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EVALUATION OF ENAMEL THICKNESS OF UPPER ANTERIOR TEETH IN DIFFERENT AGE GROUPS BY DENTAL CONE BEAM COMPUTED TOMOGRAPHY SCAN *IN VIVO*

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ABSTRACT
The purpose of this study was to examine *in vivo* the possible variation in enamel thickness among upper anterior teeth. Linear enamel thickness of central, lateral incisors and canines was measured on 3D CBCT data, subsequently image refinement. Filters and parameters of the radiographic image were optimized for the protocol of measurements. Twenty-four patients, from 21 to 75 years, divided in three age groups were included into this study. Following selection criteria, 40 centrals, 42 laterals and 41 canines were examined. Teeth were segmented and then (3D) three-dimensionally reconstructed. Tooth enamel volumes were calculated from 492 CBCT crosscuts. On each clinical crown, 14 enamel spots of interest were selected and 1722 linear enamel measurements were obtained. Statistical evaluation was performed with t-test and One-way Analysis of Variance (ANOVA) at a confidence level of 95% ($p = 0.05$). The mean value of enamel decreased statistically significantly over time (Young 846μm, Middle 758μm, Aged 705μm). Enamel thickness was influenced by gender, type of tooth (only for canines), but not by the respective quadrant (right vs. left). Enamel, was also influenced by the area of measurement (thinnest in the palatal areas and thickest on the incisal edge). The greatest decrease over time was observed in the incisal edge. 0.5 mm is considered a safe amount of enamel reduction in proximal and buccal areas. Great care in needed when enamel removal is performed in incisal and palatal areas while the same amount of reduction could expose dentin.

INTRODUCTION

Enamel thickness has been the topic of dental investigations for almost a century. Many studies have measured enamel volume or obtained linear measurements using different methods [1-6]. Several authors studied sections of human posterior teeth and compared enamel thickness among species, while others emphasized on the phylogenetic significance of changes in relative enamel thickness [7-10]. Such measurements have provided important insight into primate taxonomic status [11, 12] and dietary adaptations [13, 14]. Other studies have examined the functional implications of enamel thickness and distribution [15-17]. Other investigators have focused on measuring the thickness of enamel or enamel and dentin in various portions of permanent dentition and their main objective was to evaluate the amount of enamel shell that

can be removed for the purpose of making dental restorations [18, 19].

The role of enamel in tooth shade has also been investigated by examining extracted human teeth and a correlation between the sizes of HA crystals and tooth shade has been revealed [20]. In addition, interaction between light and dental tissues is affected by the thickness of dentin and enamel [21]. Tooth crowns consist internally of pulp tissue surrounded by a dentin core and an enamel coating. It is well known that the three main factors that influence the overall visual perception are the lighting conditions, the tooth itself and the human eye and brain that finally perceive the color [22-25]. Both enamel and dentin have an influence on the light transmission and obviously play a significant role in the complex phenomenon of color and appearance of teeth. Investigating the degree of correlation has to do, among others, with quantifying enamel and dentine volume and obtaining linear measurements of the substrates, ideally *in vivo*.

Enamel thickness was assessed in the past using linear measurements of exposed enamel in fractured or worn teeth [26, 27]. Moreover, several studies of enamel thickness were performed on molar sections using histological methods or aggressively by grinding the tooth to the location of the desired plane and polishing that surface [28-30]. Fossil or extracted teeth are considered fragile structures and difficult to work with. Other researchers, in order to quantify dental tissues, have used lateral radiography [31] and medical computed tomography [32]. However, the previously mentioned methods of visualization have been demonstrated to result in inaccurate measurements of enamel thickness [33].

Nondestructive diagnostic procedures become commonplace and recent advances in three-dimensional medical imaging and multiple forms of high-resolution computed tomography have yielded accurate measurements of dental tissue thickness [34]. Three dimensional (3D) dental radiography and especially Cone-Beam Computed Tomography (CBCT) imaging is an important diagnostic adjunct to the clinical assessment of the dental patient, presenting many advantages and providing multiple head and neck applications [35].

The advantages over the conventional CT are the lower levels of radiation, lower operating time and cost, high resolution imaging of hard tissues and availability in smaller dental offices. The main disadvantages are inferior visualization and differentiation of soft tissue, streaking metal artifacts and the effect of patient motion on the resulting image sharpness, caused by heartbeat or breathing [36], even though image capturing is performed in apnea conditions. Originally, surgeons placing dental implants adopted CBCT imaging in order to visualize the volume and quality of alveolar bone and to assess the relationship with surrounding anatomical structures [37]. Clinical applications of CBCT also expand in the fields of dento-alveolar and maxillofacial surgery, orthodontics and endodontics [38-42].

The purpose of this research study was to examine in 3D the possible variation in enamel thickness among upper frontal teeth in vivo. The specific aims of this research were to measure the enamel thickness of central and lateral incisors and canines as well, in patients of different age groups using Cone Beam Computed Tomography realized for other purposes. The null hypothesis was that there were no statistically significant differences in the enamel thickness of the investigated teeth.

MATERIALS AND METHODS

The CBCTs of twenty-four patients, aged from 21 to 75 years, were studied in this research (Table 1). Patients were divided in three age groups, with equal number of males and females in each group. Age group A (up to 30 years old) had 8 patients, with age ranging from 21 to 30 years (mean age = 26 y). Age group B (from 31 to 50 years old) had 8 patients, with age ranging from 33 to 45 years (mean age = 39 y). Finally Age group C (more than 51 years old) had 8 patients, with age ranging from 51 to 75 years old (mean age = 60 y). CBCTs were chosen according to two obligatory selection criteria's. The first one was to have CBCT data from another dental treatment (i.e. implant treatment, complicated extraction of wisdom teeth etc.). The second was to have at least 3 intact vital upper anterior teeth without malformations, significant intrinsic colorations, fissures or restorations in the canine-to-canine region. Because of the second criterion, 21 teeth were excluded from the current study, meaning that instead of the anticipated 144 upper frontal teeth (48 centrals, 48 laterals, 48 canines) only 123 teeth were finally examined (40 centrals, 42 laterals, 41 canines).

