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CONTENTS

MICROMETRIC MEASUREMENTS BY SCANNING ELECTRON MICROSCOPE (SEM) FOR DENTAL AGE ESTIMATION IN ADULTS

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ABSTRACT

Determining age from dental structure is a well documented forensic procedure and the data may be used as auxiliary information in identification. In this study 20 measurements of different variables in incisor teeth were obtained using a SEM micrometric scaler and the results were stastically correlated with age by the multiple regression method. A formula was then derived from the calculations for age estimation which gave statistically acceptable results. Gender differences were also investigated and when separated delivered even stronger correlation. **(J Forensic Odontostomatol 2000; 18:22-6)**

Key words: Teeth, SEM, micrometric measurements, age determination, forensic odontology

INTRODUCTION

Age estimation in forensic identification cases following crime and accidents may be accomplished by several methods where teeth are the only remaining evidence.[1] In cases of juvenile teeth age estimation can be carried out by direct dental examination,[2] examination of dental casts[3] and radiographic evidence of various developmental stages of teeth.[2] These do not of course apply to the adult dentition because of the completion of dental maturation[2] but other clues to age can be obtained from macroscopic and microscopic examination of the changes that occur with ageing.

Existing macroscopic procedures include direct oral and dental cast examination and correlating the data with dental records, which has proved to be reliable.[4,5] Existing microscopic and molecular studies include scanning electron microscopy with energy disperse x-ray,[6,7] periapical radiographic parameters,[8] chromatographic analysis[9] and dental pulp DNA testing,[10] all useful in identification.

The aim of the present study was to measure 20 aspects of longitudinal cross-sections of incisor teeth and to develop a formula for calculating age.

MATERIALS AND METHODS

Twenty-four newly extracted human incisor teeth without caries from donors of known age and gender, made up from six from each of four groups in the age range 35 to 70 years were studied (Table 1).

All teeth were embedded in elastometric impression materials* in a steel mould and sectioned labio-lingually in the centre from crown to root by means of a diamond separating disc,** guided by a longitudinal groove in the mould. The sections were coated with gold-palladium alloy and observed by SEM† at 100x magnification11 with micrometric scaler (Fig.1).

The tooth landmarks used for measurement are shown in Fig.2.

Least squares multiple regression modelling was used to estimate age using the dental parameters described in Fig.2. Statistical assumptions of constant variance and the predictors were checked for colinearity by VIF (Variance Influence Factor).

* Optosil P and Xantropen VL, Bayer Dental Levunkesen, Germany
** Comet Dental Gbr. Brasseler Gmbh & Co. KG Lemgo, Germany
† Joel, JSM-6400 Scanning Microscope, Tokyo, Japan

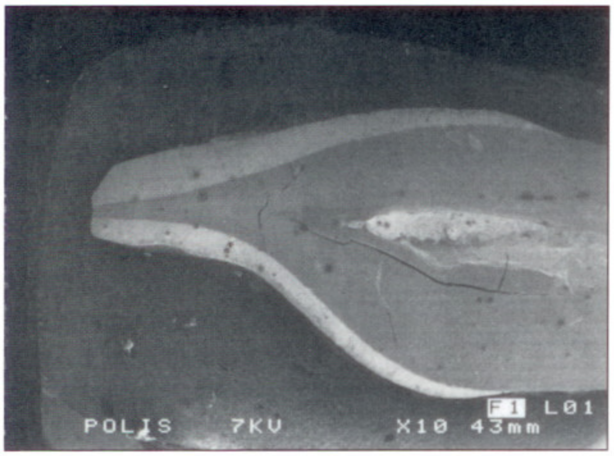

Fig.1: A sagittal section of a tooth from a male subject aged 25 years

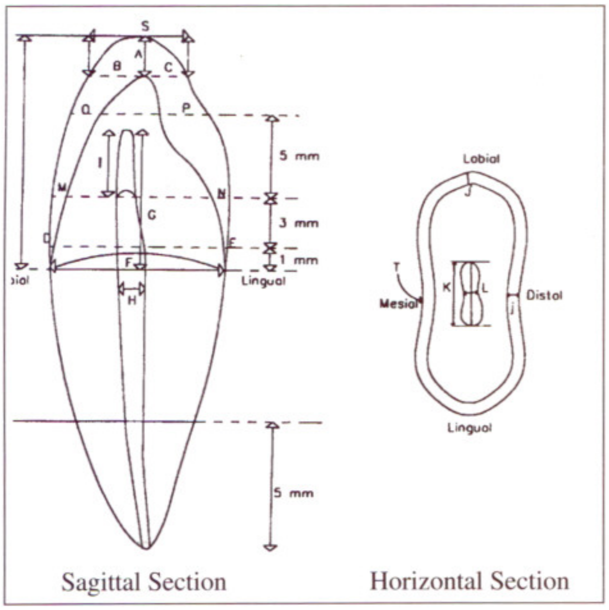

Fig.2:

A: Incisal enamel thickness
B: Incisal, labial enamel thickness
C: Incisal, lingual enamel thickness
D: Labial enamel thickness, 1mm above cervical line
E: Lingual enamel thickness, 1mm above cervical line
F: Cervical total thickness
G: Pulp height from cervical line
H: Pulp width at cervical line
I: Height of predentine over pulp tissue
J: Labial cementum thickness at 5mm over root apex
K: Labio-lingual pulp width at 5mm over root apex
L: Mesiio-distal pulp width at 5mm over root apex
M: Labial enamel thickness, 3mm above cervical line
N: Lingual enamel thickness, 3mm above cerrvical line
O: Labial enamel thickness, 5mm above cervical line
P: Lingual enamel thickness, 5mm above cervical line
R: Maximum crown length
S: Maximum thickness of incisal edge (labio-lingual)
T: Countable cementum layers at 5mm over root apex

RESULTS

The micrometric measurement results for specific points on the teeth were statistically analysed and the descriptive statistics given in Table 2.

The measurements were analysed by the multiple regression method for age estimation and the results for variables T,G,H,F,R,M,O and S showed R^2=0.90 accuracy with the following formula being derived: Age=21.5+2.82T+3.51 G-0.00292 H+3.91 F-2.75 R+0.0278 M-0.00919 O-2.96 S. Standard error prediction of the reported equation is S=4.147. Standard errors and significance of these coefficients are given in Table 3.

When some variables are omitted it is still possible to obtain statistically significant results for age estimation. Thus, when variables T,G,H,F,R and S (R^2=0.882) T,G,H,F and S (R^2=0.857) were tested T,G,H and S (R^2=0.848) and T and S (R^2=0.798) were tested a statistically significant level of $p<0.01$ was obtained.

The reliability of the statistical results may be strengthened by omitting the non-measurable variables and zero measurements i.e. the age dependent factors such as the worn incisal edges or receded pulp. Four cases were then discarded and statistical analyses were repeated for the rest. This time the multiple regression analyses gave R^2=0.95 accuracy for variables T,G,H,F,K,M,D and S ($p<0.01$), R^2=0.84 with variables T,G,H,F and S ($p<0.01$), R^2=0.83 for variables T,G,H and S ($p<0.01$), R^2=0.81 for variables T,G and S ($p<0.01$) and R^2=0.77 for variables T and S ($p<0.01$).

The age estimation calculations were also done for female and male groups separately. The multiple regression analyses for 12 females with the points T,D,S,F,E,O and C showed R^2=0.99 accuracy ($p<0.01$) with the points T,D,S,F,E and D it was R^2=0.99 ($p<0.01$) and with T and D it was R^2=0.85 ($p<0.01$).

The multiple regression analyses for 12 males with the points T,G,H,R,N,M and O was R^2=0.99 ($p<0.01$), with the points T,G,H,R and N was R^2=0.99 ($p<0.01$), and then with the points T and G it was R^2=0.89 ($p<0.01$).

Table 1: *Ages and genders of 24 donors*

Age Groups (years)	35-44						45-54					
Ages	35	36	37	42	42	43	47	54	51	54	54	54
Genders	M	F	M	M	F	M	F	F	F	F	F	M
Age Groups (years)	**57-64**						**65-74**					
Ages	57	57	57	59	60	63	65	65	67	68	70	70
Genders	F	M	M	F	F	M	M	M	M	F	F	M

Utilising the above data the following formula were derived and the age estimation female regression equation is:

Age: 13.6+1.41 T-0.117 D+12.3 S+7.17 F-0.0402 E+0.00749 O+0.00636 C. Standard error prediction of the reported equation is S=1.297. Standard errors and significance of these coefficients are given in Table 4.

Age estimation for male regression equation is:

Age: 51.6+2.16 T+3.24 G-0.0177 H-3.70 R+0.0405 N-0.0169 M+0.0159 D. Standard error prediction of the reported equation is S=1.377. Standard errors and significance of these coefficients are given in Table 5.

Table 2: *The descriptive statistics of specific tooth points within the age range of 35-70 yrs*

VARIABLES RECORDED**	N	MEAN	MEDIAN	TRMEAN	STDEV	SEMEAN	MIN	MAX
B	24	787.6	734.2	786.3	225.5	46	295.3	1310
C	24	504.6	503,4	502.4	110.4	22.5	323.4	784.4
D	24	280.3	273.7	279.7	61.2	12.5	168.6	405.9
E	24	238.5	245.8	239.5	71.2	14.5	84.3	370.8
F	24	5.3796	5.445	5.3859	0.4527	0.0924	4.47	6.15
G	24	2.976	2.805	3.002	1.342	0.274	0	5.37
H	24	785.4	734.1	770.9	450.3	91.9	0	1890
I	23*	800	590	747	724	151	0	2710
J	24	108.77	112.21	107.69	46.31	9.45	49.18	192.15
J'	24	77.89	78.32	72.22	47.59	9.71	27.2	253.39
K	24	914.8	837.2	915.7	353.9	72.2	169.1	1640
L	24	184.8	158.5	179.9	79.5	16.2	68.9	407.6
M	24	501.7	522.5	509	107.8	22	169.1	674.6
N	24	420	417.5	419.6	91.8	18.7	224.7	623.9
O	24	726.3	793.9	746.4	254.5	52	0	1010
P	24	442.6	483.9	455.2	156	31.8	0	609.1
R	24	6.659	6.705	6.651	1.386	0.283	3.48	10.01
S	23*	2.14	1.92	2.12	0.652	0.136	1.15	3.55
T	24	7.667	7	7.591	2.988	0.61	4	13
AGE	24	54.58	55.5	54.77	10.86	2.22	35	70

* (I) and (S) could not be measured in 2 subjects

** For key, see Fig.1

DISCUSSION

In this project the differentiating factors required for age estimation were identified and a formula proposed based on human incisor teeth extracted for periodontal reasons. It is however suggested that teeth extracted for other reasons such as from cadavers and forensic cases might reflect biological variations better than homogeneous sources.[12] In this study, in order to establish an age calculation formula and to ensure standardisation, it was preferred to measure tooth layers from the same teeth which were extracted for the same reasons.[13]

Measurements of enamel, dentine, predentine, pulp layers of the crown portion and dimensions of root pulps and cementum layers and in addition the number of cementum layers were recorded. The age correlation formula derived from the statistical analysis of these results varied from R2=0.77-0.94 when gender variables were omitted ($p<0.01$), R^2=0.90 when B,C,D,E,I,K,L,N and P variables were also

Table 3: *Standard errors and significance of coefficients*

Predictor	Coefficient	Standard Error	Significance
Constant	21, 46	14, 28	0, 155
T	2, 8226	0, 4889	0, 001**
G	3, 5137	0, 9958	0, 003**
H	-0.002916	0, 002924	0, 335
F	3, 906	2, 250	0, 105
R	-2.745	1, 405	0, 071
M	0, 02780	0, 01631	0, 110
O	-0.009193	0.006854	0.201
S	-2.957	3, 140	0, 362

** p<0.01

Table 4: *Standard errors and significance of coefficients*

Predictor	Coefficient	Standard error	Significance
Constant	13,619	8.762	0.195
T	1,4142	0,2374	0,004**
D	-0.1171	0,01067	0,001**
S	12,282	1,944	0,003**
F	7,173	1,046	0,002**
E	-0.040184	0.007352	0,005**
O	0,007492	0,003385	0,091
C	0,006361	0,005147	0.284

**p<0.01

Table 5: *Standard errors and significance of coefficients*

Predictor	Coefficient	Standard error	Significance
Constant	51,626	4,875	0,001**
T	2,1573	0,1903	0,001**
G	3,2418	0,4242	0,002**
H	-0.017748	0.00255	0,002**
R	-3.6996	0,6406	0,004**
N	0.040468	0,009251	0.012
M	-0.016859	0.008257	0,111
D	0,015949	0,009976	0.185

**p<0.01

omitted and R^2=0.94 when additionally J,O and R variables were omitted.

