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Non-destructive dental-age calculation methods in adults: intra- and inter-observer effects

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Abstract

The aim of the present study was to obtain data on the reliability and reproducibility of two non-destructive dental-age estimation methods in adults by calculating inter- and intra-observer effects. Both a morphological and a radiological technique available in the scientific literature were evaluated on a number of recently extracted teeth: the morphological technique was evaluated on a total of 160 teeth by two examiners, while three examiners applied the radiological technique on apical radiographs of 72 extracted teeth. Paired *t*-tests were used to calculate intra- and inter-observer differences.

For the morphological method, both examiners were able to produce dental-age estimations that did not differ significantly from the real age of the teeth, obtaining a mean error between 0.5 and 1.8 years and a standard deviation of this error between 9.0 and 11.3 years. When using the radiological technique according to the original protocol, all three examiners produced age estimations that were statistically comparable to the real age of the teeth with a mean error of 0.5–2.5 years and a standard deviation of 4.6–9.8 years. For both techniques, intra-observer differences were observed.

Based on the results of this study, it can be concluded that both non-destructive dental-age estimation techniques were able to produce reasonably accurate dental-age estimations, at least when these techniques were applied appropriately. However, the forensic odontologist is recommended to use different age estimation techniques and perform repetitive measurements in order to verify the reproducibility of the calculations performed.

Keywords: Age estimation; Forensic odontology; Non-destructive; Teeth

1. Introduction

Estimation of age in individuals has received considerable attention in the forensic scientific literature as well as in archaeological cases. Some age estimates are based on the degree of closure of the sutures and epiphyseal plates [1], others on the degree of dental attrition [2,3]. Many other techniques have been reported but scientific statistical methods or specific dental methods are rarely used. However, forensic odontologists have a number of such methods available for use in their discipline. The search for optimal age estimation procedures has continued over the years until the present day. Age estimation in individuals, dead or alive, will continue to be of tremendous importance in various circumstances such as in the case of adult refugees.

Several techniques are available for dental-age estimation in children and because of the presence of a multitude of developing teeth, the accuracy of dental-age estimation in children is much greater (standard error (S.E.) ±1–2 years) [4–7] compared to adults (S.E. ±10 years) [8]. Basically, two main groups of well-known techniques based on dental maturation are available for this purpose, the atlas-techniques [4–6] and the scoring-techniques [7,9].

The same techniques may be applied in estimating the age in disaster victims. Any dental examination of a deceased person whose identity is unknown should include an estimation of age. In cases where there is no indication of the identity, age estimation may be crucially important for the

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progress of the investigation. It narrows down the search within the missing persons' files and enables a more efficient approach.

For adults, the literature offers different possibilities, such as morphological [10–15] and radiological techniques [16,17]. Among these techniques are very fine and relatively accurate methods that nowadays are available for forensic odontologists [18]. Some of these methods do not require grinding or sectioning of teeth and as such do not cause destruction of tooth substance. This may be important since the forensic odontologist does not always have permission to produce tooth sections in order to make an age estimation. Clearly tooth sectioning is not appropriate for living adults. Especially when determining the criminal responsibility of a young adult, the individual's chronological age might be out of the age range of the techniques developed for children. Non-destructive techniques for adults could then be used. As a first step, the experienced forensic odontologist should perform a visual age estimation of the tooth, teeth or dentition to be evaluated. This seemingly unscientific approach has shown to be of value in dental-age estimation, especially after obtaining conflicting and contradictory results when using different techniques [8]. The visual age estimation should be complemented by at least one other and preferably more age estimation techniques. In living individuals, only radiological techniques [16,17] may complement the visual age estimation. For deceased persons where no permission was granted for tooth sectioning, both the radiological age estimation techniques mentioned and an age estimation based on the length of the translucent zone of the tooth apex [12] are techniques that can be used in addition to visual age estimation. All these techniques are well described in the literature and offer the advantage of non-destructive age evaluation. However, questions may be raised concerning their reliability and reproducibility and whether non-experienced dentists are able to produce age estimation as accurate as their experienced colleagues.

The purpose of this study was to evaluate the inter- and intra-observer effects on two non-destructive age estimation methods in adults, one morphological [12] and one radiological [17] in order to obtain data on the reliability and reproducibility of these techniques.

2. Materials and methods

Anterior teeth and bicuspids from Belgian Caucasian origin, scheduled for extraction, were collected in the University Hospital St. Raphael (Katholieke Universiteit Leuven, Leuven, Belgium) and the University Hospital of Antwerp (University of Antwerp, Antwerp, Belgium). Each extracted tooth was stored separately in a plastic container filled with an aqueous solution of chloramine 0.5%.