The tomographic acquisition was performed using a Cone Beam Computer Tomography Device (Planmeca, Promax 3D Max, Finland) with the following acquisition parameters: All scans were output with 512 X 512 pixels per slice and 8 bits per pixel. Resultant voxels were isotropic having identical length, width and depth of 0.16 mm. The acquisition time for each slice was 12 seconds and the reconstruction time was 60 seconds at an angular increment of 0 degrees. WL/WW and CLUT parameters were manually inserted to the Osirix Image software after the following CBCT image adjustment procedure.

An intact extracted human central incisor was sent for 3D and CBCT scanning to enable the documentation of the 3D tooth dimensions and its internal morphology (enamel, dentine, pulp). After scanning procedures, the tooth was submerged in 10% hydrochloric acid under ultrasonic vibration for 18 minutes, in order to selectively demineralize enamel. The root-dentine core complex was again sent for 3D and CBCT scanning to enable the documentation of the remained 3D tooth dimensions.

The exact dimensions of the enamel core observed in 3D scanning pictures (fig 1) were obtained in the 3D

CBCT scanning images by modifying parameters and filters of the Imaging software. After defining the finest WL/WW and CLUT parameters, they were inserted in Osirix Imaging software (fig 2). Enamel linear measurements where then calculated with these acquisition parameters.

CBCT scanning data were saved in DICOM format (digital imaging and communications in medicine) and the previous mentioned professional medical imaging software (Osirix Dicom Viewer, 64-bit, MD 2.8.1, Mac OS Edition, Switzerland) was used for tissue segmentation, 3D reconstruction and enamel linear measurements.

Each clinical crown was schematically divided in thirds with two transverse and two sagittal planes on the working stone models (mean section distance was 3 mm). On the transverse sections, 4 enamel spots of clinical interest were indicated and examined (2 buccal & 2 proximal). On the sagittal sections, 3 enamel spots of clinical interest were indicated and examined (1 incisal & 2 palatal). A total number of 14 spots were indicated on each clinical crown (central, lateral, canine) in four sections (fig 3). These sections corresponded to CBCT crosscuts (fig 4a, b, c).

Finally on each transverse CBCT crosscut 4 enamel spots were found and on every sagittal CBCT crosscut, 3 enamel spots. A total number of 14 enamel linear measurements were obtained from each tooth for 123 teeth (40 centrals, 42 laterals, 41 canines).

These linear measurements were located in 4 CBCT crosscuts from each tooth. A total number of 492 CBCT crosscuts were examined for this study and 1722 enamel linear measurements were calculated. Each linear measurement was the mean value of five separate measurements at each separate spot of interest.

Data was imported for descriptive and inferential statistical analysis, using a dedicated software program (IBM SPSS Statistics for Mac, Version 20.0: SPSS, Inc, Chicago, IL). The values were statistically analyzed with t-test and One-way Analysis of Variance (ANOVA) at a confidence level of 95% ($p = 0.05$). A post-hoc Tukey's HSD (honestly significant difference) test was used for multiple pairwise comparisons between groups.

All patients were informed about the purpose of the study and signed an informed consensus for the use of the data of their existing cone-beam computed tomography for the purpose of this study, as well as for the photographic analysis and polysiloxane impressions that were necessary for the fabrication of stone models, which were also used in this study.

The Ethical Committee of the Dental School of the University of Geneva approved the research study design. The evaluation took place a few days after cleaning and polishing of the teeth and giving oral hygiene instructions to all of the patients that participated.

RESULTS

A wide range of enamel thickness values was observed (0-1671μm). Zero enamel values, which correspond to exposed dentin, were mainly located on the incisal edges (49/1722 measurements) and rarely on the palatal areas (6/1722 measurements). Most of the zero enamel values on incisal surfaces were found in aged patients (Aged: 36/49, Middle: 7/49, Young: 6/49). On the palatal surfaces, zero enamel values were observed only in aged (3/6) and middle-aged (3/6) patients, and not in young patients. No exposed dentin was recorded on the buccal and interproximal surfaces.

Important structural dimension differences in enamel thickness were identified among groups, as described in Table 2. The mean value of enamel thickness decreased over time (Young 846μm, Middle 758μm, Aged 705μm). Differences between all groups being significant ($p<0.05$).

Different genders also showed statistically significant difference between them, as shown in Table 3 ($p<0.05$). The mean value of enamel thickness was greater in females (805μm) than males (735μm).

Different types of teeth exhibited statistically significant differences between them, as shown in Table 4 ($p<0.05$). No significant differences were found between central and lateral incisors (CI=734μm, LI=745μm), but both were significantly different from canines (828μm) ($p<0.05$).

Right and left side did not exhibit significant differences, as shown in Table 5. There was no difference between right and left side central incisors (RCI=728μm, LCI=740μm), lateral incisors (RLI=737μm, LLI=753μm) or canines (RC=827μm, LC=829μm). Similar to the aforementioned comparison of different types of teeth, also when studying all different teeth individually, there was no significant difference between all four incisors. Canines were significantly different from both central and lateral incisors ($p<0.05$).

Different enamel spots exhibited significant differences between them, as shown in Table 6 ($p<0.05$). Enamel spot No 12, which corresponded to the distal incisal spot, showed the highest enamel thickness (916mm). It was significantly different from all other enamel spots ($p<0.05$). Enamel spots No 14 (670μm), No 11 (693μm), No 13 (706μm) and No 10 (730μm), which were located in the palatal area, exhibited significantly thinner enamel thickness than enamel spots No 2 (797μm), No 3 (802μm), No 7 (808μm) and No 6 (812μm), which were located on the buccal area ($p<0.05$).