The determination coefficient 'R^2' showed that this method was close to the accepted age estimation results obtained by Gustafson[14] where R^2=0.83 (r=0.91) or others at R^2=0.76 (r=0.87),[15] R^2=0.72 (r=0.85),[16] and R^2=0.64 (r=0.80)[17] and R^2=0.83 (r=0.91).18 Bang and Ramm[18] further found R2=0.37-0.69 (r=0.61-0.83) for intact teeth and R^2=0.25-0.86 (r=0.50-0.93) for sectioned teeth (r is given as a correlation factor).

The age estimation calculations in this research were also applied to the male and female groups separately where the multiple regression analyses for 12 females were applied using the measurement point T,D,S,F,E,O and C showed R^2=0.99 accuracy ($p<0.01$). The gender variables were chosen, depending on the statistical determination coefficient, derived from different gender sample measurements. Where the gender is known, statistical analysis makes the age estimation becomes more accurate and in female tooth samples the following measurements were important: T (countable cementum layers), D (labial enamel thickness 1mm above the cervical line), S (maximum thickness of incisal edge), F (cervical total thickness), E (lingual enamel thickness 1mm above the cervical line), O (labial enamel thickness 5mm above the cervical line), C (incisal, lingual enamel thickness). In male tooth samples: T,G,H,R,N,M and D were specific measurements, where T (countable cementum layers), G (pulp height from cervical line), H (pulp width at cervical line), R (maximum crown length), N (lingual enamel thickness 3mm above the cervical line), M (labial enamel thickness 3mm above the cervical line), and D (labial enamel thickness 1mm above the cervical line).

These results showed that a minimum of 8 variables were essential for an accurate age estimation calculation. However, some of the current age correlation formulae use only 3-6 variables[14,17] which probably means that some important information is overlooked. This survey established that labial and incisal enamel thickness at different levels, width and height of pulp tissue amount, and the number of cementum layers must be taken into account when age is to be determined. However this research is performed on 24 samples, and should be regarded as a pilot study.

CONCLUSIONS

1. Human age can be determined from SEM measurements of sections of incisor teeth, by using a formula proposed in this study.
2. When gender is known two different age estimation formulae can be used, which will deliver more reliable statistical results.
3. The ageing factors of teeth i.e. changes of enamel layer thickness, amount of pulp tissue recession, and apposition of cementum layers, may be reliable age estimation factors.

The variables directly related to the ageing of teeth, i.e. changes in enamel layer thickness, amount of pulpal recession, and deposition of cementum may be reliable indicators in age estimation.

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COMPARISON OF BITEMARKS LEFT IN FOODSTUFFS WITH MODELS OF THE SUSPECTS' DENTITIONS AS A MEANS OF IDENTIFYING A PERPETRATOR

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ABSTRACT

In a recent court case, a comparison was made between an impression of marks left in cheese at a murder scene and a set of study models of one of the suspects. The court was reluctant to accept the validity of the pattern-associated comparison that was used in the identification.

This study compared marks made in cheese, butter and cooked potato with study models taken from volunteers. Pattern-associated comparison was the method used. Eighty pair-wise comparisons were made by two odontologists. The examiners correctly identified all the true matches from among the eighty comparisons as well as selecting the dental models for which there were no corresponding silicone impressions. In the absence of identifiable fingerprints or DNA samples, the method can be employed for matching left in foodstuffs to the dentitions of suspects. **(J Forensic Odontostomatol 2000;18:27-31)**

Keywords: Identification, bitemarks, foodstuffs

INTRODUCTION

In a recent court case, the State verses Shabangu, the prosecution attempted to prove the identity of the murderer, based on an impression made from a piece of cheese with bitemarks found at the scene of the crime. A pattern-associated comparison between the cheese impression and a study model of the suspect was performed. The court was only prepared to accept the match between the cheese bitemarks and the suspect as substantive evidence, whereas fingerprints found at the scene of the crime were used as the definitive evidence in the final judgement.

This study was carried out to test the method used in identifying the perpetrator in the above court case. The science of fingerprint analysis was officially adopted in 1901 when Sir Edward Henry successfully matched a suspect to fingerprints found at a crime scene.[1] Today, twelve concordant fingerprint features are universally accepted as a minimum to establish identity.[2, 3] DNA technology has a discrimination potential of 1 in 303 billion and eight-locus Tandem Repeat (STR) multiplexing DNA analysis has become an important method of accurately matching an individual to any DNA contaminated object.[4] No DNA evidence was found at the crime scene, and therefore was not considered in the above court case.

Simply stated, a bitemark is a patterned injury produced by teeth on animate or inanimate objects[5] and is caused by small enamel defects on the incisal edge of incisor teeth creating individual characteristics during the biting process.[6] The concept of comparing the "mark" made by the dentition of an individual in skin with the dentition of the suspected perpetrator has been well accepted by forensic odontologists, law enforcement officers, and trial and appellate level courts.[7] Bitemarks found in certain foodstuffs, such as cheese, chocolate and potato have also contributed in implicating perpetrators.[8] Bitemark identification relies on the individual characteristics of each bite pattern, the quality of bite registration and the stability of the object bitten. It is easier to demonstrate that a person's dentition could not have caused a particular bite mark than to conclude the presence of an exact match.

The individuality of a bitemark stems from the uniqueness of the human dentition.[9,10] Regressive changes, namely attrition, abrasion and erosion will cause changes to the dentition with time[11] but as these changes occur relatively slowly, their influence will only be a serious complicating factor when the match takes place several months or years after the bite took place. Dental procedures, for example extractions, restorations and occlusal adjustments will also cause changes to the dentition.

The aim of this study was to match bitemarks made in foodstuffs with the models taken from a selection of individuals and to test the reliability of the method

MATERIALS AND METHODS

A random sample of 16 persons (8 oral hygiene students from the Dental Faculty, University of Pretoria, and 8 patients presenting for treatment at a private dental practice in Middelburg, Mpumalanga, South Africa) were chosen as volunteers for this study. Their ages ranged between 7 and 47 years of age and no person who had a history of orthodontic treatment was included. The oral hygiene students were volunteers who had to take impressions of each other and cast models as part of their practical studies. Eight impressions and models were selected by the first author from the class of 24 oral hygiene students. The patients were randomly selected by the first author, and after prior consent, an alginate impression was taken of each person's upper and lower dentitions by the practitioner's oral hygienist. A set of plaster study models for each impression was cast by the laboratory assistant. All the plaster study models were laid out in random order and sequentially numbered 1 to 16. The laboratory assistant kept a record of the relationship between plaster study model number and individual identity so that the study would be blind.

Blocks of cheese, butter, and cooked potato of size 40 x 20 x 20mm were prepared and each one of the 16 participants was asked to bite into one or more of the different foodstuffs with his or her front teeth. A total of 20 bite samples were made. The foodstuffs were left for 1 day before impressions were taken of the bite patterns using hydrophilic vinyl polysiloxane, type 1, low viscosity Reprosil* impression material.[12] The time delay was introduced to simulate the usual sequence of crime investigation.

The 16 pairs of models and 20 impressions were randomly divided into four groups of four and five respectively. Both odontologists (HB & TS) had previous experience in evaluating bitemarks. Each received two groups of models/impressions. Each group was subjected to comparison, i.e. in each of the four cases twenty pair-wise comparisons were made. A total of 80 pair-wise comparisons were made between sixteen study models and twenty silicone impressions.

The protocol tested is based on the principle of the double blind experiment. To prevent any subjective bias the forensic odontologists were presented with numbered models and silicone impressions, i.e. they did not know the names of the persons producing them. There were no identifying marks, colours or features on either the impressions or models that could give any clue as to their identity. To prevent matching by a process of elimination they were not told whether every study model had at least one corresponding silicone impression. To ensure that they did not stop after finding a positive match they were told that there was at least one instance where two impressions corresponded to one study model.

In order to perform a pattern-associated comparison, the examiners (HB & TS) were instructed to take a set of study models and a silicone impression, juxtaposing the two, and to compare them feature by feature. They were requested to hold the study model and silicone impression in close proximity and to work interactively back and forth between the two. If at any stage of the comparison an obvious discrepancy was found, a mismatch was declared. If a positive match was suspected, every possible point of concordance was documented.

To ensure that each examination was comprehensive, nine features (Table 1) were used as a guide during

*Caulk Dentsply. Dentsply International Inc, Milford, USA.

the comparison. The features chosen were those seen in previously examined bite mark cases. Any other distinctive features found were also documented under "Other", e.g. rotations.

Table 1: *The ten classes of features used to examine the study models and impressions*

A	Significant difference in incisal height between 11/21 and 31/41
B	Anterior teeth placed out of arch (state tooth number)
C	Anterior open bite (state position and expanse)
D	Chip on anterior teeth (state tooth number and position)
E	Midline status (use mandible and state L,R or no deviation)
F	Abnormal incisal slope(state tooth number)
G	Anterior teeth missing (state tooth number)
H	Diastema present (state position and whether wide or narrow)
I	Crossbite present (state position)
J	Other (e.g. rotation)

The data from this comparison (i.e. comparative double blind) was tabulated (Table 2) documenting the most obvious discordant feature in each of the 80 pair-wise comparisons. In those cases where no obvious discrepancy could be seen a star was placed and a further detailed analysis was carried out. Each of these candidate pairs was carefully examined using the nine categories of features (Table 1) and the results recorded separately. Table 3 provides an example on one such result.

Table 2: *Obvious discordant features (asterisk indicates no discordant features and therefore possible match*

MODELS	MODEL 11	MODEL 2	MODEL 7	MODEL 3
Impression no				
2_(butter)	Even lower teeth	Even lower teeth	Diastema 11/21	Diastema 11/21
3 (cheese)	23 present	No diastema 11/21	Diastema 32/31	*
9 (cheese)	Position 41/42	No diastema 11/21	Position of 11/21	No diastema 42/42, 31/32
11 (cheese)	Even lower teeth	No diastema 11/21	Even front teeth	Even 11/21
12 (butter)	Position 31/41	No diastema 11/21	*	Prominent d. edge 11
19 (potato)	Position 12 and 33	No diastema 11/21	Position 41/31	No diastema 43/42, 31/32
20 (cheese)	Diastema 11/21	*	Diastema 11/21	Diastema
22 (butter)	*	No diastema 11/21	Position 11 and 31	No diastema lower teeth
41 (potato)	Position 41/41	No diastema 11/21	Position 21	*
200 (potato)	Position 41/42	No diastema 11/21	Angle of 42	Rotated 42

RESULTS

Odontologists HB and TS correctly identified the fifteen true matches from among the 80 pair-wise comparisons as well as selecting the dental models for which there were no corresponding silicone impressions. Neither odontologist found any false positives (i.e. said that there was a match when in fact there was none).

