ISSN 2219-6749

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The Journal of Forensic Odonto-Stomatology

Volume 16, n. 1 - Jun 1998

OFFICIAL PUBLICATION OF THE
INTERNATIONAL ORGANIZATION FOR
FORENSIC ODONTO-STOMATOLOGY

CONTENTS

MACROSCOPIC AND MICROSCOPIC ASPECTS OF INCINERATED TEETH

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ABSTRACT

Fifty-eight premolars, extracted for orthodontic reasons, were incinerated for one hour in a furnace at temperatures varying from 150°C to 1150°C, increasing at 100°C intervals. Between 150°C and 700°C the teeth changed from light yellow to bluish-white passing through brown. The scanning electron microscope showed that at 150°C cracks appeared in the enamel, increasing in number as the temperature rose. At 450°C the internal surface detached from the dentine and showed the same cracked surface as the exterior. Above 1100°C the enamel fragments had a prismatic structure difficult to identify due to their melted structure. Dentine retained its tubular structure up to 1150°C even when the tubules' diameter decreased at 700°C. The cementum cracked as the temperature increased. Certain cracked areas actually detached at 600°C giving a corroded aspect to the cementum and the residual zones became irregular when the temperature rose and eventually at 1150°C the cementum was no longer identifiable. **(J Forensic Odontostomatol 1998; 16: 1-7)**

Keywords: Forensic odonto-stomatology, burnt teeth, enamel dentine, cement

INTRODUCTION

After a fire involving loss of life, police investigation assisted by forensic expertise is necessary in order to establish the circumstances surrounding the event. Among the elements generally found on site, teeth are important as they are particularly heat resistant and their role in the field of identification is recognized.[1-3] In addition, macroscopic examination of burnt teeth can reveal clues as to their "thermal history", a useful parameter in understanding the chain of events during the fire. Examination under scanning electron microscope (SEM) of severely burnt teeth is also useful, further enhancing the investigation.[4,5] The limited amount of research on burnt teeth has, however, not included the possibility of determining the temperature of a fire from the state of teeth. [2,6-9] With this goal in mind we studied the macroscopic and microscopic alterations of teeth burned in a variety of temperatures.

MATERIALS AND METHODS

Fifty-eight premolar teeth, free from any pathology and extracted from adolescents for orthodontic reasons, were burned after having been kept for less than a week in water. Twelve temperatures were applied: 150°C, then from 200°C to 1100°C in 100°C increments, and 1150°C. In each case a tooth was placed for one hour in a dental ceramic furnace* at the required temperature. After heating, the tooth was removed and left to cool in open air, after which four teeth from each temperature level were observed macroscopically for changes in colour. Each tooth was then mounted on a metal sample carrier using a double-sided adhesive and silver-rich glue, to be vacuum-metallized by cathodic gold spraying. All teeth were then observed by SEM** at magnifications varying between 20x and 2000x. In order to validate this method, and as a control, a tooth was fired at 50°C, a temperature at which it would not normally undergo deterioration, and cooled under the same conditions. Finally, after observing 49 burnt premolars, nine more were submitted to temperatures varying from 410°C to 490°C for one hour in increments of 10°, in order to determine the temperature at which enamel and dentine separate.

* KAVO EWL type 5645
** JEOL 35 J.S.M.

RESULTS

1. Macroscopic aspect of the tooth

At 150°C enamel showed crazing but retained its normal colour. After having lost its gloss at 200°C it crazed further with an increase in temperature, initially at the neck and then over the crown. A few frank cracks then developed and the root, which was light yellow at 150°C, took on a calcinated aspect with the onset of numerous small bubbles at 300-400°C. It then displayed a crazed structure which was restricted to the coronal one-third and which, above 800°C, progressed to the apical third, initially having a granular appearance. At 400°C an increased separation of the dentine from the enamel was observed with the formation of a ledge at the cemento-enamel junction, and the total separation of the enamel shell occurred at 450°C. The colour of the enamel was between brown and dark grey at this temperature, after having passed through a very light brown colour. Its change in colour appeared always to begin at the neck, with the onset of spots, and then with crazing on its internal surface, it fractured above 500°C into fragments which became consistently smaller as the temperature rose. The colour of the fragments was dark grey. The coronal dentine, which was exposed at 450°C, initially appeared grey. Cracks appeared at 600°C before disintegration from 800°C onwards, without conversion into powder as was the case with enamel. Concomitantly, it became white after passing through a bluish-grey shade. It was only at 1150°C that the root became totally white and took on a chalky aspect (Fig.1).

Figure 1. *Colouring changes of burnt teeth.*

a.	*150°C.*	*Crown: Normal colour and shiny appearance* *Root: Off-white to light yellow*
b.	*200°C*	*Crown: light brown dull surface*
c.	*300°C.*	*Crown: Light brown to greyish with dark brown spots* *Root: Black*
d.	*400°C*	*Crown: Dark brown/grey* *Root: Shiny black*
e.	*500°C*	*Crown: Enamel (dark brown/grey) separated from dentine (grey)* *Root: Grey/brown*
f.	*600°C*	*Crown: Dark grey enamel and blue-tinted grey dentine* *Root: Blue-tinted grey with white spots*
g.	*700°C*	*Crown: Very dark grey enamel and blue-tinted white dentine with navy blue cuspid tips* *Root: Blue-tinted white to white*
h.	*800°C*	*Crown: Very dark grey enamel with white spots and white dentine* *Root: White with grey spots* *Cracked appearance of the dentine starting at 800°C*
i.	*900°C*	*Crown: White dentine* *Root: White with dark grey to black spots at cemento-enamel junction*
j.	*1000°C*	*Crown: White dentine* *Root: White except at the apex where the root is slightly pinkish*
k.	*1100°C*	*Crown: Chalky white appearance* *Root: Chalky white appearance*
l.	*1150°C*	*Crown: Chalky white appearance* *Root: Chalky white appearance*

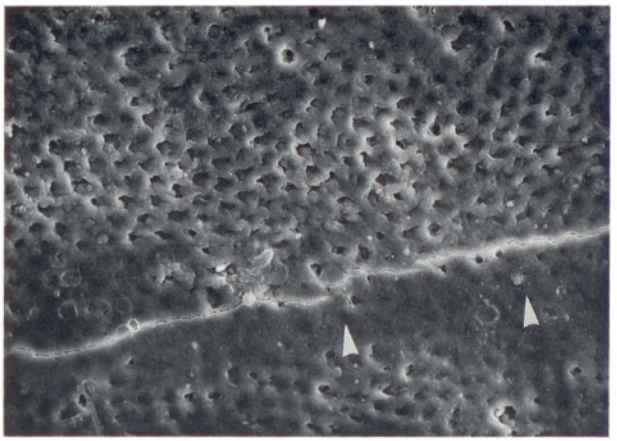

Figure 2. *T = 150° (x500): Superficial modification of the enamel showing a probable interference with the striae of Retzius leading to the creation of fine lines (perikymata) (arrowed).*

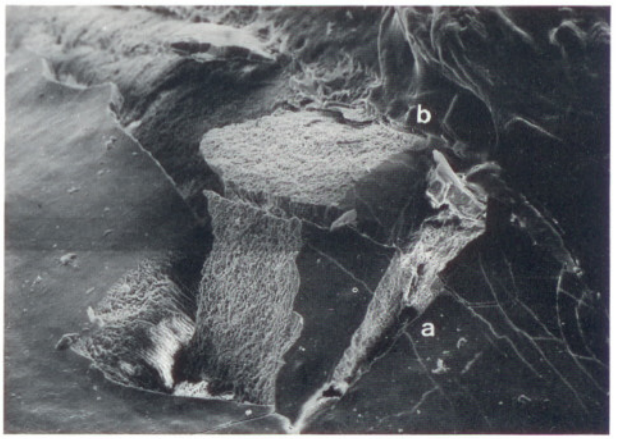

Figure 3. *T = 200° (x100): The crazing (a) develops from the cemento-enamel junction (b). The enamel appears to be normal.*

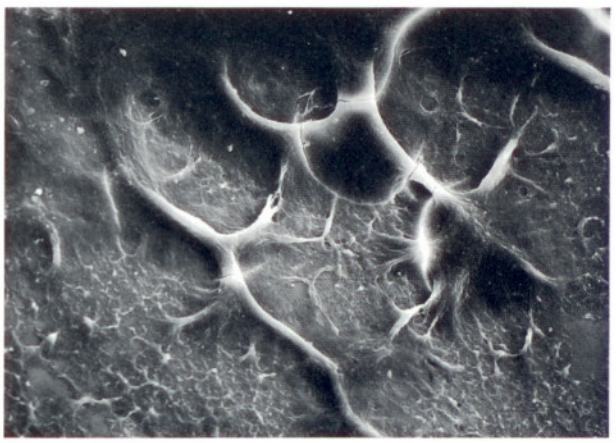

Figure 4. *T = 300° (x500): A melted appearance standing proud of the enamel surface (0,2 to 2 mm in length).*

2. Microscopic aspect of the enamel

At 150°C the enamel displayed superficial changes (Fig.2) confined to a few small crazing patterns and an approximate 100µm crack containing clumps of prisms with a normal structure and oriented perpendicularly to the surface. The crazing lines were more numerous and more pronounced at the level of the cemento-enamel junction (Fig.3). At 300-400°C a few zones with a "molten" aspect were noted, which got smaller as the temperature rose (Fig.5). Crazing patterns and cracks multiplied with the rise in temperature, leading to a chequered look of the enamel at 400°C (Fig.4). After having separated from the dentine at 450°C, the enamel shell showed the same appearance on its internal surface. Furthermore, in certain areas it was covered with fragments of dentine (Fig.6). The enamel fragments, which became consistently smaller with the increase in temperature, presented the typical prismatic structure up to 1100°C (Fig.7), but after that the prisms melted and lost their recognizable form (Fig.8).

