

# Correlation between human chronological age and dentin volume of maxillary canine

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## ABSTRACT

**Introduction:** One of the identification methods in forensic odontology is to estimate age through tooth characteristics. With age, odontoblast cells that line the pulp cavity will continue the process of secondary dentin deposition at a lower rate. Generally, maxillary canines are teeth that can survive in the oral cavity until old age and are less likely to wear out. This study aimed to determine the correlation between human chronological age and the dentin volume of the maxillary canines using CBCT radiographs. **Methods:** An analytic study of the Pearson correlation test was conducted on 114 samples of CBCT radiographs of the maxillary canines from the Radiology Unit of the Rumah Sakit Gigi dan Mulut, Faculty of Dentistry, Universitas Padjadjaran. The inclusion criteria for this study were canines with closed apex, no caries, no attrition, no restorations, no orthodontic treatment, and generally good health. Maxillary canine dentin volume was measured using the application ITK-SNAP version 3.8.0. The results of dentin volume were analyzed using the Pearson correlation test using SPSS IBM Statistics 25. **Results:** Pearson correlation analysis showed the correlation coefficient value ( $r = 0.270$  and  $r = 0.427$ ) for the male and female samples, respectively. **Conclusion:** The correlation between chronological age and maxillary canine dentin volume in this study was not strong enough to be used as a single parameter in the age estimation method. Future research is expected to expand the sample size with a homogenous age distribution to obtain more consistent and accurate results.

**Keywords:** age estimation; dentin volume; maxillary canines; ITK-SNAP; CBCT.

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## INTRODUCTION

Age estimation through dental characteristics is one of the identification methods in forensic odontology.<sup>1</sup> The identification process through fingerprints and DNA often cannot be carried out

in cases of mass disasters due to the condition of the victims who suffered serious injuries or were found in an incomplete condition.<sup>2,3</sup> The age estimation method in mass disaster cases is one of the methods that can help simplify the identification process by grouping the ages of the

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victims. Age identification in law enforcement proceedings can help determine the punishment to be meted out to perpetrators who do not have clear identification documents.<sup>4,5</sup> One example of the role of age estimation in law enforcement process is in the murder case in North Nias District, a 15-year-old defendant was finally released from the death penalty after going through age identification process through the teeth using two age estimation methods and it was scientifically proven that YT was underage when the case occurred in 2012.<sup>6</sup>

Age estimation by teeth can be done through clinical, radiographic, histological, and biochemical examinations. Several age estimation examination techniques, such as histological examination, provide more accurate results compared to radiographic examinations, but to obtain these results requires the process of taking the dental tissue so it is considered an invasive method, so it cannot be performed on living individuals.<sup>7</sup> The age estimation method using radiographic images is a non-invasive method and is often used to this day.<sup>8</sup> Radiographic examination can produce two-dimensional images such as periapical radiographs, bitewing, panoramic, cephalometric or three-dimensional images such as CBCT radiographs.<sup>9</sup>

CBCT or Cone-Beam Computed Tomography is a type of radiographic examination that produces a three-dimensional image which includes the axial, coronal, and sagittal planes and provides a more detailed image than two-dimensional radiographic. CBCT radiography was used in this study because it can provide a more accurate volumetric assessment. Canines are considered as the object of age estimation research because canines are one of the teeth in the oral cavity that has one cusp and is the longest tooth among all teeth so that it can survive better from mastication and fracture forces.<sup>10,11</sup>

Maxillary canines can generally survive in the oral cavity until old age, and are less likely to experience wear and tear. Maxillary canines are teeth with a single root that have the largest pulp chamber space.<sup>10</sup> Odontoblast cells that are in the walls of the pulp cavity will continue the secondary dentine deposition process at a lower rate. This causes a reduction in the size of the pulp chamber space with age.<sup>12-14</sup> A study conducted by Arora

et al<sup>15</sup>, showed that the decreasing size of the pulp chamber with age is due to the formation of secondary dentine.<sup>15</sup> Andrade et al.<sup>12</sup> conducted a study on the correlation between age and sex based on pulp chamber volume. The result of this study showed a high correlation between chronological age and pulp chamber volume in these two types of teeth.<sup>12</sup> Pinchi et al<sup>16</sup>, conducted a study using 148 CBCT samples with a geometric approach method on various tooth areas. The result of this study indicates that the narrowing of the pulp chamber is a reliable parameter for determining the age of an adult individual.<sup>16</sup> Secondary dentin deposition which can affect dentin volume has the opposite effect on the pulp chamber which shrinks with age, so that in this study it is hoped that dentine volume can also be measured as an indicator of estimated age.

