Dentomaxillofacial Radiology (2022) 51, 20210368

birpublications.org/dmfr

RESEARCH ARTICLE

Three-dimensional verification of the radiographic visibility of the root pulp used for forensic age estimation in mandibular third molars

[1]Dilara Nil Gunacar, [2]Seval Bayrak and [3]Enver Alper Sinanoglu

[1]*Department of Oral and Maxillofacial Radiology, Faculty of Dentistry, Recep Tayyip Erdogan University, Rize, Turkey;*
[2]*Department of Oral and Maxillofacial Radiology, Faculty of Dentistry, Bolu Abant İzzet Baysal University, Bolu, Turkey;*
[3]*Department of Oral and Maxillofacial Radiology, Faculty of Dentistry, Kocaeli University, Kocaeli, Turkey*

Objectives The aim of this study was to assess the reliability of panoramic radiography (OPG) using age estimation method using cone beam computed tomography (CBCT) verification in the evaluation of radiographic visibility of root pulp (RPV) of mandibular third molars for age estimation.
Methods CBCT and OPG images of 429 mandibular third molars from 290 patients were evaluated. RPV of fully mineralized mandibular third molars was evaluated as stages 0, 1, 2, and 3 for both imaging methods. Descriptive statistics were performed separately for stages by age for both genders. The consistency of these scores with chronological age was evaluated for both imaging methods. The reliability of OPG evaluation was also analyzed with CBCT scores.
Results Spearman's rho correlation demonstrated a positive correlation between RPV and chronological age for both genders and for OPG and CBCT evaluation. Considering the minimum ages of both imaging methods, all stages were above the age of 18 except for the female group of Stage 2 and all Stage 0. For the comparison of OPG and CBCT RPV Staging scores, the κ score was found to be 0.312 ($p < 0.001$), indicating a fair agreement.
Conclusion In conclusion, the absence of Stage 3 at age of 18 and under might have a forensic value for RPV age estimation method. Considering the fair agreement in the verification of OPG scores, it is not possible to determine the exact age with the RPV detected in OPG images, the use of CBCT for the RPV evaluation is recommended to available cases.
Dentomaxillofacial Radiology (2022) **51**, 20210368. doi: 10.1259/dmfr.20210368

Cite this article as: Gunacar DN, Bayrak S, Sinanoglu EA. Three-dimensional verification of the radiographic visibility of the root pulp used for forensic age estimation in mandibular third molars. *Dentomaxillofac Radiol* 2022; **51**: 20210368.

Keywords: cone-beam computed tomography; dental pulp; forensic dentistry; forensic sciences; panoramic radiography; third molar

Introduction

Age estimation of living individuals has an important role in legal requirements.[1] Due to the increasing number of refugees in Turkey as well as in the world, identification requirement has recently gained utmost importance.[2] Globally, the age range of being an adult varies between 15 and 21.[3] In Turkey, legislation defines a child as an individual under the age of 18 in accordance with the United Nations Children's Charter.[3,4] To this end, the international and interdisciplinary "Study Group on Forensic Age Diagnostics (AGFAD)" established in 2000 recommended physical examination and anamnesis, including the recording of body measurements and an evaluation of signs of sexual maturity, X-ray examination of the left hand, and dental examination

Correspondence to: dr Dilara Nil Gunacar, E-mail: dt.dilaranil@gmail.com
Received 17 August 2021; revised 16 September 2021; accepted 02 November 2021

which records dentition status with the assessment of conventional panoramic radiography (OPG) for age assessments of adolescents and young adults if there is a legal justification for X-ray examinations without a medical indication.[5–8]

Radiographic evaluation of the teeth is a common and widely used age determination method for individuals under the age of 16.[9] This estimation investigates the root formation and can be achieved via Demirjian method,[10] Nolla method,[11] Cameriere method,[12] Schour and Massler method,[13] and Gleiser and Hunt method.[14] After the age of 16, since the root apices of all other teeth are closed, the only one in the developmental stage for age estimation may be the third molar. On the other hand, considering cases with early completed roots, it will not always be correct to establish the 18-year-old threshold by evaluating the apical development of the third molar teeth.[15] On the other hand, considering cases with early completed roots, it will not always be correct to establish the 18-year-old threshold by evaluating the apical development of the third molar teeth.[15]

Investigation of the secondary dentin ratio of the existing root is another radiographic evaluation method used for age determination. As a lifelong process, secondary dentin is accumulated in the root cavity. First, pulp horns disappear, and this process is followed by a narrowing in size of the pulp chamber and narrowing of the root canals.[16,17] Many studies correlated with this secondary dentin accumulation, and significant results were obtained.[18–20] In this respect, Olze et al evaluated the radiographic root pulp visibility (RPV) of third molars and proposed a classification for forensic purposes.[21] This classification is based on the degree of mineralization of the root cavity and evaluated by the analysis of the third molar appearance on the OPG. This method has been used by numerous researchers in various populations since 2010.[1,2,22–28]