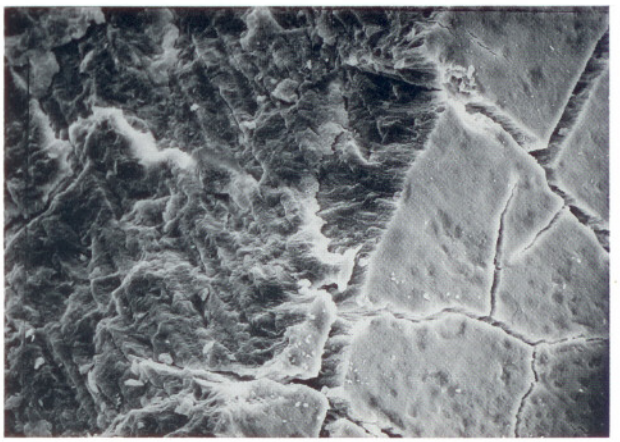

Figure 7. *T = 800° (x500): Cracked enamel fragment displaying a prismatic structure which has not been modified.*

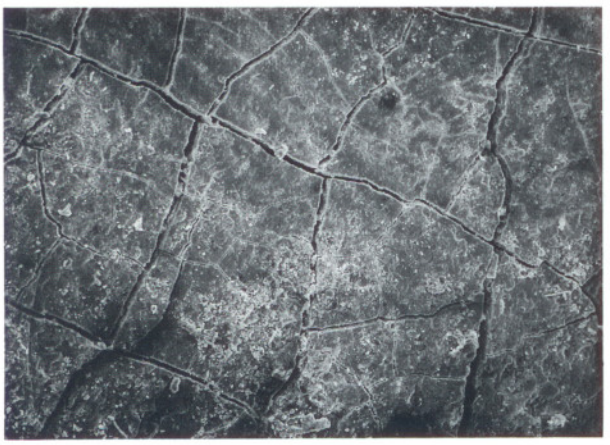

Figure 5. *T = 400° (x100): The enamel exhibits numerous cracks which are perpendicular to each other together with minor cracks constituting a crazing pattern.*

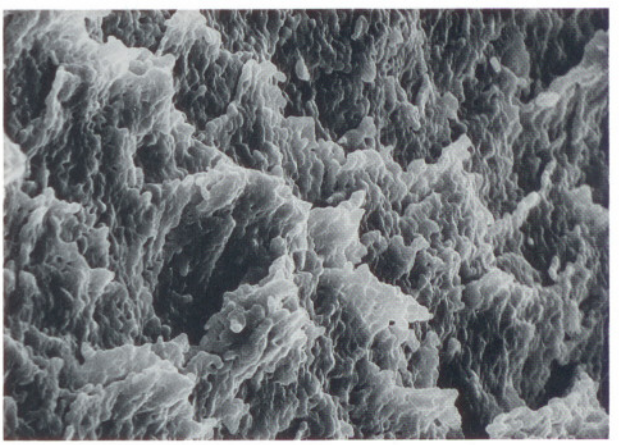

Figure 8. *T = 1150° (x1200): The enamel structure is difficult to recognise. Further, interprismatic enamel is absent.*

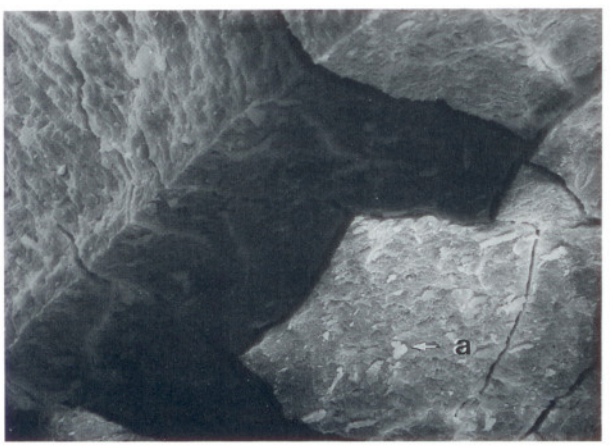

Figure 6. *T = 450° (x500): Crazed internal face of the enamel shell showing adhesion of dentine (a).*

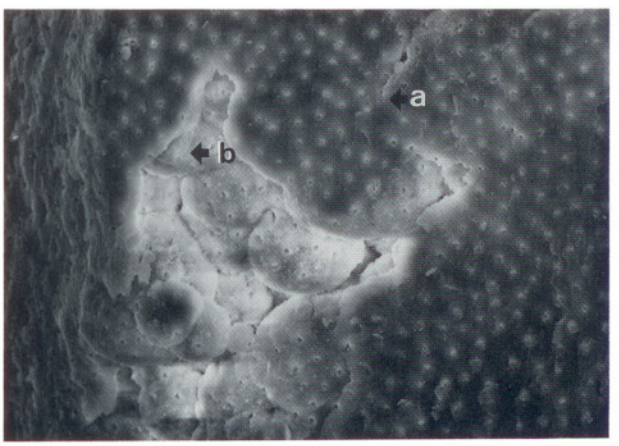

Figure 9. *T = 500° (x500): The crown area dentine is crazed on its surface (a) and displays zones where the dentinal tissues have been torn off (b).*

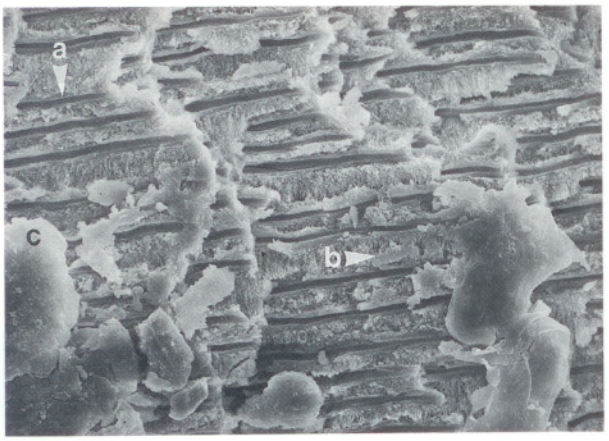

Figure 10. *T = 600° (x1000): The tubules are parallel to each other. Peritubular (a) and intercanalicular (b) dentine have a normal structure. Aggregations of fusional debris are present (c).*

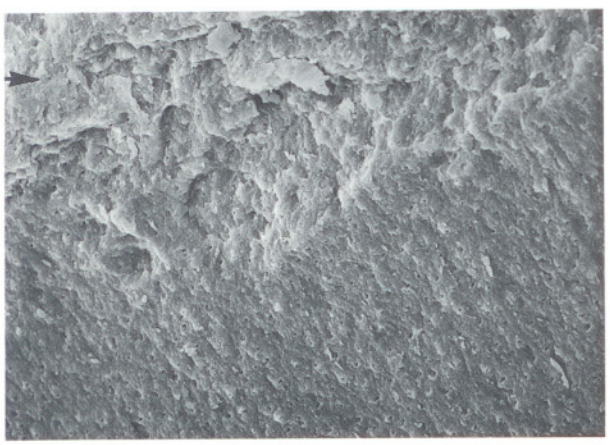

Figure 11. *T = 700° (x500): The dentine close to the occluding surface displays tubules of a smaller diameter (arrowed).*

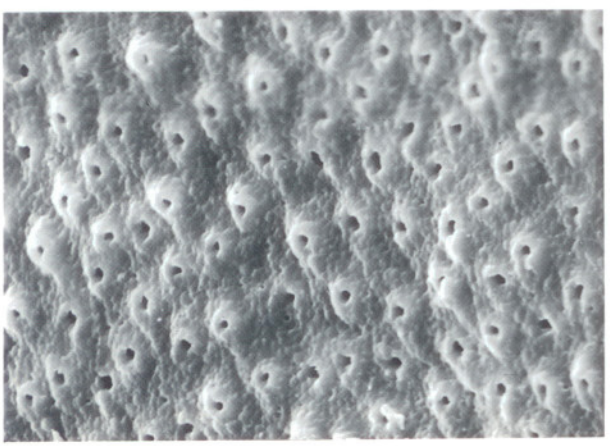

Figure 12. *T = 1100° (x2000): Tubular diameter is reduced as seen in crosssection.*

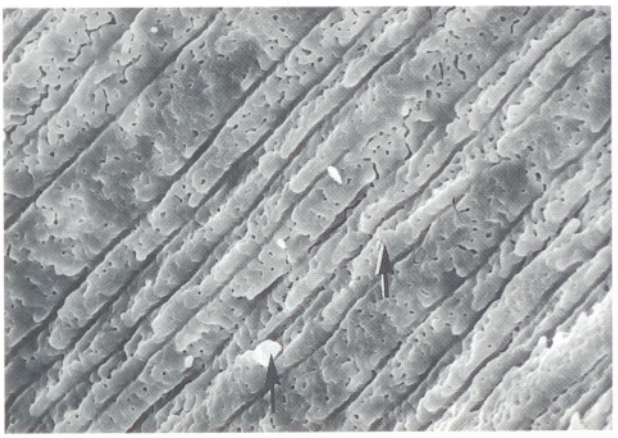

Figure 13. *T = 1150° (x1200): In longitudinal section the tubules are covered with matter in the form of granules, grains or a coating layer (arrowed).*

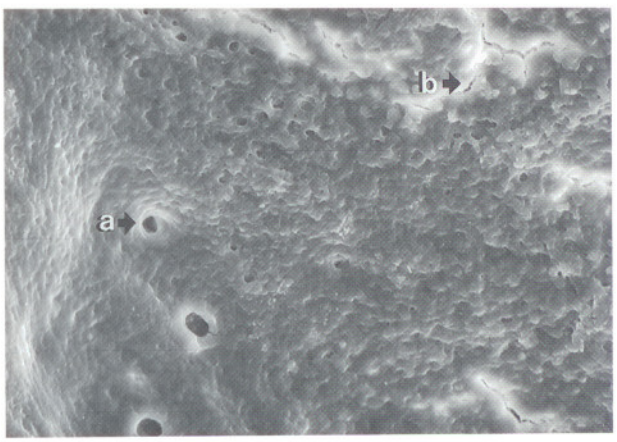

Figure 14. *T = 150° (x500): The cellular cement, situated close to the apex, displays a particular orientation of the layers; it suggests a centrifugal mineralistion of the extrinsic collagenic fibrillae which would be included in this case in the cement of the matrix. A few foramina (a), with a diameter of 5 to 10μ, are present as crazing develops (b).*

