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Assessment Of The Correlation Between Dental Crypt-To-Tooth Size Ratio And Chronological Age In Pediatric Population— A Cross-Sectional Analysis Using Radiographic Imaging.

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Abstract
Background: *The forming tooth is enclosed within a bony crypt, and its dimensions change in response to tooth's positional shifts during different phases of eruption. The amount of space within the alveolar bone available for tooth initiation plays a crucial role in determining its development timing. By examining the association between the crypt-to-calcified tooth area ratio and chronological age, an age prediction regression model can be formulated.*
Materials and Methods: *This research employed a cross-sectional design, analyzing a randomly drawn retrospective sample of 60 panoramic digital radiographs from pediatric patients between 4 and 16 years old. In this study, ImageJ software was utilized to measure bony crypt and the developing right mandibular second molar across 60 orthopantomograms. An analysis was conducted to assess the relationship between the CTR and the children's actual ages. Statistical methods were applied to examine this relationship, and linear regression equations were formulated to create a model for estimating age based on the observed correlation between CTR and age.*
Results: *A highly significant and strong inverse relationship was found between age and crypt-to-tooth ratio in males, females, and all subjects combined.*
Conclusion: *This new approach could serve as an additional tool for estimating age, complementing existing age estimation methods for children.*
Key Word: *Bony tooth crypt, Mandibular second molar, Crypt to tooth ratio, Age estimation by Cypt to tooth ratio.*

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I. Introduction

Evaluating children's overall progress and maturation by considering both systemic and oral developmental stages, along with tooth development, is essential for precise clinical evaluation and management planning in pediatric dental care.[1] To monitor bodily maturation, various developmental markers are assessed, including calcified structures like bones and teeth, indicators of sexual maturity such as testicular growth and menarche, and physical attributes such as height and weight.[2] Although many physiological characteristics can be influenced by external environmental conditions, teeth are particularly valued as maturation markers because of their relative resistance to effects of systemic nutrition and illness.[3]

Tooth development starts with the formation of the dental lamina and progresses as tooth develops within a designated region of the jaw.[4] The tooth germ's mineralization is typically divided into stages that involve tooth crown,tooth root, and apical tip. The development of the tooth crown starts with initial mineralization observed at the tips of the cusps, followed by fusion of cusps and the completion of the cusp outline (Moorrees et al., 1969).[5] Once the crown is fully developed, root formation starts with the growth of

Hertwig's epithelial root sheath (HERS). With occlusal migration of the tooth germ, root elongation occurs, allowing Hertwig's epithelial root sheath (HERS) to grow in a downward vertical direction. From the perspective of forensic odontology, various invasive and non-invasive age prediction approaches based on dental parameters have been formulated. (Nolla,1963, Demirjian et al.,1973; Moorrees et al., 1963, Cameriere et al.,2006).[6]

The primary objective of this research was to examine crypt-to-tooth ratio (CTR) by measuring imaging-based size of the forming right mandibular second molar along with its adjacent osseous cavity, and subsequently relate this ratio to subject's actual age. We propose that with increase in tooth's development area, size of the surrounding bony crypt will correspondingly decline.[7]

Figure no 1: Crypt surrounding developing tooth

II. Material And Methods

This cross-sectional study utilized a convenience sample of 60 retrospectively selected digital panoramic radiographs (OPGs), randomly chosen from children aged 4 to 16 years, including 33 males and 27 females. The average chronological age of the participants was 9.5 years. The radiographic images, taken for treatment, were retrospectively analyzed for the study. The study included OPGs of children with parental consent obtained for their use. Institutional Ethical committee approval (302/IEC/SS/2025) was taken for this study.

***Study Design*:** Restropective cross-sectional study

Study Location: Department of Pediatric and Preventive Dentistry, Government College of Dentistry, Indore, Madhya Pradesh, India

***Study Duration*:** December and January 2024-25

***Sample size*:** 60 patient's Orthopantomograms

***Sample size calculation*:** The sample size is calculated using Sample Size Calculators for designing clinical research.