The dentists who performed the extractions were given instructions on how to code the containers. They made lists of the assigned codes and wrote down the corresponding tooth number, the patient's gender, date of birth and the date of extraction.

This blind study design was intended as a pilot study for performing age estimations of 100 teeth. A sample of 160 teeth was collected. Only 100 of them were useful to estimate the age based on the formulas provided by Kvaal et al. [17]. These formulas were restricted to upper central and lateral incisor and second premolar, the lower lateral incisor, canine and first premolar. Taking two or more teeth (maximum six) from the same individual might influence statistical analysis, but since all the tooth types (upper central and lateral incisor and second premolars, the lower lateral incisor, canine and first premolars) were collected from different individuals, we evaluated the statistics as each tooth type derived from one individual. In order to perform the age estimations, three examiners applied the morphological method [12] to all 160 teeth and the radiological method [17] to 100 specific teeth. The selection was based on the different tooth types that were needed. Since they were only given the tooth number and gender of the patient corresponding to the different assigned codes, they were able to calculate the age of the patient independently and objectively. Two examiners (A and B) measured the transparent zone at the apex of each tooth according to the morphological method [12]. Examiner B repeated the observations with an interval of more than 2 months in order to check for possible intra-observer differences. Three examiners (A, B and C) collected data according to the radiological method [17] using a stereomicroscope. Examiners B and C repeated the measurements with an interval of more than 2 months for the same reason as mentioned earlier. All three examiners evaluated the same radiographs made by examiner B according to the specific instructions [17] and using correct viewing conditions [19] in order to obtain reliable results. Measurements were made in a dim room in order to improve concentration on the radiographs. Bright white backlight transillumination and masking facilities were used. In order to acquire the maximum possible information it was necessary to use an X-ray viewing box with facility to mask extraneous light not passing through the radiograph. Additionally, examiner B made the measurements with the naked eye and digital calipers and examiner C performed the same measurements with the naked eye and digital calipers but made use of a low power magnifying glass.

The morphological technique using translucency [12] consists of measuring the length of the transparent zone at the apex of an intact root. Bang and Ramm [12] included intact, restored and decayed teeth. Only root-filled teeth were excluded. The outcome of the measurements from 926 teeth, which they obtained, was statistically analyzed and age has been computed as the dependent variable from both a first- and a second-degree polynome. This was done after dividing the material in different groups based on two parameters, firstly whether the tooth was sectioned or not

and secondly, whether the transparent zone was smaller (and equal to) or larger than 9 mm. For age estimation based on teeth with a transparent zone less than or equal to 9 mm, a second-degree polynome should be used and in excess of 9 mm a first-degree polynome. Bang and Ramm [12] were able to define first- and second-degree polynomes for upper and lower centrals, laterals, cuspids, first and second premolars and first molars with a distinction for all tooth types between left and right.

The second technique evaluated in the present study is a radiological technique [17]. This method is based on apical radiographs of a selection of 100 patients with an age range between 20 and 87 years and a mean age of 42.6 years. The parameters evaluated on the radiographs are the length of tooth, root and pulp and the width of root and pulp at three different levels, namely cemento–enamel junction (level A), at one quarter root length from level A (level B) and in the middle of the root (level C). Based on these measurements different length and width ratios as well as correlations with age were calculated. Principal component analysis and regression analysis resulted in regression formulas for a selection of six teeth (left or right), three maxillary teeth (central and lateral incisors, second premolars) and three mandibular teeth (lateral incisor, cuspid, first premolar) [17].

The 100 teeth assembled for this pilot study correspond to an age range from 26 to 85 years of age with a mean age of 50 years. The teeth were extracted in 29 patients, 12 females and 17 males. Since the original article [17] reported a mean age of 42.6 years and a relatively small number of patients above 60 years of age, all teeth belonging to patients above 60 years were eliminated from the radiological study. Seventy-two instead of 100 teeth remained resulting in an age range of 26–60 years with a mean age of 44.9 years.

The results were statistically evaluated by means of a univariate procedure of the SAS statistical program (SAS Software Package, SAS Institute, Cary, NC, USA). This allowed application of a paired *t*-test in order to determine whether the age estimates significantly differ from the real age of the patient. Paired *t*-tests were also used to evaluate intra- and inter-observer variations. For these evaluations, α was set at 0.01. The mean error as well as its standard deviation between the estimated age and the real age was calculated for those results that did not significantly differ from the real age.

