Age estimation for children and young adults by volumetric analysis of upper anterior teeth using cone-beam computed tomography data

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Background: The aim of this study was to investigate the association between chronological age and the pulp/tooth volume ratio (PTR) of specific teeth using cone-beam computed tomography (CBCT) enhanced with Materialise-Mimics Research software 21.0 in children and young adult population from Eastern China. **Materials and methods:** CBCT scans of 230 patients (119 males, 111 females), aged 8.18–19.92 years were analysed by two well-trained examiners in this retrospective study. The intraclass correlation coefficient value was calculated to test the intra- and inter-examiner agreement. The volumetric analysis of the pulp and calcified tissues was performed on the maxillary left central incisors and canines. The correlation and regression analyses were then performed.

Results: The Pearson correlation analysis showed a strong coefficient of correlation (r) for maxillary left canines (-0.81 for girls and -0.88 for boys) as compared to central incisors (-0.63 for girls and -0.70 for boys). Regarding performance, the canine model was more powerful than the central incisor model. The derived regression equation from maxillary left canines had high coefficients of determination (Age = $21.979 - 105.42 \times PTR$, $R^2 = 0.69$).

Conclusions: Our study proved that the PTR value of canines had a negative correlation relationship with a subadult's chronological age and volumetric analysis of CBCT scans using the software may become an efficient method to estimate the chronological age of children and young adults. (Folia Morphol 2020; 79, 4: 851–859)

Key words: dental age estimation, Mimics Software, Chinese children, cone-beam computed tomography, volumetric analysis

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INTRODUCTION

Age estimation has vital importance in civil and criminal proceedings and clinic work and was generally utilised in identity verification of human remains, adjudication of criminal responsibility and determination of clinical treatment plans [15, 18]. Dental age is an efficient method to estimate the chronological age of humans. The advantages of dental age assessment over other methods are its simplicity and less time consumption. In many circumstances, tooth tends to be the only remained tissue of human remains we could utilize to estimate the chronological age quickly, since dental enamel is the hardest tissue in the human body with exceptional resistance to physical and chemical forces and tooth development is regulated under strict genetic control [7, 13, 16, 21].

Systematic research continued for several decades [23], and many methods were proposed to estimate the dental age of children. The most widely used methods included the gualitative methods that relied on the manual assessment of the shape of the developing tooth by the naked eye, such as Demirjian's method [9] and Willems's method [27], and qualitative method, such as Cameriere's method [6]. As most of the techniques were based on samples from a foreign population, many scientists attempted to test their applicability to the Chinese population. Ye et al. [28] selected 941 orthopantomograms of 7-14-year-old children from Eastern China and found that the Willems's method was more accurate in estimating the dental age than the Demirjian's method [28]. For the 11–18-year-olds from Eastern China, the Demirjian's method was more reliable compared to the Willems's method according to Wang et al. [26]. They conclude that the Demirjian's method was unsatisfactory for 11-18-year-olds from Eastern China [26]. Evidently, several researches showed different, even contradictory results. This could be because manual observation of the shape of the tooth was readily influenced by the personal interpretation.

The shape of a tooth changes throughout life, especially during its developing stage. The apical foramen becomes smaller in diameter; the crown develops with the formation of enamel. Under this circumstance, Cameriere et al. [6] estimated the dental age of children quantitatively by the extent of the apical foramen's opening (the Cameriere's method). They assessed the maturity of teeth by recording the number of teeth with completely closed apical foramen, the distance between the inner point of the open apical foramen and the length of the teeth [6].

In fact, even after completion of tooth development, the tooth's morphology continues to show age-related changes. With the deposition of secondary dentin along the inner walls of the pulp chamber, the pulp/tooth ratio (PTR) reduces continually [4, 17]. Hence, there is an obvious inverse correlation relationship between chronological age and the PTR value and many studies started using this parameter to estimate the chronological age of adults [1-3, 8, 10-12, 14, 20]. Cameriere et al. [6] collected orthopantomograms of 100 Italian, White, Caucasian subjects, between 18 to 72 years of age. PTR, tooth length, pulp/tooth length ratio, pulp/tooth area and pulp/root width ratios of maxillary right canines at three different levels were computed and the ratio between pulp and tooth area correlated best with age $(r^2 = 0.85)$ [5].