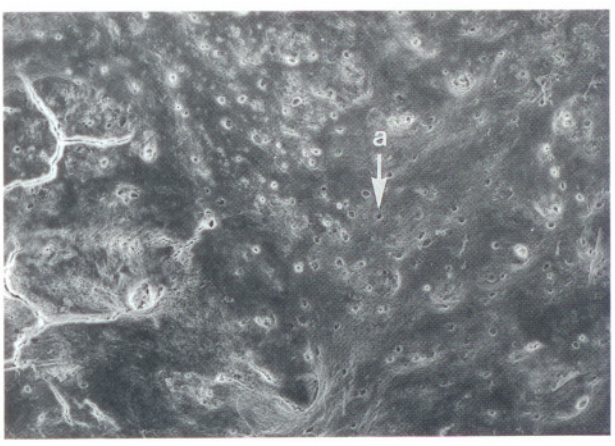

Figure 15. *T = 600° (x100): In some zones at the level of the apical third, the cement has disappeared to reveal dentinal tubules (a).*

3. Microscopic aspect of the dentine

The separation of the enamel shell at 450°C revealed a slightly crazed dentine. Dentinal tear-off zones were observed (Fig.9) and at 600°C the presence of aggregations of fusional debris was noted covering the tubules of the radicular dentine adjacent to some fracture lines (Fig.10). At 700°C a reduction in the diameter of the tubules, which was a sign of an elevated temperature was observed (Fig.11) and was concomitant with an increased number of granules, which gave a molten aspect to the structures (Figs.12 and 13).

4. Microscopic aspect of the cementum

Cracks appeared initially at 150°C (Fig.14) and they multiplied rapidly with the rise in temperature. At 600°C some of them separated (Fig.16). Cementum located at the level of the apical third seemed to melt, leading to the previously observed appearance of the underlying dentine (Fig.15). Some of the teeth continued to crack near the cemento-enamel junction, leading to a honeycomb appearance of these zones (Fig.17). At 100°C, the few residual zones seemed to granulate under the effect of the heat and could no longer be recognized (Fig.18).

DISCUSSION

The teeth used in the experiments described should all be of the same type in order to enable comparison between observations at different temperatures. Our choice was thus to use premolars which had to be extracted as part of orthodontic treatment and also because they were free from decay. Further, this choice appears appropriate as premolars belong to the pluriradicular group and are better preserved in a fire than incisor teeth, which are rapidly destroyed because of lip withdrawal.[10] The time needed to cremate human remains has been determined to be 60 mins and this was utilised to study burnt bones at different temperatures under SEM.[11] These authors selected this period of time as it also equates with the average duration of a fire[1] and furthermore, corresponds with the time required to incinerate an adult subject in a cremation at a temperature of 800°C.[3] It is important to remember that temperatures are highly variable depending on the type and the location of the fire (open or closed), the nature of the oxidant and the combustion medium but temperatures reach 1000°C and over only at the centre of the fire.[3] In our experiments we placed the teeth directly into the furnace at the desired temperature in order to subject them to a thermal shock similar to that created by a fire.

The enamel reacts to heat at 150°C by crazing. This consistent observation, as already described, is on the other hand not very significant.[6] Observing the colour is more difficult as it can sometimes require the careful preliminary cleaning of the tooth found at the site of a fire. The presence of a superficial metallic layer, resulting from the dry distillation of organic matter, can lead to observational errors. After one hour's heating at 150°C the colour of the crown was unchanged, contrary to that of the root which appeared slightly darker. A tooth burnt at this temperature thus appears normal, but then it passes through different shades of brown. More rapidly acquired

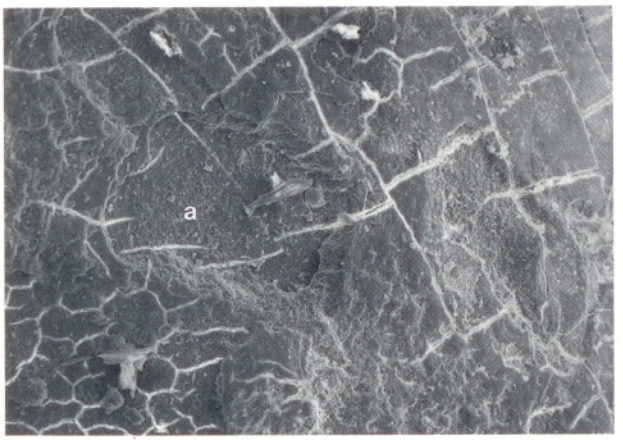

Figure 16. *T = 700° (x100): The cement crazed and in some zones reveals the underlying dentine (a). At the boundaries of these zones the cement has a variable thickness, from 20 to 50µ.*

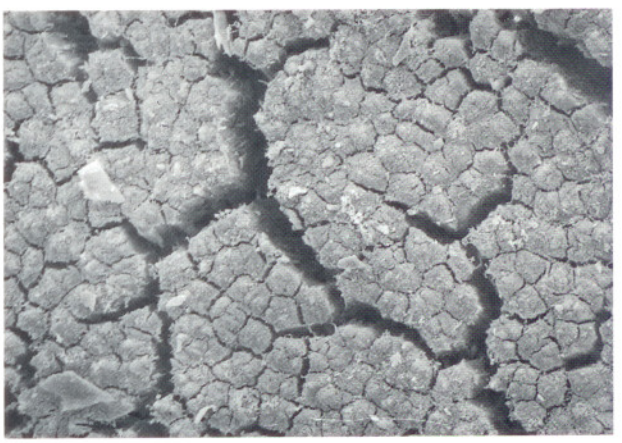

Figure 17. *T = 900° (x1000): This hexagonal "honeycomb" structure makes the cement difficult to recognise.*

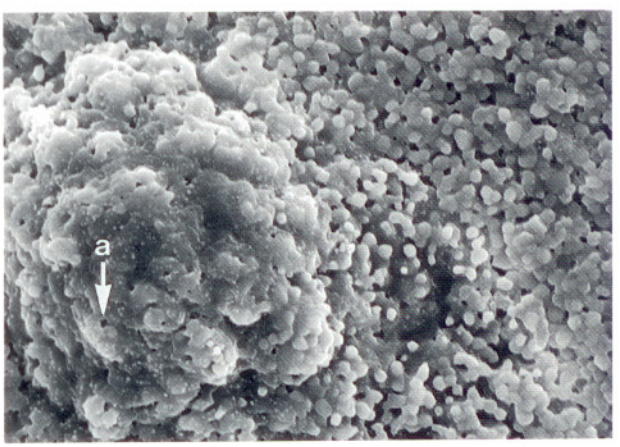

Figure 18. *T = 1150° (x2000): The cement can no longer be identified in the presence of these vesicular shaped granules. A few smaller diameter tubules are visible (a).*

and consistent at the level of the root, this discolouration appears initially in the form of spots near the neck before covering the whole surface of the enamel at 400°C. Present observations are thus different from those who describe a grey enamel even if the teeth displayed grey shades at 300°C.[6-8] The sequence in which these spots developed also leads to the speculation that this could be accompanied by a multiplication of crazing patterns initially located close to the cemento-enamel junction at 300°C, before increasing in extent at a distance from this junction in accordance with the elevation in temperature. The calcinated aspect already described at 300-400°C is specific to the root[6,7] and in association with the presence of numerous small holes, suggests a significant destruction of the cement.

Dental tissues seem to react differently in the course of heating. In the same way, the separation of the enamel shell between 400°C and 500°C could be explained by a lower rate of shrinking under the effect of heat on enamel, the mineral phase of which, at 96-98%, is the most significant factor in its reaction. In contrast, the dentine would shrink more because of its water content of 12%, which on evaporation[12] would bring a force to bear on the enamel and cause it to crack more and faster in its thinner zone close to the neck. Botha[1] explains this phenomenon as the boiling and vapourisation of the 8 to 10% of water contained in the tubules, which would then separate from the enamel. It was during the second stage, when different teeth were heated at temperatures between 400°C and 500°C, that the separation of the enamel at 450°C was able to be observed. Results thus confirm the findings made by some authors at 500°C.[9,13] Conversely, they differ from those obtained by other researchers at 300-400°C[6] or 500-700°C,[8] in the first case where it was found that exposing the teeth to a gradual rise in temperature could explain this difference while in the second, only a different tooth storage method after extraction and/or the smaller number of temperatures used could be significant, the teeth otherwise having been subjected to the same experimental conditions. Another explanation, that it was caused by the lower mineralisation of the teeth extracted for orthodontic reasons in young subjects, seems to be unlikely. Once isolated, the enamel broke down into consistently smaller and more crazed pieces, in line with the elevation in temperature, its grey shade remaining unchanged. The coronal dentine, which has been stripped, crazed rapidly and the colour changed from grey to white at 800°C after passing through bluish shades. After the disappearance of the calcinated and bubbly phase,[6] at 300°C - 400°C, the root took on a crazed aspect, initially at the level of the neck and then, at 800°C, down to the apex where it had a slightly granular appearance.[6] At 600°C the root was almost the same colour as the crown, but changed to a bluish colour up to 800°C.[6,7] After that the root differed, became grey-black and then developed pink patches between 800°C and 1100°C. In this respect, the present observations were different from other studies[8,9] where a totally white root at 900°C was described. In contrast, they refer to descriptions of a pink tinge in the white tooth at 1000°C - 1100°C.[6,7]

The root separates from the crown and breaks down into more numerous fragments with the rise in temperature. We were thus able to confirm that teeth fragment into larger and then smaller fragments at temperatures of 900°C and 1100°C respectively, the small pieces originating in the crown.[8] It is not unusual to find teeth in the ash from a fire which only consist of their roots under an open pulp chamber surrounded by irregular dentine, to which a few enamel fragments adhere.[7] On the other hand, we do not agree with the claim that teeth severely burnt to high temperatures burst, unless they have been totally dehydrated.[2] It should be borne in mind that the experimental teeth used in this study were stored in water.