OPG is the most common examination and a low-cost technique that is widely used during a routine dental examination. Nonetheless, it can be inadequate to define root and canal morphology due to its limitations of two-dimensional imaging such as superposition, magnification, and distortion.[29] Regarding the anatomical position of the mandible, the mandibular molar inclination towards the posterior increases gradually, which forms the Wilson curve, the occlusal inclination in the coronal plane.[30,31] Additionally, root dilaceration and position anomalies may occur in third molar teeth which may hinder evaluation in two-dimensional radiographs, such as OPG. In the presence of these conditions, the visibility of the pulp chamber of the root can be affected.[32] As a kind of examination technique, cone beam computed tomography (CBCT) is an emerging imaging method with a low radiation dose with a sub millimeter resolution. Due to the accurate and comprehensive assessment advantage of this three-dimensional imaging method, subtle changes, such as the reduction of the pulp cavity caused by secondary dentine formation can be analyzed in detail.[33]

Considering those above-mentioned superiorities of three-dimensional imaging in the evaluation process of RPV and the absence of studies investigating Olze et al's method[21] from this perspective, the purpose of this study was to assess the reliability of this OPG using the age estimation method via CBCT verification. Additionally, we aimed to investigate the relationship between chronological age and radiographic visibility of root pulp with two imaging modalities in a Turkish population.

Methods and materials

Study group selection

This retrospective study was conducted at Bolu Abant Izzet Baysal University, Faculty of Dentistry with ethical approval granted by the Ethical Committee. According to the principles described in the Declaration of Helsinki, the study protocol was carried out including all amendments and revisions. From the database of the Department of Oral and Maxillofacial Radiology between January 2015 and April 2020, images of patients who had both OPG and CBCT scans with a time-lapse no more than 3 months were retrieved. The images with at least one mandibular third molar tooth with fully developed roots and closed apices were included in the study. And also, the reason why the maxillary third molars were not included in this study, included only mandibular third molars, is that it is difficult to evaluate RPV due to factors such as anatomical noise (*i.e.* presence of maxillary sinus, tuber maxilla, zygomatic bone, etc.), geometric distortion and superposition of oral structures. Every mandibular third molar fulfilling the inclusion criteria was evaluated separately as one case.

Cases with any systemic disease that has the potential to affect growth and images with insufficient diagnostic quality were excluded from the study. Third molars presenting developmental or acquired dental anomaly, pulpal calcification, carious lesion, inflammation or root canal treatment, fused single root, narrowed furcation, or malformed roots, which appear to have extended root pulp laterally and in embedded position, were also excluded. Additionally, any third molar teeth associated with a cystic or tumoral pathological entity, affected by orofacial trauma and present in the region with surgical intervention, such as the recipient or donor site for bone graft were not selected as well.

Imaging procedure and evaluation

CBCT images were obtained with I-CAT 3D Imaging System (Imaging Sciences International, Hatfield, PA) with parameters of 5 mA and 120 kVp with different fields of views of 16 cm × 6 to 13 cm. Images were evaluated with I-CAT Vision software (Imaging Science International) at 0.3 mm slice thickness and 0.3 mm

slice interval. Standardized digital OPGs were obtained using Soredex (Cranex Novus, Tuusula, Finland) with parameters of 70 kVp and 10 mA. In order to prevent undesired horizontal and vertical magnifications, patient positioning was done in a standard for each patient in accordance with the user instructions. The patients were positioned such that the Frankfurt plane was parallel to the floor and the sagittal plane was parallel to the vertical plane of the dental panoramic machine. The magnification of the OPG device is x1.25, but the examiner was also permitted to use enhancements and orientation tools such as magnification, brightness, and contrast to improve visualization. The resolution of images is 1024 × 768 pixels and the pixel size of the digital image receptor is 96 microns. Images were evaluated through a medical diagnostic monitor. No time limit was imposed during the evaluation process of the observer.

The chronological age was calculated by subtracting the date of birth from the date of radiological data acquisition and the gender of the cases were recorded. Then, all OPG and CBCT images were paired and anonymized with case numbers. The cases were separated into two groups of OPG and CBCT afterward.

The selected images were evaluated by an experienced oral and maxillofacial radiologist (SB). The images of the OPG group were evaluated in random order. After that CBCT group was scored separately again in random order. For CBCT assessment, each root of the third molar was orientated in all three planes, and the sagittal sections were selected for the evaluation. Additionally, the toolbar was moved continuously from the floor of the pulp chamber to the apex in axial sections to determine the visibility of the root lumen if needed. Following this analysis, CBCT scores were matched with OPG scores. 2 months after the evaluation, 100 randomly selected OPG-CBCT pairs were re-evaluated for investigation of intra observer agreement.

