Zagazig Journal of Forensic Medicine and Toxicology 2025; 24 (1): 1- 10

Zagazig Journal of Forensic Medicine and Toxicology

Home page: https://zjfm.journals.ekb.eg/

Journal homepage: https://zjfm.journals.ekb.eg/

Original article

Crypt-to-Tooth Ratio of the Permanent Second Mandibular Molar as an Indicator of Chronological Age in Egyptian Children: A Radiological Study

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ARTICLE INFO

Article history

Received 21 - 6 - 2025

Revised 18 - 7 - 2025

Accepted 2 – 8 - 2025

Keywords:

Crypt-to-tooth ratio, Age estimation, Egyptian children, Second molar, Dental radiographs

Abstract

Background: Dental development is a reliable method to estimate the chronological age in forensic investigations. Crypt-to-tooth ratio (CTR) has emerged as a quantitative tool for age estimation as tooth crypt decreases in size during tooth development. However, its applicability has not been explored in Egypt. Using dental radiographs, this study aimed to evaluate the reliability of CTR of the permanent mandibular second molar as an age estimation tool for Egyptian children and establish population-specific reference data.

Methods: This retrospective cross-sectional study analyzed 200 digital panoramic radiographs from Egyptian children aged 5–17 years (97 males, 103 females; mean age 11.14 ± 3.09 years). CTR was calculated using ImageJ software by measuring the crypt area and mineralized tooth area. Reliability was assessed through re-measurement of 20 radiographs.

Results: A Strong negative correlation was observed between chronological age and CTR values ($r = -0.975$, $p < 0.001$). Mean CTR decreased consistently from 2.66 ± 0.28 in the youngest group to 1.00 ± 0.06 in the oldest group. No significant sex differences were found. Linear regression yielded the equation: age = 19.95 - 5.6 (CTR) with $R^2 = 0.856$ and standard error of 1.175 years. Validation testing showed no significant difference between estimated and chronological ages ($p = 0.670$), with most estimates falling within ± 2 years of actual age.

Conclusion: CTR of the mandibular second molar demonstrates high reliability for age estimation in Egyptian children and provides a valuable population-specific tool for forensic applications with acceptable accuracy ranges.

I. Background

Age estimation is a critical component in forensic and anthropological fields, particularly when legal documentation is absent or disputed. In forensic investigations, determining the age of unidentified minors is essential for addressing criminal responsibility, immigration conflicts, and human trafficking cases (Taranilla Castro *et al.*, 2023). Anthropologically, it aids in reconstructing biological profiles of historical populations (Kitpatanasombat *et al.*, 2025).

Legal systems following the United Nations Convention on the Rights of the Child require accurate age estimation for protection of those under 18. When age assessment fails, severe consequences follow, such as wrongful legal charges and inadequate medical care. This makes non-invasive, precise methods essential (Koch *et*

DOI: 10.21608/zjfm.2025.396527.1219

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al., 2025). However, current age estimation methods face significant limitations when applied across different populations. Thus, population-specific validation and calibration are necessary to ensure accuracy in diverse demographic contexts (Mbonani *et al.*, 2025).

Given these limitations, dental development has been used as a robust indicator for age estimation. Tooth formation follows a well-documented sequence from crown calcification to root completion. Skeletal maturation varies with nutritional status and systemic diseases, but dental development remains more or less stable across diverse populations (Sarac *et al.*, 2024). Panoramic and periapical dental radiographs offer non-invasive visualization of developing teeth. Thus, clinicians and anthropologists can assess developmental stages without physical disruption (Cheong *et al.*, 2023).

Among dental age estimation methods, crypt-to-tooth ratio (CTR) has emerged as a novel quantitative tool for age estimation. As the tooth matures, the crypt diminishes in height while the tooth elongates, creating a dynamic ratio inversely correlated with age. CTR quantifies the spatial relationship between the residual crypt and the developing tooth. Compared to traditional subjective staging systems such as the Demirjian method, CTR provides continuous measurement scales that capture subtle developmental variations missed by staging approaches (Pillai & Babu, 2022).