Our observations did not confirm those of Harsanyi[8] who saw no modification in structure up to 300°C. At 150°C a superficial erosion of the enamel was observed, probably resulting from the thermal shock (Fig.2) and was not due to ageing because the teeth were all from young subjects. Furthermore, the enamel started to crack at the same temperature, the cracks appeared to multiply from the cemento-enamel junction (Fig.3) and at 400°C, the crown had a chequered appearance (Fig.4).[8] On the other hand, we also observed zones with a molten configuration at 300°C and 400°C which are not referred to by other investigators (Fig.5).[6,7,9] It is supposed that they correspond with areas covered by soft tissues torn off in the course of extraction and then burnt. With the elevation in temperature, they would have been completely calcinated and would disappear as the dimensions of these zones diminish. At 450°C the enamel crown separated from the dentine core and this enabled us to observe its juxtadentinal surface, which was found also to be cracked. It was interesting to note that the contraction of the enamel shell drew on the dentinal tissue (Figs 6 and 10) while the prismatic structure of the enamel remained perfectly identifiable up to 1100°C (Fig.7). Above this temperature it remained recognisable despite its molten aspect (Fig.8) and these results therefore did not confirm those of Harsanyi,[8] who stated that at 700°C - 900°C the enamel structure could no longer be differentiated because of granular formations, which melted rapidly and led to a "liquified" appearance.

With the loss of the enamel shell, the coronal dentine, which was no longer protected, immediately crazed (Fig.9). This reaction of the dentinal tissue to heat was however limited and the number of crazing patterns remained small, despite their increase related to that of the temperature. Contrary to the behaviour of enamel or cementum, therefore, dentine would never acquire a chequered appearance and its structure would remain protected, as has already been noted.[8] These results thus differ from those studies[9,14] which had utilised a method now considered to be inaccurate. At 600°C the dentine was sometimes covered with accumulations of fusional debris close to the fracture lines, which indicated that fusion would come before the fracture unless the dentine melted on the surface at the level of the fracture line (Fig.10).

At higher temperature the onset of a reduction in the diameter of the tubular openings was observed,

confirming Harsanyi's findings that at 700°C smaller dentinal tubules were present (Fig.11).[8] This reduction in size of the tubules would be progressive up to 1150°C, and seems to have been related to the development of the numerous granules which reached a diameter of 1 to 2 µ (Figs.12 and 13). These have already been observed[8] at 1000°C, when their diameter did not exceed 0.1 to 0.2 µ. The shrinking of the tubules could thus have resulted from an increase in the number and size of the granules as the temperature rose. Despite both granulating and melting, and regardless of the temperature reached, the structure of dentine remained readily identifiable. The retention of its tubular structure at 1150°C was probably due to the presence of hypermineralised pericanalicular dentine (Fig.13).

In this study, where the root was directly exposed to the heat source without protection from the bone, the cementum cracked quickly (Fig.14), giving the root a chequered appearance. Because the root decreases in size progressively nearer to the apex the effect of heat at 600°C was different. Either it appeared to melt when the apical third was observed (Fig.15), or it separated when located in the coronal two thirds, thus showing a "gnawed" appearance (Fig.16). In both cases, however the dentinal tubular orifices were exposed at temperature levels of 300°C and above.[8] These observations were explained as caused by the evaporation of water and the consequent lifting of the cementum from the dentine. Not all these cementum fragments separated at 600°C, however. From that point on, they continued to crack and acquired a honeycomb/hexagonal structure which made the identification of cementum very difficult at 900°C (Fig.17). Granules also built up on the surface at 800°C which had been previously observed at 700°C.[8] With the increase in temperature, they will multiply and increase their volume, reaching a diameter of about 1µm at 1150°C, and consequently make it impossible to identify the cementum (Fig.18).

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DISCRIMINATION BETWEEN RESTORATIVE DENTAL MATERIALS BY THEIR RADIOPACITY MEASURED IN FILM RADIOGRAPHS AND DIGITAL IMAGES

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ABSTRACT

The aim of this study was to investigate the possibility of differentiating between various dental restorative materials by means of their radiopacity. Ten extracted molars and ten canines/anterior teeth were selected for the study. In the molar group a class II cavity and in the canines/anterior teeth group a class III cavity were cut by airrotor. The cavities were coated with vaseline before filling with five molar- and three anterior tooth restorative materials in the following sequence: for molars: amalgam, light-cured composite, glass ionomer cement, reinforced glass ionomer cement and light-cured composite. After each filling sequence radiographs were taken of the teeth on conventional film (Ektaspeed Plus) and by two digital systems: a storage phosphor plate (Digora) and a ccd-based sensor (Sidexis). Density was measured in the films with a densitometer in three areas of "air", in three areas of the class II fillings and one area of the class III fillings. The same areas were measured in the digital images where the program calculated automatically the mean grey shade values. The density values obtained from the filling areas were taken as a proportion of the values obtained from the areas of "air". Statistically significant differences in material density when related to "air" density (Wilcoxon's test) were observed between all materials in film ($p<0.01$ for molars and $p<0.02$ for canines/anterior teeth). For Digora only half of the materials differed significantly which was also the case for the Sidexis system (none of the CF materials were significantly different). In conclusion, the molar filling materials could be distinguished with a high probability in film while the two digital systems were less reliable. The results may be useful in forensic dentistry. **(J Forensic Odontostomatol 1998; 16: 8-13)**

Keywords: Dental materials, radiography, dental, computer-assisted, forensic dentistry

INTRODUCTION

The change in dental filling materials from amalgam to composites and glass ionomers has resulted in a change in the radiopacity (radiodensity) of restorations. At the beginning of the composite era resins could not be clearly recognized in dental radiographs as observers could not distinguish between such materials and empty cavities.[1] Of 18 composites tested, only one was more radiopaque than enamel[2] and it was suggested that heavy salts be added to the composite and glass ionomer materials in order to enhance their radiopacity.[1] In recent years manufacturers of composites have increased the radiopacity of the materials in order to make them more radiopaque than dentine or even enamel.[3-5]

For glass ionomer cements it has been demonstrated that cements which were less radiopaque than dentine should not be used as bases or liners under dental restorations,[6] and as a consequence many of the more recent resin-modified glass ionomers now possess a radiopacity higher than enamel.[7] The glass ionomer cements are not a homogeneous group and therefore the radiopacity varies depending on the added component (silver alloy, zinc, strontium, barium).[7]

The radiopacity of filling materials has been of interest to clinicians mainly in relation to diagnosis of secondary caries and marginal defects.[8-10] In forensic work comparison between ante- and post-mortem radiographic images of these restorations plays a significant role in determination of identity[11] but during simulated ante- and post-mortem radiographic matching exercises in non-treated individuals a number of errors were made.[12-14] The presence of traditional dental restorations minimizes the risk of mismatching radiographs but a decline in dental caries occurring over the last twenty years especially in the younger age groups[15] has brought about a rapid decrease in the number of complex restorations and this will decline further in future populations. If the types of materials used for restorations could be determined from post-mortem radiographs it would be an additional aid in identification and moreover where ante-mortem radiographs may not always be available the documentation of tooth fillings is usually to be found in the patient's dental record. It may therefore be of interest to investigate the possibility, on the basis of one radiograph of a restored tooth, of determining which dental material was used for the restoration.

In recent years direct digital intraoral radiography has found its way into dental practice.[16] The new image receptors can be either the ccd-based (charge-coupled-device) sensor or the stimulable phosphor plate,[17] and they differ in such aspects as their dynamic range (dose in relation to density). The radiopacity of dental materials has never been investigated using digital image receptors.

It has been the aim of this study to evaluate the radiopacity of some common dental filling materials in class II and class III cavities using a conventional intraoral radiographic film, a storage phosphor plate and a ccd-based sensor as the image receptor, and further, to investigate the possibility of discrimination solely on the basis of radiopacity of the materials in a simulated clinical situation.

Material	Product	Manufacturer	Filling	Symbol
non-2 amalgam	Dispersalloy Konstanz, Germany	Dentsply, De Trey,	Class II	A
light-cured composite	Herculite	KERR Corp., Orange, CA, USA	Class II	B
glass ionomer cement	Ketac-Molar	ESPE, Dental-Medizin, Seefeld, Germany	Class II	C
"re-inforced" glass ionomer cement	Miracle-Mix	G.C. Industrial Corp., Tokyo, Japan	Class II	D
chemically cured composite	P-10	3M Dental Corp., St. Paul, MN, USA	Class II	E
light-cured glass ionomer cement	Fuji II LC	G.C. Industrial Corp., Tokyo, Japan	Class III	F
light-cured glass ionomer cement	Photac-Fil Quick	ESPE, Dental-Medizin, Seefeld, Germany	Class III	G
light-cured composite	Herculite CA, USA	KERR Corp., Orange,	Class III	H

Table 1. *Materials used in Class II and Class III fillings*

MATERIALS AND METHODS

Ten clinically intact, extracted human molars and ten anterior teeth (6 canines, 4 incisors) were included in the study.

In the molars a class II (MOD) cavity and in the anterior teeth a class III cavity were cut by airrotor. The teeth were filled with different restorative materials and in order to facilitate replacement of the materials the cavities were coated with vaseline before filling. Five different materials were used in the molars and three in the anteriors (Table 1) in the following sequence: molars: amalgam, Dispersalloy*, light-cured composite, Herculite (class II)*, glass ionomer cement, Ketac-Molar* reinforced glass ionomer cement, Miracle-Mix*, and chemically cured composite, P-10*; anterior teeth: light-cured glass ionomer cement, Fuji II LC* and Photac-fil Quick* and light-cured composite Herculite (classIII)*. The symbols A-H were used to identify the materials (Table 1) and changing the filling materials was done manually without the use of the airrotor to avoid the risk for removing additional tooth substance.

