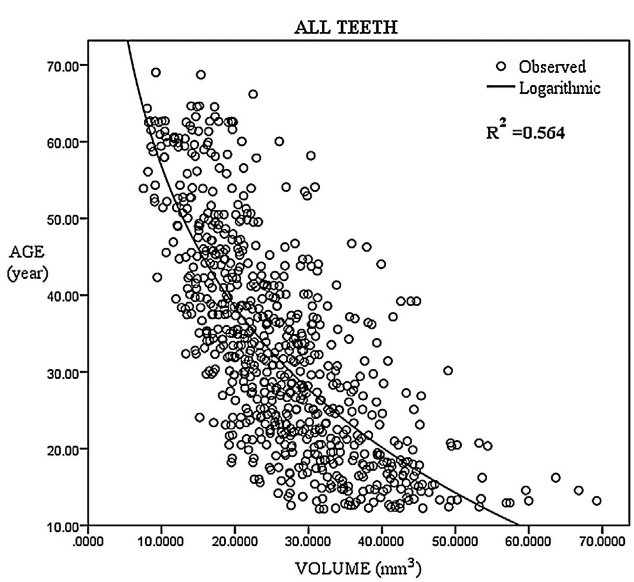

Fig. 3. Scatter diagram shows the relationship between the volume of the pulp chamber and age for all teeth.

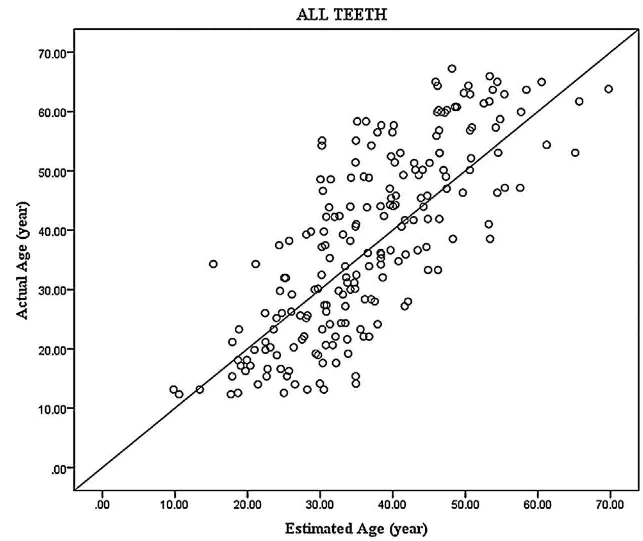

Fig. 4. Plots of actual age versus estimated age with logarithmic regression model for all teeth.

There was a significant difference between the pulp volumes obtained from Micro CT and CBCT image ($p = 0.024$) with an average difference of 2.3%.

Plots of actual age versus estimated age with logarithmic regression model for all teeth is shown in Fig. 4.

Mean absolute error (MAE) and root mean square error (RMSE) between the actual age and estimated age for all teeth, female maxillary first molars, male maxillary first molars and female mandibular first molars, male mandibular first molars in different age groups are shown in Table 4.

6. Discussion

Due to the apposition of secondary dentine, the decreased volume of pulp chamber was generally accepted as an age estimation indicator. Although couples of 2D imaging methods like panoramic radiography [20] and periapcial radiography [21] have been applied to analyzing the real 3D decrease of pulp chamber restrictively, it seems that the use of 3D images which demonstrate the real morphological change is the most suitable one in this kind of dental age estimation methods.

CBCT is a newly developed 3-Dimensional imaging modality. It uses a 2D X-ray detector and a cone- or pyramid-shaped X-ray beam to reconstruct isotropic high-spatial-resolution 3D images [32]. Advantages of CBCT imaging are its easy accessibility, ease of handling, and ability to offer (from a single scan) a dataset of multi-planar cross-sectional and 3D reconstructions [33]. To acquire three-dimensional datasets, CBCT seems having some privilege over micro-CT or medical CT. First, CBCT can provide a relatively large scanning area while Micro CT only has a confined scan area in which one extracted tooth can be scanned at a time. Second, the radiation dose is high in a high resolution Micro-CT image [34]. Moreover, extracted teeth are needed for a Micro CT scan, which is not acceptable for a live person. CT imaging can be acquired for a live person but it needs relatively high cost and radiation dose compared with CBCT [35,36].

