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Age estimation by canines' pulp/tooth ratio in an Iranian population using digital panoramic radiography

Mahdieh Dehghani[a], Elaheh Shadkam[b,*], Farzaneh Ahrari[c], Mahboobe Dehghani[c]

[a] Department of Oral and Maxillofacial Radiology, Faculty of Dentistry, Shahid Sadoughi University of Medical Sciences, Yazd, Iran
[b] Department of Orthodontics, School of Dentistry, Mashhad University of Medical Sciences, Vakilabad Blvd., Mashhad, Iran
[c] Dental Research Center, Mashhad University of Medical Sciences, Mashhad, Iran

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ABSTRACT

Objective: Age estimation in adults is an important issue in forensic science. This study aimed to estimate the chronological age of Iranians by means of pulp/tooth area ratio (AR) of canines in digital panoramic radiographs.
Materials and methods: The sample consisted of panoramic radiographs of 271 male and female subjects aged 16–64 years. The pulp/tooth area ratio (AR) of upper and lower canines was calculated by AutoCAD software. Data were subjected to correlation and regression analysis.
Results: There was a significant and inverse correlation between age and pulp/tooth area ratio of upper and lower canines ($r = -0.794$ for upper canine and $r = -0.282$ for lower canine; p-value < 0.001). Linear regression equations were derived separately for upper, lower and both canines. The mean difference between actual and estimated age using upper canine was 6.07 ± 1.7.
Conclusion: The results showed that the pulp/tooth area ratios of canines are a reliable method for age estimation in Iranians. The pulp/tooth area ratio of upper canine was better correlated with chronological age than that of lower canine.

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1. Introduction

Age estimation is a very important field in forensic science. It can be applied for identifying victims, criminals and those damaged in accidents, and it is sometimes useful in other purposes such as signing up for school, recruitment, and some social activities [1,2]. Dental maturation has been recently considered as a valuable index for age estimation in dead and living persons [3–5]. In contrast to bone mineralization, the incremental pattern of tooth formation is not significantly affected by diseases, drug intake or endocrine status [6]. Furthermore, teeth are resistant to mechanical, chemical and thermal stimuli as they are the hardest structures in human body. Therefore, numerous methods have been applied to estimate chronological age based on dental tissues [6–8]. These methods can be broadly classified as morphological and radiological methods. Some methods based on morphologic changes of teeth are invasive and warrant tooth extraction, which is not possible in living individuals [7,9]. However, it is simple and practical to use methods that include analysis of radiographic images for age estimation in living subjects [10,11].

The analysis of radiographic images is a relatively accurate procedure for age estimation in children, because the developmental stage of teeth, as observed in periapical or panoramic radiographs, is well correlated with chronological age [12]. However, radiographic-based techniques cannot be easily applied for determining age in adults [13]. Indeed, after third molar development, only aging process and regressive changes in teeth can be employed to estimate chronological age [12]. It has been clarified that with increasing age, the size of the dental pulp cavity reduces as a result of continual apposition of secondary dentin by odontoblasts [14,15]. In 1995, Kvaal et al. [16] introduced a non-destructive dental method for calculating age of adults based on measurements of teeth and their radiographs. They measured pulp length and width as well as root length and width on the radiographs and found a negative correlation between age and the ratios between the pulp and the root width. In 2004, Cameriere et al. [17] detailed a radiographic method for age estimation of adults using single rooted teeth. They measured different dimensions (length, width, area) of pulp, root and tooth in right maxillary canines of 100 Italian Caucasian patients and revealed that the ratio between pulp and tooth area was best correlated with age ($r^2 = 0.85$). The correlation between age and the radiographic measurement of pulp to tooth area ratio (AR) has been the subject of interest for many investigators and the applicability of this

* Corresponding author.
E-mail address: shadkame941@mums.ac.ir (E. Shadkam).