After placing each material radiographs were taken of the teeth (rectangular tube, 70kV, 7mA, soft tissue scatter 12mm acrylic) on conventional film (Ektaspeed Plus)** and by two digital systems: a storage phosphor plate (Digor)*** and a ccd-based sensor (Sidexis)#. For the two digital systems exposure time was approximately 20% of that for film. The teeth were positioned in direct contact with the receptor with the position of the tooth and the focus-film distance consistent before exposure. The conventional film was developed in an automatic developer (Dürr 1330 ac 245L)## in fresh chemicals and the phosphor plate was scanned in the dedicated Digora scanner.[17] For the Sidexis system, there is no image development as the x-radiation is converted directly to digital values and transferred from the sensor to a computer. One image in the Photac-Fil Quick material group taken with the Sidexis system was lost due to an error in the recording. In both digital radiography systems the image was displayed on the PC monitor.

Radiopacity of the materials in the films was measured with a densitometer (RM22)### in three areas outside the tooth, and in three areas within the filling for the molars and in one area within the filling for the anterior teeth. Tracings of the teeth with the fillings as seen in the films were made on a transparent sheet and were then photocopied in a magnification that fitted the size of the digital images as displayed on the monitor. This procedure ensured that the same areas outside the tooth and in fillings in the same tooth were measured in the films and in the digital images. The measurements by densitometer were performed three times in each area in each film and the means of the readings calculated and used in further data treatment.

For the digital images, the program automatically calculated the mean grey shade values in the areas which were drawn with a mouse directly on the images, on the monitor, using a dedicated image analysis program (wdens)+. A digital image consists of pixels (picture elements) which are aligned in rows and columns constituting the matrix of the image, and each pixel is

* Names of manufacturers and addresses may be found in Table 1.
** Eastman Kodak Corporation, Rochester, NY
*** Soredex, Orion Corporation, Helsinki, Finland
\# Siemens, Bensheim, Germany
Bietigheim-Bissingen, Germany
Helioprint AS, Denmark
\+ Torben Jørgensen, Lystrup, Aarhus, Denmark

defined by a value corresponding to a particular shade of grey.[18] The number of grey shades available in the digital image is given by the number of binary digits (bits) used to define a pixel. This is termed the bit depth resolution and the difference between the highest and the lowest grey shade available determines the image contrast. In intraoral digital radiography systems today the bit depth is eight bits which equals 256 possible shades of grey and the darkest grey shade is usually defined by the value zero while the lightest has a value of 255. In opposition to density measures in conventional films the filling areas will thus register higher values than the dark "air" areas in the digital images.

Data treatment

All measurements were entered into a statistical program (SPSS for Windows) and the median and range of the density in filling and in "air" were calculated in each material group. The density values obtained from the filling areas were thereafter calculated as a percentage of the values obtained in "air" for film (filling/air x 100) and opposite for the digital images (air/filling x 100). Differences between the filling materials using this percentage were tested by Wilcoxon's signed rank test for molars and anterior teeth separately. Differences between the materials were tested within each radiographic modality, but not between the three. Probability values <0.05 were considered statistically significant.

RESULTS

The medians and ranges in density for all materials can be seen in Table 2 together with the densities in "air". There was some variation in the overall density of the film radiographs as the median density values in "air" ranged from 1.8 to 2.7. Similarly, there was a difference in background density in the Digora images, the median grey shades in "air" ranging from grey value 41 to 82 while the Sidexis images were more homogeneous in density with grey values in "air" ranging from 17 to 21 (Table 2).

The median and range in densities for all materials as a percentage of the density in "air" are illustrated in Table 3 and Fig.1 for the three radiographic modalities. For the molar materials in film, amalgam had the highest density followed by Herculite, Miracle-Mix, P-10 and Ketac-Molar which had the lowest. For the anterior teeth Herculite had the highest density followed by Fuji II LC and Photac-Fil Quick and with the Digora, Ketac-Molar had a higher density than P-10. With the Sidexis system, there were only small differences between the densities of the materials. The density differences were statistically significant between all materials measured in film ($p<0.01$ for molars and $p<0.02$ for anterior teeth). Using the Digora system, amalgam was not significantly different from Herculite, Ketac-Molar not different from Miracle-Mix in the molars and Fuji II LC was not different from Herculite in the anterior teeth ($p<0.05$). Using the Sidexis

		A		B		C		D		E		F		G		H	
		fil	"air"	fil	"air"	fil	"air"	fil	"air"	fil	"air"	fil	"air"	fil	"air"	fil	"air"
	min	0.22	2.3	0.39	2.3	0.62	1.7	0.49	2.3	0.88	2.6	0.70	1.9	0.60	1.6	0.62	2.4
Film	**med**	0.37	2.4	0.45	2.4	0.70	1.8	0.54	2.4	1.02	2.7	0.83	2.4	0.78	1.8	0.71	2.5
	max	0.42	2.6	0.79	2.5	0.78	1.8	0.59	2.6	1.08	2.9	1.07	2.6	0.91	1.8	1.03	2.6
	min	231	29	225	33	223	45	236	69	196	62	185	36	200	48	192	36
Digora	**med**	239	41	231	46	231	76	242	82	202	74	209	54	221	76	211	46
	max	243	60	236	93	236	80	244	88	212	85	225	57	225	90	216	67
	min	231	15	211	17	191	17	222	16	176	18	153	19	158	19	140	15
Sidexis	**med**	233	17	222	19	213	19	228	17	187	19	205	21	219	21	206	19
	max	234	18	225	222	224	25	233	22	200	21	220	24	225	32	219	20

Table 2. *Median, minimum and maximum values for radiodensity of the filling materials (A-H) and of "air" measured by three radiographic modalities (Symbols A-H, see Table 1).*

		A	B	C	D	E	F	G	H
	min	9.1	16.8	36.5	21.1	32.8	27.4	37.0	24.2
Film	**med**	15.2	19.5	40.0	22.5	36.8	35.4	46.8	30.7
	max	17.2	22.4	42.8	25.0	38.7	42.7	52.0	40.8
	min	12.4	14.2	20.2	28.8	31.6	17.6	22.3	17.0
Digora	**med**	18.1	20.6	33.0	34.6	37.3	26.5	35.4	22.4
	max	26.1	39.6	34.5	37.4	43.0	29.1	40.7	31.3
	min	6.5	7.5	7.8	7.0	9.1	9.4	8.6	7.5
Sidexis	**med**	7.3	8.6	9.0	7.7	10.1	10.4	10.6	9.6
	max	7.9	10.0	13.5	9.4	11.5	12.7	30.0	13.8

Table 3. *Median and range for the density of fillings as a percentage of "air" for the 5 materials in molars (A-E) and the 3 in canines/front teeth (F-H) (Symbols A-H, see Table 1).*

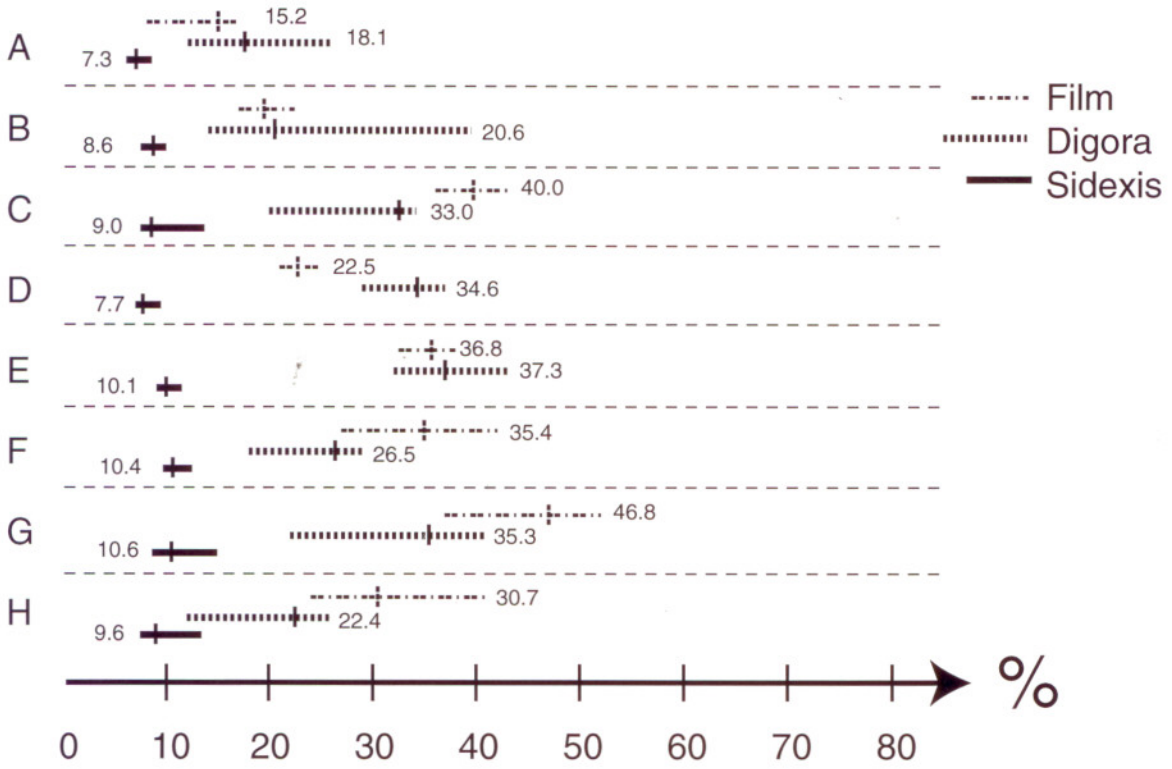

Figure 1.: *Distribution with median values of the density of fillings as a percentage of "air" for the materials tested with three radiographic modalities (Symbols A-H, see Table 1.)*

system, amalgam and Miracle-Mix were not significantly different and neither were Herculite and Ketac-Molar, nor Ketac-Molar and P-10 ($p<0.1$). None of the filling materials in anterior teeth were significantly different.