Mesial and distal areas exhibited significant differences between them, as shown in Table 7 ($p<0.05$). Mesial area (757μm) exhibited significant thinner enamel than distal area (782μm) ($p<0.05$).

Different areas of teeth exhibited significant differences between them, as shown in Table 8 ($p<0.05$). Palatal area (700μm) had the thinnest enamel, which was significantly different from all other areas ($p<0.05$). Proximal area (769μm) did not show significant difference

from the buccal area (805μm), but was significantly different from the incisal area (839μm), which had the thickest enamel ($p<0.05$).

Different age groups exhibited significant differences in different types of teeth, as shown in Table 9 ($p<0.05$). The mean value of enamel thickness decreased with age in all teeth. Young patients had significant more enamel in comparison to the other age groups in all tooth types ($p<0.05$). Middle age patients had significantly more enamel in canines, but not in central or lateral incisors. (CI, Young 825 μm > Middle 710 μm ≠ Aged: 666 μm; LI, Young 813 μm > Middle 721 μm ≠ Aged 716 μm; C Young 892 μm > Middle 836 μm > Aged 740 μm) ($p<0.05$).

Different age groups showed significant differences of enamel thickness in the studied areas of different types of teeth, as shown in Tables 10 a, b, c, d ($p<0.05$). The mean value of enamel thickness decreased progressively with age in all areas in canines only.

In the palatal area (Table 10a) the middle age group was significantly different from the young age in all teeth ($p<0.05$). All age groups exhibited significant differences between them in canines only ($p<0.05$).

In the proximal area (Table 10b) significant differences were found between all age groups in central incisors and canines ($p<0.05$). In lateral incisors significant differences were found between the young and the other two age groups ($p<0.05$), without significant difference between the latter two.

In the buccal area (Table 10c) significant differences were found between all different age groups in all the teeth ($p<0.05$).

In the incisal area (Table 10d) significant differences were found between all age groups in central incisors only ($p<0.05$). Young and aged groups were significantly different in lateral incisors and canines ($p<0.05$). Middle age group was not significantly different from the other age groups in lateral incisors ($p<0.05$). Middle age group was significantly different from aged group but not from the young age group in canines ($p<0.05$).

DISCUSSION

There is only little 3D quantitative information available about enamel thickness in humans in clinical conditions in the literature, even though it has been for a long time considered of importance by several researchers and clinicians as well. The purpose of the present study to quantify the enamel volume of upper anterior teeth in various age groups, was well justified, as important findings were observed. The use of 3-D Cone Beam Computed Tomography proved useful and its nondestructive nature could enable the monitoring of the patients in the future, in order to see the loss of enamel over time. Significant 3D differences in enamel thickness in permanent dentition were documented among three age groups in a wide age range (age range 21 to 75). At the time of the present research, no similar methodology on three-dimensional data was found to compare these findings with other studies. Cone Beam Computed Tomography scanning technology is a very powerful tool and could overcome many limitations of previous evaluation methods. One of the most significant advantages is considered the possibility of examining in a completely nondestructive way the same patients and the intraoral condition of tooth structures at different periods of time. As CBCT is becoming more common, patients with CBCT obtained for other purposes than just studying enamel could be analyzed in large time intervals with this research protocol, in order to quantify changes in enamel on the long term. Very important conclusions could be drawn and considerations such as aging, wear and the physiologic loss of enamel, could be investigated in a more objective and effective manner.

The accuracy of CBCT has been investigated by Wang et al., who evaluated the accuracy of CBCT for volumetric measurement of teeth using Micro-Ct as the reference standard [43]. After examining 27 teeth before and after extraction, it was concluded that the CBCT method used for the volumetric measurement of teeth *in vivo* was comparable to the micro-CT method *in vitro*.

CBCT scanning technology could be proposed as an alternative method of measuring tooth crown dimensions. The specific technology offers the possibility of three-dimensional data and is indicated even in cases of overlapping and/or crowded teeth, without influencing the precision of the calculations. Young patients with orthodontic problems could be included in a research study such as the present one.

Magne *et al.*, described a methodology of measuring tooth crown dimensions but the destructive protocol used limited its application to extracted teeth. As a consequence, human maxillary anterior teeth of unknown origin and without real age assignment were used [44]. The method used in that research was considered accurate, as a caliper could be used for measuring the size of the extracted teeth. Nevertheless, that methodology would be difficult if not impossible to perform in patients with overlapping or crowded teeth. These difficulties are overcome with the use of CBCT technology and by applying the evaluation protocol used in the current study.

The present study demonstrated that age contributed to human enamel loss and confirms in a quantitative way the assumption of aging considerations and physiological loss of enamel. The three age groups examined presented significantly different enamel thickness values, which decreased with increasing age. Similar conclusions were drawn by Atsu *et al.*, [45], who demonstrated, by using regression analysis, that enamel thickness depends on chronologic age. Limitations of the aforementioned study were that only extracted human maxillary central incisors were examined, and that the results were obtained using a sectioning technique, which is destructive for the fragile teeth.

The present study showed that gender is a factor that influences enamel thickness, as females presented higher values of enamel thickness than males. Several authors demonstrated that males have larger tooth dimensions on average than females but at the same time it was shown that the width dentin portion of the crown was significantly larger also in males. This is the explanation of the findings of the present research together with the lower masticatory forces in females, which may contribute to less enamel loss. Other studies on permanent molars demonstrated that significantly higher mean enamel thickness in females is the result of greater enamel cusp volume together with smaller dentin volume.