All third molars were investigated in accordance with the classification of the stages of root pulp visibility as described by Olze et al[21] and modified by Lucas et al.[27] The RPV of the mandibular third molars was categorized into four stages:

Stage 0 = visible lumen of all root canals up to the root apex.

Stage 1 = the lumen of one root canal is not entirely discernible up to the root apex.

Stage 2 = two root canals with incompletely visible lumen to the apex, or one canal might be virtually not visible in the entire length.

Stage 3 = the lumen of two root canals is virtually not visible in the entire length (Figure 1).[21]

Statistical analysis

The statistical analyses were performed using IBM SPSS 8 v. 23 (SPSS Inc., Chicago, IL) with a level of significance of 5% ($p < 0.05$). For evaluation of intraobserver agreement, Cohen's κ was used. The descriptive analysis for each CBCT stage and OPG; stage of RPV, mean age with standard deviation, minimum age, maximum age and lower, median and upper quartiles were calculated. Spearman's ρ correlation was performed to investigate the relationship between age and stages of pulp visibility.

Results

After the evaluation of the OPG and CBCT data, a total of 429 third molars (211 on the left side and 218 on the right side) of 290 patients (164 males and 126 females

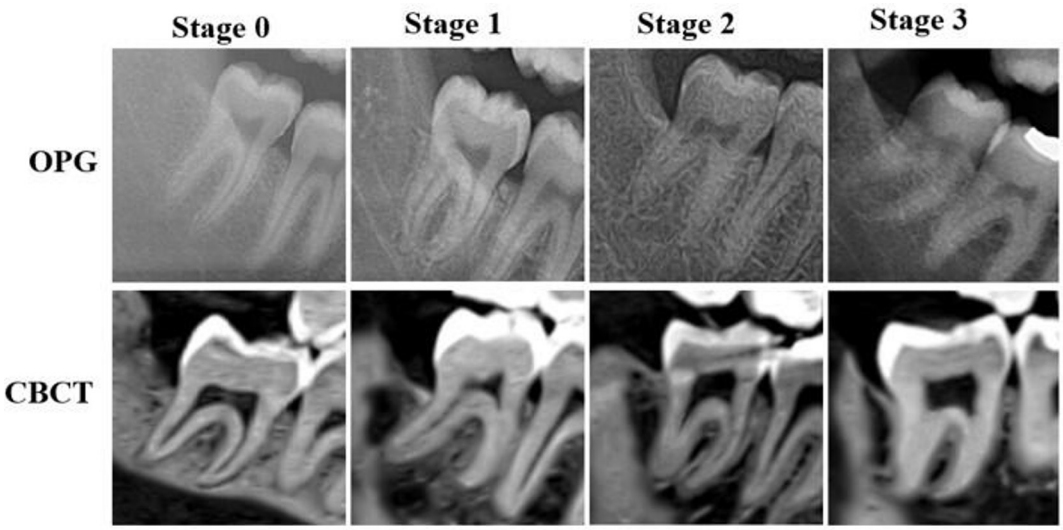

Figure 1 Root pulp visualization stages as proposed by Olze et al[21] in OPG and CBCT images. CBCT, cone beam CT; OPG, panoramic radiography.

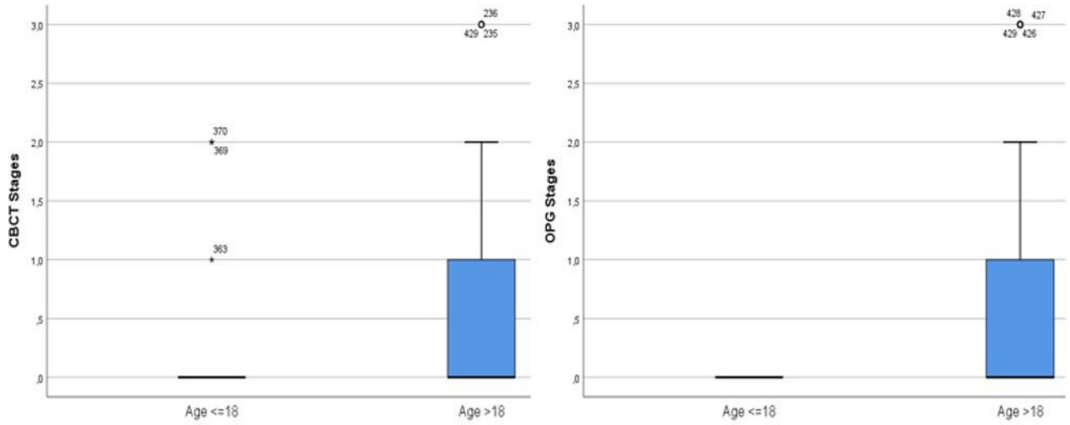

Figure 2 Median and quartiles for CBCT and OPG for age groups (Age groups for cut-off 18). CBCT, cone beam CT; OPG, panoramic radiography

aged between 16 and 68) fulfilled the criteria and were enrolled in the study. Of the 290 patients in the study group, 9 were 18 years old, 203 were 18–40 years old, and 78 were over 40 years old (Figure 2). The mean ages ± standard deviations of the study group were 33.7 ± 9.7 years old.