In addition, they did not find any false negatives (i.e. missed a match). Both HB & TS had similar numbers of features for categories A to D and H but had very different frequencies for categories E to G and I and J (Table 1). The major differences were in categories E (midline status) and J (rotated teeth). HB made positive matches using more features than TS. It should be remembered that the two operators did not examine the same sets of models and impressions.

Table 3: *Example of comparison of one set of models with an impression*

Identification: Model 3 with bite No.41
(bold print indicates features present,
normal print indicates features not present)

Difference in height between 11/12 and 31/41
Teeth placed out of arch 11
Chip on anterior teeth
Midline status Lower jaw deviates left
Abnormal incisal slopes
Anterior missing teeth
Diastemas present 43/42, 31/32 and 32/33
Abnormal incisal slopes
Crossbite present
Other - Rotated teeth 11 and 32
- 4 + features

DISCUSSION

Each examiner positively matched all models with their respective silicone impressions. This method was based on the three dimensional structure of each tooth as well as its relationship to the surrounding teeth. The relationship of the maxilla to the mandible could also be compared. Both examiners used the features in Table 1 as a guide in the identification process and as the individual features could not be weighted in any way, their individual occurrence (common, uncommon or very uncommon)[13] did not contribute substantially in the final identification. More research is needed to establish the probability of occurrence of selected dental features in the population in which the case occurred. Without this data a full statistical analysis of this method cannot be made.

This method can be compared to a puzzle piece that has to be placed in an exact position to complete a picture. Even though there are several pieces that could fit into a specific space in the puzzle, only one piece will have the right shape and picture. Another example of three-dimensional recognition is that of one's own child among hundreds or thousands of other children. All the children have common features i.e. one nose, two eyes, two ears, one mouth etc, but it is the relationship of each of the features to one another that gives us the ability to recognise our specific child.

Pattern-associated comparison was used in preference to metrical comparison. Unreliable dynamic changes which occur during the shrinkage period[10] in most foodstuffs, would create problems for the expert witness during cross-examination if metrical analysis were used.

The quality of the bite marks left in different foodstuffs will vary according to the physical characteristics of the material. The more pronounced the individual traits of a dentition are, the less important the quality of the bite marks becomes and vice versa. It was found that the movement of the mandible during the biting process produced longer bite marks with the frequent presence of scratch marks, than that of the maxilla. The firmer the foodstuff, the more reliable the midline status seemed to be when examining the silicone impressions taken from the different foods. Inanimate objects such as cheese, butter and potatoes have a limited shelf-life and the impressions should be made as soon as possible. The bitten objects will change shape in time, depending on humidity, temperature and storage quality[14] and such changes make metric analysis unreliable. Only pattern association may be demonstrated between an impression and the suspect's dentition.

Notwithstanding the imperfections of the proposed technique, it is possible to match bitemarks left in foodstuff to the dentition of the person who produced the bite marks. However, it should be remembered that bitemarks must be clearly visible. The nearer the suspect's dentition is to "perfect", the more difficult it will be to match.

Information on the probability of occurrence of dental characteristics specific to particular populations is not available for the population in which this study took place. The frequency of occurrence of dental features would allow the presence/absence of that feature in a model to be weighted.

CONCLUSION

In the absence of identifiable fingerprints or DNA samples, the method can be employed for matching bitemarks left in foodstuffs to the dentitions of suspects. Until such time as statistics exist on the probability of occurrence of dental features in a specific population, multiple concordant features in the absence of any unexplainable discrepancies should be present to express the likelihood that a given suspect has made a sledge mark. If such features are especially characteristic, less features would need to be present. However, the highly reliable identification methods of finger printing and DNA analysis remain the procedures of choice and bite mark recognition cannot equal them at present[15].

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POSTMORTEM TOOTH LOSS IN HUMAN IDENTIFICATION PROCESSES

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ABSTRACT

Teeth provide essential data for human identification. However, they are frequently lost during the process of skeletonization or under manipulation during exhumation. Because of the high frequency of postmortem tooth loss, this phenomenon was examined in three different samples, in an attempt to simulate the actual circumstances experienced in the forensic processes of identification of human remains. The method employed aimed to describe and quantify dental spaces, distinguishing between loss after death or before by extraction. The results showed a high prevalence of postmortem tooth loss: 41.37% in sample A, 56.83% in sample B and 3.96% in sample C. These losses were most frequent in the maxillary incisor group in all three samples analysed. These data underline the need for redoubled care in recovery, transportation and storage of crania, so that teeth are not lost, which could seriously compromise the identification process. **(J Forensic Odontostomatol 2000;18:32-6)**

Keywords: Forensic Dentistry, postmortem tooth loss, human identification

INTRODUCTION

The process of human identification can be undertaken in two ways. In the reconstructive approach there are no antemortem data available and the aim is to establish generic elements for a general identification. In the second or comparative approach, records from before the individual's death, including dental charts, are used as the basis for individual identification.

Teeth can provide decisive information for human identification[3]. Although they may be missing, it is still possible to examine the remaining alveolar bone to establish whether loss occurred before or after death. If loss was by extraction shortly before death, the period can be estimated by using alveolar remodelling chronology.[2]

Once these observations have been made, comparative analysis of the remaining anatomical and restorative aspects of the teeth can begin. This is made possible by obtaining satisfactory ante-mortem dental records including radiographs, the importance of which cannot be overestimated because they are often the most reliable way to identify an individual.

The use of anatomical structures and dental restorations is so important that the material must be preserved at all costs even though identification by DNA is becoming more prevalent and reliable. It cannot however outweigh or rule out the use of the former.[5]

Teeth may be missing at the time of identification and this will hamper the process. Tooth loss occurring after death may be due to factors relating to the natural process of skeletonization that destroys the attaching periodontal tissues and the conical nature of the roots of single rooted teeth makes them particularly susceptible. Careless handling in the collection, transportation, preparation, packing and dispatch for examination of human remains from crime scenes or in exhumations further contribute to tooth loss.

Another risk factor for postmortem tooth loss is the age of the individuals at death. Whittaker and Molleson[7] examined 92 crania of known age and found a greater prevalence of postmortem tooth loss in older individuals, suggesting that greater care be taken during exhumation if age is known.

The environment where the body was found can also contribute to this process. Places which are highly exposed to the sun, places in shade provided by trees, remains which have been embalmed, or kept in plastic bags for long periods, or submerged in water, as well as changes in the weather (in countries with marked differences between the seasons), all influence postmortem tooth loss. For this reason the study of tooth loss in conjunction with the environmental conditions can provide supplementary clues for an estimation of time of death.[4]

Given the importance of tooth loss in hampering human identification, the present study aimed to analyse the incidence of postmortem tooth loss in three samples and draw attention to the resulting problems.

MATERIALS AND METHODS

Three samples were studied in order to explore different circumstances encountered during the identification process.

The first sample (Sample A) comprised 151 crania from the Museum of Human Anatomy of the University of São Paulo's Institute of Biomedical Sciences. The collection was compiled between 1920 and 1960 and has been used exclusively for research purposes in recent times. The material was considered suitable because it is curated by the University and is handled and conserved appropriately. Secondly, owing to its age, it would have undergone thoroughly the process of skeletonization and therefore, in their current state, the crania studied would provide a fair approximation of the various conditions encountered in real forensic circumstances.

Sample B comprising 100 crania, was obtained from a cemetery in the city of Salvador, state of Bahia, which routinely stores material from exhumations in tagged sealed plastic bags until the legal and administrative terms have expired, at which point they are handed over to the family to be suitably disposed of, or if unclaimed, reburied in a common grave. By its nature this material also replicates the typical features of a forensic examination.

Sample C was obtained from the University of São Paulo's Museum of Archaeology and Ethnology. Five crania, three complete, two without mandibles and five isolated mandibles were examined. This material was collected from archaeological sites on the littoral of the State of São Paulo, occupied between approximately 2980 BC and 75 AD. These sites were excavated in the 1970s and the material has been in the custody of the Museum since then.

All examinations were carried out to observe and record the presence or absence of teeth. Missing teeth were classified as extracted during life or lost postmortem depending on whether the alveolar sockets showed signs of tissue healing, in which case they were considered lost postmortem.

The classification of Whittaker and MacDonald[6] was used to identify postmortem tooth loss, differentiating it from antemortem dental extraction. According to these authors, when the tooth is lost after death the bone surrounding the socket will not have reacted to the loss of the tooth and the margins of the socket would have retained a sharp profile. On the other hand antemortem tooth loss includes blood clot formation in the socket which rapidly organizes into granulation tissue, new bone is progressively laid down on the walls of the socket and within the granulation tissue and as the new bone mineralises the socket will have developed a granular appearance.

In order to estimate the reproducibility of the observation, we calibrated the dentists that performed the examinations, and we measured inter- and intra-examiner concordance by re-examining 9.9% and 6.6% respectively of Sample A. The set of observations was subjected to statistical analysis using SPSS 8.0, 1997.

RESULTS

Figs. 1 and 2 show alveolar sockets not occupied by teeth, with and without signs of alveolar healing, corresponding respectively to classifications of teeth extracted during life and postmortem tooth loss.

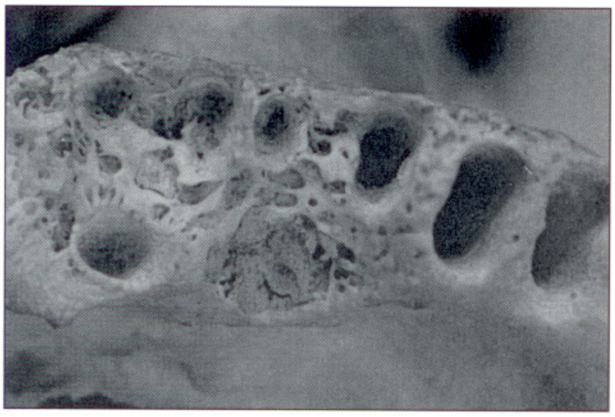

Fig.1: *Molar region with evidence of alveolar healing, characteristic of antemortem tooth loss*

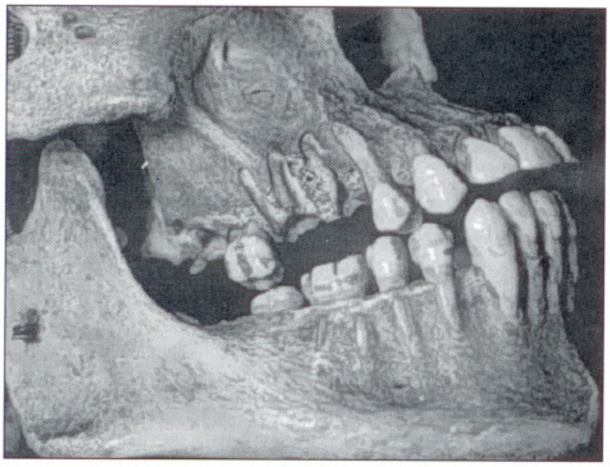

Fig.2: *Molar and pre-molar region without alveolar healing, characteristic of postmortem tooth loss*

Table 1 presents the set of analysed teeth and dental spaces for each sample. "Dental spaces" were subsequently classified as "present tooth", "ante mortem extracted tooth" or "postmortem tooth loss". Tables 2, 3 and 4 show the total number of "present teeth" and dental spaces in samples A, B and C respectively that did not undergo extraction during life, as well as the percentage of dental spaces classified as postmortem tooth loss.