The second molar is ideal for age estimation in children under 17 years because its development spans from early childhood through adolescence, with crown formation starting around ages 2–3 and root completion by 14–17 years. Therefore, it provides continuous and reliable developmental markers during this period. Compared to first molars, which mature too early, and third molars, which are highly variable and frequently absent, second molars exhibit more consistent development across populations and clearer radiographic visibility without common complications like impaction or ectopic positioning (Yellapurkar *et al.*, 2018).

Egypt, with its unique demographic and environmental conditions, lacks validated CTR standards for age estimation. Existing studies in the Middle East and North Africa (MENA) region, such as those by El-Bakary et al. (A. A. El-Bakary *et al.*, 2019) and Moness et al. (Moness Ali *et al.*, 2019), have focused on third molar development or Demirjian's staging, overlooking molar crypt dynamics. This gap undermines forensic practices in Egypt, where accurate age estimation is increasingly demanded due to rising legal disputes involving minors.

To our knowledge, no study has explored the applicability of CTR as an age indicator in Egyptian children. Thus, this study aimed to evaluate the reliability of CTR of the mandibular second molar in dental X-rays as an age estimation tool for Egyptian children aged 5–17 years. By correlating radiographic measurements with chronological age, this research sought to establish population-specific reference data, addressing the current reliance on foreign standards.

II. Subjects and Methods

1 Study design and sample selection

This retrospective cross-sectional study was conducted upon 200 digital panoramic radiographs (OPGs) from pediatric patients. The study population consisted of 97 male and 103 female subjects ranging from 5 to 17 years with a mean chronological age of 11.14 ± 3.09 years. These radiographs were obtained from patients seeking treatment at the Oral Medicine and Periodontics Department, Alexandria Faculty of Dentistry using images that had already been taken for diagnostic purposes. Ethical approval was obtained from the Ethics Committee of Alexandria Faculty of Medicine (IRB no. 12098, FWA no. 18699). Due to the retrospective nature of the study, the need for informed consent was waived.

Eligibility criteria were digital OPGs of patients aged 5–17 years with clear and high-quality images showing the developmental stages of mandibular second molars in both quadrants and complete demographic data (age and sex). Exclusion criteria were radiographs with poor image quality or artifacts, cases with congenital anomalies, previous orthodontic treatment, systemic conditions affecting dental development, pathological lesions in the mandibular second molar, or incomplete patient information.

Chronological age (CA) was calculated in years using Microsoft Office Excel 2019 by determining the interval between birth date and radiograph acquisition date. To prevent observer bias, all radiographs were coded to mask demographic information from the investigators.

2 Age categorization and measurement protocol

Subjects were divided into six distinct age groups at 2-year intervals: Group I (5–7 years, n = 25), Group II (7.01–9 years, n = 41), Group III (9.01–11 years, n = 37), Group IV (11.01–13 years, n = 43), Group V (13.01–15 years, n = 34), Group VI (15.01–17 years, n = 20). Digital OPGs were analyzed using ImageJ software. The crypt outline was traced using the draw function, and the area measurement was recorded in pixels through the Analyze-Measure toolbar. Subsequently, the calcified portion of the developing second molar was similarly outlined and measured to the nearest pixel value (Figure 1). Both measurements were documented in Excel, and the CTR was calculated.

Measurement reliability was assessed by re-measuring 20 randomly selected OPGs and comparing these values with original measurements through statistical analysis. The correlation between CTR values and chronological age was examined statistically.

3 Statistical analysis

Statistical analysis was conducted using R software (Version 4.3.3). Continuous variables (CA and CTR) are presented as mean ± standard deviation (SD). Categorical variables are reported as number and percentage. Distribution normality for quantitative data was evaluated using the Shapiro–Wilks test. Measurement consistency and reliability were assessed using intraclass correlation coefficient (ICC), paired t-test, and Dahlberg formula. Sex differences in CTR values were assessed using the Mann–Whitney test. Analysis of variance (ANOVA) was applied to examine CTR value differences across the six age categories. Spearman correlation coefficient determined the strength of linear relationship between chronological age and CTR values across all groups. Linear regression analysis was performed with age as the dependent variable and CTR as the independent variable to develop the predictive equation. Statistical significance was set at $p < 0.05$.