Input parameters
α-two tailed- 0.05
β (1-power)- 99%
r= 0.588 (Based on previous research by Pillai JP & Babu R, 2022)

Output parameters
Sample size= 43

Thus, the minimum required sample size is 43 but as we could collect more samples we have taken sample size of 60.

Inclusion criteria:
1. OPGs of children in the age range of 4 to 16 years.
2. OPGs demonstrating clear radiographic details of the development stage of right permanent mandibular second molar.

Exclusion criteria:
1. OPGs of children showing dental developmental anomalies and defects.
2. OPGs with artifacts.

3. OPGs with missing permanent mandibular second molars.

Procedure methodology

The real age in years and months was obtained from the OPGs and corresponding records. The OPGs were anonymized to ensure the investigators did not have access to the children's demographic details. Digital OPG images were examined using ImageJ software, employing threshold tool to improve the visibility of calcified region of the growing second molar. The perimeter of crypt was traced using drawing tool, and its pixel area was calculated through the Analyze > Measure feature.

In the same way, the mineralized section of maturing second molar was delineated, and its area was determined using the same method. All measurements taken in ImageJ were recorded with precision to the closest pixel. The collected data were compiled in an Excel spreadsheet, and the crypt-to-tooth area ratio was subsequently calculated. (Fig 2)

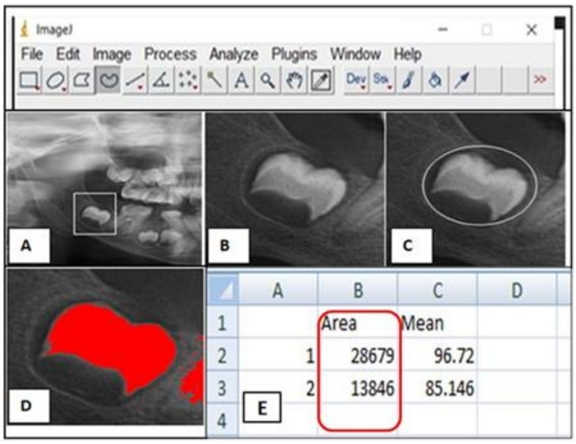

Figure no 2. Application of ImageJ tools to measure crypt area and tooth area of mandibular second molar. A) Selecting target tooth from the digital OPG; B) Applying image enhancement tools to improve the visibility of the mineralized portion of the tooth; C) Utilizing elliptical tool to outline the bony crypt; D) Adjusting threshold tool to isolate the mineralized crown of second molar E) The areas of both crypt and tooth were measured using the Measure function.

The children's actual chronological ages were compared to their crypt-to-tooth ratios (CTR), and statistical analyses were carried out. Regression equations were established by analyzing the direct correlation between CTR values and the individual's chronological age.These equations were subsequently cross-validated using the same samples, with the null hypothesis positing that there is no significant difference between the actual age (CA) and the estimated age (EA) obtained from the recently established age determination formula.

Statistical analysis

Data were recorded in Microsoft Excel and analyzed using IBM SPSS Statistics software, version 25.0 (IBM Corp., Chicago, IL). The data was assessed for normality using the Kolmogorov-Smirnov test to determine its probability distribution. Descriptive statistics was performed. The bivariate correlation was analysed using Spearman's correlation coefficient. Comparisons between groups were performed using One-way ANOVA or the Kruskal-Wallis test, with a p-value less than 0.05 considered statistically significant.

III. Result

Crypt area

Table 1 shows the median crypt area, along with the interquartile range (IQR), for males, females, and the overall sample. The crypt area was marginally greater in males compared to females, but this difference did not reach statistical significance. (p-value>.05)

Table no 1: Description of crypt area (in mm^2) among study subjects.

