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References should be listed at the end of the paper in alphabetical order. Articles in preparation or articles submitted for publication, unpublished observations, personal communications, etc. should not be included in the reference list but should only be mentioned in the article text (e.g., A. Kingori, University of Nairobi, Kenya, personal communication). Journal names are abbreviated according to Chemical Abstracts. Authors are fully responsible for the accuracy of the references.

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Full Length Research Paper

Gender determination using primary teeth: A polymerase chain reaction (PCR) study

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Accepted 6 September, 2013

The aim of this study was to assess the effect of various environmental factors on the preservation of pulp tissue in primary teeth as a source of DNA and its use for sex determination using polymerase chain reaction (PCR). 120 samples were grouped into 4 groups of 30 teeth each. Group I samples were kept immersed in a bucket of drainage water, group II, III and IV samples were buried in sand taken from seashore, burial ground and desert, respectively for a period of 2 months. Pulp tissue was collected from each sample and DNA was isolated. PCR amplification was performed and sex determination was done by detection of X and Y chromosome-specific alphoid centromeric repeat sequences. In group I, 86% of the samples exhibited correct gender interpretation by PCR amplification. In groups II, III, and IV, all the samples showed correct results indicating a significant difference in scores between group I and the remaining three groups. Teeth stored in dry conditions can serve as a better source of DNA as compared to the teeth stored in moist conditions and co-amplification of both X and Y specific sequences by PCR is a fast, specific, sensitive and reliable method providing sex determination.

Key words: DNA, gender, polymerase chain reaction (PCR), forensics

INTRODUCTION

The rights of children and their aspirations are of paramount importance in our demonstration towards an inclusive and equitable society. Investment in the well-being of children is an investment in the future of the country. Eradication of malnutrition and improving general and dental health is receiving topmost priority. At the same time issues relating to child protection are high on every government's agenda.

Child abuse is shrouded in secrecy and there is a conspiracy of silence around the entire subject. Certain kinds of traditional practices that are accepted across many countries, knowingly or unknowingly amount to child abuse. Existing socio-economic conditions also render some children vulnerable and more at risk to abuse, exploitation and neglect. It is time that this is recognized and appropriate remedial measures taken. Lack of empirical evidence and qualitative information on the dimensions of child abuse and neglect makes it difficult to address the issue in a comprehensive manner.

In a study conducted by Ministry of Women and Child Development, Government of India (2007), state-wise break up of overall incidence of physical abuse revealed that in all the 13 states covered under the study, the reported incidence of physical abuse was very high, in fact uniformly above 50%. Annually, large numbers of children go missing and there is little attempt to track them or trace them.

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The global scenario is also not much different as compared to the Indian situation, according to an overview given by the United Nation Secretary General's Study on violence against children, WHO estimated that almost 53,000 children that died in 2002 were due to child homicide (Ministry of Women and Child Development, Government of India, 2007).

In the past, there were many efforts made by numerous professionals to detect and document various cases of child abuse, but unfortunately most of them focused on the children who are living. But definitely there will be a group of children who could have lost their life due to the same barbaric act which went unnoticed, and these unfortunate children also deserve justice as much as the living children do. Determination of the victim's identity is the first step in any kind of forensic investigation and determination of gender is one of the important dimensions of this process. During the past few decades physical evidence has become increasingly important in criminal investigations. Courts often view eye witness accounts as unreliable and biased. Physical evidence such as DNA, fingerprint and trace evidence may independently and objectively link suspects to a crime, disprove an alibi, or develop important investigative leads. Gender identification can also provide valuable information about past human societies, culture and their life styles.

There are no reliable methods available for sex determination in primary teeth using morphometric analysis (Murakami et al., 2000). With the improvement of technology, increased speed, sensitivity and specificity, DNA analysis has revolutionized the field of forensics and PCR method is one of them. Bones and teeth are the only material available in markedly decayed/skeletonized bodies. The most important variables upon which identification and availability of DNA depend are time after death and type of soil (pH, humidity, temp, bacteria, etc) where in the bodies are buried (Dragan, Unpublished).

If found reliable in tissues exposed to such conditions, PCR method of sex determination can contribute immensely in gender identification in forensic as well as anthropological, archeological and paleontological researches.

The aim of this study was to assess the effect of various environmental factors on the primary teeth in the preservation of pulp tissue as a source of DNA and its use for sex determination using PCR.

MATERIALS AND METHODS

Sample selection

A total of 120 non carious primary canines extracted for serial extraction or various other purposes were used as the study samples. Written informed consent was taken from the parents of the children whose teeth were selected for the study (by the co-investigator). The teeth showing apical resorption beyond apical 1/3 were not included in the study.