Regression equations obtained from the linear relationship between CTR measurements and chronological age were validated on an independent sample of 34 panoramic radiographs from a different cohort (15 males, 19 females). This validation cohort was selected from a separate group of children who attended the same department six months after the completion of the primary data collection period to test the generalizability and accuracy of age estimation equations derived from the original sample.

The sample size for the validation cohort was determined based on the rule of thumb for validation of regression models, requiring approximately 15–20% of the original sample size (Bujang, 2021). With 200 subjects in the primary sample, a validation cohort of 30–40 subjects was appropriate. Thus, we selected 34 radiographs that met the same inclusion and exclusion criteria as the primary study. The null hypothesis stated no significant difference would exist between CA and estimated age (EA) using the newly formulated age estimation equation.

III. Results

1 Sample characteristics

The total study sample comprised 200 subjects with a mean age of 11.14 ± 3.09 years. There was a relatively balanced distribution across age groups, with Group IV (11.01–13.00 years) having the highest representation at 21.5% and Group VI (15.01–17.00 years) having the lowest at 10.0% (Table 1).

2 Reliability testing

Good reliability was demonstrated between the CTR measurements in the test and retest samples. Table 2 shows the results of the reliability tests. The ICC was 0.779 with a mean difference of -0.014 ± 0.184. The Dahlberg's formula value was 0.127, and the paired t-test revealed no statistically significant difference between measurements ($p = 0.732$), confirming adequate measurement consistency.

Table (1): Distribution of the study subjects (N = 200) according to age range.

Group	Age range (years)	N (%)
I	5.00 – 7.00	25 (12.5)
II	7.01 – 9.00	41 (20.5)
III	9.01 – 11.00	37 (18.5)
IV	11.01 – 13.00	43 (21.5)
V	13.01 – 15.00	34 (17.0)
VI	15.01 – 17.00	20 (10.0)