Group	Median (inter-quartile range)	Mean rank	Z value	p-value
Male	27389.0 (25318.5- 30037.5)	33.58	-1.508	.132
Female	25828.0 (21906.0- 28963.0)	26.74		
All subjects	26968.5 (23630.75- 29279.25)			

Tooth area

Table 2 shows the median (inter-quartile range) tooth area among males, females and all subjects. The tooth area was somewhat larger in males than in females; however, this difference was not statistically significant. (p-value>.05).

Table no 2: Description of tooth area (in mm^2) among study subjects.

Group	Median (inter-quartile range)	Mean rank	Z value	p-value
Male	14847.0 (10271.5- 24659.5)	32.42	-.944	.345
Female	12533.0 (10871.0- 17074.0)	28.15		
All subjects	13751.0 (10702.5- 22292.0)			

Mann-Whitney U test.

Crypt to Tooth ratio

Table 3 shows the median (inter-quartile range) crypt to tooth ratio among males, females, and all subjects. The ratio of crypt area to tooth area was slightly larger in females than males; however, the variation was statistically insignificant (p-value>.05).

Table no 3: Description of crypt-to-tooth ratio among study subjects.

Group	Median (inter-quartile range)	Mean rank	Z value	p-value
Male	1.8616 (1.1457- 2.3220)	29.09	-.691	.490
Female	2.0529 (1.3173- 2.3116)	32.22		
All subjects	2.0089 (1.2538- 2.3057)			

Mann-Whitney U test.

Age and CTR

Table 4 shows the age was found to have a statistically significant very strong negative correlation with crypt-to-tooth ratio among males, females, and all subjects (p-value<.05, ρ= >0.9 to 1).

Table no 4: Correlation between age and CTR.

Group	Spearman's correlation coefficient	p-value
Males	-.917	<.001*
Females	-.905	<.001*
All subjects	-.930	<.001*

*p-value<.05 was considered statistically significant.

Linear regression

Table 5 and 6 shows the linear regression analysis revealing a significant association between age and CTR among male, female, and total study population (p-value <.05).The regression model used to predict age based on CTR has been derived.

Table no 5: Regression analysis between age and CTR (among males).

Model		Unstandardized Coefficients		Standardized Coefficients	T	p-value
		B	Standard Error	Beta		
1	(Constant)	16.595	.869		19.087	<.001*
	CTR	-3.423	.397	-.840	-8.629	<.001*
a. Dependent Variable: Age						

Table no 6: Regression analysis between age and CTR (among females).

Model		Unstandardized Coefficients		Standardized Coefficients	T	p-value
		B	Standard Error	Beta		
1	(Constant)	13.219	.844		15.655	<.001*
	CTR	-1.775	.334	-.728	-5.307	<.001*
a. Dependent Variable: Age						

The equation for the prediction of age using CTR(for males):

$$Age = -3.423(CTR) + 16.595$$

The equation for the prediction of age using CTR(for females):

$$Age = -1.775(CTR) + 13.219$$

Cross-validation

Among all subjects, males, and females, the calculated and estimated age was not found to differ significantly (p-value>.05).

Table 7 shows that the amount of difference between calculated and estimated age was non-significant between males and females (p-value>.05) showing that CTR is equally useful in males and females.

Table no 7. Comparison of estimated age and calculated age among all subjects.

Group	Variable	Median (inter-quartile range)	Mean rank	Z value	p-value
Males	Calculated age	10.0 (6.0- 13.0)	14.86	-.563	.574
	Estimated age	10.2227 (8.6468- 12.6733)	20.75		
Females	Calculated age	9.0 (7.0- 12.0)	13.43	-.024	.981
	Estimated age	9.5803 (9.1188- 10.8760)	14.62		
All subjects	Calculated age	9.5 (6.25- 12.75)	30.37	-.029	.977
	Estimated age	9.7339 (9.0292- 11.5264)	30.63		