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technique has been validated in different ethnic groups. Several studies indicated that pulp/tooth area ratio is a reliable and simple technique for age estimation in adults [2,18–20]. Canine teeth have been used for age estimation in most studies, as they are single-rooted teeth with the largest pulp area which makes the radiographic analysis easier. Furthermore, they have lower chance of wear than other anterior and posterior teeth and are frequently present in the mouth of older subjects [21]. Other studies found that AR ratio of other single-rooted teeth can also be employed for estimating age of adults [22,23].

The present study was conducted to assess the chronological age of Iranian adults using pulp/tooth area ratio of maxillary and mandibular canines in panoramic radiographs. The aim was to derive a population-specific formula based on Cameriere's method for age estimation.

2. Materials and methods

The protocol of the study was reviewed and approved by the Ethics Committee of Research Deputy of Shahid Sadoughi University of Medical Sciences, Yazd, Iran and the study was conducted in accordance with the Declaration of Helsinki.

The sample consisted of 300 digital panoramic radiographs belonging to patients referred to the Department of Oral and Maxillofacial Radiology of Yazd Dental School for routine dental treatments. The subjects consisted of 156 males and 144 females with age range of 16–64 years. The inclusion criteria were the presence of right or left maxillary and mandibular canines in the mouth with fully formed roots in panoramic radiographs. The radiographs were excluded from the sample in the presence of the following conditions: canines with root fillings or any pathology such as caries, periodontitis or periapical lesions, rotated canines with large areas of enamel overlap with neighboring teeth, canines with any prosthetic fittings and orthodontic appliances, fractured canines, canines showing severe attrition secondary to parafunctional habits, and canines with any developmental anomalies. The patient's ID number, date of birth and sex were recorded at the start of the study.

All the radiographs were taken using a digital device (Proline, Planmeca, Finland) under the following conditions: voltage: 66–70 kV, intensity: 8–10 mA and duration: 10–12 s. Radiographs were saved as high resolution JPEG files on a computer. Images were then imported to AutoCAD software (2013). In order to measure the pulp/tooth area ratio of canines, 20 points were identified on the tooth outline and 10 points were marked on the pulp periphery (Fig. 1).

Afterwards, the points on pulp and tooth outlines were connected to calculate the area of the tooth and pulp chamber.

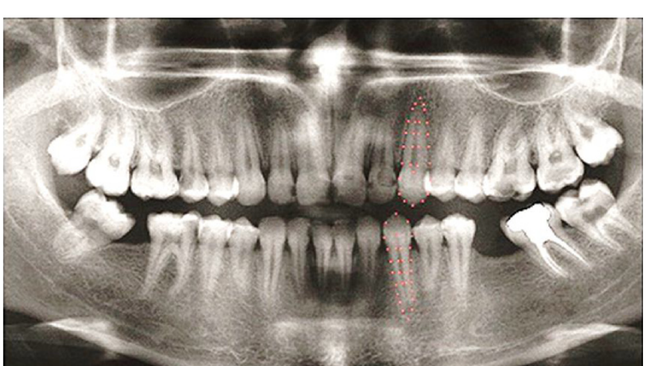

Fig. 1. A panoramic image showing twenty points on tooth outline and 10 on pulp outline of left upper and lower canines to measure tooth and pulp areas by AutoCAD software.

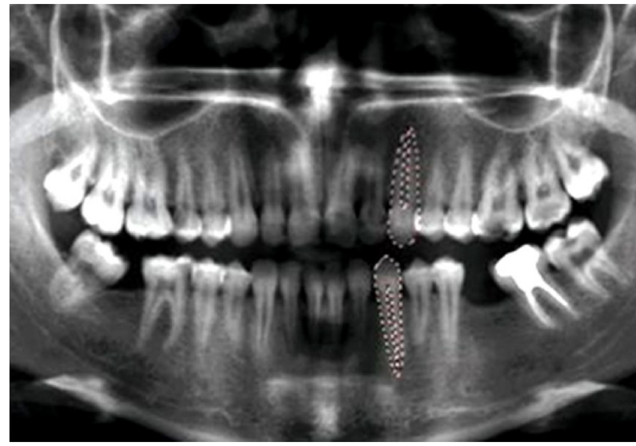

Fig. 2. A panoramic image showing tracing of upper and lower canines pulp and tooth outline to measure pulp/tooth area ratio (AR).