The present study also showed that permanent central and lateral incisors present very close mean values of enamel thickness (CI: 734μm, LI: 745μm) but both of them are significantly thinner than canines (C: 828μm). The shape of teeth is influenced mainly by their position and use in the dental arches and the differences of the external dimensions of teeth have been well documented and described in dental anatomy books [46]. The present study examined variations of the thickness of enamel only. The difference of initial shape and their function in the oral environment certainly plays a role in the differences of enamel thickness observed in this research.

This research found no difference between central and lateral incisors, which can be explained by the fact that they erupt into the mouth within a relatively close time period, leaving thus similar period of time for development (7-9 years). On the other hand, both incisors were different from canines, which may be explained by the fact that canines erupt into the oral environment much later (11-13 years), so there is more time available for enamel formation.

The present study showed that there was no difference between the left and the right side of the patients in the anterior dentition, even though patients do not use symmetrically their teeth for chewing or para functional activities.

It was also shown that distal enamel was slightly thicker than mesial enamel when taking into account all mesial (757μm) and distal measurements (782μm), confirming a previous study [47]: Using standardized bitewing radiographs and the parallel technique the enamel thickness was assessed in maxillary incisors in a young group of patients (age range from 13 to 17 years old). The investigators measured the maximum mesio-distal crown width, the maximum distal enamel thickness and the maximum mesial enamel thickness. It was found that enamel thickness was greater of about 0.1 mm (100 μm) on the distal than on the mesial side of each tooth (both central and lateral). Mean values were of 0.91 mm and 1.05 mm respectively. The present 3D study measured two enamel spots, one located above and one below the maximum tooth width, with (3mm distance between them, on the mesial and on the distal area of each tooth. Even though statistical evaluation of the mesial (No1 and 5) and distal (No4 and 8) proximal enamel spots did not show significant differences between, the distal measurements presented higher mean values. The aforementioned two-dimensional study calculated enamel thickness in one spot, located on the maximum mesio-distal crown width, either mesially or distally. This was not accounted for in this research protocol, so the results are not directly comparable. Nevertheless, the related group to the previous study was the young group of patients' that exhibited mean mesial enamel thickness of 786.5 microns, while mean distal was found 810 microns (No1=819μm, No4= 754 μm, No5=754μm, No5= 866 μm).

Patients ask today for esthetic restorations with natural appearance and many researchers are focusing on this field [48-50]. Direct restorative procedures have become a common treatment performed in daily clinical practice [51]. Composite resin is considered the material of choice for minimally invasive direct techniques [52]. Layering techniques are probably the most predictable procedures to mimic the internal architecture of natural teeth, especially when restoring anterior dentition [53]. The goal of these techniques is to be able to mimic the internal structure and the different optical properties of dentin and enamel, and by using equivalent amounts of enamel and dentin materials to accomplish naturally appearing esthetic restorations. The present study showed interesting data on the morphology of the anterior teeth that can be applied in case of restoring class III, IV, V lesions and veneering.

Contemporary stratification or layering techniques often start from the palatal area, using a silicone index, and placing a thin layer of enamel material to restore the enamel tissue loss. Findings of the present study demonstrate that enamel thickness in the palatal area is approximately 0.6-0.7mm for central and lateral incisors and approximately 0.7-0.8mm for the canines. The lower values always correspond to the aged group of patients. In the layering process, proximal areas follow the palatal area. Proximal areas are important in direct restorations. Clinicians shouldn't exaggerate when placing the restorative enamel material in order to avoid a greyish appearance. Findings of the present study showed that for centrals and laterals a thickness of approximately 0.7-0.85mm is indicated. For canines a thickness of approximately 0.75mm-0.9mm was measured. Once again, the clinician should take into consideration the decreased enamel volume in aged patients.

Proximal areas are also of a paramount importance for orthodontic treatments. Cosmetic re-contouring and stripping in orthodontics are considered conservative esthetic procedures [54] and many orthodontists have increasingly focused on such non-extraction treatments [55]. Enamel reduction in the proximal areas is unavoidable and the elimination of the black triangles becomes easier [56]. Previous studies refer to potential iatrogenic effects including the increased frequency of caries, periodontal disease and temperature sensitivity [57-60]. The amount of enamel reduction while

stripping is questionable. It has been suggested that in case of mandibular anterior crowding with mesio-distal crown reduction of 0.3 of tooth structure of the four lower incisors and 0.4 mm of the enamel surfaces of the canines, approximately 4.0 mm of mesio-distal crown reduction may be obtained [61]. Findings of the present research confirm that if the same enamel reduction is applied on the proximal surfaces of the upper anterior teeth (central, lateral, canine) then no dentin will be exposed in any of the different age or gender groups examined.

Knowledge of enamel thickness in the buccal areas is important for both direct and indirect restorations. In the case of direct restorations the amount of enamel material should be less that 1 mm to avoid a greyish effect of the restoration,. According to the results of the present study, it should be approximately 0.75-0.85mm for centrals and laterals and 0.8-0.9mm for canines. The lower values refer to the aged group of patients. Other significant clinical applications are the indirect restorative procedures when preparing for partial (laminate veneers) and full coverage crowns. Different tooth preparation techniques have been described without the considerations of aging, wear and the physiologic loss of enamel. The ability of bonding to tooth structures has benefit tooth conservation for all the preparation techniques available, because the mechanical retention principles that were described in the previous years are no longer of great importance. It is well documented that bonding to tooth substrate is stronger in on enamel than on dentin [62].

Possible clinical risks and failures are combined with bonding in dentine especially in the case of ceramic veneers. It has been assumed that an average of 0.5mm (0.3-0.7mm) intra-enamel tooth reduction is considered acceptable [63-66]. Moreover other researchers claim that a tooth reduction of 0.5 mm might expose dentin [67, 68], which is considered one of the major risks of clinical failure of restorations [69]. Comparing previous results with the findings of the present study, average enamel thickness reduction in the buccal area of 0.3-0.7mm may be clinically acceptable. Nevertheless, in the present study the enamel thickness was not measured near the clinical cervix of the tooth, an area that is critical in the clinical practice.