N: number of subjects

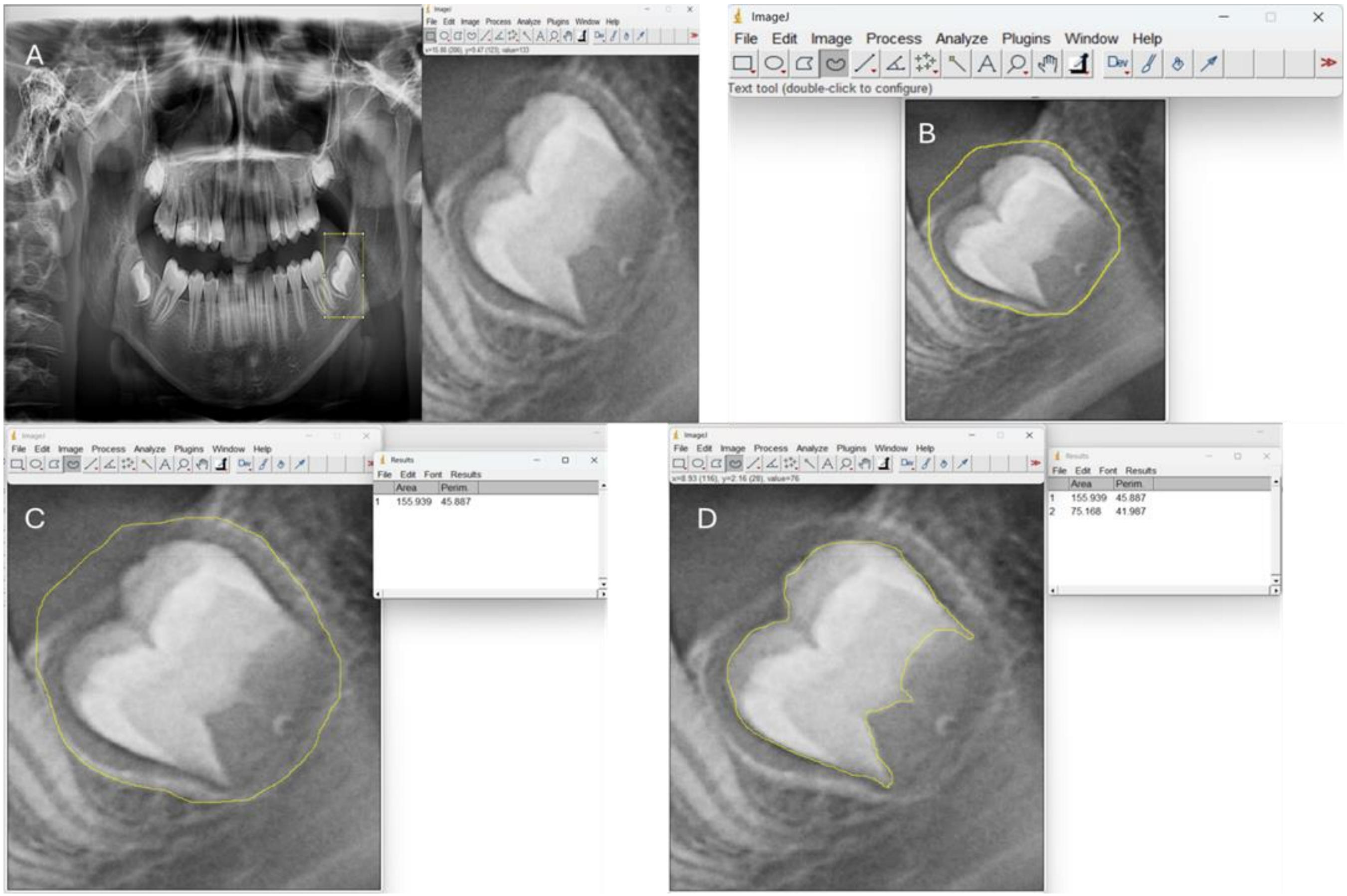

Figure (1): Using ImageJ tools to calculate the crypt and tooth areas of the mandibular second molar. (A) The developing tooth is identified within the radiographic image. (B) Tracing both the mineralized and follicular areas using the draw tool. (C) Area quantification of the traced region is performed through the Analyze-Measure function. (D) Tracing only the developing tooth using the draw tool, and the area is measured again using the same method.

Table (2): Reliability results for CTR values (N = 20).

Variable	Mean value	SD	Mean difference	SD	Dahlberg's formula value	P-value[1]	ICC
CTR (first time)	0.258	0.236	-0.014	0.184	0.127	0.732	0.779
CTR (second time)	0.272	0.302					

[1]Paired t-test
CTR: crypt-to-tooth ratio, SD: standard deviation, ICC: intraclass correlation coefficient

3 Correlation between age and CTR

The mean CTR measurement for all subjects was 1.57 ± 0.51. A statistically significant difference in the CTR values was observed between the six age groups (Table 3), with a significant negative correlation between the chronological age and the CTR measurements across all groups ($r = -0.975$, $p < 0.001$). The mean CTR value decreased consistently as age increased, from 2.66 ± 0.28 in Group I to 1.00 ± 0.06 in Group VI. The strongest correlation between age and CTR was observed in Group

III (r = -0.899, p <0.001), while the weakest correlation was found in Group V (r = -0.546, p = 0.038).

Table (3): Correlations between means of age and CTR according to age group (N = 200).