(Fig. 2) In this way, the morphological variable i.e. pulp/tooth area ratio (AR) was obtained. When bilateral canines were present, one of them which was easier for measurement was selected for the analysis.

The measurements were performed by the same investigator who was blinded to the chronological age of the subjects. In order to test the intra-observer reproducibility, a random sample of 30 radiographs was reevaluated after an interval of one month.

2.1. Statistical analysis

The morphological variable, i.e. pulp/tooth area ratio (AR) was determined for upper canine, lower canine and both upper and lower canines. The actual age of the patient was calculated by subtracting patient's birth date from the date of radiograph. The morphological variables (AR), age and gender of the subjects were entered into Microsoft Excel (2007). The subjects were divided into 5 age groups; each of them consisted of an interval of 10 years as follows: group I (16–24 years), group II (25–34 years), group III (35–44 years), group IV (45–54 years) and group V (55–64 years) (Table 1).

The Kolmogorov–Smirnov test confirmed the normal (Gaussian) distribution of the data ($P > 0.05$). Student t-test was run to determine any difference in morphologic variables (AR) between male and female subjects. Pearson's correlation coefficient was calculated to determine the association between pulp/tooth area ratio (AR) and age. Linear regression equations were developed and applied to the sample to estimate the age of Iranians according to the AR of upper, lower and both canines. Intra-examiner reproducibility of measurements was determined by paired sample t-test. The statistical analysis was performed by statistical Package for the Social Sciences (SPSS version 0.16) and $p < 0.05$ was considered significant.

Table 1
Age and gender distribution of the study groups.

Group	Age	Male	Female
Group I	16–24	25	29
Group II	25–34	33	30
Group III	35–44	30	33
Group IV	45–54	30	34
Group V	55–64	15	12
Total		130	141

Table 2
Pearson's correlation between chronological age and pulp/tooth area ratio of upper, lower and both canines.

Tooth	r	p
Upper canine	−0.794	<0.001
Lower canine	−0.282	<0.001
Both canines	−0.685	<0.001

3. Results

In the present study out of 300 panoramic radiographs, those belonged to 29 subjects had low image quality and were discarded from the sample. Therefore, a total of 542 upper and lower canines from 271 subjects (141 women and 130 men) were finally analyzed. Table 1 indicates age and gender distribution of individuals in the present study.

The paired sample t-test revealed no statistically significant intra-observer differences between the first and repeated measurements of orthopantomographs ($p = 0.336$). There were no significant differences in pulp/tooth area ratio of upper/lower canines between male and female subjects ($p > 0.05$), indicating that gender had no significant effect on the morphologic variables (AR).

Pearson's correlation coefficient showed a significant and inverse correlation between morphological variable i.e. pulp to tooth area ratio (AR) with chronological/actual age ($r = -0.794$ for upper canine, $r = -0.282$ for lower canine and $r = -0.685$ for upper and lower canine; p-value < 0.001) (Table 2).

Figs. 3 and 4 depict scattered spot lines showing correlation between age and AR of upper and lower canines, respectively. The upper canine had a greater correlation coefficient than the lower canine.

Linear regression equations were developed to estimate the chronological age according to AR of upper and lower canines (Table 3).