For the re-evaluation of OPG and CBCT images, the weighted κ coefficients were found to be 0.864 and 0.882, respectively, with good intraobserver reliability.[34]

Spearman's ρ correlation demonstrated a positive correlation between RPV and chronological age for both genders and for both radiological evaluation methods (Table 1). Additionally, the mean ages for CBCT groups were as follows: 30.46 and 30.47 for Stage 0, 38.55 and 37.63 for Stage 1, 41.91 and 37.71 for Stage 2, and 40.40 and 36.50 for Stage 3 for males and females, respectively. The mean ages of OPG groups were 30.48 and 30.76 for Stage 0, 38.94 and 37.79 for Stage 1, 42.20 and 35.94 for Stage 2, and 37.58 and 39.63 for Stage 3 for males and females, respectively.

Identifying age distributions of the RPV stages based on gender for CBCT and OPG groups are shown in Tables 2 and 3. In CBCT, Stage 0 was first seen at the age of 16 in males and at the age of 17 in females. For the OPG group, Stage 0 first appeared at the age of 16 for both genders. For CBCT, for males and females respectively Stage 1 first occurred at ages 19 and 18, while for the OPG group it was 24 and 20. For the CBCT group, Stage 2 was seen at the age of 31 in males and at the age of 16 in females, while it was first recorded at the age of 27 in males and at the age of 25 in females for the OPG group. For CBCT, Stage 3 was first observed at the age of 32 in males and at the age of 33 in females, and those were 25 and again 33 for OPG group. Interestingly, Stage 0 was seen at the ages of 60 and 68 for CBCT and OPG groups, respectively.

Table 1 Correlation between RPV Stages and chronological ages for both imaging methods

	Male		*Female*		*Total*	
	r	*p*	*r*	*p*	*r*	*p*
CBCT	0.459	<0.001	0.335	<0.001	0.429	<0.001
OPG	0.431	<0.001	0.335	<0.001	0.401	<0.001

CBCT, cone beam CT; OPG, panoramic radiography; RPV, root pulp visibility.

The distribution of RPV stages and gender under and over the 18-years-old thresholds is given in Tables 4 and 5. In the CBCT group, all subjects who were classified as Stage 3 were older than 18 for both genders, whereas Stage 1 and Stage 2 were not attained in males. For the OPG scores, Stages 1, 2, and 3 were not seen below 18 for both genders.

Table 6 presents the comparison of OPG and CBCT scores for RPV staging. After evaluation, κ values were found to be 0.260, 0.255, and 0.312 ($p < 0.001$) for males, females and both genders indicating a fair agreement. The number of matching scores of OPG and CBCT for Stage 0 was 240. For Stage 1, it was 25, for Stage 2 it was 13, and for evaluation of Stage 3, it was 2. Considering the incompliant results, there were five cases scored as Stage 3 with OPG and Stage 0 with CBCT.

Discussion

Recently, it has become increasingly important to determine the age of living individuals.[8]

Various methods have been developed to estimate chronological age based on tooth development. This complex process takes place from early fetal life to adulthood affecting dental tissue and tooth morphology.[35] After the root development is completed, root pulp disappears over time due to the secondary dentin, which continues to be produced for life, and age determination can be made radiographically by determining RPV.[21] There is also another dentin formation, the tertiary dentin, which is a reparative reaction to different external

Table 2 Identifying age distributions of RPV stages for CBCT group based on gender

Stage	*Sex*	*N*	*Mean*	*SD*	*Min*	*25th percentile*	*Median*	*75th prcentile*	*Max*
0	**Male**	171	30.46	9.490	16	22.00	28.00	38.00	60
	Female	118	30.47	8.290	17	23.75	32.00	38.00	53
1	**Male**	44	38.55	9.672	19	31.00	40.00	45.00	68
	Female	30	37.63	8.244	18	30.75	38.50	43.00	55
2	**Male**	35	41.91	6.099	31	34.25	45.00	48.75	60
	Female	24	37.71	8.903	16	30.00	33.00	42.75	51
3	**Male**	5	40.40	11.502	32	37.00	40.00	41.00	60
	Female	2	36.50	4.950	33	34.50	39.50	44.50	40

CBCT, cone beam CT; RPV, root pulp visibility.

stimuli and does not correlate with age. Identification of tertiary dentin can be challenging and requires clinical correlation such as the presence of different external stimuli such as chemical irritants, caries, and wear. The distinction between secondary and tertiary dentin can only be made histologically.[36] Therefore, as an important radiological differential of secondary dentin, teeth with exposure to such stimuli should be excluded from RPV studies. Since our study was a retrospective radiological study, teeth with any dental pathology were excluded from the study group.