Comparative analysis of the data obtained for sample A showed that the group of incisors presented significantly higher prevalence of postmortem tooth loss than other types of teeth ($p<0.05$), while premolars came second, with significantly higher prevalence of postmortem tooth loss than molars and canines ($p<0.05$). With regard to the latter two tooth groups, no significant statistical difference was observed ($p=0.795$).

A significant difference was observed in comparing the upper arch with the lower arch ($p<0.05$), with a greater percentage of postmortem tooth loss for maxillary teeth.

A significantly higher rate of postmortem tooth loss was found in Sample B than in Sample A ($p<0.001$) but no significant differences were observed between maxillary and mandibular teeth ($p=0.840$). The only dental group recording a significantly higher frequency of postmortem tooth loss was the incisors ($p<0.05$), an observation which ties in with that in Sample A.

Complete absence of postmortem tooth loss in mandibular teeth was observed in Sample C, as well as a low frequency of loss in maxillary teeth, virtually restricted to incisors. This observation may be due to the greater care with which archaeological research is carried out, including delimitation of the surrounding area and the sieving of the earth in places where remains are found, factors that enable the recovery of dental elements which could otherwise have been lost.

Re-examination of 6.6% of Sample A by the same examiner and 9.9% of the same sample by another examiner in the team was carried out to assess the precision of the method employed and to check intra- and inter-examiners concordance. Re-examination by the same examiner scored 98.07% observed agreement (Kappa coefficient = 0.978), while re-examination by a different examiner scored 99.11% observed agreement (Kappa coefficient = 0.959). These figures prove that the method is reproducible and easily interpreted.

DISCUSSION

There was a higher prevalence of postmortem tooth loss in the upper arches in Sample A. This observation can partly be ascribed to the positioning and the exposure of the crania during storage over several years. To achieve greater stability the crania were rested on their mandibles, on the masseteric processes and the occipital bone, which subjected the maxillary teeth to forces of gravity and consequent dislodgement. Another influence may have been that the bone density of the maxilla generally is less than in the mandible.

Table 1: *Total of teeth and dental spaces ("present tooth", "ante mortem tooth extraction", "postmortem tooth loss") analysed in the three samples*

Dental type	Sample A	Sample B	Sample C	Total
Maxillary Molars	906	600	30	1,536
Maxillary Premolars	604	400	20	1,024
Maxillary canines	302	200	10	512
Maxillary incisors	604	400	20	1,024
Maxillary teeth	2,416	1,600	80	4,096
Mandibular Molars	906	600	48	1,554
Mandibular Premolars	604	400	32	1,036
Mandibular canines	302	200	16	518
Mandibular incisors	604	400	32	1,036
Mandibular teeth	2,416	1,600	128	4,144
Molars	1,812	1,200	78	3,090
Premolars	1,208	800	52	2,060
Canines	604	400	26	1,030
Incisors	1,208	800	52	2,060
Total teeth	4,832	3,200	208	8,240

Of all groups of teeth analysed, incisors presented the highest frequency of postmortem tooth loss, a fact which may be explained by their anatomical characteristics. The incisors can be considered generally to be single rooted teeth but canines, on the other hand, although they usually possess only one root, have the largest of the dental roots which are often slightly curved, and this tends to hook them in place. Premolars in general present a single twinned root, although bifurcation is common in the upper teeth. Molars in general possess three roots, except for the third molars which display varying morphology.

Our observation of greater prevalence of postmortem tooth loss in the incisors ties in with the findings of Borrman et al.[1] who analysed an approximately 9800 year-old human skeleton, finding postmortem loss of seven teeth, of which **five were incisors.**

In Sample B, many toothless dental arches were found, which may be due to the widespread use of total extractions as a form of dental treatment, and as the subjects were from an urban cemetery may also link them to an underprivileged socio-economic and cultural stratum. Another observation was the lack of pattern of dental loss and with no differences between the upper and lower arches, which may be ascribed to the method of storage of these crania in plastic bags along with the rest of the skeleton, and piled one on top of the other. A significantly higher prevalence of postmortem tooth loss was observed than in Sample A, which may be explained by the handling and storage conditions of this material in

Table 2: *Frequency of postmortem tooth loss in Sample A by dental type and potential number of teeth*

Dental types	% postmortem loss	Potential number of teeth
Maxillary Molars	29.528	352
Mandibular Molars	30.189	355
Molars	29.865	707
Maxillary Premolars	38.583	467
Mandibular Premolars	36.752	461
Premolars	37.705	928
Maxillary canines	28.829	254
Mandibular canines	32.394	247
Canines	30.575	501
Maxillary incisors	79.636	378
Mandibular incisors	48.813	447
Incisors	61.774	825
Maxillary teeth	44.611	1,451
Mandibular teeth	38.328	1,510
Total teeth	41.368	2,961

the cemetery, or may even be possible evidence of a deplorable practice which has not yet been eradicated in Brazil, that is the illegal trade in teeth for instructional purposes.

In Sample C a low incidence of antemortem dental extraction was observed – only six teeth out of a total 208 examined dental spaces – which seems to be characteristic of the specifically archaeological material usually targeted for research. The low percentage of postmortem tooth loss for this sample (3.96%) is indicative of the skilled and professional care normally taken in collecting and preserving this material during research. This degree of care should be adopted in the practice of forensic dentistry.

CONCLUSION

1. A high percentage of tooth loss was detected. It was possible to distinguish, with satisfactory accuracy, whether this occurred before or after the death of the individual by means of visual parameters (signs of alveolar healing).

2. In Sample A, postmortem tooth losses presented a higher pattern of prevalence in the upper arches and incisors.

3. In Sample B, there was no significant difference in postmortem tooth loss between the upper and

Table 3: *Frequency of postmortem tooth loss in Sample B, by dental type, and potential number of teeth*

Dental type	% postmortem loss	Potential number of teeth
Maxillary Molars	42.609	115
Mandibular Molars	35.000	100
Molars	39.070	215
Maxillary Premolars	44.737	76
Mandibular Premolars	53.846	156
Premolars	50.862	232
Maxillary canines	66.667	66
Mandibular canines	53.719	121
Canines	58.289	187
Maxillary incisors	71.795	117
Mandibular incisors	70.168	238
Incisors	70.704	355
Maxillary teeth	56.417	374
Mandibular teeth	57.073	615
Total teeth	56.825	989

Table 4: *Frequency of postmortem tooth loss in Sample C, by dental type, and potential number of teeth*

Dental type	% postmortem loss	Potential number of teeth
Maxillary Molars	3.57	28
Mandibular Molars	0.00	45
Molars	1.36	73
Maxillary Premolars	0.00	19
Mandibular Premolars	0.00	32
Premolars	0.00	51
Maxillary canines	0.00	10
Mandibular canines	0.00	16
Canines	0.00	26
Maxillary incisors	35.00	20
Mandibular incisors	0.00	32
Incisors	13.46	52
Maxillary teeth	10.38	77
Mandibular teeth	0.00	125
Total teeth	3.96	202

lower arches but a greater frequency was observed in incisors.

4. In Sample C, there was a very low prevalence of tooth loss, virtually restricted to the maxillary incisors.

5. Enhanced care should be taken in the handling of human remains. The standards of care normally employed in archaeological excavations should be extended to routine forensic dentistry in order to avoid postmortem tooth loss which can make identification more difficult.

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TOOTH ROOT COLOUR AS A MEASURE OF CHRONOLOGICAL AGE

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ABSTRACT

The purpose of this study was to assess a possible colour shift in the root surfaces of adult human teeth and if so, whether this colour change is related to chronological age.

Teeth extracted from persons of known age and gender were obtained from Ontario dental practitioners and grouped into five-year age ranges. Three experiments were undertaken: (1) to identify a possible difference in yellow colouration between the four surfaces of tooth roots (mesial, distal, lingual, and buccal), (2) to investigate the difference in yellow colouration of tooth roots between non-molar teeth and molar teeth and (3) to assess the correlation between the age of teeth and root colour saturation for yellow, magenta, cyan and black. The teeth in all investigations were scanned by a flat-bed digital colour scanner with a Kodak* colour scale control and viewed on a colour computer monitor. In the first two experiments the yellow colour saturation of the root surfaces was measured at six points on each root using Photoshop 5.0** software. A significant difference was observed in the percentage yellow colour saturation between the mesial and the other three anatomical surfaces ($p<0.01$), and between the root surfaces of non-molar and molar teeth ($p<0.01$) (ANOVA with Bonferroni post-test). The authors then randomly assigned tooth surfaces to select an equivalent number of posterior and anterior teeth in the study, assessing the relationship between age and root colouration. Four points of colour measurement on 40 teeth (sample size permitting, see Table 1) for each known age and gender were assessed for colour saturation (cyan, magenta, yellow and black). The correlation of chronological age to colour saturation was linear for all colours, with correlation coefficients ranging from $r = 0.81$ to $r = 0.94$. The high correlation values strongly support the conclusion that chronological age is related to increased root colouration. **(J Forensic Odontostomatol 2000;18:37-45)**

Keywords: forensic odontology, age determination, teeth, cementum, colouration

INTRODUCTION

Police, coroners or pathologists consult forensic odontologists when there is a need to derive forensically significant information from teeth or dental restorations.[1,2] Gustafson[1] introduced the measurement of six dental regressive changes to estimate age 50 years ago. These regressive changes in dental tissue are still commonly used to estimate the age of adults.

Gustafson's[1] method of aging teeth involved cutting them into 250 to 350 m thick slices and ranking the following characteristics from 0-3: attrition, apical migration of periodontal attachment, secondary dentine formation, cementum apposition, root resorption and root-end translucency. The characteristic ranks were added and an estimated age was extrapolated.[1] Drawbacks to Gustafson's method include: a) none of the criteria can be used alone; b) when only one tooth is available the standard deviation increases greatly; c) expert training in dental histology is required; d) root translucency assessment must be made on sections 1 µm thick; e) the standard deviation increases significantly on teeth over 50 years-old; f) anterior teeth should be used whenever possible and g) the police often require rapid results whereas sectioning requires significant time and expense.[3,4]

* Kodak colour block, Eastman Kodak Corporation, Rochester NY, USA

** Adobe Photoshop 5.00, Apple Computer Inc., USA

The age at death for sub-adults can be estimated by referring to charts indicating age of eruption of teeth. There are, however, differences between races and the genders.[5] Environmental factors and disease can also cause individual variations to be too large to allow accurate estimation of age.[6] Using charts depicting age of root crown mineralization is a good method of estimating age because development is comparatively unaffected by nutritional, endocrine and other factors which impact on the rate of dental maturity.[7]

Since Gustafson[1] introduced his method for age assessment, several similar methods have been performed.[8-15] Of the six criteria introduced by Gustafson[1], the variable with the strongest correlation to age is apical translucent dentine.[13-15] The canine teeth provide the most reliable estimate using this variable.[15]

More recently, the racemization rate of D and L enantiomers of amino acids in enamel, dentin and cementum has been explored.[16-18] The racemization reaction proceeds at a constant rate in dentin and cementum, confirming that cementum like dentin has a low metabolism and is stable throughout an individual's life.[16-18] Therefore, age estimation is possible using the racemization reaction in teeth.[16-18]

Ten Cate *et al.*[3] showed that teeth could be aged by comparing the root colour to a known standard. Their experiments further showed that untrained individuals trying to estimate age of teeth by visual comparison were not consistent but results from trained personnel were more accurate. They also measured optical density of colour using photographs and transmission densitometry. Yellow was measured using a Wratten blue 47 filter and a regression line relating age in terms of optical density was constructed, with the results being better than visual matching. However, a densitometer can only measure a small root surface at any one time, and the processing time is lengthy. Using reflective spectrophotometry, Solheim[19] reported that crown and root dentin colour was related to age. The procedures outlined by Ten Cate *et al.*[3] and Solheim[19] have advantages over Gustafson's method in that any tooth may be used, no sectioning is required, and it is relatively easy to do. Since the studies of Ten Cate *et al.*[3] and Solheim[19] colour measurement has become simplified using colour scanning and computer assisted measurement.