The regression equation to predict chronological age from AR of upper canine was as follows:

$$\text{Age} = 71.48 - 314.04 \times \text{AR}_{(\text{upper canine})}$$

When lower canine was taken into account, the regression model resulted in the following linear regression formula:

$$\text{Age} = 50.08 - 111.77 \times \text{AR}_{(\text{lower canine})}$$

The regression model predicting chronological age from pulp/tooth area ratios of both canines yielded the following linear regression formula:

$$\text{Age} = 60.93 - 211.63 \times \text{AR}_{(\text{both canines})}$$

Table 4 shows the mean difference between actual and estimated age using upper canine, lower canine and both canines for all subjects in this study. The mean difference between actual and estimated age was lower for upper canine compared to lower canine (6.07 ± 1.7 for upper canine versus 10.30 ± 3.45 for lower canine). Among the five age groups studied, the best result for age estimation was observed in group II (25–34 years) with the mean difference between actual and estimated age of 1.39 ± 0.75 years (considering upper canine).

4. Discussion

Several methods have been proposed for age estimation using dental radiographs [12]. The apposition of secondary dentin reduces the area and volume of pulp chamber, thus the analysis of the pulp/tooth area ratio can be employed as a reliable biomarker for ascertaining age [16,24,25]. In the present study, the pulp/tooth area ratio (AR) of canines (upper, lower and both) was measured in 271 digital panoramic radiographs to determine chronological age of Iranian adults. Panoramic radiographs were used because these are routinely taken for dental examinations and provide the possibility of assessing both upper and lower teeth in a single radiograph. Furthermore, a digital orthopantomograph can be acquired using a standard technique with high reproducibility, while the acceptability of intraoral radiographs is greatly dependent on the technique used and the practical training of the personnel [10]. One of the main source of measurement errors in radiographic techniques is the difficulty in detecting the reference points on tooth periphery and subsequently difficult drawing of reference lines for measurements [26]. However, in our study the radiographs had adequate resolution, and there was no significant

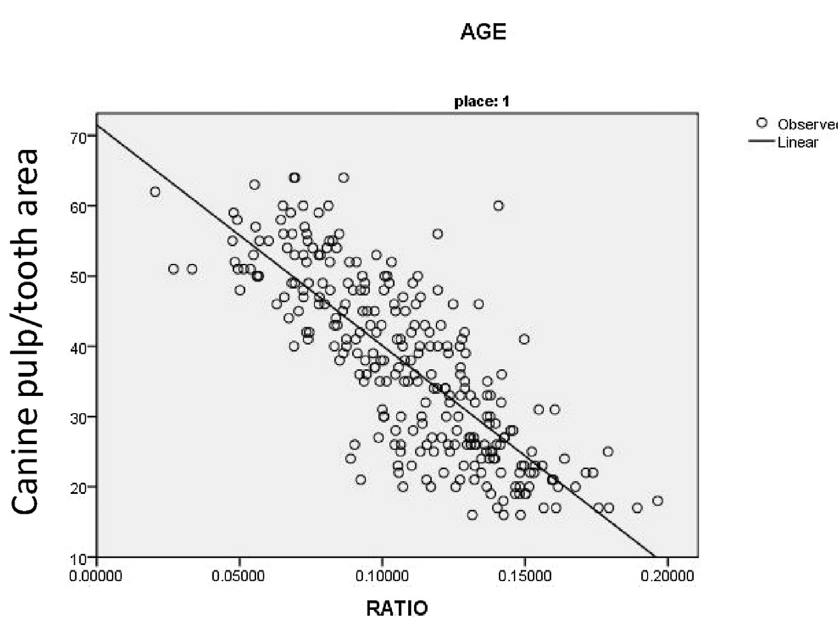

Fig. 3. A scattered spot line showing correlation between age and pulp/tooth area ratio of upper canine.