The use of dental radiographs for age estimation is based on the observation of the morphologically distinct stages of mineralization, the degree of formation of root and crown structures of the teeth, the evaluation of eruption, and the intermixture of primary and adult dentitions.[35,37] As a fast, simple, and economical dental radiograph, OPG is commonly used in studies investigating the RPV of third molars for forensic age determination.[1,2,22–28] On the other hand, OPG may not precisely display the vertical spatial relationships due to the caudocranial tilt in its projection geometry. Additionally, the external oblique ridge may form a prominent protrusion in the third molar teeth region resulting in the superposition.[17] Therefore, inclusion and exclusion criteria should be carefully planned while conducting such age estimation studies evaluating RPV in OPG. While some researchers considered these inherent limitations of two-dimensional imaging and excluded positional, dental, and developmental anomalies,[2,23,24] in some studies, those criteria were not described in detail.[1,21,25,27] In our study, we formed our criteria from this perspective and excluded cases that may complicate evaluation.

Due to forensic concerns, the 18 year threshold has been an important age marker point for many studies for age estimation. In many studies, it is stated that individuals are more likely to be above the 18 year threshold for RPV Stage 2 and Stage 3.[1,2,21,24–28] Additionally, Stage 0 is not expected after this threshold as well.[22] On the other hand, some studies report different results for the 18 year threshold. In a study by Kumar et al, it was reported that males were seen below the age of 18 for Stage 2.[23] Perez-Mongiovi et al also reported females below the age of 18 for Stage 1.[28] In our study, Stage 1 and Stage 2 were found in CBCT groups, whereas Stages 1, 2, and 3 were not found in OPG groups below the age of 18. In accordance with the majority of cases, Stage 3 was not seen under the age of 18 for either the OPG group or the CBCT group in the present study. But interestingly, Stage 0 was seen for both OPG and CBCT groups after the age of 18. The different progression rates of the maturation and eruption of the third molar from other molar teeth (*i.e.* the first molars) examined in other age estimation studies may be responsible for that delay. Detection of Stage 0 in older people has also been reported by Timme et al with the age of 51.4.[26] Gök et al reported this maximum age as 40 in a Turkish population aged between 15 and 40.[1] In accordance with their results, Stage 0 was detected in individuals aging over 60 in our study.

Table 3 Identifying age distributions of RPV stages for OPG group based on gender

Stage	*Sex*	*N*	*Mean*	*SD*	*Min*	*25th percentile*	*Median*	*75th percentile*	*Max*
0	**Male**	165	30.48	9.913	16	23.00	29.00	37.00	68
	Female	122	30.76	8.593	16	24.00	29.50	36.25	53
1	**Male**	51	38.94	8.640	24	30.00	39.50	45.75	60
	Female	28	37.79	8.248	20	32.25	39.00	42.25	51
2	**Male**	20	42.20	8.307	27	38.00	40.00	45.00	60
	Female	16	35.94	8.629	25	33.25	39.00	45.25	55
3	**Male**	19	37.58	5.491	25	32.00	39.00	49.50	44
	Female	8	39.63	5.125	33	33.00	36.50	N/A	47

OPG, panoramic radiography; RPV, root pulp visibility.

Table 4 Distribution of RPV stages for 18-years-old thresholds for female

	CBCT					*OPG*				
	<=18 age		**>18 age**		**Total**	**<=18 age**		**>18 age**		**Total**
Stage	**N**	**%**	**N**	**%**	**N**	**N**	**%**	**N**	**%**	**N**
0	5	4.2	113	95.8	118	8	6.6	114	93.4	122
1	1	3.3	29	96.7	30	0	0	28	100.0	28
2	2	8.3	22	91.7	24	0	0	16	100.0	16
3	0	0	2	100.0	2	0	0	8	100.0	8

CBCT, cone beam CT; OPG, panoramic radiography; RPV, root pulp visibility.

In the present study, we evaluated Olze et al.'s method from a different perspective.[21] To the best of our knowledge, there are no studies investigating the use of OPG in this age estimation method with CBCT. When evaluated separately, our CBCT and OPG results seemed consistent with age determination and both demonstrated a positive correlation between RPV stages and chronological age. But when both data were compared, matching scores of both evaluations were low. Regarding the minimum and maximum ages of all RPV stages, 12 of 16 age limit values were different for both OPG and CBCT evaluation. Regarding the RPV evaluations, for Stage 3, OPG was able to predict only 2 of 7 cases and evaluated the remaining cases as Stage 1 and 2. For Stage 0, evaluation with OPG was unable to predict 49 cases of 289 cases and evaluated 5 cases as Stage 3.