However, all the methods mentioned above are based on the two-dimensional (2D)-panoramic radiographs and there is always some detailed information of teeth is lost.

With the development of medical technology, cone-beam computed tomography (CBCT) has become a valuable tool in dental clinics. This is due to its lower radiation dose and cost-efficacy as compared to traditional medical computed tomography. Moreover, it has greater metric accuracy with isotropic voxel resolutions [19]. The CBCT image analysis can be enhanced by Materialise-Mimics Research software 21.0. Using its segment function, we can now generate the three-dimensional (3D) model of the scanned structure and acquire its superficial area and volume [26], then the pulp/tooth ratio could be calculated.

In addition, many previous studies focusing on the relationship between chronological age and the pulp/tooth volume ratio included samples of all age groups from young to old, not a specific age group [2, 3, 12, 29]. Thus far, no study to date has used PTR value to estimate dental age of children.

So, our interest was aroused that if the PTR value derived from CBCT data could be utilized to estimate the chronological age of children.

As the canines are the most stable teeth in situ and subject less wearing [8, 24], the central incisors had been studied in many researches which are convenient to be compared and both teeth types are mono-radicular. So, we selected the maxillary left canines and the central incisors to examine the efficacy of the CBCT technique for dental age estimation of children from Eastern China.

MATERIALS AND METHODS

Ethics issues

The research was approved by the independent ethics committee of the Shanghai Ninth People's Hospital affiliated with Shanghai Jiao Tong University, School of Medicine (No: 2017-284-T212).

Sample and eligibility criteria

Cone-beam computed tomography data were randomly selected from 8–19-year-old patients at Shanghai Ninth People's Hospital between January 2015 and May 2019. All CBCT data was obtained for diagnosis and therapeutic purposes, and no subjects suffered additional radiation exposure.

The inclusion criteria were Han Chinese between 8 to 19 years of age and clear CBCT image.

The exclusion criteria were severe jaw defects or deformities, missing maxillary central incisors or canines, severe caries or defects on maxillary central incisors or maxillary canines, and severe systemic disease.

Data collection

All the CBCT data were acquired using the same Planmeca ProMax 3D Max machine. The pixel size was 200, and the slice thickness was 0.2 mm. The selected scans had an exposure parameter of 96 kV, 8.0 mA, and the exposure time was approximately 12 s. The dose area product was 1289 mGy \times cm².

A total of 230 children (aged 8–19 years) were included in our study. The samples were divided into 12 groups, according to age. Ten boys and ten girls were included in each group to ensure balance in data component distribution. For each subject of our study, we collected the following details: age, sex, date of birth, date of CBCT scan, and the DICOM information of the CBCT data. Our study included 228 maxillary left incisors and 229 maxillary left canines. The specific composition of our samples is shown in Tables 1 and 2.

Measurements

For the training group, 10 samples were randomly selected to calculate the intraclass correlation coefficient (ICC). The PTR values of these 10 maxillary left canines were measured separately at the same time by two examiners to test the inter-examiner agreement

Age	Boys	Girls	Total
8	10	8	18
9	8	8	16
10	10	10	20
11	12	7	19
12	8	10	18
13	11	10	21
14	9	8	17
15	10	9	19
16	12	9	21
17	9	10	19
18	9	11	20
19	11	11	22
Total	119	111	230

 Table 2. The distribution of examined teeth according to age group and sex

Age [years]	Toot	h 21	Toot	h 23	Total
	Boy	Girl	Boy	Girl	-
8	10	8	10	8	36
9	8	8	8	8	32
10	10	10	10	10	40
11	12	7	12	7	38
12	8	10	8	10	36
13	11	10	11	10	42
14	9	8	9	8	34
15	10	9	10	9	38
16	12	9	12	9	42
17	7	10	9	9	35
18	9	11	9	11	40
19	11	11	11	11	44
Total	117	111	119	110	457

and remeasured by one examiner with 2 weeks' interval to test the intra-examiner agreement. Then we measured the PTR value of the maxillary left central incisor and canines for each sample. If the maxillary left central incisor or canine was missing or could not be analysed due to severe caries or defects, its counterpart on the other side was analysed.