In incisal areas the loss of enamel was more pronounced as age progressed, which can be explained by chewing and possible para functional habits. Thus, more patients in the aged group were found with zero enamel values, with exposed dentin. Detailed results of the enamel thickness of the incisal area, which are reported in table 10d illustrate that a large range of values was recorded. This finding confirms the clinical reality that clinicians face everyday when restoring Class IV restorations.

In relative intact teeth, when restoring the incisal edge with enamel-like restorative material, care should be given to place the appropriate amount of resin composite, in thickness not thinner than 1.1mm. In case of preparation for indirect restorations a reduction of less than 1.1mm would risk the dentin exposure. In the middle aged and aged group of patients these values decrease to 0.25mm for centrals, 0.7mm for laterals and 0.8mm for canines.

Conservation of tooth structure is very important and maintaining an enamel substrate for both direct and indirect restorations guarantees adequate bond to the tooth structure and precludes postoperative sensitivity.

Table 1. Data of patients and teeth that were examined

Group Age A (up to 30 years old)												
Cd	**Name**	**Sex**	**Age**	**Canine 13**	**Lateral 12**	**Central 11**	**Central 21**	**Lateral 22**	**Canine 23**	**Total Canines**	**Total Laterals**	**Total Centrals**
A1	C.I.	M	27	✓			✓	✓	✓	2	1	1
A2	C.I.	M	28	✓	✓	✓	✓	✓	✓	2	2	2
A3	N.D.	M	29	✓		✓	✓		✓	2	0	2
A4	V.K.	M	29	✓	✓		✓	✓	✓	2	2	1
A5	S.Z.	F	21	✓	✓	✓	✓	✓	✓	2	2	2
A6	N.K.	F	22	✓	✓	✓	✓	✓	✓	2	2	2
A7	L.T.	F	25	✓		✓	✓	✓		1	1	2
A8	M.M.	F	27	✓	✓	✓	✓	✓	✓	2	2	2
Totals				8	5	6	8	7	7	15	12	14
Group Age B (between 30–50 years old)												
B1	G.A.	M	33	✓	✓		✓	✓	✓	2	2	1
B2	L.H.	M	36	✓	✓	✓	✓	✓	✓	2	2	2
B3	B.D.	M	42	✓	✓	✓	✓	✓	✓	2	2	2
B4	P.P.	M	44		✓	✓		✓		0	2	1
B5	K.S.	F	35	✓	✓	✓	✓	✓	✓	2	2	2

B6	K.E.	F	37	✓	✓	✓	✓	✓	✓	2	2	2
B7	M.L.	F	41	✓	✓	✓		✓	✓	2	2	1
B8	B.D.	F	45	✓	✓	✓		✓	✓	2	2	2
Totals				7	8	7	5	8	7	14	16	12
Group Age C (more than 50 years old)												
C1	K.I.	M	59	✓	✓	✓	✓	✓	✓	2	2	2
C2	T.G.	M	59		✓	✓	✓	✓	✓	1	2	2
C3	C.G.	M	61	✓	✓	✓	✓	✓	✓	2	2	2
C4	B.P.	M	75	✓	✓	✓	✓	✓	✓	2	2	2
C5	C.S.	F	51	✓	✓	✓	✓	✓	✓	2	2	2
C6	C.M.	F	53	✓	✓	✓		✓	✓	2	2	1
C7	G.A.	F	62	✓		✓	✓			1	0	2
C8	B.C.	F	63		✓	✓	✓	✓		0	2	2
Totals				6	7	8	7	7	6	12	14	15
				21	20	21	20	22	20	41	42	41

Table 2. Enamel thickness measurements of different Age Groups (in microns)

Age Group	N	Mean	SD	Min	Max	F	Statistical Significance*
Young	574	846	190	0	1617	71.05	A
Middle	574	758	180	0	1523		B
Aged	574	705	234	0	1403		C
Total	1722	769	211	0	1617		
*Age Groups with different letters exhibited statistical difference at the level of 0.05 statistical significance.							

Table 3. Enamel thickness measurements of Males & Females (in microns)

Gender	N	Mean	SD	Min	Max	F	Statistical Significance*
Male	868	735	225	0	1617	48.63	A
Female	854	805	189	0	1523		B
Total	1722	769	211	0	1617		
*Gender Groups with different letters exhibited statistical difference at the level of 0.05 statistical significance.							

Table 4. Enamel thickness measurements of different type of Teeth (in microns)

Tooth	N	Mean	SD	Min	Max	F	Statistical Significance*
Central Incisor	560	734	214	0	1407	35.41	A
Lateral Incisor	588	745	197	0	1593		A
Canine	574	828	209	0	1617		B
Total	1722	769	211	0	1617		
*Teeth with different letters exhibited statistical difference at the level of 0.05 statistical significance.							

Table 5. Enamel thickness measurements of upper frontal Teeth (in microns)

Tooth	N	Mean	SD	Min	Max	F	Statistical Significance*
Right Central Incisor	280	728	202	0	1407	14.43	A
Right Lateral Incisor	280	737	208	0	1593		A
Right Canine	294	827	196	0	1617		B
Left Central Incisor	280	740	226	0	1370		A
Left Lateral Incisor	308	753	186	0	1283		A
Left Canine	208	829	222	0	1563		B
Total	1722	769	211	0	1617		
*Teeth with same letters did not exhibit statistical difference at the level of 0.05 statistical significance.							