Group	N	Mean age (SD)	Mean CTR (SD)	Correlation coefficient (r)	P-value[1]
I	25	6.16 (0.75)	2.66 (0.28)	-0.857	<0.001*
II	41	8.54 (0.45)	1.84 (0.13)	-0.700	0.004*
III	37	10.35 (0.63)	1.53 (0.17)	-0.899	<0.001*
IV	43	12.36 (0.63)	1.32 (0.08)	-0.653	0.017*
V	34	14.29 (0.5)	1.15 (0.05)	-0.546	0.038*
VI	20	16.2 (0.63)	1.00 (0.06)	-0.759	<0.001*
Total	200	11.14 (3.09)	1.57 (0.51)	-0.975	<0.001*

[1] One-way analysis of variance (ANOVA) test

CTR: crypt-to-tooth ratio, SD: standard deviation, N: number of subjects, * significant p-value

4 Sex comparison

The study included 97 males (48.5%) and 103 females (51.5%). The mean CTR value in males (1.59 ± 0.52) was slightly higher than in females (1.56 ± 0.50); however, this difference was not statistically significant. The CTR values showed significant negative correlations with age in both males (r = -0.913, p <0.001) and females (r = -0.938, p <0.001). Age-specific correlations revealed that both sexes demonstrated significant negative correlations between CTR and age across all six age groups. In males, the strongest correlations were observed in Groups I and III (r = -0.873 and -0.864, respectively, both p <0.001), while the weakest correlation was found in Group IV (r = -0.637, p = 0.003). Among females, the strongest correlation was observed in Group I (r = -0.829, p = 0.001), whereas the weakest correlation occurred in Group II (r = -0.531, p = 0.034) (Table 4).

Table (4): Distributions and correlations of age and CTR according to sex and age group (N = 200).

Group	N	%	Age (years)		CTR		Correlation coefficient	
			Mean	SD	Mean	SD	r	P-value
Males								
I	10	5	5.90	0.74	2.82	0.23	-0.873	<0.001*
II	22	11	8.67	0.41	1.82	0.14	-0.735	<0.001*
III	17	8.5	10.18	0.60	1.56	0.19	-0.864	<0.001*
IV	19	9.5	12.31	0.65	1.35	0.09	-0.637	0.003*
V	15	7.5	14.30	0.59	1.17	0.05	-0.731	0.002*
VI	11	5.5	16.29	0.61	0.99	0.06	-0.789	0.004*
Total	97	48.5	11.10	3.13	1.59	0.52	-0.913	<0.001*
Females								
I	15	7.5	6.33	0.72	2.55	0.27	-0.829	0.001*
II	16	8	8.33	0.43	1.88	0.19	-0.531	0.034*
III	20	10	10.48	0.63	1.52	0.14	-0.731	<0.001*
IV	24	12	12.40	0.62	1.30	0.06	-0.706	<0.001*
V	19	9.5	14.29	0.43	1.13	0.04	-0.665	0.002*
VI	9	4.5	16.10	0.68	1.02	0.05	-0.753	0.019*
Total	103	51.5	11.18	3.07	1.56	0.50	-0.938	<0.001*

CTR: crypt-to-tooth ratio, SD: standard deviation, N: number of subjects, * significant p-value

5 Regression analysis

Linear regression using the CTR value as independent variable generated similar regression equations for both sexes (Table 5). The equation for males was age = 19.83 - 5.5 (CTR) with an R^2 value of 0.834 and a standard error of estimate (SEE) of 1.283. For females, the equation was age = 20.07 - 5.7 (CTR) with an R^2 value of 0.879 and an SEE of 1.072. The combined equation for the overall sample was age = 19.95 - 5.6 (CTR) with an R^2 value of 0.856 and an SEE of 1.175.

6 Validation of the regression formulas

The regression formulas were tested on a separate sample of 34 subjects (15 males and 19 females) with a mean age of 11.84 ± 3.69 years. Table 6 presents the descriptive statistics of the validation sample. The mean CTR value for the validation sample was 1.58 ± 0.65, with no statistically significant difference between males and females (p = 0.790).