Many studies have been conducted on the correlation between chronological age and pulp volume using CBCT radiographs, but so far there has been little research on the correlation between chronological age and dentin volume in maxillary canines using CBCT radiographs. This study aim to analyzed the correlation of dentin volume in maxillary canines with chronological age using a volumetric segmentation system was carried out with the hope of contributing to the field of forensic odontology in the application of age estimation methods on the field.

## **METHODS**

An analytical study was carried out on 114 CBCT radiographs of maxillary canines from Radiology Unit RSGM Padjadjaran University. This study uses Pearson correlation test. The study population included patients who came to the RSGM Unpad Radiology Unit from 2013 to 2019. The age criteria for sampling was determined from the period when the roots of the canines were fully formed, which is from 13 years of age.

The sampling technique used was purposive sampling with sample inclusion criteria; Patients with an age range of 13-59 years, canine teeth with closed apex, no caries, no attrition, no restoration, and no orthodontic treatment. This study uses secondary data in the form of CBCT radiographs of the maxillary canines from the Vatech E-woo EPX impla (by Vatech Korea 2006)

and the OP300 maxio instrumentarium which had been exported to DICOM file format.

The instrument used in this research is the open-source software *ITK-SNAP* 3.8.0. to measure dentin volume. Dentin volume measurements were performed using the active contour segmentation mode (Figure 1 and 2). Data from dentin volume measurements were then recorded in Microsoft Excel and statistical analysis would be carried out using *SPSS IBM Statistics 25* software with the

Pearson correlation test to see the correlation between human chronological age and dentine volume of maxillary canines. This research was conducted at the Radiology Unit of the Dental and Oral Hospital, Padjadjaran University in March - June 2021. This study was approved by the Ethics Committee of the Faculty of Medicine, Padjadjaran University - Bandung, West Java, Indonesia with ethical number 214/UN6 .KEP/EC/2021 on March 22, 2021.

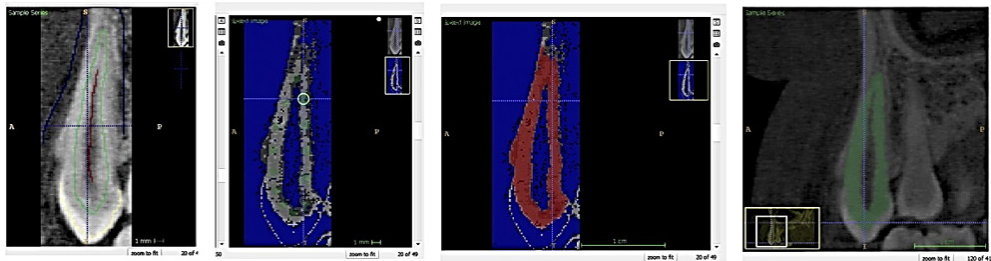


Figure 1. Steps of dentin segmentation using *ITK-SNAP*.  
(Source: personal)



Figure 2. Dentin segmentation result on *ITK-SNAP*.  
(Source: personal)

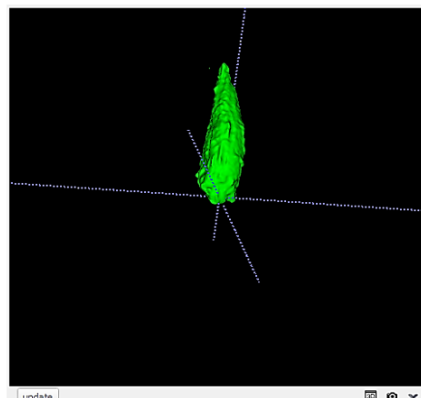


Figure 3. Three-dimensional view of dentin tissue  
(Source: personal)

## RESULTS

Table 1. Research sample distribution

|                   | Female  | Male  | Total |
|-------------------|---------|-------|-------|
| Sample Size       | 73      | 41    | 114   |
| Age Range (years) | 13 - 59 | 13-59 |       |

The reliability test was carried out before conducting research on the dentine volume of maxillary canines by conducting *intra-observer* and *inter-observer reliability tests*.