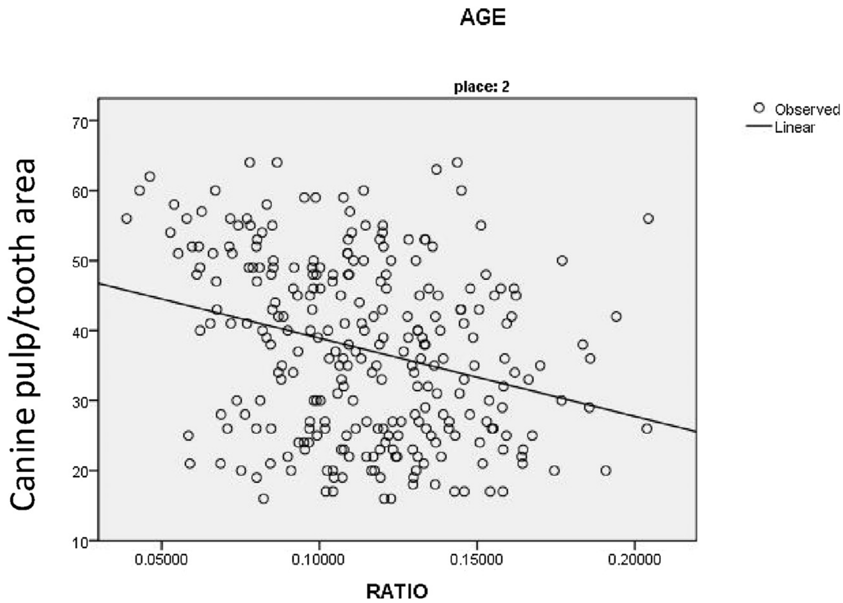

Fig. 4. Scattered spot line showing correlation between age and pulp/tooth area ratio of lower canine.

intra-observer difference between the repeated measurements. Similarly, other studies found high intra-observer agreement in measuring pulp and tooth areas, indicating high reproducibility of the measurements [2,18,25,27]. Canines were chosen for age estimation in this study, as they are single-rooted teeth with the largest pulp area and thus the easiest teeth to analyze. In addition canines are not easily lost even in older subjects and undergo less wear than other anterior and posterior teeth.

This study revealed that gender had no significant effect on the pulp/tooth area ratio of canines. This finding is in accordance to the similar studies carried out by Jeevan et al. [27] and Singaraju et al. [28] who found that gender had no significant influence on age estimation regression equations.

The teeth were chosen either from the left or right side, because it has been clarified that there is no significant difference between permanent teeth from the left or right side of the jaw [16].

Pearson's correlation coefficient between age and morphological variable (AR) of upper canine, lower canine and both canines showed a significant and inverse correlation, suggesting that measuring AR could be used as a valuable indicator for age estimation. Ubelaker and Parra reported that correlation coefficients were in the range of 0.69 to 0.86 for maxillary and mandibular central incisors in different regions of Peru [29]. The negative correlation between age and AR confirms the outcomes of previous authors that the apposition of secondary dentin throughout life results in reduction of pulp area relative to tooth area and this reduction is directly related to chronological age of subjects. This finding is in agreement with the results of several investigations published by Cameriere et al. regarding the applicability of this age estimation method [18,25,30]. Similarly, Juneja et al. [31] and Misirlioglu et al. [20] repored that AR of maxillary canine was significantly correlated with age in Indian and Turkish populations, respectively.

The correlation coefficient of upper canine was about three times greater than that obtained for lower canine indicating better accuracy of predicting chronological age from upper canine model. When both upper and lower canines were included in the model, the result was slightly weaker than the coefficient of estimation when upper canine was included alone, but better than that of lower canine alone. The outcomes of this study were in accord to those of Brkic et al. [32] who concluded that teeth of both jaws are reliable for age estimation; however, the correlation between chronological age and maxillary teeth was stronger. The result of a similar study in Indian population showed that pulp/tooth area ratio of upper canines was better correlated with chronological age than lower canines [27]. It seems that the rate of secondary dentine deposition progress more consistently in upper than lower canine throughout life. Fancy [33] stated that the growth layers in the maxillary teeth are more regular and distinct compared to those of mandibular teeth.