When applying the sex-based formulas, the estimated ages were 11.32 ± 3.60 years for males and 11.54 years ± 3.76 for females, with an overall mean EA of 11.44 ± 3.63 years. The mean difference between EA and CA using sex-based formulas was -0.81 years ± 1.31 for males and -0.06 ± 1.25 years for females, with an overall mean difference of -0.39 ± 1.31 years. This difference did not reach statistical significance (p = 0.103).

Using the general regression formula, the overall mean EA was 11.10 ± 3.63 years, with mean differences between EA and CA of -0.84 ± 1.32 years for males and -0.65 ± 1.25 years for females. The overall mean difference was -0.74 ± 1.26 years, which did not demonstrate statistical significance (p = 0.670).

Table (5): Regression equations to calculate age using CTR.

Sex	Regression equation	R^2	SEE	P-value
Males (N = 97)	Age = 19.83 - 5.5 (CTR)	0.834	1.283	<0.001*
Females (N = 103)	Age = 20.07 - 5.7 (CTR)	0.879	1.072	<0.001*
Total (N = 200)	Age = 19.95 - 5.6 (CTR)	0.856	1.175	<0.001*

CTR: crypt-to-tooth ratio, SEE: standard error of the estimate, N: number of subjects, * significant p-value

Table (6): Sex-based descriptive statistics of the validation sample (N = 34).

Variables	Males (N = 15)		Females (N = 19)		Total (N = 34)		P-value[1]
	Mean	SD	Mean	SD	Mean	SD	
CA	12.13	3.64	11.60	3.81	11.84	3.69	0.681
CTR	1.55	0.65	1.61	0.66	1.58	0.65	0.790
EA (sex-based formula)	11.32	3.60	11.54	3.76	11.44	3.63	0.867
EA - CA (sex-based formula)	-0.81	1.31	-0.06	1.25	-0.39	1.31	0.103
EA (general formula)	11.29	3.66	10.95	3.69	11.10	3.63	0.790
EA - CA (general formula)	-0.84	1.32	-0.65	1.25	-0.74	1.26	0.670

[1] One-way analysis of variance (ANOVA) test

CA: chronological age, CTR: crypt-to-tooth ratio, EA: estimated age, N: number of subjects, SD: standard deviation

Figure (2) demonstrates the distribution pattern of the validation sample according to the mean difference between EA (calculated using the general formula) and CA. The plot illustrates that the majority of the EAs fell within ± 2 years of the actual CA. Younger children (5–8 years) showed positive EA-CA differences, indicating age overestimation, while older children (>12 years) showed negative differences, indicating age underestimation. The crossover occurred around 10–11 years. This pattern appeared in all six groups and both sexes, with some variations between individual groups.