DISCUSSION

In the true forensic situation dental restorations may constitute an important basis for victim identification. A post-mortem radiograph can often be taken of the teeth and may be compared to an ante-mortem radiograph of a suspected victim. If only simple tooth restorations, or no restorations are present, the matching of such radiographs is fraught with errors[12,14] and furthermore there may not necessarily be an ante-mortem radiograph. A dental chart of a suspected victim will, on the other hand, most frequently provide information on which filling material (amalgam, composite, glass ionomer) has been used for a specific restoration. Forensic work in cases of drowning for example, might be assisted if post-mortem radiographs were able to establish the identity of the material with a high probability. Heat induced colour changes of composite and glass ionomer materials may also be able to establish their identity, but this technique is mainly applicable to fire victims.[19]

In order to simulate the real life situation as closely as possible in this study, the density of the filling materials was calculated in absolute values and as a percentage of the density values in the "air" part of the same radiograph. Previous calculations of density of materials in relation to density of a known reference for example 1mm of dentine or aluminium in laboratory experiments,[20-22] were aimed usually at investigating the radiopacity of the material in relation to caries or restoration defects of enamel or dentine.

In this study it was attempted to approximate the overall density in the film radiographs to that in "air", and which should be within the usual diagnostic area for radiograph density. As in the clinical situation there were, however, variations in overall density in the individual radiographs which were taken into account by assuming the density of the filling as a percentage of the density in "air".

For the molar filling materials in this study it was interesting to note that amalgam, composites (Herculite, P-10) and glass ionomers (Miracle-Mix, Ketac-Molar) could be differentiated in film with a high probability, as only very little overlap was seen between their radiodensity distributions. More overlap was observed in the materials used for the front teeth. The materials in the anterior teeth could thus not be differentiated with as high a probability as those in the molars.

For the digital systems the differences between the distributions of the grey shade values for the various materials were smaller than for film. In particular the ccd-based sensor produced images of quite similar density within the fillings irrespective of the material. The digital systems operate with an absolute (discrete) number of available grey shades (256) in contrast to the continuous density curve in the analog film image and therefore the limited number of grey shades in every exposure will be sought used. Though the human eye is not capable of discriminating small differences in the analog density curve an objective quantification of the density level as is carried out by a densitometer can be expected to record such differences.

The digital systems have previously been demonstrated to be as accurate as film for detection of primary caries,[23-25]

but in only one study so far has the accuracy for diagnosing caries under various filling materials been investigated.[26] The advantages of the digital systems are, in addition to immediate image capture and avoidance of processing chemicals, their wide dynamic range and their increased sensitivity to radiation exposure.[17] A widespread use of these systems may be expected in the future.

In conclusion, molar filling materials (amalgam, composites and glass ionomers) could be differentiated with a high probability, and anterior tooth materials with somewhat lower probability, in conventional film radiographs while the two digital systems were not valuable in differentiating the tested materials. Conventional film radiography should therefore be preferred over digital radiography for the assessment of radiodensity of filling materials and the identification of a filling material from a radiograph may be used as an adjunct to other methods in borderline cases in forensic dentistry. The present *in vitro* experiment warrants further *in vivo* studies but continuous changes in the composition of restorative materials may reduce the differentiating capacity of the present method. However, it is significant, in spite of the usefulness of radiographs, that dentists keep their patient records as detailed as possible, recording not only the type of filling material, but also the brand.

ACKNOWLEDGEMENTS

Ali Said and Davood Rezai are thanked for filling the teeth and taking the radiographs.

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AN INCLUSION TECHNIQUE FOR MARKING DENTURES

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ABSTRACT

The importance of placing identification markers in dentures is well documented and this paper describes a simple and inexpensive technique for doing it. Twenty marked upper acrylic simulated dentures and 20 controls (unlabelled) were constructed in a standardized mould. Clear laminated labels were produced in a P-touch 300 electronic lettering system and contained both the patient's identification number and suffix ZA (international code for South Africa). The markings were both clear and aesthetically acceptable. Strength tests were carried out to establish the effect of the mark with the result that no significant difference (p70.05) was demonstrated. The routine marking of all dentures by this method is advocated. **(J Forensic Odontostomatol 1998; 16: 14-6)**

Keywords: denture marking, inclusion technique, compressive strength, South Africa.

INTRODUCTION

Over the years, many different denture marking systems have been described. For simplicity, they are divided into two systems: surface identifiers and inclusion methods.[1] Distinctions are also made between markings placed during and after denture fabrication.[1,2] The use of the patient's national identity number plus letters indicating the country of origin e.g. ZA, instead of inserting the patient's name, were first described by Thomas.[3] Berry *et al.*[2] compared various materials and methods used for denture identification: engraved or stamped metal or acrylic strips, discs, simple engraving of denture base, microchips and printed paper strips. The above authors recommended a postfabrication technique in which laser printed white paper labels were inserted into any flat, aesthetically acceptable position in the denture. A technique in which soft rolled metal bands were buried in fabricated dentures has been described[4,10] which is aesthetically superior to other inclusion methods but is of no value in day-to-day identification of dentures as the marking is not visible externally. A T-shaped clear resin bar has been used[5] as a method to place identification markers in existing removable dental prostheses, its advantage being that all dentures, both new and used, can be marked, although the technique is cumbersome. Ibrahim[6] was able to include multiple items of information to make up a graphic image which was then photographed by polaroid, but the marking was only inserted after completion of the dentures.

Routine marking of acrylic dentures does not take place in most private practices and government clinics in South Africa. A survey of 23 laboratories and 14 dental surgeries, in seven of the nine provinces of South Africa, showed that no routine marking of dentures takes place. With the increase in crime and air disasters, as well as the awareness of cross infection in institutions, the need for denture marking has increased. It is important for the forensic identification of corpses,[1-9] identification of dentures in the laboratory and in the dental practice, identification of lost dentures, and of dentures in hospitals and other institutions for the aged and mentally handicapped.[1,7,8] The expenditure incurred in placing denture identification markers is extremely low compared to the cost of replacing a lost denture or the loss of quality of life associated with being dentureless.[7]

The aim of this study was to develop a technique for marking dentures which would contain the patient's national identification number plus an internationally accepted code suffix, and furthermore to develop an aesthetically acceptable label, technically easy to insert, inexpensive for the patient and one which would not weaken the denture in any way.

MATERIALS AND METHODS

The labels were produced in a P-touch 300 electronic lettering system* (Fig.1). Clear laminated tape with 2mm letters was used and a thirteen digit patient identification number plus the suffix ZA (international code for South Africa) was printed onto the labels. The laminated tape is 103µm thick and consists of six layers, an outer polyethylene protective layer, a layer of adhesive acrylic into which the ink is imbedded, a base film of polyethylene, a colouring base, another acrylic layer and

***Figure 1:** The labels were produced in a P-touch 300 electronic lettering system.*

* Brother, U.K.

	Marked Group	Unmarked Group
Mean	1509.18N	1437.49N
Standard deviation	206.66	218.58
Coefficient of variation	13.69%	8.59%

Table 1. *The mean, standard deviation and coefficient of variation of the compressive strengths measured in Newton (N).*

an outer separator which is removed before use. The tape is non-toxic, fade-resistant and will retain its properties up to a temperature of 365°C.[10] Twenty Royale denture base polymer acrylic** simulated dentures containing identification labels, and 20 unmarked dentures were manufactured by a private dental laboratory*** from a single mould. The investing and processing was done according to conventional methods, including the use of a separating layer of plastic for "trial packing". After "trial packing" the flasks were opened, the separating plastic removed and the labels placed 3mm anterior and parallel to the post-dams marked on the casts. The labels were covered by a small amount of acrylic which had been set aside specifically for this purpose. The flasks were closed slowly to prevent label movement, curing carried out and the dentures were de-flasked, finished and polished. An important property of any denture mark should be that it not weaken the denture. The 40 simulated dentures were therefore subjected to a crushing test in order to compare the strengths of the 20 labelled and 20 unlabelled specimens. Compression testing equipment (Zwick Z1010)# was used to break the dentures sagitally, across the mark, and the mean values of compressive strengths statistically analysed by Student's t test for uncorrelated data.

RESULTS

Analysis of the compressive strength tests is shown in Table 1 and the Student's t test revealed that the mean compressive strengths of the two groups did not differ significantly (p=0.347). The cost of marking a denture was inexpensive (less than ten South African Rand/two US Dollars), and technically simple and quick to implement. The markings were clearly visible.

DISCUSSION

In the past high costs and lengthy techniques have deterred dentists and technicians from placing patient identification markers in dentures. On the other hand, the technique described in this report is cost effective, quick and easy to implement and in contrast to Dippenaar's method,[4] the marks resulting from this method are readily visible to the unassisted eye, and can be read by lay personnel e.g. in old age homes on a day-to-day basis. The marking is obvious, and no scanner, ultraviolet light or destructive interference with the denture (drilling) is necessary to expose it. The risk of overlooking a marked denture is thus reduced. Although Ryan's technique[5] is cumbersome, it has the advantage that new and used dentures can be marked.

Although primarily aimed at new dentures the technique described in this study can also be applied when a used denture is relined. As the marking is situated nearer to the fitting surface of the denture, loss of the label due to wear of the acrylic during the normal lifespan of a denture is unlikely.