Considering the legal importance of the 18 year threshold, this misinterpretation may be problematic. Additionally, for OPG, κ values indicated a fair agreement with the CBCT evaluation. As the reason for this, the 2D nature of OPG, anatomical noise (*i.e.* presence of surrounding bone), geometric distortion, and superposition of oral structures include difficulties in its assessment. In the third molar teeth region, the external oblique ridge may be superimposed on the pulp tissue and the RPV evaluation may be incorrect. And the perpendicular to the sagittal plane X-ray beam is not always orthogonal for the third molar teeth due to the caudocranial tilt. CBCT can be used to easily visualize the roots of third molars without any magnification or distortion, with superior spatial resolution, 3D examination, as well as the buccolingual inclination

Table 5 Distribution of RPV stages and 18-years-old thresholds for male

	CBCT					*OPG*				
	<=18 age		**>18 age**		**Total**	**<=18 age**		**>18 age**		**Total**
Stage	**N**	**%**	**N**	**%**	**N**	**N**	**%**	**N**	**%**	**N**
0	11	6.4	160	93.6	171	11	6.7	154	93.3	165
1	0	0	44	100.0	44	0	0	51	100.0	51
2	0	0	35	100.0	35	0	0	20	100.0	20
3	0	0	5	100.0	5	0	0	19	100.0	19

CBCT, cone beam CT; OPG, panoramic radiography; RPV, root pulp visibility.

Table 6 Comparison of OPG and CBCT scores for RPV staging

			OPG stages				
			0	***1***	***2***	***3***	***Total***
Male	**CBCT stages**	**0**	137	22	7	5	171
		1	18	13	4	9	44
		2	10	13	7	5	35
		3	0	3	2	0	5
	Total		165	51	20	19	255
Female	**CBCT stages**	**0**	103	11	4	0	118
		1	9	12	6	3	30
		2	10	5	6	3	24
		3	0	0	0	2	2
	Total		122	28	16	8	174
Total	**CBCT stages**	**0**	240	33	11	5	289
		1	27	25	10	12	74
		2	20	18	13	8	59
		3	0	3	2	2	7
	Total		287	79	36	27	429

CBCT, cone beam CT; OPG, panoramic radiography; RPV, root pulp visibility.

of each tooth and root positions in the alveolar bone. For example, in our study results, since OPG is a two-dimensional imaging method, the root canal pulp can be viewed uninterruptedly, and but in the axial sections obtained by CBCT, it determined that the visibility of the root pulp may be interrupted with the buccolingual direction is partially filled with secondary dentin. Therefore, Stage 0 on OPG images was detected at a minimum age of 16 years in females and at a minimum age of 17 years on CBCT images. For this reason, we recommend if the individual has a recent CBCT and OPG, the RPV evaluation of the mandibular third molar should be supported with CBCT.

The main limitation of this study was the lack of uniformity in the distribution of individual numbers in each age range and each gender group. The lack of more than one observer in this study, which was based on a single observer, and the fact that inter observer agreement could not be evaluated is also a limitation. Another limitation can be considered as the absence of the verification of CBCT evaluation. Especially for the teeth with occlusion, there is always a chance of tertiary dentin formation due to the stimulating effect of post-eruptive changes, such as the functional wear leading to minor attrition.[38] Such minor changes may be radiologically and clinically overlooked and may be evaluated as the secondary dentin. Therefore, a histological examination would differentiate the secondary dentin and be a gold-standard for CBCT. But this would be possible only with the extraction of the teeth, which would not be applicable for the age estimation of the living individuals. Also, further RPV staging studies investigating the radiological evaluation methods are needed with larger and homogeneously distributed age and gender groups.

Conclusion

In conclusion, the absence of Stage 3 at age of 18 and under might have a forensic value for Olze's age estimation method. The presence of Stage 3 can be used to identify individuals over 18. The use of CBCT facilitated the investigation of root canals in detail. Considering the fair agreement in the verification of OPG scores, the use of CBCT for RPV evaluation is recommended for available cases.

Competing interests

The authors declare that they have no conflict of interest.

Funding

No funding to declare.

Ethics approval

The study design was approved by the Ethics Committee of the Bolu Abant Izzet Baysal University Faculty of Medicine.