*Intra-observer* and *inter-observer* determined by conducting the *Intraclass Correlation Coefficient* (ICC) test. The ICC output for *intra-observer* was 0.991 and *inter-observer* was 0.855 (Table 2).

Table 2. Result of Inter- and intra-observer ICC test

| Reliability    | Intraclass Correlation Coefficient |
|----------------|------------------------------------|
| Intra-observer | 0.991                              |
| Inter-observer | 0.855                              |

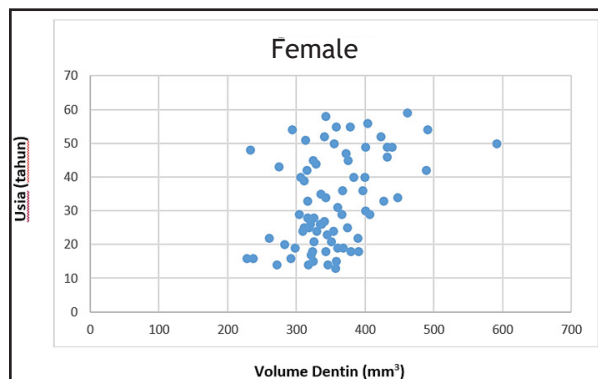


Figure 4. Dentine volume and chronological age of the female sample

This shows that there was no significant difference between the measurement results of the first observer and the second observer. The measurement results of dentin volume and

chronological age in the female sample in this study can be seen in Figure 4. The measurement results of dentin volume and chronological age in the male sample in this study can be seen in Figure 5.

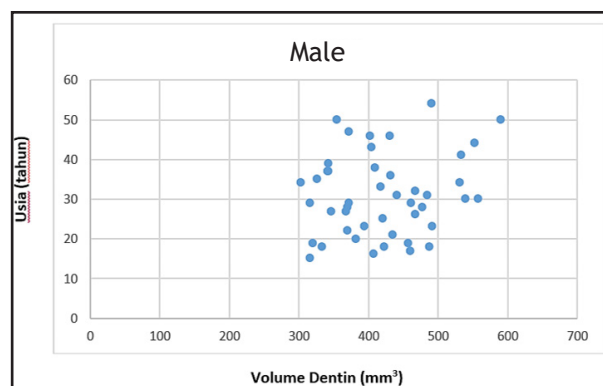


Figure 5. Dentine volume and chronological age of the male sample

Table 3. Pearson correlation test of chronological age and dentin volume of maxillary canines

| Gender    | Sig. (p) | Correlation Coefficient (r) |
|-----------|----------|-----------------------------|
| Perempuan | 0.00     | 0.427                       |
| Laki-laki | 0.088    | 0.270                       |

The result of the analysis of the female sample gave a  $p\text{-value} < 0.05$ , meaning that the correlation between age and dentin volume is significant. The value of  $r = 0.427$  also indicates that the correlation that occurs is of moderate value. The results of

the analysis of the correlation test for the male sample gave a  $p\text{-value} > 0.05$ , meaning that the correlation between age and dentin volume in the male sample was not significant. The value of  $r = 0.270$  also indicates that the correlation is weak.

Table 4. Pearson correlation test of chronological age and dentin volume of maxillary canines for all sample

| N   | Sig. (p) | Correlation Coefficient (r) |
|-----|----------|-----------------------------|
| 114 | 0.000    | 0.327                       |

Pearson correlation analysis for all sample (Table 4) provides a  $p\text{-value} < 0.05$ , meaning that the correlation between age and volume is significant. The value of the correlation coefficient

( $r = 0.332$ ) indicates that the relationship between human chronological age and dentin volume, if not taking into account the parameter of sex, is weak.

Table 5. Pearson correlation test of chronological age with dentin volume of right and left maxillary canines

| Canines | Sig. (p) | Correlation Coefficient (r) |
|---------|----------|-----------------------------|
| 13*     | 0.011    | 0.300                       |
| 23*     | 0.008    | 0.402                       |

\*13 : maxillary canine tooth in the right region

\*23 : maxillary canine tooth in the left region

Pearson correlation analysis (Table 5) for teeth 13 sample showed a  $p\text{-value} < 0.05$  and for teeth 23 showed a  $p\text{-value} < 0.01$ , meaning that the correlation between age and dentin volume of the maxillary canines in the right and left regions is significant. The correlation coefficient value of the right maxillary canines ( $r = 0.300$ ) indicates that the correlation is weak. The correlation coefficient value of the left maxillary canines ( $r = 0.402$ ) indicates that the correlation is moderate. These results indicate that there is a difference in correlation between the right and left maxillary canines with the maxillary canines in the left region having a stronger correlation than the maxillary canines in the right region.