The purpose of this study was to assess a colour change in the root surfaces of adult human teeth and if so, whether this is related to chronological age. We hypothesized that there may be a difference in the colour between the anterior and posterior teeth as well as between the four anatomical surfaces of the roots. In addition, we hypothesize that there is a linear relationship between tooth root cyan, magenta, yellow and black colouration and chronological age.

MATERIALS AND METHODS

A sample of extracted teeth of known age and gender was obtained from Ontario dental practitioners, cleaned with pumice and stored in corked glass jars in cupboards. The teeth were pooled by gender into five-year age groups: 15-19, 20-24, 25-29, and so on until 80-84[3] and three experiments were performed.

Experiment 1: Comparison of yellow saturation of the four anatomical surfaces of the tooth roots

From each of the 20-24 and 70-74 female age groups 21 teeth were scanned by AGFA Arcus II Scanner# using Fotolook## software and then viewed on their mesial, distal, buccal and lingual sides (4 separate scans) by Adobe Photoshop 5.0.** Each scanned image of a particular tooth's root surface was measured at six points for percent yellow saturation – two points from the cervical third, two points from the middle third and two points from the apical third of the root. Areas of environmental staining were avoided. A Kodak colour block* served as a control for variations in the percent yellowness from image to image. The data from each side were entered into Instat 2.0† and a comparison of the percentage yellow between the four sides was tested by ANOVA. A Bonferroni post-test determined the percentage yellowness variation from surface to surface of a tooth's root.

Experiment 2: Comparison of yellow colour saturation of anterior vs. posterior tooth roots

Agfa-Gevaert AG, Mortsel, Belgium
Fotolook 2.0, Microtek Corporation, California, USA
† Instat 2.0, Graphpad Software, Inc., San Diego, CA, USA

Table 1: *Columns depicting how many teeth were used for each age group and both genders in Experiment 1*

AGE GROUP	Number of specimen teeth	
	Female	Male
15-19	38	22
20-24	40	40
25-29	40	40
30-34	40	40
35-39	40	36
40-44	40	40
45-49	40	40
50-54	40	33
55-59	40	40
60-64	40	33
65-69	40	26
70-74	26	10
75-79	6	8
80-84	11	10
85-89	10	

Table 4: *Summary of Experiment 2 data compares the difference in tooth root yellowness between the four anatomical root surfaces between non-molars and molars of 20-24 aged females. Values connected by underlines are not significantly different. Values not underlined are significantly different at least 0.01 according to Bonferroni post hoc test for multiple comparisons.*

Yellow value	54.37±3.97	53.33±3.60
Surface	Mesial Anterior	Mesial Posterior
Yellow value	55.53±6.52	57.04±4.85
Surface	Distal Anterior	Distal Posterior
Yellow value	58.72±5.1	54.61±5.06
Surface	Lingual Anterior	Lingual Posterior
Yellow value	58.90±5.98	54.45±4.89
Surface	Buccal Anterior	Buccal Posterior

Table 2: *Summary of experiment 1 data compares the difference in tooth root yellowness between the four anatomical root surfaces of the 20-24 aged female group. Values connected by underlines are not significantly different. Values not connected by underlines are significantly different at least 0.01 according to Bonferroni post hoc test for multiple comparisons.*

Yellow value	52.82±3.8	56.32±5.72	56.56±5.49	56.57±5.86
Surface	Mesial	Distal	Lingual	Buccal

Table 3: *Summary of experiment 1's comparison of the difference in tooth root yellowness between the four anatomical root surfaces of the 70-74 aged female group. Values connected by underlines are not significantly different. Values separated by underlines are significantly different at least 0.01 according to Bonferroni post hoc test for multiple comparisons.*

Yellow value	67.81±4.63	69.18±5.31	69.63±4.40	69.90±5.55
Surface	Mesial	Distal	Lingual	Buccal

The female 20-24 years data from Experiment 1 were used, but data from both non-molar and molar (10 non-molars, 11 molars) were grouped by tooth surface only (mesial, distal, buccal, and lingual). Thus, the mean female 20-24 mesial anterior tooth yellowness was compared to the mean female 20-24 mesial posterior tooth yellowness and similarly for the other three sides. The data were entered into Instat 2.0† and the means compared by ANOVA and a Bonferroni post-test determined the percentage yellowness of root variation between anterior and posterior teeth.

Experiment 3: Measuring colour change of tooth roots with age

Forty teeth (20 non-molars and 20 molars) [sample size permitting, see Table 1] from each age group and gender were placed randomly on the scanner, the images were scanned by Fotolook 2.0## and viewed by Adobe Photoshop 5.0**. Four points on each tooth's root were selected at random (two above the midline of the root and two below). Once again environmentally stained areas were avoided and Photoshop 5.0** was used to measure the percentage cyan (c), magenta (m), yellow (y), and black (k) at each of the four points. Instat 2.0† performed an analysis of the mean values and confidence intervals for each age and gender group. The mean and standard deviation with a 95% confidence level for each of the colour values, age groups and genders were determined and plotted. The mean

†† Cricket graph 1.3.2, Computer Associates International, Islandia, NY, USA

and 95% confidence levels of the various colour values and age groups were then graphically displayed using Cricket graph 1.3.2[††] for display.

RESULTS

Experiment 1

The one way analysis of variance with Bonferroni post-test for multiple comparisons in experiment 1 yielded unexpected results. The anatomic surfaces of the teeth for the female 20-24 year group revealed a significant difference between the percentage yellow of the mesial surface of tooth roots when compared to the other three root surfaces (Table 2). The p value was < 0.01 between the mesial and distal surfaces and < 0.001 between the mesial and both the buccal and lingual surfaces, indicating that the mesial surface of the roots were less yellow than the other three surfaces.

The female 70-74 year data (Table 3) indicated a significant difference in colour value between the mesial surface and the buccal and lingual, but failed to show a significant difference between the percentage yellow of the mesial and distal surfaces. This finding is unlike the female 20-24 year category which indicated a significant difference between the mesial and the other three surfaces. However, the measured t value (2.18<2.65) for the comparison between the percentage yellow of the mesial versus distal root surfaces indicated a non-significant, linear trend.

Experiment 2

The second experiment's results indicated that there was a significant difference in the percentage of yellowness between two of the four root surfaces (Table 4). The one way ANOVA with a Bonferroni post-test multiple comparison gave a $p<0.001$ for the comparison between lingual and buccal surfaces on anterior versus posterior root surfaces. The difference between the anterior and posterior root surface yellowness is therefore not due to chance. Comparing mesial and distal anterior with posterior surfaces, however, gave $p>0.05$ resulting in a failure to show a statistically significant difference between these surfaces.

Experiment 3

Figures 1 to 8 depict the relationship between the percentage cyan, magenta, yellow, black and the age of the genders. All of these figures indicate a positive increase in the percentage of the measured colour with age. A correlation value of 0.93 between the percentage cyan and age for males was calculated (Fig. 1) while values of 0.93, 0.90 and 0.94 were determined for the correlation between age and the percentage magenta, yellow, and black in males. The slope of the male data graphs ranged from 1.16 to 1.96% colour/5 years, the smallest slope being 1.16% magenta/5 years, while yellow and cyan produced slopes of 1.20 and 1.23 respectively. Figure 4 depicts the change in percentage black with age for males, which yielded the steepest slope of 1.96% black/5 years.

The linear model fitted the data well. In all the male graphs (Figures 1-4) the straight line model intersected more than half of the values within their 95% confidence intervals.

Figures 5-8 show the correlation between female age and percentage cyan, magenta, yellow and black respectively. The correlation values for the female data ranged from 0.81 to 0.93, the lowest and highest correlation values being attributed to magenta and cyan in figures 6 and 5 respectively. This was unlike the male data which indicated that the lowest and highest correlation values were from yellow and black respectively. The female slopes ranged from 0.83 to 1.30 and in agreement with male data, the females' magenta graph in Fig. 6 gave the lowest slope of 1.30, while the black gave the greatest slope of 1.30. The slopes for the cyan and yellow were 0.99 and 1.18, which were different from the male data.

Another difference between female and male graphs in Figs. 1 to 8 is that all the female graphs have a higher y-intercept values of 25.43, 23.65, 64.81 and 5.22 for percentage cyan, magenta, yellow and black respectively whereas the male y-intercepts for the graphs of cyan, magenta, yellow and black versus age were 24.26, 21.41, 54.77 and 1.63 respectively. For females the straight line model fitted the data well but there were less cases when compared to the male graphs of the line intersecting a majority of the measured values within their 95% confidence interval.

DISCUSSION

The results of experiment 1 indicate that there is an uneven colouration of the four anatomic root surfaces. The percentage yellow was similar on three of the four surfaces, and considerably less on the mesial surface of the female 20-24 year teeth. This may indicate that less cementum is deposited on the mesial surfaces which coincides with the phenomenon known as "mesial drift", defined by Jablonski[20] as the gradual movement of the teeth mesially as a consequence of natural interproximal tooth wear and which could result in cementum deposition on the other 3 surfaces. Similarly, the 70-74 year group data indicate a significant difference among three of the four surfaces. Our study was not specifically concerned with the cause of the difference in percentage yellow, but this difference will be the subject of further research.

For the purpose of the current analysis of root colouration as an indicator of chronological age, the results of experiment 1 encouraged us to expand our sample size to 40 teeth. In addition, randomly selected tooth surfaces were scanned due to the observed varying colouration between mesial and other root surfaces.

A significant difference in the degree of yellowness between buccal and lingual non-molar versus molar teeth was observed. However, no significant difference was observed among the other two surfaces and these results coincide with the observations in experiment 1 where tooth roots have an uneven distribution of cementum, among non-molar and molar teeth on certain anatomical surfaces. The cause of this variation requires further research. This data indicated that a similar number of non-molars and molars would be necessary in experiment 3 to give a meaningful colour measurement.

The data in experiment 3 confirm conclusively that there is a relationship between tooth root colour and age, in which all four colour measurements correlated well. This change in colour with age is due to continued cementum deposition through life[3,8] or perhaps due to some undefined intrinsic change in the dentine.[21] The authors have not attempted to determine this. With the lowest correlation value of 0.806 and the majority of the values above 0.9, these data clearly indicate an important and indisputable relationship between root colouration and age and from a forensic dental viewpoint this correlation could prove to be quite useful when the age of found remains needs to be estimated.

Variation between the genders was present, yet small. In all of the graphs the females had higher y-intercept values which may coincide with the observed sexual dimorphism between male and female tooth eruption times. Since female teeth, with the exception of third molars erupt earlier than in males, these higher values may be due to slightly longer cementum deposition time or development of coloured substances in the dentine.[21] This argument contrasts with Solheim's[22] finding that there is less cementum present on female teeth and further, that less cementum on female teeth may be due to their smaller tooth size or the weaker masticatory force applied. Another difference between the male and female graphs of colouration with age is that all the male graphs had greater slopes. Could it be possible that there is dimorphism in the rate of deposition of cementum or other coloured products? Once again, the biological mechanism behind the increased colouration with age needs to be explored further.

The advantage of this technique over other methods is that it does not require long and expensive laboratory time and can be performed with a minimal knowledge of dental anatomy and computer skills. The tooth is not destroyed and it is inexpensive to perform. The disadvantage and limitations of this technique however are that teeth need to be extracted, the standard regression equation may not apply to all populations, racial groups and conditions of the remains, whether a body has been burned or buried, and for how long, all may have an effect on tooth root colouration. It may however be possible to make new standard correlation equations for each of these conditions using the same technique.