3. Results

The results of the statistical analysis of the age estimations are displayed in Tables 1–4. The *P*-values for the morphological method [12] are listed in Table 1. No significant differences were found between the real age of the tooth and the estimated age. This was true for both examiners A and B. On the other hand, intra-observer evaluation for examiner B showed a significant difference between the two measurements (B1 versus B2, Table 1). Inter-observer evaluation resulted in no significant statistical differences (A versus B1 and A versus B2). The three measurements made (A, B1 and B2), resulted in a mean error and standard deviation for the difference between the age estimation and the real age of, approximately 0.50–2.00 years and of 9.04–11.26 years, respectively (Table 2).

Table 1
Inter- and intra-observer differences (*P*-values) of age estimations performed by two examiners (A and B) according to the morphological method [12]

	RA	A	B1	B2
RA	–	0.53	0.09	0.52
A	–	–	0.09	0.04
B1	–	–	–	**
B2	–	–	–	–

RA: real age; A, B: examiners A or B; 1, 2: first or second measurement.
** $P < 0.001$.

Table 2
Mean error and standard deviation of the age estimations performed by both examiners A and B according to the morphological technique [12] in relation to the real age

Examiner	Mean error (years)	S.D. (years)
RA-A	−0.55	9.04
RA-B1	−1.84	11.26
RA-B2	0.64	10.27

RA: real age; A, B: examiners A or B; 1, 2: first or second measurement.

When performing the radiological method, *t*-tests showed for all examiners significant statistical differences between their age estimates and the real age of the patient with a *P*-value of less than 0.01. Inter-observer evaluations resulted in significant differences between the three examiners. Also significant intra-observer differences were found.

Table 3 lists the *P*-values for the radiological technique [17] based on teeth from patients under the age of 60 years. Using a stereomicroscope, examiner A made age estimations that did not really differ statistically from the real age. Examiner B, using his naked eye, did not succeed in making sound and reproducible age estimations, since for all inter- and intra-observer evaluations highly statistically significant differences were found. Examiner C (using naked eye with help of low power magnifying glass) made age estimations comparable to those made by examiner A. However, the estimates of examiner C did differ significantly from the real age of the patient. Examiner C was also not able to reproduce his measurements accurately since a significant intra-observer effect was found when measuring with the naked eye.

Using a stereomicroscope, examiners B and C succeeded in producing age estimations that were statistically comparable to the real age of the individual, which was a remarkable

Table 3
Inter- and intra-observer differences (*P*-values) of age estimations performed by all the three examiners (A, B and C) according to the radiological method [17]

	RA	A (st)	B1 (ne)	B2 (ne)	B (st)	C1 (ne + mg)	C2 (ne + mg)	C (st)
RA	–	0.03	***	***	0.54	***	**	0.02
A (st)		–	***	***	0.11	0.19	0.55	***
B1 (ne)			–	***	***	***	***	***
B2 (ne)				–	***	***	***	***
B (st)					–	***	***	0.03
C1 (ne + mg)						–	***	***
C2 (ne + mg)							–	***
C (st)								–

Teeth used belonged to individuals beneath the age group of 60 years. RA: real age; A, B, C: examiners A or B or C; 1, 2: first or second measurement; st: stereo microscope; ne: naked eye; mg: magnifying glass.
** $P < 0.001$.
*** $P < 0.0001$.

Table 4
Mean error and standard deviation of the age estimations performed by all three examiners (A, B and C) according to the radiographical technique [17] in relation to the real age

Examiner	Mean error (years)	S.D. (years)
RA-A (st)	2.54	9.80
RA-B (st)	−1.28	4.61
RA-C (st)	0.48	6.60

RA: real age; A, B, C: examiners A or B or C; st: stereomicroscope.

improvement of the results compared to not using a stereomicroscope. Table 4 lists the mean error and standard deviation (in relation to the real age) of the age estimations performed by all three examiners according to the radiological technique [17] and using a stereomicroscope.

4. Discussion

In some studies, the extracted teeth have been stored in tap water at room temperature for up to 6 weeks or more, without disinfection or sterilization [20]. Pulp tissue and components of dentin may be degraded and bacteria may alter the pulpo–dental complex. The same applies to teeth stored in physiological saline solution [21].

In order to avoid infection and preserve tooth structure, teeth have been stored either in buffered 10% formalin [22,23] or in a 70% alcohol solution [24] for various periods. These solutions may also have some negative influence on the organic components of the dentin. This may also be true for teeth stored in water and Thymol solution [25] or a 1% chloramine solution [26]. Storage of teeth in chloramines has been recommended when freezing facilities are not available [27]. Previous research concluded that no major changes occurred in the mineral phase of the teeth stored for 4 months in water containing 0.5% chloramines at 4 °C, but that storage might result in loss of the peritubular dentin [28,29]. This storage medium is not expected to interfere with the analyses carried out in this study.