## DISCUSSION

Teeth are the hardest part of the body and have the ability to withstand damaging factors, such as high temperatures, chemicals, and trauma.<sup>17,18</sup> The unique dental characteristics of each individual make identification through teeth a reliable identification method. Dentin is the largest part of the tooth which is formed by the odontoblast layer that lines the pulp chamber. Once the process of tooth root formation is complete, secondary dentine deposition continues to occur with age. Secondary dentin is formed throughout the life of the tooth as a result of physiological factors,

namely age and tooth eruption.<sup>19</sup> Physiological secondary dentin formation is one of the factors that can be used for age estimation methods using dentition.<sup>17</sup> This is supported by previous studies which found a close relationship between chronological age and secondary dentine.<sup>15,20</sup>

The Intraclass Correlation Coefficient (ICC) test was used in this study to test the intra-observer and inter-observer. ICC is also used to assess the reliability and validity between two or more observers in samples that have ratio data. ICC test value for intra-observer in this study is 0.991, which means that the reliability of each measurement is satisfactory because the value is above 0.9.

ICC value for intra-observer in this study indicates that the results of the first researcher's calculations are stable. ICC value of intra-observer also indicates that measurements using *semi-auto-segmentation* with the ITK-SNAP provide measurement results that are consistent with the role of the researcher in determining the area of image segmentation. ICC test value for Inter-observer was 0.855, meaning that there was no significant difference between the results of the first and second observer measurements. This shows that the first and second observer have the same perception in measuring volume using the ITK-SNAP application. The results of the Pearson correlation test analysis on the female sample

showed a correlation coefficient ( $r=0.427$ ). This shows that there is a moderate correlation between dentin volume and chronological age in the female sample. Pearson correlation test analysis on male sample showed a correlation coefficient ( $r = 0.270$ ) with a  $p\text{-value} > 0.05$ , this shows the correlation that occurred was not significant. The result of this study indicates that there are differences in the value of the correlation coefficient in both sexes with the correlation in the female sample being stronger than in the male sample.

This shows that gender can be a factor that influences age estimation, therefore gender must be differentiated in age estimation to get more accurate results. The results of this study are inconsistent with the study conducted by Biuki N et al.<sup>21</sup> who used pulp/tooth volume ratio in anterior teeth as an age estimation method with a stronger correlation in male samples and the study by Ge et al.<sup>22</sup> which showed a stronger correlation between maxillary canine pulp volume and age in the male sample. The result of this study indicates that there were significant differences in the correlation values between the female and male sample, which could be due to the less extensive distribution of the male sample and the non-homogeneous age distribution.

The result of this study indicates that the dentin volume was larger in the male sample. In this study, it was found that the mean volume of dentin in the male sample was  $417.88 \text{ mm}^3$ , while in the female sample it was  $349.37 \text{ mm}^3$ . This can be caused by sexual dimorphism in female and male teeth.<sup>23,24</sup> Several studies have found that sexual dimorphism in teeth is characterized by differences in dentin thickness.<sup>23,24</sup> Dentin tissue in males was found to be thicker than in females.<sup>23,24</sup>

Sexual dimorphism in these teeth can occur due to the influence of the X and Y chromosomes on the formation of dental hard tissue. The amelogenin gene is located on the X chromosome, it was found from several studies that the X chromosome affects the formation of enamel only, while the Y chromosome affects the formation of enamel and dentin.<sup>23</sup> Research by Raija et al.<sup>25</sup> found that in female subjects with chromosome 47,XXX, there was an increase in dentin growth in the root of the mandibular teeth indicating that the X chromosome had an effect on dentin growth. A history of abnormalities in

the production of the hormone estrogen can also affect dentin formation. Several studies have found that mice with OVX (ovariectomy) have thin predentin structures. The group with estrogen deficiency was found to have a lack of expression of proteins related to the mineralization process, such as RUNX2, which has an important role in dentine formation and mineralization.<sup>26,27</sup>

This shows that the lack of the hormone estrogen can affect the process of dentinogenesis. Research conducted by Son et al.<sup>28</sup> found that the potential for odonto/osteoblast differentiation in human dental tissue increases after being given E2 supplementation which could affect the growth of dentin tissue. This is in line with the study conducted by Woo et al.<sup>29</sup> Therefore, it is recommended that male and female samples have a different age estimation formula.