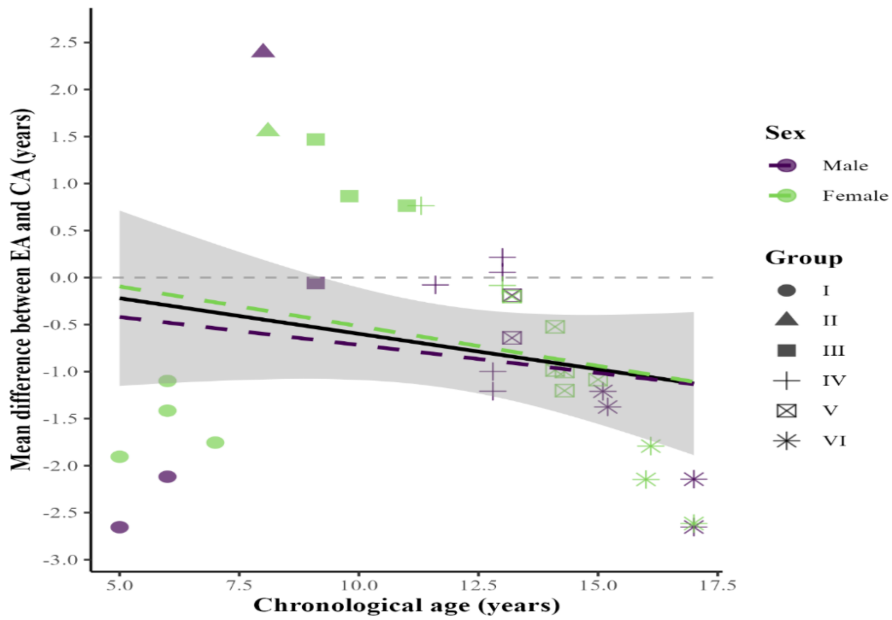

Figure (2): Scatter plot showing the distribution of the test sample (N = 34) according to the mean difference between estimated age (EA, calculated with the general formula, and chronological age (CA). Negative values indicate age underestimation while positive values indicate age overestimation.

IV. Discussion

Age estimation in children is essential in forensic and legal settings, especially when documentation is absent or disputed. Compared to skeletal methods, dental development offers a stable biological marker to determine the chronological age and remains relatively unaffected by nutritional and systemic influences (Rojas-Torres *et al.*, 2025). Previous studies in Egypt have applied qualitative dental methods such as Demirjian staging (A. El-Bakary, 2021); however, none has tested the CTR as a continuous metric for age estimation. This study is the first to evaluate the reliability of CTR of the mandibular second molar in an Egyptian pediatric sample and to validate a population-specific regression equation. The aim was to establish a quantitative, reproducible, and non-invasive tool for age estimation in Egyptian children aged 5–17 years using routine panoramic radiographs.

CTR is based on the inverse link between tooth mineralization and bony crypt height. As tooth development advances, the mineralized crown and root grow while the surrounding crypt shrinks. The mandibular second molar was strategically chosen because its crown begins forming around age three and root completion occurs near age 17 (Thomas *et al.*, 2021). This span aligns with our 5–17-year target.

ImageJ, a free image processing software, was utilized to trace and measure the crypt and tooth areas of the mandibular second molar on digital OPGs. The implementation of such image analysis tools for radiographic evaluation eliminates subjective observer bias noted in previous studies using dental developmental stages for age estimation (Yu & Klein, 2020).

In the present study, CTR showed a very strong inverse relationship with age (r = -0.975), confirming its developmental logic. The highest correlation appeared during active crown formation (ages 5–7 years) when crypt reduction and tooth growth occur rapidly. Correlation weakened modestly in the 11–13-year group, likely because root bifurcation introduces variability in the crypt outline as the tooth erupts (Pillai & Babu, 2022).

On average, CTR decreased gradually with age starting by 2.66 in the youngest age group and reaching about 1 in the oldest age group at which complete apical closure and root formation occurred. Sex did not affect the results as males and females had nearly identical mean CTR. Separate regressions for each sex yielded the same formula. Thus, a single combined equation applies to both sexes in this age range. Validation on an independent test group (N=34) confirmed the equation's accuracy, with a non-significant mean difference of -0.74 ± 1.26 years between estimated and actual age. In practice, this meant most individual estimates were within about ±2 years of the actual age, which meets typical forensic acceptability.

Direct comparison with an Indian study conducted by Pillai and Babu (Pillai & Babu, 2022) reveals population-specific variations in dental development. Although they used identical ImageJ measurement protocols for measuring CTR of the mandibular second molar, their correlation coefficient ($r = -0.898$) was lower than that found in the current study ($r = -0.975$). The regression formula in the Indian study (age = 17.192 - 3.855 × CTR, $R^2 = 0.807$) differed substantially from that in the present study (age = 19.95 - 5.6 × CTR, $R^2 = 0.856$). This may be attributed to the significant population-specific variations in crypt-tooth developmental dynamics.