Applying the morphological method [12] resulted in comparable findings between examiners A and B, although it is clear that examiner A scored best. The intra-observer evaluation for examiner B pointed out statistically significant differences between his first and second measurement (Table 1), although when compared to the real age of the patient the difference of each individual measurement was not statistically significant. The reason for this finding might be explained by the experience both examiners have with this technique. Examiner A was a very experienced forensic odontologist familiar with the age estimation technique. Examiner B had very limited experience which might have reduced the precision and the accuracy of his measurements. The difference in level of experience might explain why there was an intra-observer error. The alternative is that the method is not objective enough to prevent such errors.

Reducing the number of teeth to a group of patients aged less than 60 years of age was judged to be justified. Kvaal et al. [17] note that in the older age groups (above 60 years of age), it was difficult to find patients who retained all the six teeth that were measured in their study. Not only was the older age group less represented because of the smaller number of patients available, the patients selected did not all present the six permanent teeth studied. This fact might have influenced the dental-age calculation procedures resulting in a less accurate dental-age estimation for the older age group. It is also clear with any method that the variation in older persons is larger than in younger persons. That might explain why introducing the age limit of 60 years in the present study results in several significant findings, as reported in Table 3.

Although, the measurements made according to this radiological method [17] by examiners A and C were statistically comparable, the estimates produced by examiner C differed from the real age while the estimates

produced by examiner A did not (Table 3). Both experience and the use of a stereomicroscope when performing the measurements might explain this difference. Examiner A made length and width measurements using a stereomicroscope as described in the original article probably resulting in more accurate and reproducible results. Examiner C used a low power magnifying glass. This is frequently done by forensic odontologists not having access to a stereomicroscope. This technique might be prone to more inaccurate measurements than when using the stereomicroscope. Performing the linear measurements with the naked eye (examiner B) seems to render the least accurate results. It is concluded that in order to produce reliable results with this radiological technique, a stereomicroscope should be used for performing the linear measurements accurately. This was proven in this study by examiners B and C through repeating the measurements while using a stereomicroscope. Similar results were found comparable to the results obtained by examiner A. When using a stereomicroscope for the radiological technique as described [17], the three different examiners obtained age estimates that were statistically comparable to the real age.

The mean error and standard deviations found and reported in Table 4 should always be taken into account in view of the biological variation that exists in nature. In view of the different levels of experience of all three examiners it is concluded that even with little experience, the radiological technique performs well when the original protocol is followed.

In this respect, it might be worthwhile to produce a calibrated digital image of the apical radiograph in order to be able to perform digital linear measurements. This might deliver the most accurate measurements since we believe that digital stereomicroscopic magnification of the calibrated image would increase the accuracy of the measurements and thus result in more accurate age estimations.

It might appear that the magnifying glass technique used by examiner C, although inferior to the stereomicroscopic technique, was able to produce acceptable results. These estimations were not statistically different from the estimations performed by examiner A (Table 3) who used the stereomicroscope. However, examiner C was not able to reproduce these measurements a second time, resulting in a significant intra-observer effect. It is concluded that the use of the stereomicroscope is critical in the procedure of this radiological age estimation technique.

The results of other studies confirm the present findings [30,31]. These studies report inter- and intra-observer differences when using dental methods for age estimation in children [30] as well as inter-observer differences when assessing age related factors in teeth [31]. Some authors suggest that care should be taken not to rely too much on the results of one single examiner, pointing out the importance of obtaining a second opinion. The same is true for the age estimation methods. Some methods may not be as reliable as they claim to be and the conscientious forensic odontologist should always use several dental-age estimation methods in order to check the obtained findings [18,32].

5. Conclusion

Two techniques for non-destructive age estimation described in literature have been compared in this pilot study. The morphological technique [12] resulted in accurate dental-age estimation regardless of the experience of the forensic odontologist. The radiological technique makes accurate age estimations possible in living adults, at least when the original protocol is carefully followed. This necessitates the use of a stereomicroscope. Further research is needed to evaluate whether the development of new digital technology can result in a reliable and reproducible dental-age estimation.

An important aspect in dental-age estimation is that the investigator should apply a number of different techniques available and perform repetitive measurements and calculations in order to improve reproducibility and reliability of the age estimation.

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