Analysis of the maxillary canines with a total of 72 teeth in the right region and 42 teeth in the left region with an age range of 13-59 years was carried out in this study. The result of this study indicates that there is a stronger correlation for teeth in the left region than the right region. The habit of mastication on one region can lead to differences in the physiological changes of the teeth and can lead to differences in the level of correlation between the left and right maxillary canines.<sup>30</sup>

Secondary dentin is deposited first on the palatal part of the pulp chamber then continues on the labial or incisal wall of the pulp chamber in the maxillary anterior teeth. This is influenced by the style of mastication.<sup>31,32</sup> The results of this study are in line with the results of the study by Li, Zhao et al.<sup>31</sup> who used the Kvaal method found that the maxillary canines in the left region had a stronger correlation than the right region.

The results of this study are not in line with the research conducted by Kazmi et al.<sup>33</sup> which showed a strong correlation between the pulp volume of the maxillary & mandibular canines, and the study by Asif et al.<sup>34</sup> which showed no significant difference in the correlation coefficient of the maxillary canines in the right and left regions. Pearson correlation test analysis on all sample with a correlation coefficient value ( $r=0.332$ ) which indicates that the correlation that occurs is weak. The uneven sample distribution between the right and left maxillary canines



may affect the result of the weak correlation between chronological age and dentin volume of maxillary canines. The results of this study are not in accordance with a study by Dehghani et al.<sup>35</sup> which showed a strong correlation in the maxillary canine/pulp ratio with a correlation coefficient value ( $r=0.794$ ). The sample used in this study have various FOVs with different pixel densities so that they can affect the accuracy of segmentation and the resulting volume when using ITK-SNAP application. Another possibility can be caused by radiopaque images (fillings, root canal treatment) on other teeth which can affect the image quality of the canines on CBCT radiographs.

Differences between secondary and tertiary dentin cannot be seen on CBCT radiographs. The history of orthodontic treatment in the subjects of this study cannot be known because the sample is in the form of secondary data. The inconsistent increase in dentin volume in this study could be caused by a history of orthodontic treatment which could lead to the formation of tertiary dentin which could cause changes in dentin volume. A study by Venkatesh, S et al.<sup>36</sup> demonstrated a reduction in the pulp chamber caused by the formation of tertiary dentine as a result of orthodontic treatment.

The inconsistent increase in dentin volume in this study could also be due to a history of drug use which could not be traced due to the use of CBCT radiograph as secondary data sample. A history of treatment with corticosteroids may contribute to inconsistent dentin volume measurements. Glucocorticoids / corticosteroids treatment have an effect on dentin formation. Some researchers found that research subjects who were given corticosteroids treatment experienced significant reduction of the pulp space compared to those who were not given corticosteroids. Increased dentin deposition was found in the mature teeth of subjects who were given corticosteroids.<sup>37,38</sup>

The open-source ITK-SNAP software was used in this study as a tool for measuring dentin volume of maxillary canines. Previous study has shown that ITK-SNAP provides accurate and efficient three-dimensional radiograph measurement results.<sup>14</sup> One of the advantages of using ITK-SNAP for three-dimensional radiograph analysis is good segmentation control, meaning errors can be minimized in volumetric analysis.<sup>39</sup>

This study shows that volumetric measurements using the ITK-SNAP provide stable results with an intra-observer of 0.990. The result of this study are similar to those of Gomes et al.<sup>40</sup> with an inter-observer ICC result of 0.9991,  $p<0$ .

This study uses secondary data as a research sample, so there is a lack of information regarding factors that can affect dentin volume, such as the patient's dental history, history of drug consumption, history of orthodontic treatment, dietary habits, bruxism, and systemic disorders that can affect dentin deposition. Research with large and homogeneous samples can provide more optimal results. Further research is recommended to test volumetric measurements using other segmentation software that can increase the level of accuracy and provide more stable measurement results. It is hoped that this research can contribute to the field of forensic odontology in the application of age estimation methods on the field.

## CONCLUSION

The correlation between chronological age and dentin volume of maxillary canines in this study was not strong enough to be used as a single parameter in age estimation method. Future research is expected to expand the sample size with a homogeneous age distribution in order to obtain more consistent and accurate results.

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