Compressive strength tests showed no significant difference between the marked and unmarked dentures. The heat resistance of the labels was not tested as it has been repeatedly found that a denture which remains in the mouth during incineration is very well protected[11-13] and any mark which is situated posteriorly will have a good

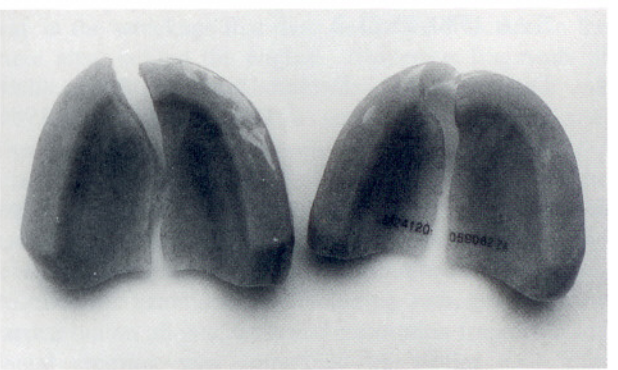

Figure 2: *Marked and unmarked simulated dentures after compressive strength determination.*

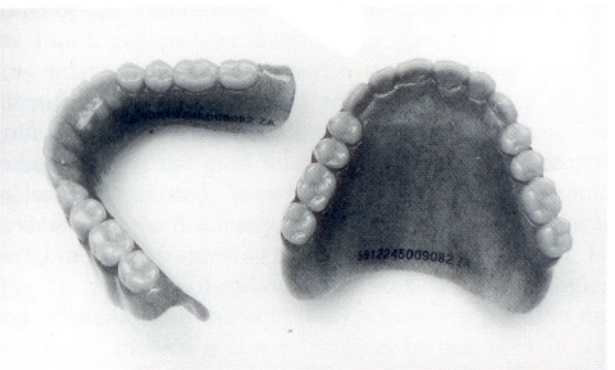

Figure 3: *The aesthetically acceptable result of marking in complete maxillary and mandibular dentures.*

** Meadway Dental Supplies, England
*** Highveld Dental Laboratory, Middelburg, R.S.A.
# Wirsaw Scientific and Precision equipment, Johannesburg, R.S.A.

chance of remaining undamaged. Labels produced by this method have proved to be aesthetically acceptable to patients in the private practice of one of the authors (Fig.3). Routine marking of all new dentures by the described method is advocated. This inclusion technique represents the amalgamation of the best features of several previous techniques, with improvements.

ACKNOWLEDGEMENTS

The authors wish to express their thanks to Professors A.J. Ligthelm and W.F.P. Van Heerden for their support and guidance, Dr. P.J. van Niekerk for initiating the concept, Professor S.J. Botha for carrying out the compressive strength tests and statistical analysis, and Royale Dental Supplies for providing the acrylic for the manufacture of the specimens.

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PERSON IDENTIFICATION BY MEANS OF A SINGLE UNIQUE DENTAL FEATURE

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ABSTRACT

The combination of restorative procedures, developmental or acquired defects, or abnormalities of teeth enables the forensic dentist to make a meaningful comparison of ante- and postmortem data in search of the identity of a person. Dental records are not always sufficiently accurate to supply 12 concordant points from which a positive identification can be made. Under certain circumstances a single feature may be so extraordinary or unique that it alone can be sufficient to make a positive identification. Two cases of identification in which only one unique feature was used are reported. **(J Forensic Odontostomatol 1998; 16: 17-21)**

Keywords: forensic odontology, identification, points of concordance, unusual single features.

OPSOMMING

Die kombinasie van herstellende prosedures, ontwikkelings of verworwe defekte, of ander abnormaliteite stel die forensiese tandarts in staat om by wyse van sinvolle vergelyking van ante- en postmortem gegewens, die identiteit van 'n persoon vas te stel. Onvoldoende tandheelkundige rekordhouding veroorsaak dikwels dat die vereisde 12 punte van ooreenstemming nie verkry kan word nie. 'n Enkele kenmerk mag egter onder sekere omstandighede uitsonderlik of uniek genoeg wees ten einde positiewe identifisering te verseker. Hierdie artikel behandel twee sulke gevalle waar 'n enkele unieke eienskap voldoende was. Die uniekheid van 'n besondere eienskap word bespreek.

INTRODUCTION

A universally accepted minimum number of concordant points for positive dental identification has not been established. In Europe a minimum of 12 concordant points for identification by means of fingerprints has led to the same number of dental characteristics being suggested for dental identification.[1] The South African courts of law however accept seven concordant points for fingerprint identification,[2] but the number of corresponding features for identification remains 12. The criteria for determining which features or characteristics are important for comparison are also subject to interpretation and may vary greatly between examiners. Keiser-Nielsen[1] states that "the aim of comparison is to examine data on the same jaw sector, single tooth or even tooth surface for concordance - a tooth surface being the smallest unit of discrimination".

Quantification of concordant points remains important, and a single feature should not be over-emphasized. However, depending on the circumstances that prevail some authors suggest that a single feature may be unique enough to be solely responsible for positive identification.[3,4]

This paper presents two cases where a single unique feature in each was used to positively identify the individual, and discusses those circumstances or conditions relevant to such a feature.

Case 1:
In 1989 a light aircraft carrying the pilot and three passengers crashed in the mountains near Montague in the South Western Cape, South Africa. On the 19th the forensic investigation was conducted at the crash site by a forensic pathologist accompanied by this co-author (VMP). One of the bodies, only superficially charred and numbered no.1, was found outside the aircraft. While the remains of the other three victims (labelled nos.2,3 and 4) were badly charred and fragmented. As quoted from forensic report: 'These remains were positioned in such a way in the wreckage that their heads were located in the same area, behind the engine. Numerous fragments of skulls, maxillae, mandibulae, teeth, roots of teeth and restorations were found in this small area at the crash site. The fragments were extremely brittle as a result of the intense heat, and were carefully placed in labelled plastic bags". Antemortem records of the four persons alleged to have been on board were collected, were interpreted and re-written onto standardized Antemortem Record Forms. Postmortem examinations, including radiographs, were carried out on all jaw, teeth and restoration fragments and dental charting was completed on victim no.1 after resection of the maxilla and mandible.

Subsequent comparison of the ante- and postmortem data revealed the identity of two of the victims having 23 and 13 concordant points respectively. The third person had some dental restorative work in fragments of jaws while the fourth person was identified by exclusion; the only concordant point in this victim was his blood group. Identification of victim no.3 was by means of a single gold inlay restoration. No antemortem dental records or radiographs of the gold inlay on the upper right central incisor (11) existed, knowledge of which only became evident with the discovery of a radiograph of the cervical vertebrae clearly showing an opaque restoration on the 11 (Fig.1). A series of radiographs taken at various angles and orientations of the gold inlay which was found in the wreckage, showed a shape identical to that seen in the radiograph of the cervical vertebrae (Fig.2). The insurance company in this instance had been reluctant to compensate the widow financially until a positive identification was made.

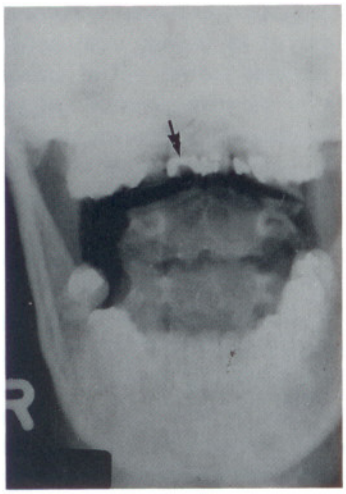

Figure 1: Radiograph of the superior cervical spine of Case 1. Note the shape of the restoration in the right maxillary central incisor (arrow).

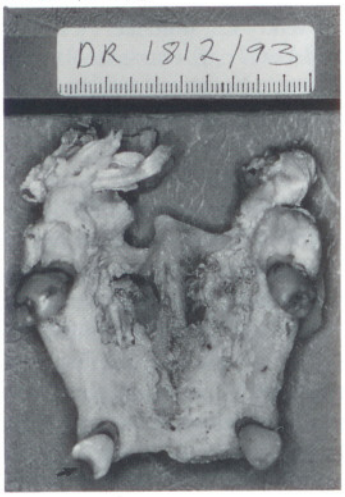

Figure 3.: The resected maxilla of Case 2. Note the worn labial surface of the left canine tooth (arrow).

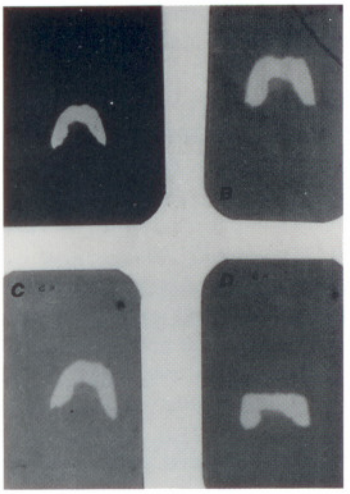

Figure 2.: A series of small radiographs (A-D) showing the variation in the shape of the image of the gold inlay. Radiograph D shows the same morphology as the restoration seen in Figure 1.

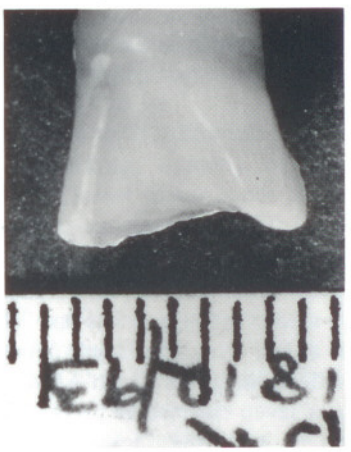

Figure 4.: The labial view of the left maxillary canine showing the unusual attrition of the tooth.

Case 2:

The badly decomposed remains of an adult male were presented for dental identification. No antemortem records were available and postmortem charting of the fractured maxilla and mandible showed four teeth present in the maxilla, i.e. left and right canines, and left and right third molars (Fig.3); eight teeth were present in the mandible, i.e. from left second premolar to right canine. The labial surface of the upper left canine (23) displayed the unusual feature of having undergone severe attrition (Fig.4) as a result of the peculiar class 3 relation with the lower left canine. This tooth (23) was removed from the jaws and shown to the wife of a reported missing person; she immediately recognized it as belonging to her husband.

In this instance the peculiar feature of the canine wear pattern was sufficient to make a positive identification. Although identification was visual and without documented proof, there was no reason to doubt the credibility of the identification. The case was subsequently found to be one of a series of gang murders.