Table 6. Enamel thickness measurements of different Enamel Spots (in microns)

SPOT	N	Mean	SD	Min	Max	F	Statistical Significance*
14	123	670	125	0	990	11.32	A
11	123	693	93	419	930		AB
13	123	706	140	0	1122		AB
10	123	730	126	0	1084		ABC
5	123	746	110	552	1096		ABC
1	123	758	88	521	970		BC
9	123	762	469	0	1433		BC
4	123	776	118	509	1154		BC
8	123	795	139	441	1318		C
2	123	797	71	614	1034		C
3	123	802	75	531	998		C
7	123	808	88	364	1128		C
6	123	812	79	584	1084		C
12	123	916	468	0	1617		D
Total	1722	769	211	0	1617		

*Enamel spots with different letters exhibited statistical difference at the level of 0.05 statistical significance.

Table 7. Enamel thickness measurements of Mesial & Distal areas (in microns)

Area	N	Mean	SD	Min	Max	F	Statistical Significance*
Mesial	861	757	201	0	1433	6.09	A
Distal	861	782	219	0	1617		B
Total	1722	769	211	0	1617		

*Gender Groups with different letters exhibited statistical difference at the level of 0.05 statistical significance.

Table 8. Enamel thickness measurements of different Areas (in microns)

Tooth	N	Mean	SD	Min	Max	F	Statistical Significance*
Palatal	492	700	123	0	1122	33.28	A
Proximal	492	769	116	441	1318		B
Buccal	492	805	78	364	1128		BC
Incisal	246	839	474	0	1617		C

*Areas with different letters exhibited statistical difference at the level of 0.05 statistical significance.

Table 9. Enamel thickness measurements of different Age Groups in different types of teeth (in microns)

Tooth	Age Group	N	Mean	SD	Min	Max	F	Statistical Significance*
Central Incisor	Young	196	825	171	0	1407	32.25	A
	Middle	154	710	152	0	1170		B
	Aged	210	666	256	0	1111		B
Lateral Incisor	Young	168	813	179	0	1593	14.50	a
	Middle	224	721	183	0	1293		b
	Aged	196	716	214	0	1350		b
Canine	Young	210	892	206	0	1617	27.14	α
	Middle	196	836	171	536	1523		β
	Aged	168	740	222	0	1403		γ

*Age groups with same letters did not exhibit statistical difference at the level of 0.05 statistical significance.

Table 10a. Enamel thickness measurements of Palatal Area (in microns)

Area	Tooth	Age Group	N	Mean	SD	Min	Max	F	Statistical Significance*
Palatal	Central Incisor	Young	56	701	65	582	863	5.80	A
		Middle	44	641	78	494	803		B
		Aged	60	670	110	395	894		AB
	Lateral Incisor	Young	48	715	123	484	1122	7.15	a
		Middle	64	636	131	0	778		b
		Aged	56	691	81	540	989		a
	Canine	Young	60	820	109	604	1084	16.99	α
		Middle	56	737	70	542	900		β
		Aged	48	673	194	0	962		γ
*Age groups with different letters exhibited statistical difference at the level of 0.05 statistical significance.									

Table 10b. Enamel thickness measurements of Proximal Area (in microns)

Area	Tooth	Age Group	N	Mean	SD	Min	Max	F	Statistical Significance*
Proximal	Central Incisor	Young	56	837	99	633	1072	26.70	A
		Middle	44	707	83	552	916		B
		Aged	60	751	90	521	952		C
	Lateral Incisor	Young	48	781	110	550	1154	10.80	a
		Middle	64	714	90	546	1074		b
		Aged	56	689	110	441	994		b
	Canine	Young	60	879	109	709	1318	23.18	α
		Middle	56	805	111	557	1136		β
		Aged	48	743	90	538	887		γ
* Age groups with different letters exhibited statistical difference at the level of 0.05 statistical significance.									

Table 10c. Enamel thickness measurements of Buccal Area (in microns)

Area	Tooth	Age Group	N	Mean	SD	Min	Max	F	Statistical Significance*
Buccal	Central Incisor	Young	56	846	60	752	1034	31.45	A
		Middle	44	764	39	676	848		B
		Aged	60	800	52	707	925		C
	Lateral Incisor	Young	48	825	66	666	990	28.21	a
		Middle	64	740	48	617	894		b
		Aged	56	778	63	657	937		c
	Canine	Young	60	874	88	699	1128	13.46	α
		Middle	56	826	68	695	965		β
		Aged	48	788	102	364	953		γ
* Age groups with different letters exhibited statistical difference at the level of 0.05 statistical significance.									

Table 10d. Enamel thickness measurements of Incisal Area (in microns)

Area	Tooth	Age Group	N	Mean	SD	Min	Max	F	Statistical Significance*
Incisal	Central Incisor	Young	28	1007	330	0	1407	33.77	A
		Middle	22	750	348	0	1170		B
		Aged	30	224	416	0	1111		C
	Lateral Incisor	Young	24	1051	291	0	1593	4.86	a
		Middle	32	866	383	0	1293		ab
		Aged	28	693	521	0	1350		b
	Canine	Young	30	1095	79	0	1617	6.15	α
		Middle	28	1118	47	536	1523		α
		Aged	24	769	98	0	1403		β
*Teeth with different letters exhibited statistical difference at the level of 0.05 statistical significance									

Figure 1. 3D scanning images of the enamel-root (upper images) & dentin-root (lower images)