REFERENCES

1. Gok E, Fedakar R, Kafa IM. Usability of dental pulp visibility and tooth coronal index in digital panoramic radiography in age estimation in the forensic medicine. *Int J Legal Med* 2020; **134**: 381–92. doi: https://doi.org/10.1007/s00414-019-02188-w
2. Akkaya N, Yılancı H Özge, Boyacıoğlu H, Göksülük D, Özkan G. Accuracy of the use of radiographic visibility of root pulp in the mandibular third molar as a maturity marker at age thresholds of 18 and 21. *Int J Legal Med* 2019; **133**: 1507–15. doi: https://doi.org/10.1007/s00414-019-02036-x
3. Jayaraman J, Roberts GJ, Wong HM, McDonald F, King NM. Ages of legal importance: implications in relation to birth registration and age assessment practices. *Med Sci Law* 2016; **56**: 77–82. doi: https://doi.org/10.1177/0025802415590172
4. Turkish Penal CodeLaw no:5237. Official Gazette. 2004;(25611) (in Turkish).. Available from: https://www5.tbmm.gov.tr/kanunlar/k5237.html.
5. Schmeling A, Dettmeyer R, Rudolf E, Vieth V, Geserick G. Forensic age estimation: methods, certainty, and the law. *Dtsch Arztebl Int* 2016; **13**: 44–50.
6. Schmeling A, Grundmann C, Fuhrmann A, Kaatsch H-J, Knell B, Ramsthaler F, et al. Criteria for age estimation in living individuals. *Int J Legal Med* 2008; **122**: 457–60. doi: https://doi.org/10.1007/s00414-008-0254-2
7. Wittschieber D, Ottow C, Vieth V, Küppers M, Schulz R, Hassu J, et al. Projection radiography of the clavicle: still recommendable for forensic age diagnostics in living individuals? *Int J Legal Med* 2015; **129**: 187–93. doi: https://doi.org/10.1007/s00414-014-1067-0
8. Schmeling A, Olze A, Reisinger W, Geserick G. Age estimation of living people undergoing criminal proceedings. *Lancet* 2001; **358**: 89–90. doi: https://doi.org/10.1016/S0140-6736(01)05379-X
9. Cunha E, Baccino E, Martrille L, Ramsthaler F, Prieto J, Schuliar Y, et al. The problem of aging human remains and living individuals: a review. *Forensic Sci Int* 2009; **193**(1–3): 1–13. doi: https://doi.org/10.1016/j.forsciint.2009.09.008
10. Demirjian A, Goldstein H. New systems for dental maturity based on seven and four teeth. *Ann Hum Biol* 1976; **3**: 411–21. doi: https://doi.org/10.1080/03014467600001671
11. Nolla CM. The development of the permanent teeth. *J Dent Child* 1960; **27**: 254–66.
12. Cameriere R, Ferrante L, Cingolani M. Age estimation in children by measurement of open apices in teeth. *Int J Legal Med* 2006; **120**: 49–52. doi: https://doi.org/10.1007/s00414-005-0047-9
13. Schour I, Massler M. The development of the human dentition. *J Am Dent Assoc* 1941; **28**: 1153–60.
14. Hunt EE, Gleiser I. The estimation of age and sex of preadolescent children from bones and teeth. *Am J Phys Anthropol* 1955; **13**: 479–87. doi: https://doi.org/10.1002/ajpa.1330130308
15. Streckbein P, Reichert I, Verhoff MA, Bödeker R-H, Kähling C, Wilbrand J-F, et al. Estimation of legal age using calcification stages of third molars in living individuals. *Sci Justice* 2014; **54**: 447–50. doi: https://doi.org/10.1016/j.scijus.2014.08.005
16. Solheim T. Amount of secondary dentin as an indicator of age. *Scand J Dent Res* 1992; **100**: 193–9. doi: https://doi.org/10.1111/j.1600-0722.1992.tb01740.x
17. White SC, Pharoah MJ. *Oral Radiology: Principles and Interpretation*. 6th edn. St. Louis, Mo, USA: Mosby; 2009.
18. Kvaal SI, Kolltveit KM, Thomsen IO, Solheim T. Age estimation of adults from dental radiographs. *Forensic Sci Int* 1995; **74**: 175–85. doi: https://doi.org/10.1016/0379-0738(95)01760-G
19. Drusini AG. Age estimation from teeth using soft x-ray findings. *Anthropol Anz* 1993; **51**: 41–6. doi: https://doi.org/10.1127/anthranz/51/1993/41
20. Cameriere R, Ferrante L, Cingolani M. Variations in pulp/tooth area ratio as an indicator of age: a preliminary study. *J Forensic Sci* 2004; **49**: 1–3. doi: https://doi.org/10.1520/JFS2003259
21. Olze A, Solheim T, Schulz R, Kupfer M, Schmeling A. Evaluation of the radiographic visibility of the root pulp in the lower third molars for the purpose of forensic age estimation in living individuals. *Int J Legal Med* 2010; **124**: 183–6. doi: https://doi.org/10.1007/s00414-009-0415-y
22. Balla SB, Ankisetti SA, Bushra A, Bolloju VB, Mir Mujahed A, Kanaparthi A, et al. Preliminary analysis testing the accuracy of radiographic visibility of root pulp in the mandibular first molars as a maturity marker at age threshold of 18 years. *Int J Legal Med* 2020; **134**: 769–74. doi: https://doi.org/10.1007/s00414-020-02257-5
23. Kumar G Kiran, Kumar DRS, Kulkarni G, Balla S, Shyam NDVN, Naishadham Y. Olze et al. stages of radiographic visibility of root pulp and cameriere's third molar maturity index to estimate legal adult age in Hyderabad population. *J Forensic Dent Sci* 2019; **11**: 84–9. doi: https://doi.org/10.4103/jfo.jfds_40_19
24. Al Qattan F, Alzoubi EE, Lucas V, Roberts G, McDonald F, Camilleri S. Root pulp visibility as a mandibular maturity marker at the 18-year threshold in the Maltese population. *Int J Legal Med* 2020; **134**: 363–8. doi: https://doi.org/10.1007/s00414-019-02155-5
25. Guo Y-C, Chu G, Olze A, Schmidt S, Schulz R, Ottow C, et al. Application of age assessment based on the radiographic visibility of the root pulp of lower third molars in a northern Chinese population. *Int J Legal Med* 2018; **132**: 825–9. doi: https://doi.org/10.1007/s00414-017-1731-2
26. Timme M, Timme WH, Olze A, Ottow C, Ribbecke S, Pfeiffer H, et al. The chronology of the radiographic visibility of the periodontal ligament and the root pulp in the lower third molars. *Sci*