Another problem with this study is that the correlation values only apply to this sample of teeth which originate from a limited geographical site and probably contain a limited number of ethnically diverse subjects. It however creates a further data base to help cover the multiplicity of geographical regions and populations that exist.

Although the reason for each tooth's extraction was not known, one could argue that these factors might alter its root's colouration but despite this the linear trend remained true in all cases for all colours.

To establish the usefulness of this technique, further testing is required. One method would be to use these data to conduct blind tests of single or groups of teeth of known age and compare the results. In addition, field-testing could be undertaken in actual forensic cases to determine this technique's potential in age determination compared with other currently used methods.

A significant difference was observed between the colouration of the four root surfaces, as well as a significant difference between non-molars and molar tooth root colouration. The regression lines for male and female teeth from 15-89 and 15-84 years respectively indicated an excellent correlation between the measured colour and age. This technique may be valuable to forensic dentists faced with the task of estimating age.

ACKNOWLEDGMENTS

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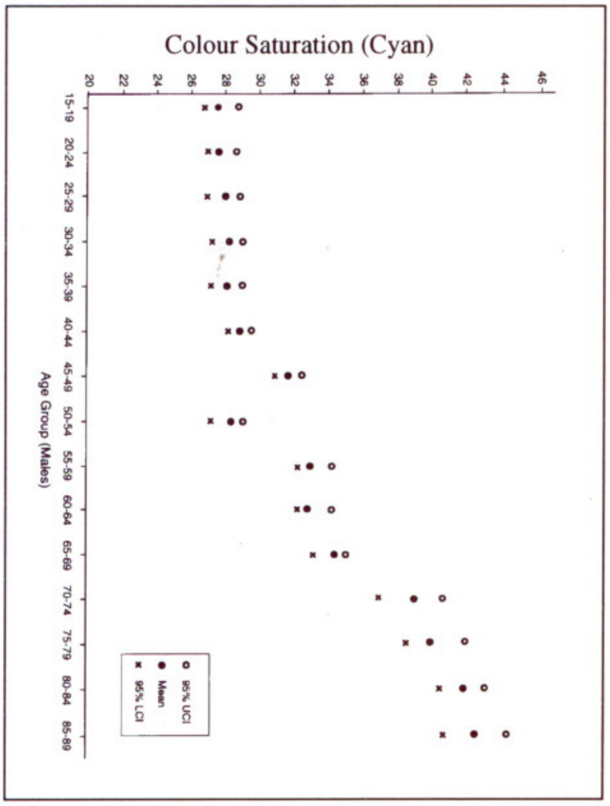

Fig.1: *The percentage of in tooth root cyan colouration as measured in teeth of male subjects displayed by age group r=0.93*

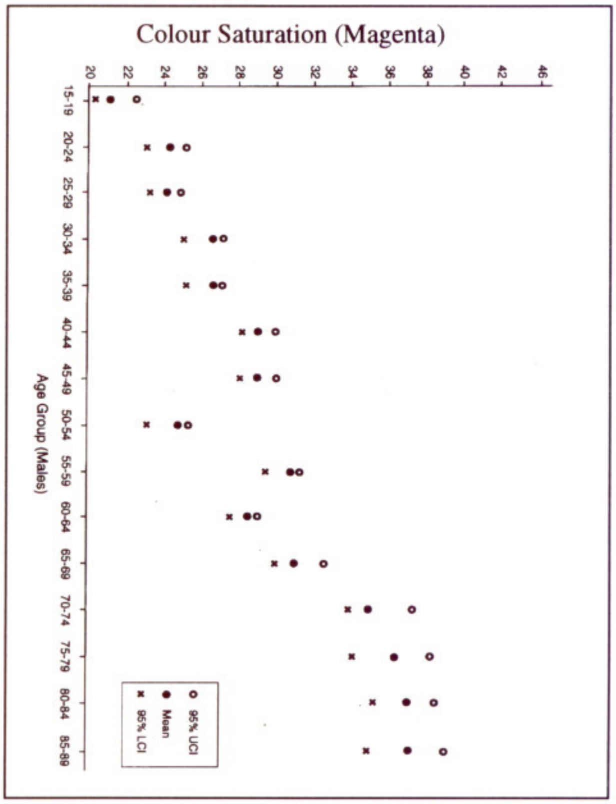

Fig.2: *The percentage of in tooth root magenta colouration as measured in teeth of male subjects displayed by age group r=0.93*

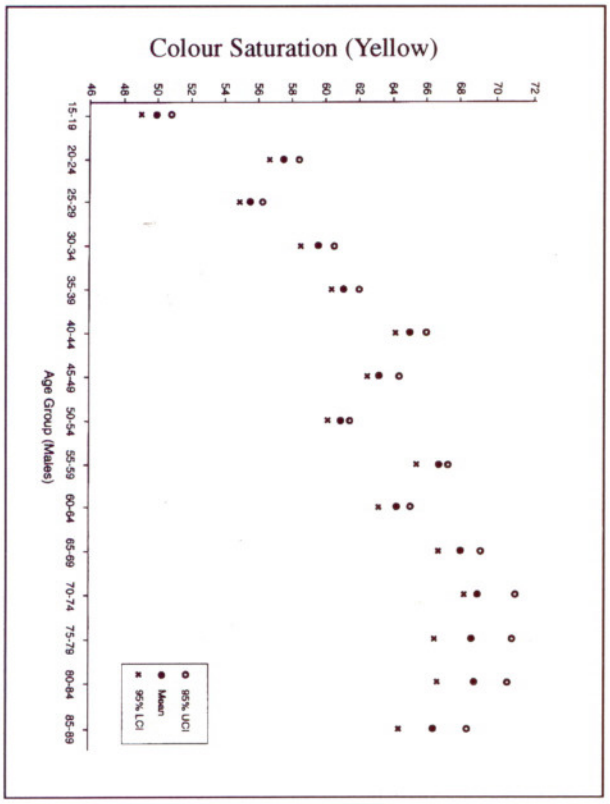

Fig.3: *The percentage of in tooth root yellow colouration as measured in teeth of male subjects displayed by age group r=0.90*

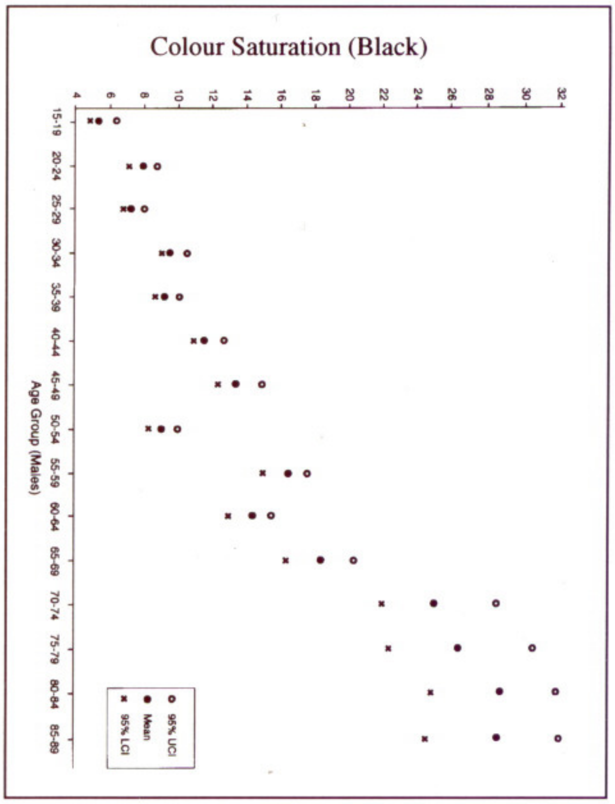

Fig.4: *The percentage of in tooth root black colouration as measured in teeth of male subjects displayed by age group r=0.94*

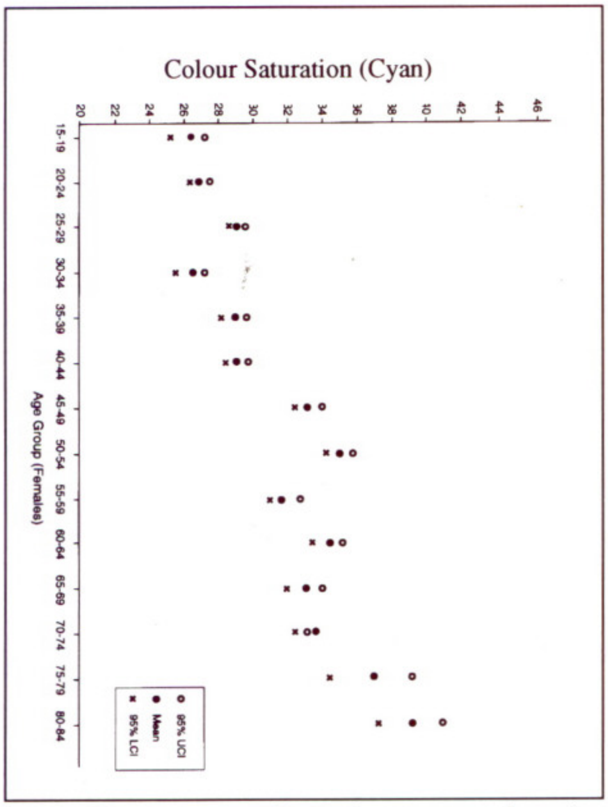

Fig.5: The percentage of in tooth root cyan colouration as measured in teeth of female subjects displayed by age group r=0.93

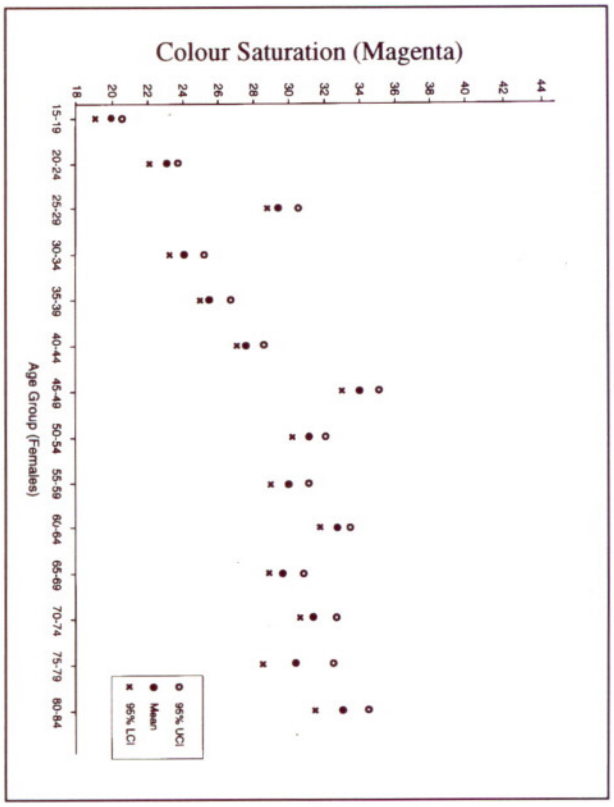

Fig.6: The percentage of in tooth root cyan colouration as measured in teeth of female subjects displayed by age group r=0.81