Firstly, the DICOM files, which contained 651 independent subfiles, were imported into the software to construct the 3D models. The number of slices was sufficient to obtain the precise volume of the hard tissue and pulp for each tooth. The more slices

Table 1. The distribution of samples according to age and sex



Figure 1. Threshold range selecting process.



Figure 2. Multiple slice editing process (axial plane).

one CBCT sample had, the smaller slice thickness it would have, and we could precisely define the tooth structure and obtain more accurate data.

Secondly, the appropriate threshold range was set to highlight the hard tissue of the tooth (Fig. 1). Once determined, it would not be altered during the entire process of 3D model construction. The false-positive highlight area around the hard tissue of the tooth, that generated due to the short width of periodontal attachment, the similar density of the bone around the tooth cementum, or other mixed pixels, was manually removed from the highlight area using multiple slice editing function in the axial plane to ensure the accuracy of the volume data obtained. (Figs. 2, 3) The region growing function was used to separate the tooth from other tissues and generated a new mask with a different colour.

Thirdly, we separated the hard tissue from the pulp cavity of the tooth by cavity fill function. The pulp cavity area of the tooth was surrounded by the highlight area of the hard tissue, which could be filled in using another highlight colour and a new mask representing the pulp cavity was then constructed. For those teeth whose apical foramen were still open, we sealed the mask in the apical foramen of the tooth using the adding functions of the multiple slice editing functions to turn the pulp cavity from an open space into a closed space. Since we analysed two teeth for each sample, the hard tissue area and the pulp cavity area of each tooth needed two highlight

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Figure 3. Multiple slice editing process (sagittal plane).

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Figure 4. Construction of the three-dimensional model and the volume measuring process.

colours, respectively. In that way, five different masks with five different colours would be created for each sample in the end.

Lastly, the four masks which represented the hard tissue and the pulp cavity of the maxillary central incisor and the maxillary canine respectively were transformed to 3D models, and the volume (mm³) of each part could be calculated by the properties function (Fig. 4). In the end, the pulp/tooth value of each tooth could be obtained.

Statistical analysis

All statistical values were analysed using Microsoft Excel 2019 and the SAS V8 and the significance level for this study was set to 5%. The following morphological variables were recorded for each sample's maxillary left central incisor and canine: pv = the volume of pulp cavity, tv = the volume of the entire tooth, and PTR = pulp/tooth volume ratio. The Pearson correlation coefficient between the PTR value and the chronological age was calculated.

Fisher Z test analysis was performed to test the statistical difference in the correlation coefficient values between the central incisor model and canine model.

Linear regression was performed using chronological age as the dependent variable and PTR value as the independent variable. The standard errors of estimate were then calculated to analyse the reliability of different regression equations.

RESULTS

The main error came from two points: 1) The threshold range selected for the segment process; 2) The multiple slices editing operation to separate the tooth from the surrounding tissues.

To eliminate the error, we calculated the interexaminer (ICC = 0.989, 95% CI(L) = 0.955, 95%CI(U) = 0.997) and intra-examiner (ICC = 0.973, 95%CI(L) = 0.973, 95%CI(U) = 0.993) agreement, which showed an excellent reproducibility of this measurement.

The sample comprised of 119 boys and 111 girls. The mean, standard deviation, minimum and maximum of the ages of all boys and girls was shown in Table 3. The mean, standard deviation, minimum and maximum of the PTR values of all the analysed teeth were shown in Table 4.