Justice 2017; **57**: 257–61. doi: https://doi.org/10.1016/j.scijus.2017.03.004
27. Lucas VS, McDonald F, Andiappan M, Roberts G. Dental age Estimation-Root pulp visibility (RPV) patterns: a reliable mandibular maturity marker at the 18 year threshold. *Forensic Sci Int* 2017; **270**: 98–102. doi: https://doi.org/10.1016/j.forsciint.2016.11.004
28. Pérez-Mongiovi D, Teixeira A, Caldas IM. The radiographic visibility of the root pulp of the third lower molar as an age marker. *Forensic Sci Med Pathol* 2015; **11**: 339–44. doi: https://doi.org/10.1007/s12024-015-9688-2
29. Bell GW, Rodgers JM, Grime RJ, Edwards KL, Hahn MR, Dorman ML, et al. The accuracy of dental panoramic tomographs in determining the root morphology of mandibular third molar teeth before surgery. *Oral Surg Oral Med Oral Pathol Oral Radiol Endod* 2003; **95**: 119–25. doi: https://doi.org/10.1067/moe.2003.16
30. Andrews LF. The six keys to normal occlusion. *Am J Orthod* 1972; **62**: 296–309. doi: https://doi.org/10.1016/S0002-9416(72)90268-0
31. Wilson GH. *A Manual of Dental Prosthetics*. Philadelphia: Lea&Febiger; 1911.
32. Pippi R, Santoro M, D'Ambrosio F. Accuracy of cone-beam computed tomography in defining spatial relationships between third molar roots and inferior alveolar nerve. *Eur J Dent* 2016; **10**: 454–8. doi: https://doi.org/10.4103/1305-7456.195168
33. Kazmi S, Mânica S, Revie G, Shepherd S, Hector M. Age estimation using canine pulp volumes in adults: a CBCT image analysis. *Int J Legal Med* 2019; **133**: 1967–76. doi: https://doi.org/10.1007/s00414-019-02147-5
34. Landis JR, Koch GG. The measurement of observer agreement for categorical data. *Biometrics* 1977; **33**: 159–74. doi: https://doi.org/10.2307/2529310
35. Limdiwala PG, Shah JS. Age estimation by using dental radiographs. *J Forensic Dent Sci* 2013; **5**: 118–22. doi: https://doi.org/10.4103/0975-1475.119778
36. Timme M, Borkert J, Nagelmann N, Schmeling A. Evaluation of secondary dentin formation for forensic age assessment by means of semi-automatic segmented ultrahigh field 9.4 T UTE MRI datasets. *Int J Legal Med* 2020; **134**: 2283–8. doi: https://doi.org/10.1007/s00414-020-02425-7
37. Avon SL. Forensic odontology: the roles and responsibilities of the dentist. *J Can Dent Assoc* 2004; **70**: 453–8.
38. Mjör IA. Dentin permeability: the basis for understanding pulp reactions and adhesive technology. *Braz Dent J* 2009; **20**: 3–16. doi: https://doi.org/10.1590/S0103-64402009000100001