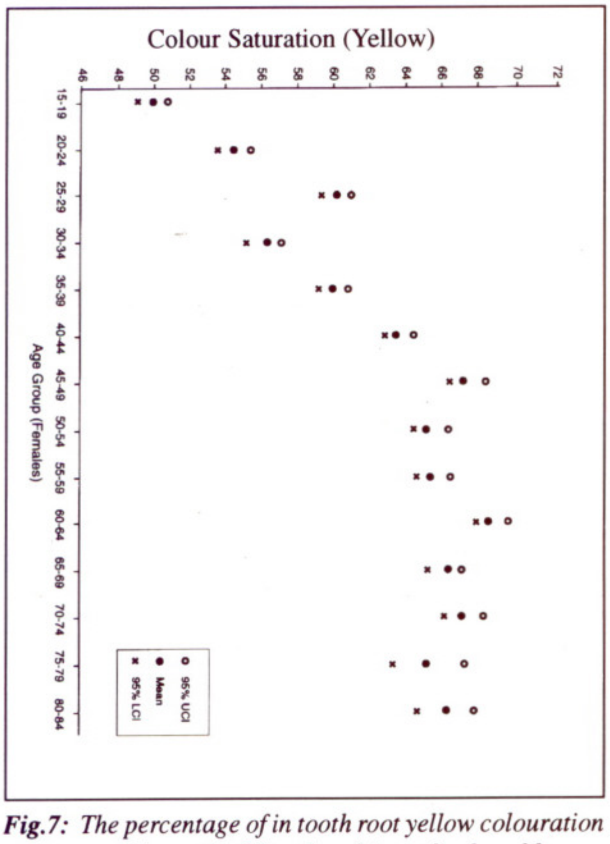

Fig.7: The percentage of in tooth root yellow colouration as measured in teeth of female subjects displayed by age group r=0.86

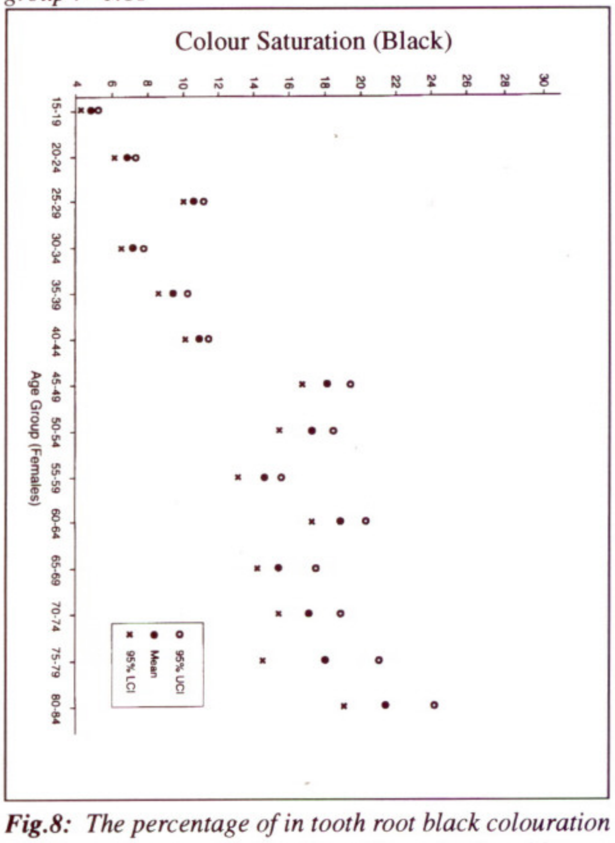

Fig.8: The percentage of in tooth root black colouration as measured in teeth of female subjects displayed by age group r=0.92

WOUNDING DYNAMICS IN DISTORTED BITEMARKS: TWO CASE REPORTS

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ABSTRACT

An essential factor involved in distortion of bitemarks on the skin is the dynamics of biting related to the location on the body. This study describes the comparison between the identification of bitemarks left on different regions of the victims' bodies in two homicide cases. The findings indicated that a stepwise dynamic comparison of serial adjacent marks with a part of the dentition in consideration of movement of the jaws and distortion of the skin was useful in identifying matching points. The identification process indicated possible wounding dynamics of biting. **(J Forensic Odontostomatol 2000; 18:46-51)**

Keywords: Forensic odontology, identification, bitemark dynamics, distortion

INTRODUCTION

A bitemark left on the victim is important evidence for identification of the assailant.[1-5] When bitemarks are not distorted, two dimensional comparison with dentitions will be useful to establish similarities and differences.[6-8] However, various distortions can be caused by the dynamics of biting particularly taking into account the location of the bite (dynamic distortion), artefacts and photographic documentation.[9-11] A simple superimposition procedure is usually insufficient to obtain a convincing result and various other methods for bitemark analysis have been reported.[12-22] An idea for the identification of bitemarks with a possible distortion may be a stepwise dynamic comparison of serial adjacent marks with a part of the dentition in consideration of movement of the jaws and distortion of the skin. The identification process can consequently be used to interpret the dynamics of biting (wounding dynamics). It may be also useful for identification of bitemarks with another kind of distortion such as photographic distortion. The concept of dynamic wounding and its analysis in bite injury has become of interest in forensic pathology,[7,10] and this study describes the identification of bitemarks left on different locations of the bodies of victims in two homicide cases, suggesting the possible wounding process.

METHODS OF BITEMARK IDENTIFICATION

Photographs: In the present cases, the photographs of bitemarks with a two-dimensional scale used for identification were taken using normal flashlight by the police officers during autopsy and printed as usual. The scaled bitemark photographs were scanned and stored in a personal computer.[23]

Superimposition: Dental casts were fabricated from the suspected assailants and also from the victim in Case 1. The occlusal surfaces of all dental casts were then pressed onto the flat surface of a plate of clay and the outlines highlighted in black ink to simulate bitemarks.[18,23] This technique provides more precise marks than direct inking of occlusal surfaces on casts, particularly considering the three-dimensional alignment of teeth. The marks were then scanned and stored in a personal computer. Superimposition for comparisons of the dentitions with the bitemarks was performed by computer and appropriate graphics software Adobe(R) Photoshop 4.0 J.*[18]

* Adobe Systems Incorporated, Mountain View, CA

Wounding Dynamics: Often during biting of a human victim the movement of the jaws and the mobility of the bitten tissues leads to distortion. This needs to be taken into consideration when matching a dentition to a bitemark and the phenomenon may be labelled wounding dynamics.

Stepwise Dynamic Comparison: This procedure is an attempt to follow the extent and direction of the bitemark distortion by identifying it and by superimposing in a stepwise or serial fashion the suspected biting dentition. Matchings come to light which, if the dentition's characteristics are unique, are reliable enough to be positive.

CASE REPORTS

Case 1

Case history

A 49 year-old female victim was found dead after a fire at her home. The immediate cause of death was carbon monoxide poisoning, possibly contributed partially by protracted asphyxia from strangulation. Two bite injuries were identified at autopsy:

a) A spindle-shaped contusion (5.5x8 cm) with arching alignment of some small triangular and rectangular abrasions (0.5x0.9 cm) on the lateral surface of the right upper arm (Fig. 1a).

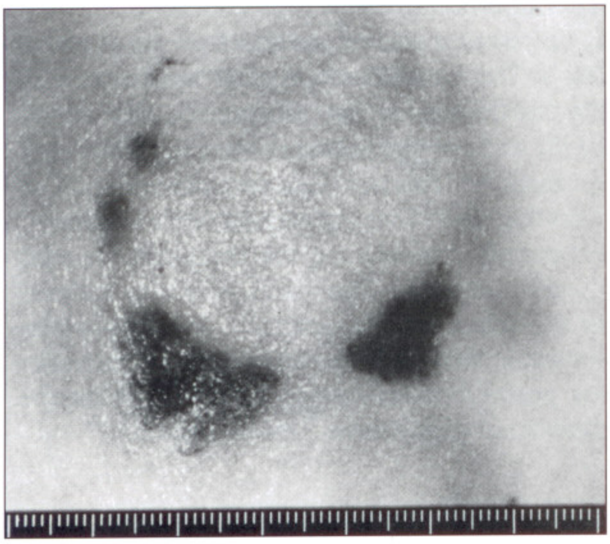

Fig.1(a): Bitemark in Case 1 (right upper arm)

b) An elliptical contusion (6x4.5 cm) surrounded by some small arching abrasions (0.5x2.5 cm) on the anterior surface of the left knee joint (Fig. 1b).

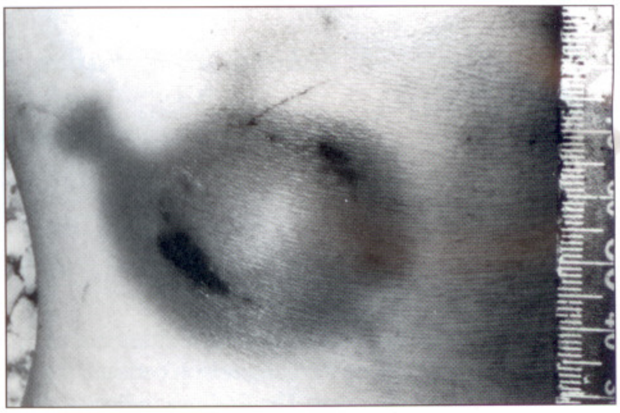

Fig.1b: Bitemark in Case 1 (left knee joint)

Bitemark analysis

Dental casts were fabricated from the suspected assailant, a 44 year-old female, and from the victim at autopsy because of the possibility of self-infliction of the knee joint bite and possible retaliatory bitemarks on the assailant. Comparisons of the dentitions with the bitemarks suggested characteristic similarities of the shape, size and alignment of the mandibular left lateral incisor, canine and 1st premolar with the bitemarks on the lateral surface of the right upper arm and of the mandibular left lateral incisor, canine and the maxillary central incisors of the suspect. Further bitemark analysis was performed using stepwise dynamic comparisons of serial adjacent marks with a part of the dentition and likely movement of the jaws concerned taking into consideration distortion of the skin (wounding dynamics) as described below. Only the suspect's dentition matched the marks at every point.

a) Bitemarks in the right upper arm (See Figs. 1, 2 and 3)

Step 1. When a cast of the mandibular left lateral incisor, canine and 1st premolar was superimposed over the middle part of bitemarks and moved obliquely upward, the labial and lingual margins matched the contours of the marks (Fig. 2a).

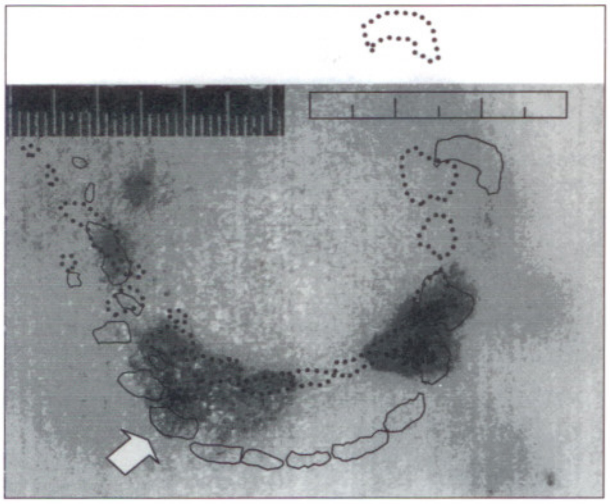

Fig.2a: Comparison of the suspect's dentition with bitemarks in the right upper arm of Case 1 (overlay of the mandibular left lateral incisor, canine and premolar)

Step 2. The right side bitemarks in the photograph were compared with the right part of the mandibular dentition. When impressions of the mandibular right canine and 1st premolar were overlaid and shifted obliquely upward, the labial and lingual margins matched the bitemark contours (Fig. 2b).