There was a significant correlation between chronological ages and PTR of central incisors or canines for both boys and girls (p < 0.01). The correlation coefficients were -0.70 (boy group), -0.63 (girl group) and -0.67 (global samples) for central incisors, and -0.88 (boy group), -0.81 (girl group) and -0.83 (global samples) for canines, respectively. Fisher Z test showed a significant difference in the correlation coefficient values between maxillary central left incisor and maxillary left canine. However, there was no significant difference in the correlation relationship between genders for both tooth positions (Tables 5, 6). In this circumstance, we combine the data and two regression equations were derived from the acquired data. The higher $R^2 = 0.69$ was found from the regression equation derived from the maxillary canines which meant that the regression equation could explain 69% of the total variance of age. The standard error of estimate (SEE) of two estimation models was calculated. The results showed that the maxillary left canine model had the smaller SEE than the central incisor model (Table 7).

The scatter plot of chronological age-pulp/tooth ratio for all three models is shown in Figures 5 and 6.

 Table 3. The mean, standard deviation (SD), minimum (Min) and maximum (Max) of the ages of all boys and girls

Sex	Mean	SD	Min	Max
Boy	13.99	3.45	8.19	19.92
Girl	14.29	3.47	8.48	19.92

 Table 4. The mean, standard deviation (SD), minimum (Min)

 and maximum (Max) of the pulp/tooth volume ratio values of

 all the analysed teeth

Sex	Mean		Mean SD		Min		Max	
	21	23	21	23	21	23	21	23
Boy	0.053	0.080	0.024	0.030	0.016	0.032	0.137	0.148
Girl	0.049	0.069	0.020	0.022	0.006	0.022	0.105	0.124
Total	0.051	0.075	0.022	0.027	0.006	0.022	0.137	0.148

Table 5. Fishers Z test to test the significant difference in the correlation coefficient between maxillary left central incisor and canine

Correlation	coefficient	Fisher	s Z test
21	21 23		P value
-0.665	-0.833	12.07	< 0.05

Table 6. Fishers Z test to test the significant difference in the correlation coefficient between genders for both tooth position

Model	Correlation	coefficient	Fishers Z test		
type	Boy	Girl	Z value	P value	
21	-0.70	-0.63	0.94	> 0.05	
23	-0.88	-0.81	1.72	> 0.05	

Table 7. The comparison of the regression equation between

 the maxillary left central incisor group and canine group

Tooth	Ν	R ²	Regression equation	SEE
21	228	0.44	$Age = 19.35 - 102.73 \times PTRI$	2.58
23	229	0.69	$Age = 21.979 - 105.42 \times PTRC$	1.91

PTRC — the pulp/tooth ratio value of canine; PTRI — the pulp/tooth ratio value of central incisor; SEE — standard error of estimate

DISCUSSION

Several scientists have studied the correlation between age and decrease in pulp volume caused by the deposition of secondary dentin and applied this phenomenon to age estimation. As the tooth is a 3D structure, dental age could not be accurately assessed from 2D orthopantomograms, especially



Figure 5. The relationship between the chronological age and the pulp/tooth ratio (PTR) value of the maxillary central incisor.



Figure 6. The relationship between the chronological age and the pulp/tooth ratio (PTR) value of the maxillary left canines.

when we could not get enough teeth from human remains [11, 20].

Many studies have proved that there exists no statistically significant difference between the right and left counterpart teeth in determining the efficacy of dental age estimation [3, 29]. Hence, we only analysed the teeth on the left side or the counterpart tooth in the contralateral quadrant, if the tooth on the left side was lost or malformed.

In our study, the correlation between the PTR and the chronological age in maxillary canines (r = -0.833) was higher than that found in maxillary central incisors (r = -0.665). The maxillary left canine estimation model, that was better than the central incisor estimation model, had a coefficient of determination of 0.77 for boys and 0.66 for girls. Gulsahi et al. [12] observed both maxillary central incisors and maxillary canines in one study, and the strongest correlation strength was found between the chronological age and PTR measured on maxillary central incisors ($r^2 = 0.532$). Asif et al. [3] also calculated the PTR ratio of maxillary canines and central incisors using CBCT data and got the similar results. Some

studies also analysed the maxillary central incisors or the maxillary canines using CBCT data but without calculating the PTR value. Asif et al. [2] analysed both the PTR and the pulp chamber/crown ratio and found that the second method had a higher R² (0.775) than the PTR method. Ignoring the tooth position, Zhang et al. [29] found that the scope of R² by CBCT images for age estimation was about 0.21–0.70.