Contrast to other studies in which pulp cavity to tooth volume ratio was used as an indicator for estimating human age, the volume of pulp chamber was employed in the present study. The reasons why we chose the volume of pulp chamber as the indicator are as follows. First, the age-related formation of secondary dentine is directly related to the decrease of pulp

Table 4
Mean absolute error (MAE) and root mean square error (RMSE) between the actual age and estimated age using models obtained from all teeth, male maxillary first molars, male mandibular first molars, female maxillary first molars and female mandibular first molars and the coefficient of determination of each model.

	All teeth		Male maxillary first molars		Male mandibular first molars		Female maxillary first molars		Female mandibular first molars	
R^2	0.564		0.544		0.562		0.684		0.612	
Age (year)	MAE	RMSE	MAE	RMSE	MAE	RMSE	MAE	RMSE	MAE	RMSE
12–20	8.172	5.604	7.44	5.51	6.206	6.028	7.161	4.799	8.843	3.961
21–30	6.009	4.363	5.722	4.474	6.261	5.005	5.822	4.043	4.874	3.932
31–40	6.707	4.451	5.092	4.305	6.451	4.343	7.188	3.839	6.059	5.072
41–50	6.851	4.899	7.667	4.255	6.922	5.493	7.221	3.573	5.836	4.417
51–60	12.163	6.402	12.607	5.413	13.68	6.606	9.009	6.614	12.328	5.296
61–67	10.838	4.844	10.424	4.583	11.765	4.614	2.215	2.356	13.64	1.361
Average	8.122	5.603	7.922	5.436	8.216	6.185	6.91	4.75	7.801	5.369

cavity volume while the volume of an entire tooth was mainly affected by the attrition of enamel. Thus, a pulp cavity/tooth volume ratio may not reflect the real change from secondary dentine apposition [24]. Second, the pulp chamber volume calculation was more accurate than the volume calculation of whole tooth because of high image contrast between dentine and pulp chamber [22].

To determine the measurement accuracy, the volume calculated from Micro CT images was used as the reference standard. Micro CT can provide highly accurate and precise assessment of internal dental structures and root canal morphology [37–40] and has been considered as a reference standard in dental liner and volumetric measurements [41–44]. In the present study, a significant difference was observed between the pulp volumes obtained from Micro CT and CBCT images ($p = 0.024$), but the average difference is relatively small (2.3%) when compared it to the difference reported in a previous study (maximally 21%) [22].

In the analysis of the previous studies using CBCT or CT images, we find a tendency that the coefficient correlation between the pulp cavity/tooth volume ratio and age increased with the sample size enlarged, for example, the coefficient of determination in one study including only 28 teeth is 0.29 [25] while in another study having 136 teeth, the coefficient of determination goes up to 0.571 [26]. In the present study, a relatively high coefficient of determination was demonstrated for the pulp chamber volume of the first molar and age (R^2 ranged from 0.544 to 0.684 based on different gender and tooth position). When compared with the present study with the studies by Someda et al. ($R^2 = 0.65$ for male, 0.77 for female in mandibular central incisors) [24], by Agematsu et al. ($R^2 = 0.67$ for male, 0.75 for female in mandibular central incisors) [27] and by Aboshi et al. ($R^2 = 0.635$ for lower first premolars and 0.703 for lower second premolars) [28], a relatively low determination coefficient was found in the present study. This may due to the fact that these studies were performed with Micro-CT, which can provide much higher spatial resolution than dose a CBCT unit and hence a much more accurate measurement. This can also explain why a significant difference was found between the pulp volumes obtained from Micro CT and CBCT images.

Opposite to the previous studies [22,23,26], an independent-sample t test in the present study showed that the difference in volume between genders was statistically significant. Someda et al. [24] and Agematsu et al. [27] performed regression analysis for age estimation on the basis of the correlation between age and decrease in pulp chamber volume using Micro-CT images of the mandibular second premolars and/or mandibular central incisors as samples, and reported that age may be more reliably estimated by selecting an estimation equation that considers sex. The results of the present study support their results and provide further evidence for a distinct sex difference. The observed relation between the volume of pulp chamber and age was stronger for female than for male in the present study, and it is in agreement with previous studies [22,24,27]. The difference between the volume of maxillary molars and mandibular molars ($p = 0.028 < 0.05$) was also statistically significant.