Study groups

The selected samples were grouped into 4 groups of 30 teeth each. Coding and decoding was done by a co-investigator. The samples in group I were kept immersed in a bucket of drainage water (collected from the main sewer channel of the town) for 2 months. The teeth samples of group II, III and IV were buried in sand taken from seashore, burial ground and desert, respectively at a depth of 30 cm for a period of 2 months (Figure 1).

Pulp sample collection

After 2 months, teeth were taken out and washed thoroughly using distilled water. The teeth were then sectioned along the long axis, through the pulp chamber using hard tissue microtome and pulp was recovered (Figure 2A and B). The recovered pulp samples were stored and carried in 100% ethanol in labeled Eppendorf tubes for DNA isolation.

Isolation of DNA from dental pulp

DNA was isolated from dental tissue obtained from the teeth by proteinase-k digestion and phenol chloroform extraction methods (Zeljka et al., 2000). Isolated DNA was then diluted with tris-ethylennediaminetetraacetic acid (TE) buffer. The isolated DNA was then utilized for PCR amplification in Amplitaq DNA polymerase buffer for various thermal cycles.

Electrophoresis and sex determination

The PCR products underwent electrophoresis in 1% agarose gel at 200 V for 1 h. Ethedium bromide staining was performed and amplified bands of X and Y sequences were examined under UV radiation (Figure 3). The sex of a subject was considered to be male when both X and Y specific sequences were detected, but female when only the X-specific sequence was detected.

Universal precautions were taken to prevent cross contamination while handling the samples which included personal protective equipments (gloves, mouthmasks, haedcaps) and PCR was done in specifically designed setup in human genetics department. Disposable devices were preferred wherever possible to rule out cross contamination either from operator DNA or amplicons from previous experiments.

Data analysis was done using Chi square test of independence. Statistical significance was set at 5% level of significance (p<0.05). Percentage distribution of positive and negative results was compared among the groups.

RESULTS

In group I (teeth stored in drainage water), 26 out of 30 teeth (86%) showed correct gender interpretation. In groups II, III, IV (teeth buried in sand taken from beach, mud taken from burial ground and sand taken from desert, respectively) all the samples (30 in each group) showed correct results indicating a significant difference in scores between group I and the remaining three groups (p<0.01, 99% significance) (Table 1).

Gender wise distribution showed up in group I out of 26 male samples, correct sex determination was possible in 22 samples (73.3%) and 4 samples gave wrong results.
Table 1. Comparisons between positive and negative results in different test groups.

<table>
<thead>
<tr>
<th>Result</th>
<th>Group I</th>
<th>Group II</th>
<th>Group III</th>
<th>Group IV</th>
<th>Total</th>
</tr>
</thead>
<tbody>
<tr>
<td>Positive Count</td>
<td>26</td>
<td>30</td>
<td>30</td>
<td>30</td>
<td>116</td>
</tr>
<tr>
<td>Within group (%)</td>
<td>86.7</td>
<td>100</td>
<td>100</td>
<td>100</td>
<td>96.7</td>
</tr>
<tr>
<td>Negative Count</td>
<td>4</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>4</td>
</tr>
<tr>
<td>Within group (%)</td>
<td>13.3</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>3.3</td>
</tr>
<tr>
<td>Total Count</td>
<td>30</td>
<td>30</td>
<td>30</td>
<td>30</td>
<td>120</td>
</tr>
<tr>
<td>Within group (%)</td>
<td>100</td>
<td>100</td>
<td>100</td>
<td>100</td>
<td>100</td>
</tr>
</tbody>
</table>

Chi-square=12.414, p<0.05.

Table 2. Chi-square tests.

<table>
<thead>
<tr>
<th>Test</th>
<th>Value</th>
<th>df</th>
<th>Asymp. Sig. (2-Sided)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Pearson Chi-square</td>
<td>12414$^a$</td>
<td>3</td>
<td>0.006</td>
</tr>
<tr>
<td>Likelihood ratio</td>
<td>11.514</td>
<td>3</td>
<td>0.009</td>
</tr>
<tr>
<td>Linear-by-linear</td>
<td>7.386</td>
<td>1</td>
<td>0.007</td>
</tr>
<tr>
<td>Association</td>
<td>120</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>No. of valid cases</td>
<td>-</td>
<td>-</td>
<td>-</td>
</tr>
</tbody>
</table>

$^a$4 cells (50.0%) have expected count less than 5. The minimum expected count is 1.00.

(13.3%). Sex was determined correctly in all 4 female samples (Table 2).

In group II, correct sex could be determined in all 23 (76.7%) male and 7 (23.3%) female samples, whereas in group III, positive sex determination was possible in all the 24 (80%) male and 6 (20%) female samples, group IV also showed correct sex determination in all 23 (76.7%) male and 7 (23.3%) female samples.