DISCUSSION

The importance of antemortem radiographs enjoys wide acceptance and often may hold the key to positive identification, being the most accurate and reliable of dental records.[1-3,5] Features such as the morphology of a restoration, anatomical landmarks like root formation and bone trabeculae,[6] and all contribute to the bank of concordant points.

The use of radiographs as a visual means to assist not only dentists confronted with a language barrier when dealing with colleagues, but also enables the dentally lay person, i.e. legal expert or police, to form an objective opinion regarding comparison of concordant points.[6] When person identification is done by comparison of dental charting alone, the minimum number of 12 concordant points as suggested by Keiser-Nielsen[1] needs to be found where no obviously extraordinary features exist. In France 17 concordant points are required,[4] as opposed to 12 points in the rest of Europe. The presence of other less frequently found features such as crowns and bridges can

reduce the number of required comparison points to as low as 6 or 7 for identification purposes.[2,7]

In an accident involving a limited number of people whose names are known, the features used for identification could also be limited to a single unique feature for distinguishing one victim from the others. In a mass disaster, however, this "unique feature" may not be extraordinary enough to identify an individual. For example in a motor vehicle accident, the discovery of a gold bridge in the jaw of one of the victims could be unique enough to make a positive identification. This feature may not be of sufficient significance in a major air disaster involving several hundred people, most of whom may have had sophisticated dental treatment.

In making postmortem radiographs it is essential to try to emulate identical projections and angulation of the x-ray tube as used for the antemortem radiographs in order to capture the same morphological characteristics of a particular restoration, root fragment, pulp chamber or bone trabeculae. In case no.1 the gold inlay had to be rotated and radiographed several times before the shape resembled that seen in the antemortem film.

Sufficient postmortem material is essential for comparison with antemortem data and in cases of gross fragmentation as found in high velocity impact fatalities, it is not always possible to obtain tooth and jaw fragments large enough to be readily used for identification. Often no teeth or only fragments of teeth and individual restorations are found.[6,8,9] The completeness of antemortem dental records then becomes extremely important as far as both dental charting and radiographs are concerned. Attention to detail of the radiographic appearance of fillings, pulp morphology, root form, etc. may then produce the distinction needed to compare ante- and postmortem data. The wider the range of antemortem data, the greater the chance of a positive identification even in cases of gross fragmentation. The number of radiographic concordant points required for positive identification have also not been established, but only one identical radiographic characteristic may be sufficient to establish an identity.[4,10,11]

Visual identification by relatives of victims is a contentious issue and can be extremely traumatic in cases of mass disaster.[1] The identification of individual features by relatives may be more reliable if these are viewed away from the mutilated body, as in case no.2 where only the tooth was shown to the wife and viewing the extremely decomposed body could be avoided.

Although 12 concordant points are regarded as the minimum number required for a positive dental identification, there are certain circumstances in which a single extraordinary dental feature may be so unusual that it may suffice for identity purposes. The forensic odontologist must be aware of these circumstances and the potential uniqueness of dental characteristics, and utilize them accordingly.

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RESPONSE TO PAPER BY OHTANI S, YAMADA Y, YAMAMOTO I. AGE ESTIMATION FROM RACEMIZATION RATE USING HEATED TEETH. J Forensic Odontostomatol 1997; 15:9-12

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Keywords: aspartic acid racemization, age estimation, heated teeth, forensic science

In a recent paper Ohtani *et al.*[1] provide encouragement for the wider use of aspartic acid racemization (AAR) as a means of estimating age at death. Previously it has been considered that the method is not applicable to burnt victims, and in an earlier study[2] the authors have demonstrated an accelerated rate in burnt teeth (see Fig.1). In this new study the authors have observed that if only the soluble protein fraction is analysed the rate does not appear to change following heating to 150°C for one hour. From this, the authors conclude "age calculated from the D/L ratio of dentinal soluble peptide from burned bodies is sufficiently accurate for this parameter to be used to estimate age in teeth from burned bodies"[1] (p.9).

This observation makes no kinetic sense. It is well known that the rate of aspartic acid racemization increases with temperature in a predictable fashion (Fig.2). The combination of time and temperature to which a corpse is exposed will lead to extreme variations in the extent of racemization of aspartic acid, asparagine and cyclic succinimides, which together comprise the observed "aspartic acid racemization" (*sic*).[7]

The authors suggest that the rate in the soluble fraction is not accelerated due to (i) dehydration during heating and (ii) an increase in the rate of conversion from D to L-enantiomers. Neither of these explanations is plausible. It is known that dehydration does slow the rate of racemization,[8,9] as spectacularly witnessed by the absence of D-amino acids in fossilised insects entombed in amber.[10] However, the pore size distribution of mineralised collagen, which is dominated by sub-10nm pores[11] and the tight hydration shells of collagen itself,[12] means that it is not possible to remove all the water by dessication. Dehydration by heating will expose the sample to considerable temperatures as the water boils off.

Racemization is observed to be a highly temperature sensitive reaction. Assuming an activation energy of 31.3kcal mole^{-1} (based on data,[13] Fig.2) the rate of racemization of aspartic acid at 150°C should be 800,000 times faster than at 37°C, i.e. 1hr at 150°C is equivalent to 92 years at 37°C. There may indeed be an increase in the rate of conversion of D to L, but there will also be an equivalent increase in the conversion from L to D. How then does the data relate to the kinetics of racemization? The authors do not give sufficient information in their paper for us to draw firm conclusions. For instance, there is no information on the concentration of the soluble protein fraction, nor its amino acid composition after heating compared with the equivalent fraction before heating. In addition there is no information on the abundance of free amino acids or N-termini after heating which may suggest peptide bond hydrolysis.

Heating will cause the collagen to gelatinise. Gelatin undergoes rapid racemization (Fig.2) whereas there is no detectable racemization in collagen purified from dentine by CNBr digestion[14] (Fig.2) or when measured in artificial diagenesis experiments.[15] Using molecular dynamic simulations of collagen and gelatin, we believe that this is due to the conformational constraint of the collagen triple helix[16] which prevents the formation of the cyclic succinimide intermediate of the reaction.

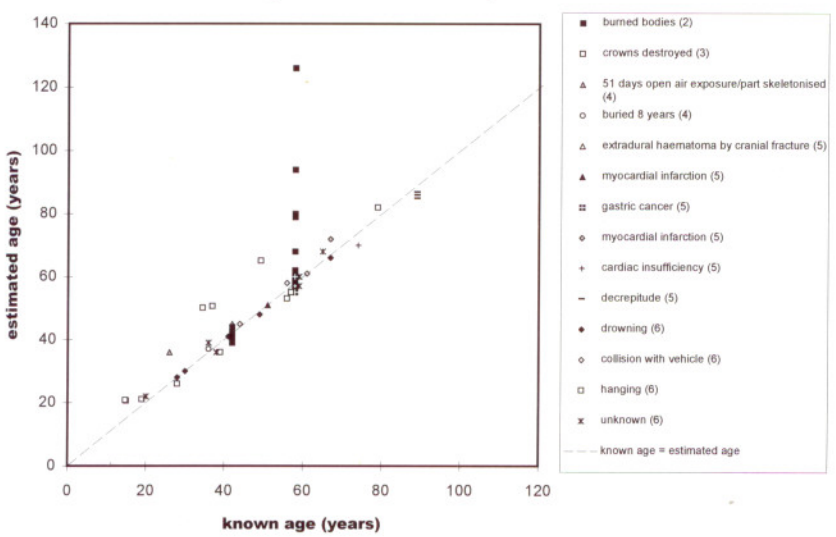

Figure 1: *Estimated age plotted against the known age determined from%D Asx in the total protein fraction of human dentine from individuals with various causes of death and post-mortem conditions, (data from [2,3,4,5,6])*

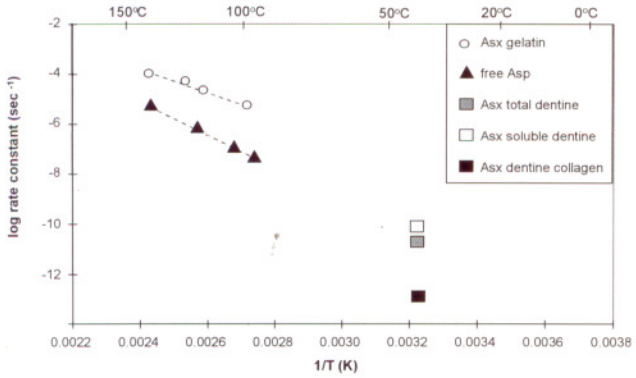

Figure 2: *The rate of racemization of free aspartic acid (▲,[8]) is much slower than gelatin (○,[8]), and approximately the same as for the total dentine fraction (❑,[1]), which comprises soluble dentine proteins (❑,[1]) and an insoluble fraction comprised chiefly of collagen (■,[9])*

By heating the sample, collagen in the insoluble protein fraction of dentine (ISF) will gelatinise and therefore contribute to the "soluble" fraction (SF). During solubilization, the combination of heat and evaporating water will lead to further rapid racemization of the SF itself. The new SF formed will comprise more highly racemized material from the original SF and less well racemized material from the originally very poorly racemized collagen and other non-collagenous proteins of the ISF. In the case of the experiments conducted here, the increase in racemization of the original SF plus the contribution of less well racemized, gelatinised ISF appear to cancel each other out to provide a new SF with approximately the same rate of racemization.

Our hypothesis is testable: we predict a greater proportion of SF/ISF after heating, and a change in the amino acid profile, a greater proportion of Gly, Pro and Hyp attesting to the increased concentrations of gelatin in this fraction. Indeed, we would propose that an amino acid profile of either the SF or ISF is always published alongside the D/L ratio to identify potential autochthonous or allochthonous contamination. To show that the authors are justified in their approach, teeth should be heated at different temperatures for varying periods of time. According to their claims, the rate of racemization in the SF should remain the same as for unheated teeth, whatever the conditions. We believe that the fortuitous findings of this study are unlikely to be repeated and conclude that the method should not be used on burnt bodies until a more thorough analysis has been conducted.

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