In the mathematical models validation test, the MAE of 8.122 and RMSE of 5.603 between the actual and estimated ages for all teeth, the MAE of 7.666 for maxillary first molars and the MAE of 8.243 for mandibular first molars were observed. Although this discrepancy between estimated and actual ages is the same as those obtained in the previous studies on the age estimation of adults by the use of single rooted-teeth, it is still relatively large [23]. Further analysis demonstrated that the MAE between the actual age and estimated age was great in the age groups of 51–60 years old and 61–69 years old. An independent sample t test indicated no significant difference in the distribution of pulp chamber volume between 51–60 year and 61–69 year in the samples used for method establishment($p = 0.254$). So the great MAE in 51–60 year old group and 61–69 year old group maybe due to the unremarkable decrease of pulp chamber volume between the groups of age 51–60 and 61–69. This is similar to the previous studies reporting that the amount of secondary dentine was larger in the young stage than in the old stage [28,45]. Meanwhile, the great MAE in 51–60 year old group and 61–69 year old group may be also because the study sample in the 51–69 years old groups is relatively small due to the fact that older people are more prone to teeth lost and molars are more prone to decay. Another possibility for the relatively large discrepancy between the estimated and actual ages may be the variability in pulp volume between individuals. The more the pulp chamber volumes of individuals within the same age group dispersed, the less precise and accurate age estimations can be obtained.

Other than previous studies in which linear mathematical models were established, a logarithmic model was developed in the present study. To compare with logarithmic models with linear models, a linear regression analysis was also conducted with age as dependent variable, pulp chamber volume as independent variable for maxillary first molars and mandibular first molars, respectively, in the present study. MAE and RMSE between the actual and estimated ages using both logarithmic and linear models for maxillary and mandibular first molars were subsequently obtained. The MAEs, RMSEs and the coefficient of determination of each model were shown in Table 5. Logarithmic model presents a little bit better results than the linear models. Further analysis using paired-t test demonstrated that the difference between the estimated ages of the 112 patients in the model validation group obtained from logarithmic model and linear model is significant in mandibular first molars ($p = 0.027$) and insignificant in maxillary first molars ($p = 0.222$).

Table 5
MAE and RMSE between the actual and estimated ages using logarithmic models and linear models obtained from maxillary first molars and mandibular first molars, and the coefficient of determination of each model.

	Maxillary first molars				Mandibular first molars			
	Logarithmic models		Linear models		Logarithmic models		Linear models	
R^2	0.66		0.607		0.604		0.542	
Age (year)	MAE	RMSE	MAE	RMSE	MAE	RMSE	MAE	RMSE
12–20	6.581	5.152	8.544	6.667	7.289	5.087	9.294	5.483
21–30	5.796	3.962	7.113	4.438	5.256	4.255	6.774	4.521
31–40	7.288	5.305	6.984	5.757	7.046	5.732	6.948	4.829
41–50	8.07	4.696	6.062	3.848	6.865	5.246	5.623	4.685
51–60	11.139	6.721	10.619	6.288	13.337	6.568	13.127	5.486
61–67	7.372	5.819	11.404	3.935	12.572	4.357	15.658	3.451
Average	7.666	5.522	8.078	5.634	8.243	6.109	8.811	5.846

One limitation of the present study is that only first molars were included. First molars are more constantly lost and restored due to caries than all the other teeth. This may limit the use of the developed model for age estimation, especially for old age people. For young persons, however, this may have little influence since there are a total of four first molars locating in each side of dentition. Rarely are all the four first molars lost or restored at a time.

7. Conclusion

The present study investigated the relationship between age and pulp chamber volume of multi-rooted first molars. The pulp chamber volume of first molar is a useful index for human age estimation with reasonable precision and accuracy. Gender and tooth position plays an important role in the estimation of age with first molars. Therefore, the use of gender and tooth position specific age estimation equations is recommended when using the pulp chamber volume of first molars for the age estimation. cone-beam CT in dental use provides plenty of 3D volume information of teeth on living individuals in the target area by a single scan but has a relatively low measurement accuracy compared with Micro CT. Future awaited ameliorations in CBCT technology with a more homogeneous age distribution of study samples may provide an optimized dental age estimation technique.

Conflict of interest statement

The authors have no relevant conflicts of interest to declare.

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