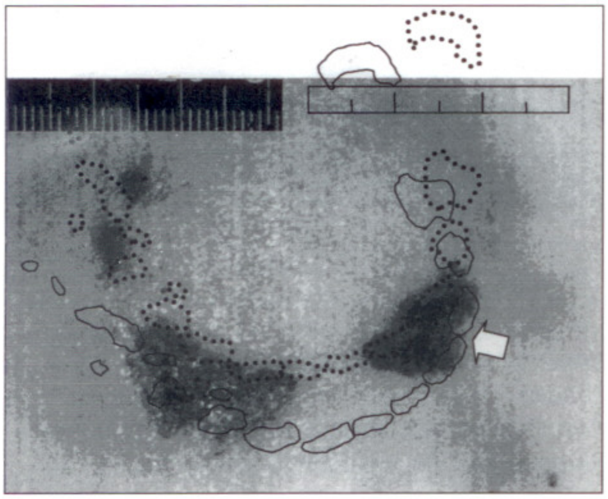

Fig.2b: Comparison of the suspect's dentition with bitemarks in the right upper arm of Case 1 (overlay of right lateral incisor and canine on the bitemarks. Matching points are shown with arrows [wounding dynamics demonstrated by shift between dotted and solid lines])

In the terminal position, the lingual margins of the left lateral incisor, canine and 1st premolar were located somewhat upward to be seen in Fig. 2b, well in parallel with the middle part of the bitemark contours, suggesting a skin surface upward distortion which thus enabled confirmation of the identity.

Step 3. When the left upper side bitemarks in the photograph were compared with the mandibular left 2nd premolar and 1st molar, a good match was obtained by a clockwise rotation of the dentition. In this case, the initial position of the left 2nd premolar coincided well with the terminal position in Step 2.

In conclusion, the bitemarks were considered to be made by the mandibular dentition, accompanied by a considerable dynamic distortion. Initially the left lateral incisor, canine and 1st premolar pressed upward, abrading and distorting the skin, causing the middle part of the bitemarks, followed by the penetrations of the right canine, 1st premolar, left 2nd premolar and 1st molar from both sides.

The process was repeated for the bitemarks in the right upper arm and left knee joint using axial and rotatory shifting, which started at the anterior teeth (incisors and canines) and finished at the molars (Figs. 2a, 2b and 3).

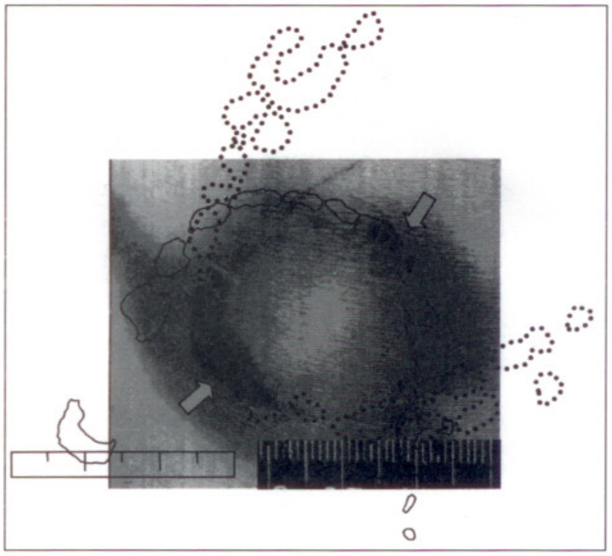

Fig.3: Comparison of the suspect's dentition with the bitemarks in the left knee joint of Case 1. Overlay of the upper (dotted line) and lower (solid line) anterior teeth on the bitemarks. Matching points are shown with arrows

b) Bitemarks in the left knee joint

The upper and lower bitemarks coincided with the impressions of the mandibular left lateral incisor and canine and the maxillary central incisors, respectively, as follows:

Step 1. The maxillary central incisors were superimposed over the lower bitemarks. An upward movement of the teeth on the round patellar surface could well explain a slightly upward distortion of the lateral parts of the bitemarks (Fig. 3). A slight abrasion was identified at the location of the right canine in the terminal biting position.

Step 2. When the mandibular left lateral incisor and canine were compared with the upper bitemarks, a good match was obtained by a rotatory shifting. An additional tracking mark of the mandibular right lateral incisor was observed proximally to the bitemarks (Fig. 3).

In conclusion, the bitemarks were considered to be made by the upward and downward movements of the maxillary and mandibular dentitions, respectively. Dynamic distortion of the bitemarks was markedly smaller at the knee joint than at the upper arm.

Case 2

Case history

A 46 year-old female victim was found dead in the grounds of a shrine. The cause of death was asphyxiation from manual strangulation and/or smothering. Three typical bite injuries of a similar size were identified on the upper abdomen: circular bruises (4-5 cm in diameter) with small, pale, rectangular impressions (0.2x0.5 cm-0.5x0.8 cm) (Fig. 4).

Bitemark analysis

Dental casts were fabricated from a 44 year-old male suspect. Superimposing the dental impressions over the bitemarks was performed as before in Case 1 and the suspect's dental casts matched the bitemarks at every point (Fig. 5).

The images of the dental impressions were dynamically compared with three bitemarks, which were located a) left-side, b) right-side and c) middle on the upper abdomen, by axial and rotatory shifting in a similar way as described above. Only minor dynamic distortion was observed in all three bitemarks.

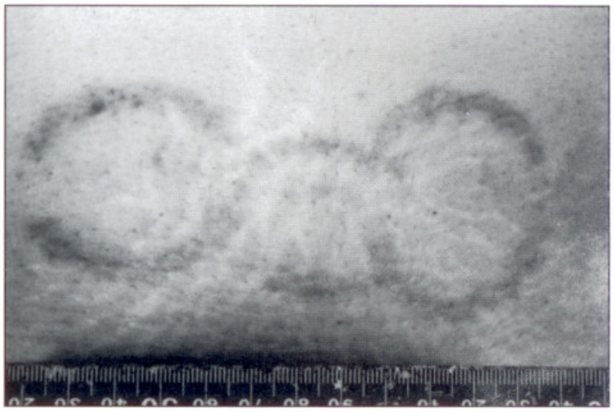

Fig.4: *Photographs of the bitemarks in Case 2*

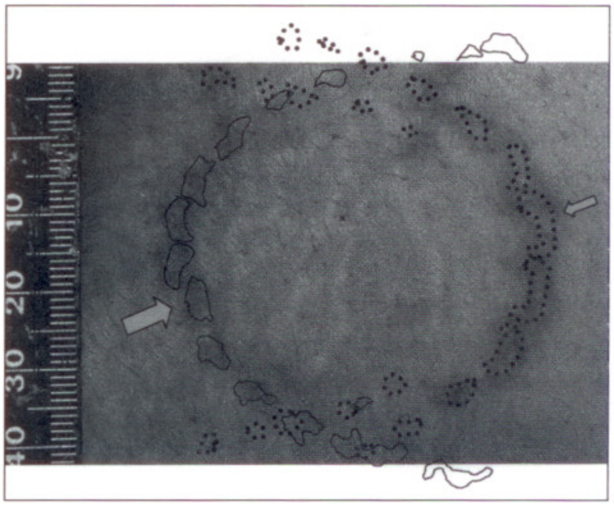

Fig.5: *Comparison of the dentition with the bitemarks in Case 2: the suspect's clay impression images matched the marks at every point (arrows) [dotted line, upper dentition; solid line, lower dentition]*

a) The left-side bitemarks

Step 1. The maxillary incisors and canines showed a good matching with the upper bitemarks by simple superimposition (Fig. 5).

Step 2. The mandibular incisors and canines also matched the lower bitemarks, and the slightly upward shift gave a good match of the right premolars.

In conclusion, the upper and lower bitemarks coincided well with the clay images of the maxillary and mandibular anterior teeth, respectively.

b) The right-side bitemarks

Step 1. The maxillary incisors showed a good matching with the upper bitemarks, and a small rotatory shift gave a match of the canines and molars.

Step 2. The mandibular incisors showed a good match with the lower bitemarks, and the canines and molars matched well after a small rotatory shift.

In conclusion, the bitemarks coincided well with the clay images of the maxillary and mandibular teeth.

c) The middle bitemarks

Characteristics of the bitemarks were not so clear as the others. No mismatching points were found by dynamic comparison with the dental impressions images.

DISCUSSION

Bitemarks on victims are usually distorted to some extent as a result of the curvature and elasticity of human skin, which mainly depends on the anatomical location and contribution of tissue composition and underlying structures. Further distortion can be caused by the dynamics of biting related to the body region, physical position of the victim, fighting or struggling, biting direction and force. This kind of distortion due to wounding process is of a great interest in forensic pathology. The shape of bitemarks may also be distorted by artefact and angles at photographic documentation. Although an exaggerated distortion may be produced when bitemarks are photographed from a markedly oblique angle,[11,23,24] this is rarely a problem where a characteristic shape of wound (bitemark) is recognizable. Minor photographic distortions do not greatly alter the view of individual dental clay impression images and alignment of adjacent teeth. The curvature of the body does not produce significant photographic distortion, and was not a problem in cases where the entire mark could be observed from one viewing angle.[10,11] Thus, it appears practical to compare a set of serial adjacent bitemarks with a part of the dentition in a stepwise dynamic comparison as a way of overcoming cases of considerable distortion especially due to the dynamics of biting.

In the present cases, convincing identification was possible using dynamic comparisons, although distortions of bitemarks due to curvature of the body surface and the dynamics of biting were observed to varying degrees. The distortion was minor in the knee (case 1) and also on the abdomen (case 2), whereas it was more marked in the upper arm (case 1). These observations supported previous studies that moderate tissue flexibility may cause a larger dynamic distortion than low and high flexibility.[5,11,23,25]

As mentioned above, a stepwise dynamic comparison of serial adjacent marks with a part of the dentition in consideration of movement of the jaws and distortion of the skin was useful to identify convincing matching points when marked distortion was suspected. The identification process indicated possible wounding dynamics of the bite.

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BOOK REVIEW

by Yvo Vermylen

Forensische Zahnmedizin

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The title of this book, Forensische Zahnmedizin means Forensic Odontology, but it is more than just that, containing in addition an extensive coverage of dental law which in its broadest sense traverses well into the medical and legal fields.

It is divided into two major parts: dental law and forensic odontology. In part one aspects of dental law cover all important topics in the field of medical and dental law. For example there is a rich documentation of literature and court proceedings and sentences, an overview of the legal principles imperative in the dentist/patient relationship (dental contract, informed consent, duty-of-care, liability, continuing education, confidentiality, duty to keep dental records), the relationship with the dental technician and medication law. A fact of dental practice is that treatments do fail and accidents happen and increasingly dentists are being brought before the courts, charged with malpractice. This book focuses on all aspects of possible malpractice including the particularly vulnerable fields of oral surgery and prosthodontics. Also dealt with in detail are the role and duties of the dental expert involved in malpractice trials.

The second part of the book is devoted to forensic odontology and starts with tooth development, the different systems of tooth numbering, dental charts and the Interpol forms for mass disaster DVI. The chapter on dental age estimation covers the topic very thoroughly and the following chapter deals with dental identification (describing such notable cases as those of Eva Braun, Martin Bormann and Adolf Hitler), gender differences, denture marking and related technical procedures. Chapter 2H covers bitemarks and gives a very good overview of all the different methods of investigation and how to handle the pieces of evidence, and the last chapter deals with lip prints.

This book is written in cooperation with Rolf Singer, Günter Seifert, Tore Solheim, Wolfgang Pilz and Dieter Leopold and this depth of expertise makes it a very valuable book that is highly recommended reading for every forensic odontologist. It is written for dentists, students, practitioners in the field of forensic sciences, lawyers, criminalists and insurance companies. The only disadvantage is that the book is written in German, but it could well be translated into English which would deservedly make it available to a much broader public. Beautifully bound and presented, it contains 110 illustrations and 45 tables and it can take pride of place in any personal or public library.