Wilcoxon sign rank test

IV. Discussion

The present study aimed to introduce a novel approach for age determination by analyzing the areas of crypt and the developing tooth. The regression formula created was validated through cross-validation within the same sample population, revealing a significant negative correlation between ratio of crypt to tooth size and actual age. This finding aligns with the results reported by Pillai JP et al. However the results demonstrated the least difference between estimated age and chronological age occurred in the 10–13-year age group, followed by the 7–10-year group. This contrasts with findings of Pillai JP et al. (2022), who found the least difference in the 4–7-year age group, followed by 7–10 years.[7]

The findings of Cameriere et al. revealed a linear connection between the dimensions of the pulp and those of the tooth (2004).[8] Additionally, Liversidge and Molleson (1999) reported a correlation between tooth length and the eruption into alveolar bone of all primary teeth, along with permanent central incisors and first molars.[9] Cameriere et al. (2006) introduced an age determination technique derived from linear measurements of the developing tooth root apices and their association with chronological age. This approach assessed all seven teeth on left side of the mandible by measuring the vertical height of forming teeth and the near transverse mesiodistal distances between the developing root apices.[10]

This research concentrated on analyzing the area of mineralised portion of erupting tooth and the surrounding bony crypt of a single tooth—the right mandibular second molar. The earliest evidence of mineralization, marked by the formation of a cone-shaped hardened area, generally appears by the age of 4. Therefore, the minimum age for inclusion in the sample was established at 4 years. Furthermore, because root apex closure typically completes by the age of 16, this age marked the highest limit for individuals considered in the study.

This age estimation technique, which uses the ratio of crypt area to tooth area, is a modification of the method proposed by Cameriere et al. (2004), who utilized regression analysis based on pulp chamber dimensions and total tooth area.[8] Additionally, the earliest sign of tooth development is often the emergence of crypt on radiographic images and throughout its maturation, there is a strong, documented correlation between root development and eruption of teeth. (Moorrees et al., 1969; Smith and Buschang, 2010).[11,12]

Comparable studies to Cameriere et al. (2004) have been performed on a representative Indian population sample, utilizing digital imaging software such as AutoCAD and Adobe Photoshop (Babshet et al., 2010; Babshet et al., 2011).[13,14]

In the current study, ImageJ tools were employed to determine mineralized area of maturing right mandibular second molar as well as surrounding bony crypt. Prior studies have also utilized ImageJ in assessment of dental development age like measuring the pulp-to-tooth ratio in upper canines and analyzing cementum growth lines(Juneja et al., 2014; Dias et al., 2010). [15,16]

ImageJ is a free, Java-based image processing program created by the National Institutes of Health (NIH) in Maryland, USA. This software allows users to visualize, modify, analyze, process, save, and print images. Additionally, it can calculate area measurements and pixel intensity data for user-defined regions.[7]

Fadili et al. conducted a study comparing volume measurements obtained through manual techniques with those derived from ImageJ software. Their results indicated no statistically significant differences between the two methods, leading them to conclude that volume measurements performed using ImageJ are both valid and precise.[17]

Several other software programs similar to ImageJ are also available. For instance, Salvarzi E et al. demonstrated that facial dimension measurements using Digimizer software yielded consistent and accurate outcomes across most parameters, suggesting it as a suitable alternative for facial anthropometry.[18] Additionally, Peñarrocha-Oltra D et al. compared three software tools—3Dicom Viewer, Adobe Photoshop, and ImageJ—for assessing peri-implant marginal bone levels, finding no significant differences in accuracy among the methods regardless of implant location.[19]

V. Conclusion

A new approach for evaluation of dental development was explored by examining crypt size and developing tooth area in digital radiographs. This method could serve as an additional tool for age estimation alongside existing, well-established approaches for pediatric assessment. Nonetheless, additional studies with larger sample groups and analysis of multiple teeth are needed to comprehensively assess the effectiveness of CTR as a tool for estimating dental age.

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