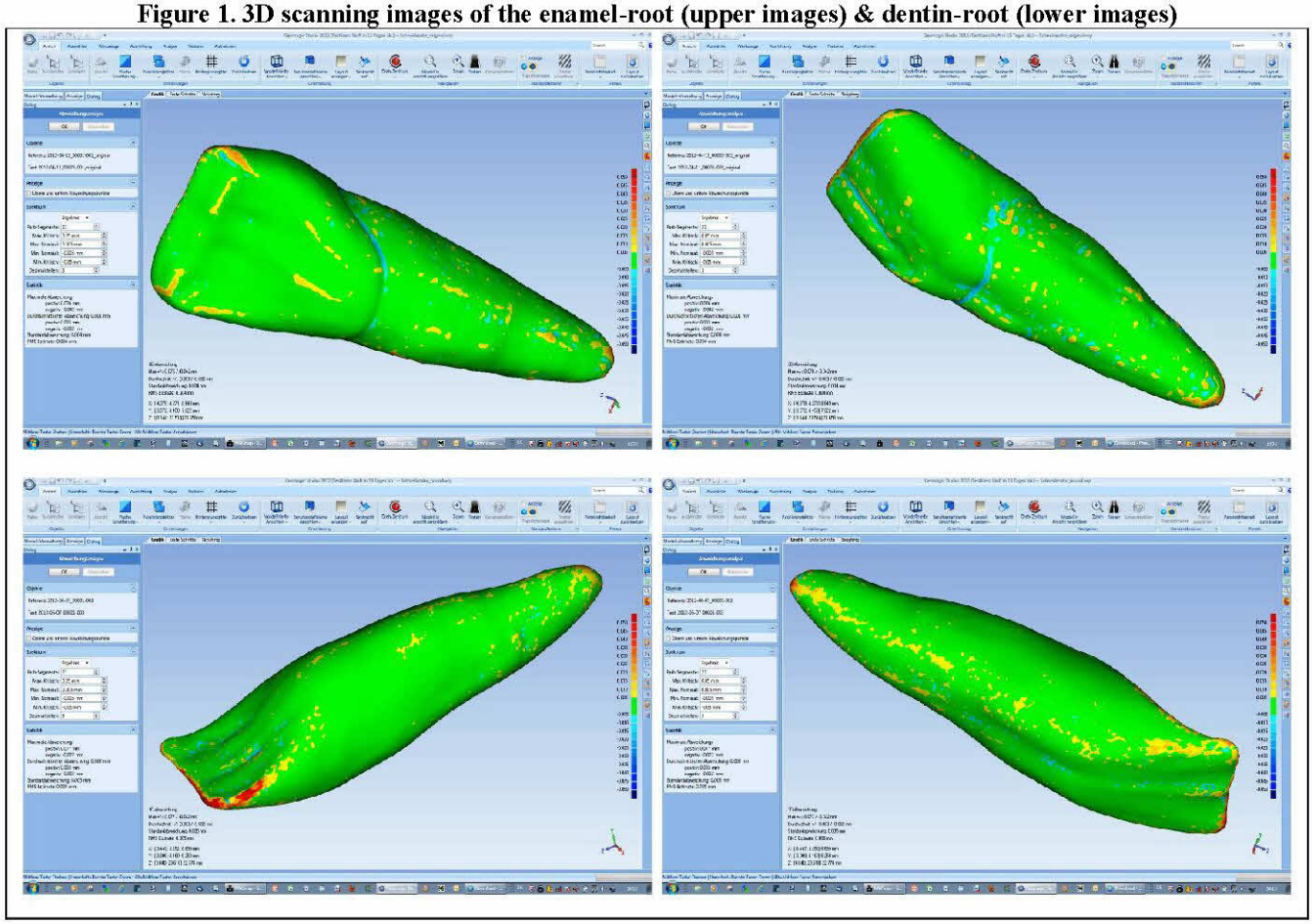


Figure 2. Measurements after reconstruction of the extracted tooth and calculating the volume of the scanned tissues (upper images). Measurements after optimizing filters and parameters of the CBCT images (lower images).

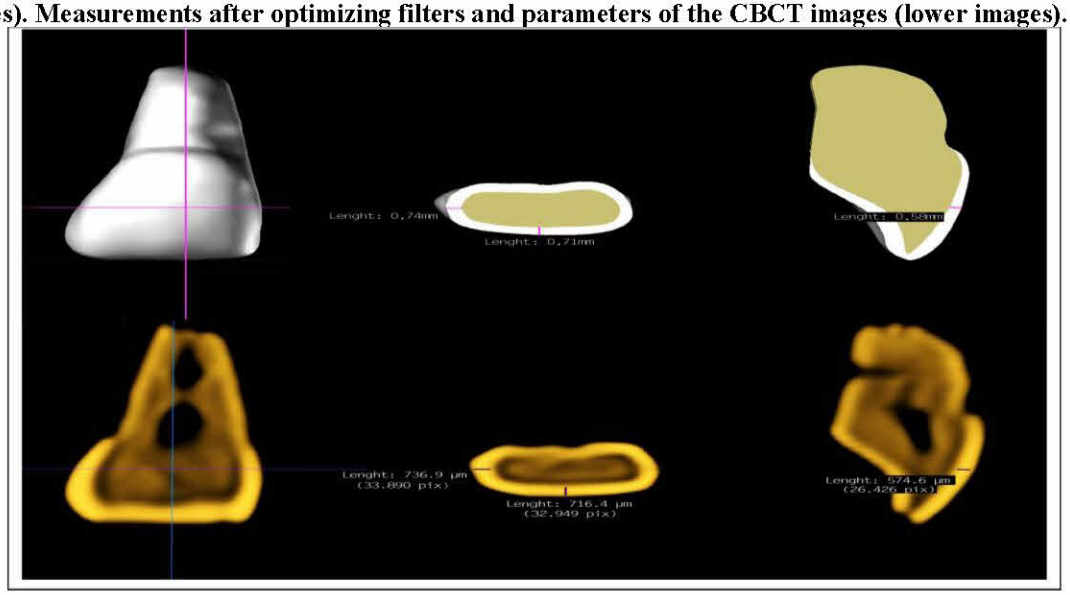


Figure 3. The selected fourteen spots of clinical interest of the enamel linear measurements