The mean difference between actual and estimated age using AR of upper canine gave a more precise result (6.07 ± 1.7 years, 95% CI: 5.86–6.27) in comparison to lower canine (10.30 ± 3.45 years, 95% CI: 9.93–10.74). Cameriere et al. [25] in their studies concluded that the accuracy of AR method for age estimation was better when canines were analyzed with the standard error of estimation (SEE) less than four years. They also found that maxillary canines (2.37 years) were slightly more predictive than the lower canines (2.55

Table 3
Regression analysis predicting chronological age using upper, lower and both canines.

Group	Effect	Regression coefficient	Standard error	p-value
Upper canine	Intercept	71.48	1.66	<0.001
	AR	−314.04	14.6	<0.001
Lower canine	Intercept	50.08	2.75	<0.001
	AR	−111.77	23.21	<0.001
Both upper and lower canines	Intercept	60.93	1.66	<0.001
	AR	−211.63	14.33	<0.001

Table 4
Mean ± SD difference (years) between actual and estimated age of study population.

Category	Upper canine	95% CI	Lower canine	95% CI	Both canines	95% CI
Group I	4.89 ± 0.68	4.70–5.07	6.25 ± 1.50	5.84–6.65	5.68 ± 1.85	5.33–6.02
Group II	***1.39*** ± ***0.75***	1.20–1.57	***2.15*** ± ***1.25***	1.84–2.45	***1.48*** ± ***0.92***	1.31–1.47
Group III	5.23 ± 1.36	4.89–5.56	7.57 ± 1.6	7.17–7.96	6.72 ± 1.06	6.53–6.90
Group IV	6.56 ± 0.89	6.34–6.77	8.20 ± 1.01	7.95–8.44	7.67 ± 1.35	7.43–7.90
Group V	8.25 ± 1.09	7.83–8.66	10.21 ± 2.5	9.26–11.15	9.45 ± 1.87	8.95–9.94
Total sample	***6.07*** ± ***1.7***	5.86–6.27	10.30 ± 3.45	9.93–10.74	8.18 ± 2.03	8.01–8.34

The **bolds** and italics values represent the best predictive variables for age estimation.

years) [30]. In another study, Cameriere et al. [19] found that the mean estimation error of this technique was ranged between 4.34 and 6.02 years when mandibular premolars were employed. It is believed that the standard error of estimate should be equal or less than 10 years to be considered acceptable for age estimation.

In this study, the regression equations gave better results in group II (25–34 years) with the mean difference between actual and estimated age of 1.39 ± 0.75 years when upper canine was considered and 1.48 ± 0.92 years when both canines were considered. Therefore, the pulp/tooth area ratio technique can be especially helpful for predicting age of young adults, in whom other radiographic methods assessing tooth developmental stage and skeletal maturation fail to be as a reliable age predictor. The greater accuracy of age-estimate formulas in young adults compared to the overall sample could be attributed to the reduction in quality of orthopantomograms with increasing age, or to the more regular secondary dentin apposition in younger than older adults.

A limitation of this study was the nature of panoramic radiography and its innate distortion. However, high quality digital orthopantomographs were used to have clear radiological images. Further studies have to be conducted using image analysis software to recognize pulp and tooth outlines precisely and thus minimizing errors of manual measurements. Investigating the correlation between age and pulp/tooth area ratio of other teeth like incisors and premolars together is suggested in larger samples to obtain more accurate age estimate equations.

5. Conclusion

From the results of the present study it can be concluded that radiography images of canines using pulp/tooth area ratio technique is useful to predict chronological age of Iranian adults. The upper canine is preferred for age estimation compared to lower canine, because it showed a higher correlation coefficient with chronological age. The age estimate equations obtained in his study were more accurate in young adults (25–34 years) as compared to the whole sample.

Conflict of interest

The authors declare no conflict of interest.

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