Validation accuracy also differed between populations. The Indian study demonstrated systematic underestimation of 1.08 ± 0.484 years with 69% of males and 54% of females showing errors exceeding one year. In contrast, validation in this study showed mean differences of -0.74 ± 1.26 years with superior individual accuracy. These disparities likely reflect genetic, nutritional, and environmental factors influencing dental maturation rates across South Asian and Middle Eastern populations. Moreover, changes in diet and habits could have some influences in altering the pattern of teeth development across years. The variations in nutritional preferences which change with time can significantly alter growth of individuals which may arise within the same population (Strumpf *et al.*, 2023).

Previous Egyptian studies have employed established dental age estimation methods and reported variable accuracy outcomes across different populations and age ranges. For instance, Elgaidy et al. (2025) examined 180 radiographs from children from Cairo aged 4–16 years using both Demirjian and Nolla approaches. They found that the Demirjian method overestimated chronological age in males by +0.37 years while underestimating age in females by -0.52 years. Nolla's method demonstrated consistent underestimation across both sexes, with errors of -0.33 years in males and -1.42 years in females (Elgaidy *et al.*, 2025).

In addition, Kaka et al. (2025) investigated Willems method in 140 children spanning 8–16 years, documenting slight underestimation patterns in both male and female subjects. Males showed mean differences of -0.20 ± 0.91 years, whereas females demonstrated -0.24 ± 1.33 years, with mean absolute errors remaining below one year across the sample (Abd Elmaguid Kaka *et al.*, 2025). Furthermore, Abdelrazik et al. (2025) conducted the most extensive validation study, applying Demirjian staging to 799 children aged 6–17 years across a broader age spectrum. They reported mean differences of +0.10 years in males and +0.30 years in females for the overall sample. However, age-stratified analysis revealed marked overestimation in younger subgroups, suggesting developmental stage-specific bias patterns within the Egyptian population (Abdelrazik *et al.*, 2025).

These validation studies demonstrate that traditional methods achieve acceptable accuracy with errors approaching or remaining below ±1 year thresholds established for forensic applications. However, they documented sex-specific bias patterns that complicate practical implementation. The CTR methodology addresses these limitations by providing comparable or superior accuracy through a single continuous measurement scale and sex-neutral regression equation.

The current study is not without limitations. The sample was drawn from Alexandria Main University Hospital which serves patients predominantly from Alexandria and surrounding Lower Egyptian governorates. Also, the study focused on the second molar, which, although appropriate for the target age range (5–17 years), excludes early childhood and late adolescence. Future research should recruit cohorts from diverse Egyptian regions to enhance generalizability and explore combining CTR measurements from multiple developing teeth to strengthen its applicability.

V. Conclusion

The present study establishes the CTR of the mandibular second molar as a novel, objective, highly reliable tool for forensic age estimation in Egyptian children aged 5–17 years. The results yielded a sex-neutral regression equation that eliminates sex-specific biases inherent in traditional staging methods while maintaining superior accuracy with validation errors of 0.74, meeting forensic acceptability thresholds. However, broader validation across different Egyptian regions and multiple teeth are warranted to strengthen the applicability of CTR as an age indicator.

List of abbreviations

CA: Chronological age
CTR: Crypt-to-tooth ratio
EA: Estimated age
ICC: intraclass correlation coefficient
OPG: Orthopantogram
SD: Standard deviation
SEE: Standard error of estimate

Declarations

Funding

This research received no specific grant from funding agencies in the public, commercial, or not-for-profit sectors.

Ethical approval

Ethical approval was obtained from the Ethics Committee of Alexandria Faculty of Medicine (IRB no. 12098, FWA no. 18699). Due to the retrospective nature of the study, the need for informed consent was waived.

Data availability statement

The datasets analyzed in the current study are available from the corresponding author on reasonable request.

Conflict of interest

The authors have no conflicts of interest to declare.

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How to cite: Haiba, M., Madian, S., & Nagah, M. (2025). Crypt-to-Tooth Ratio of the Permanent Second Mandibular Molar as an Indicator of Chronological Age in Egyptian Children: A Radiological Study. *Zagazig Journal of Forensic Medicine and Toxicology*, 24 (1), 1-10. doi: 10.21608/zjfm.2025.396527.1219