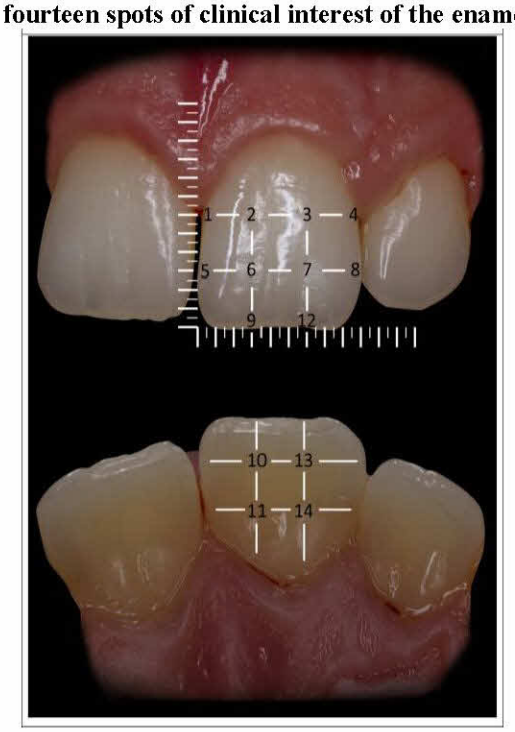


Figure 4a. A:CBCT transverse crosscut, B: CBCT sagittal crosscut, C: CBCT coronal crosscut

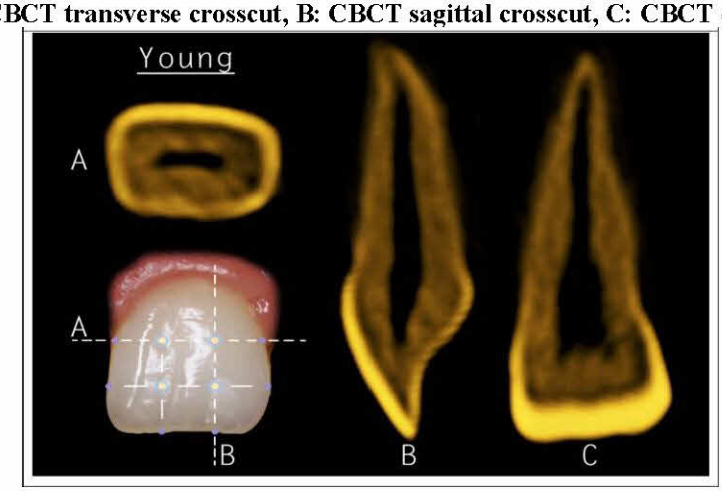


Figure 4b. A: CBCT transverse crosscut, B: CBCT sagittal crosscut, C: CBCT coronal crosscut

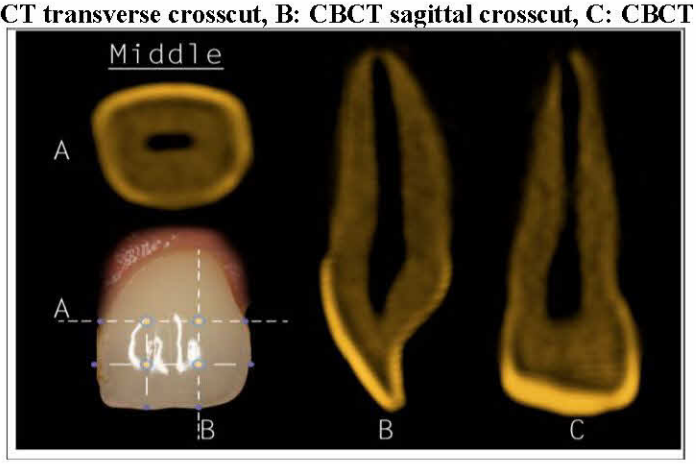
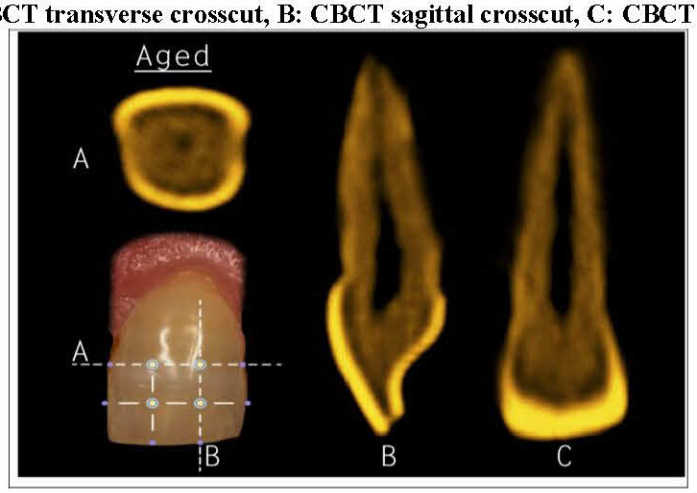


Figure 4c. A: CBCT transverse crosscut, B: CBCT sagittal crosscut, C: CBCT coronal crosscut



CONCLUSION

Within the limitations of the present study the following conclusions may be drawn:

1. Enamel thickness depends on both age and gender.
2. Enamel thickness of upper anterior teeth decreases over time.
3. No differences were found between central and lateral incisors, but both were statistically significant different from canines.
4. No difference exists between right and left side.
5. Enamel is thinnest in the palatal areas and thickest on the incisal edge
6. Great care is needed, when enamel reduction is performed, in order to avoid dentin exposure. 0.5mm enamel reduction in proximal and buccal areas should not expose dentin.

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CONFLICT OF INTEREST:

The authors declare that they have no conflict of interest.

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