In general, the strength of correlation in our study seems stronger than the previous studies. The reason may the decrease of PTR value was caused by different factors in subadults and adults. For subadults, the shape of tooth mainly changed with the deposition of enamel, dentin, and cementum and the closure of apical foramen. For adults, the shape of the tooth principally changed with the deposition of the secondary dentin. In that circumstance, it was reasonable to find different extents of correlation relationship between young people and people of all age.

Most studies showed a stronger correlation in the maxillary central incisor than the maxillary canines, which was contrary to our study. So, we may conclude that the PTR value of a central incisor was the better reference variance to estimate the age of adults comparing to a canine, while a canine is better when estimating the age of subadults. This could be because the age of apex formation for canines was 13-15 years, but for central incisors, it was around 10 years. Since the age range of our samples was 8-19 years, the variation of shape was more apparent and more sustainable in maxillary canines than the central incisors. Hence, it is guite reasonable to find that the association between PTR values and the chronological age in maxillary canines was stronger than the maxillary central incisors for children and young adults.

Our study didn't show a distinct sex dimorphism in age estimation. However, there are differences in the growth pattern between boys and girls. The growth spurt of girls precedes that of boys by approximately 2 years. Many methods for estimating chronological ages of children, such as the Demirjian's method and the Willems's method, showed significant sex dimorphism. However, most studies analysing the PTR values found no significant difference in the correlation coefficient values between genders for each type of investigated teeth [3, 12]. This might be because all the subjects were adults and the dentin deposition pattern probably the same for males and females whose growth and development had ceased. For our study, the relatively small number of samples may be the cause. In fact, the Z value (Z = 1.72) was quite close to 1.96 when we compared the R values between genders. If we had collected more samples, the result may well be different.

In addition, the method we used to acquire the pulp and tooth volume was different from other studies, and the differences were shown in the modelling setup process after the CBCT data in DICOM files were imported into the software. In previous studies, new masks were created for the pulp cavity and the entire tooth after setting different grayscale threshold values for each of the investigated teeth [3]. In our opinion, multiple sets of grayscale values meant a higher probability of error. In our study, we set the grayscale value once to separate the hard tissue of the tooth and used the cavity filling function to define the pulp area. It was not only more accurate, but also less time-consuming. Most of the studies reported that the entire time of importing the DICOM files to the software, thresholding, and 3D reconstruction took around 2-3 hours per tooth [3, 22, 25]. Pinchi et al. [20] simplified dental volume measurement through a geometric approximation of the different parts of the tooth and spent less than 15 minutes per tooth. Through one-step thresholding and multiple slice editing, we could finish the 3D reconstruction process in about 20 minutes without geometric approximation.

We couldn't compare the efficiency of our new equation with Demirjian's or Williems's method since there wasn't an independent validation group, whereas it is very important. Since the 2D technique is less time-consuming, our method merits application only if it is more accurate than the Demirjian's or Williems's method. We decided to screen samples who took CBCT and panoramic radiographs on the same day to conduct the follow-up investigation.

CONCLUSIONS

We concluded that the maxillary left central incisors and left canines had a significant negative correlation relationship with the chronological age of children and young adults. The calculation of pulp-tooth ratio value using the data acquired from CBCT might be an effective method to estimate the chronological age of children and young adults. It is more reliable to estimate the chronological age by the PTR value of the maxillary left canines than left incisors and two regression equations were established respectively using the pulp-tooth value of the maxillary left canines and central incisors as the dependent variance. The accuracy of this estimation method is considerably high, whereas it is necessary to compare the accuracy of our new regression equation with other widely used methods using independent validation samples. Hence, there is a need to carry out further studies in the future.

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