**DISCUSSION**

Children are the most vulnerable group in our society. Child fatalities due to maltreatment represent the worst-case scenario in attempts to protect children. Although the untimely deaths of children due to illness and accidents have been closely monitored, the same cannot be said of children who have died as the result of physical assault or mere neglect. Interventional strategies targeted at resolving this problem face complex challenges.

Dental evidences have continued to provoke controversy within the field of forensic dentistry. The differing views surrounding interpretation, methodologies and admissibility are cornerstones of the arguments (McNamee and Sweet, 2003).

Sex identification is the first step in personal identification in forensic medicine. In general, the sex of an unidentified body can be determined based on anatomical characteristics of the external genitalia or whether the gonads are ovaries or testis. However, bones and teeth are the only available material for sex determination in markedly decayed and skeletonized bodies.

Sex differences in dental morphometric values are not distinct except in the permanent canine teeth, and determination of the sex from a random single tooth is extremely difficult. There has been no method to distinguish the sex particularly based on milk teeth (Murakami et al., 2000). Sex determination from pulp material can be done by different methods like fluorescence Y chromosome test, Southern blot test, etc. PCR stands above all mentioned methods since the high rate of sensitivity and specificity have been noted in previous experiments (Kumar and Hegde, 2005).

Teeth are considered as a good source to obtain genetic material. This is true mainly because of their great tissue resistance against external influences. Malaver and Yunis (2003) extracted DNA obtained from dentin and cementum of 20 corpses that had been buried for at least 5 years. Pulp tissue is a loose connective tissue and it degrades easily when compared with other dental tissues. Pfeiffer et al. (1999) studied the influence of the environment on DNA degradation in teeth that were kept underneath the soil. They observed that a tooth with opened pulp exposed to external agents showed a significant degradation. Lessig and Edelmann (1995) showed that pulp can be source of DNA in teeth that had been kept or obtained under different conditions such as teeth that had been extracted when the person was alive.
Table 3. Comparisons of positive and negative results among different sexes in test groups (Chi square test was not performed because of lack of frequency distribution).

<table>
<thead>
<tr>
<th>Group</th>
<th>Positive</th>
<th>Negative</th>
<th>Total</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Count</td>
<td>% Total</td>
<td>Count</td>
</tr>
<tr>
<td>Group I</td>
<td>Male</td>
<td>22</td>
<td>73.3</td>
</tr>
<tr>
<td></td>
<td>Female</td>
<td>4</td>
<td>13.3</td>
</tr>
<tr>
<td></td>
<td>Total</td>
<td>26</td>
<td>86.7</td>
</tr>
<tr>
<td>Group II</td>
<td>Male</td>
<td>23</td>
<td>76.7</td>
</tr>
<tr>
<td></td>
<td>Female</td>
<td>7</td>
<td>23.3</td>
</tr>
<tr>
<td></td>
<td>Total</td>
<td>30</td>
<td>100</td>
</tr>
<tr>
<td>Group III</td>
<td>Male</td>
<td>24</td>
<td>80</td>
</tr>
<tr>
<td></td>
<td>Female</td>
<td>6</td>
<td>20</td>
</tr>
<tr>
<td></td>
<td>Total</td>
<td>30</td>
<td>100</td>
</tr>
<tr>
<td>Group IV</td>
<td>Male</td>
<td>23</td>
<td>76.7</td>
</tr>
<tr>
<td></td>
<td>Female</td>
<td>7</td>
<td>23.3</td>
</tr>
<tr>
<td></td>
<td>Total</td>
<td>30</td>
<td>100</td>
</tr>
</tbody>
</table>

Figure 1. Photographs showing study groups (A) group I (Drainage water), (B) group II (seashore sand), (C) burial ground mud, and (D) desert sand.
The bodies were decomposed and fragmented considerably more after death and kept in room temperature for 12 and 6 months, respectively (Da Silva et al., 2007).

In the present study, primary canine teeth were selected as the samples as these teeth are comparatively more protected teeth in the dental arch both from traumatic injuries and dental decay and compared to other primary teeth, initiation of resorption is late in primary canines. The teeth showing resorption beyond apical third were not included in the study.

It appears that the most important variables upon which identification by DNA depends are, the extent of time after death, the type of the soil in which the bodies were buried (temperature, bacterial composition, pH, etc), and the method of DNA extraction (Dragan, Unpublished).

DNA analysis of teeth for sex determination must be able to be carried out in bodies which are markedly decayed or skeletonized bodies that provide no other materials. In such bodies, dental DNA is considered to be considerably more decomposed and fragmented. Therefore, in this study highly sensitive method was followed as adopted by Murakami et al. (2000).

The Y and X chromosome-specific alphoid repeat sequences which were examined in this study were repeated several thousand times per genome, respectively and can be amplified by PCR if part of these copies remains intact (Murakami et al., 2000). Therefore, this method is more advantageous with regard to sensitivity than methods using a single copy base sequence.

In group I, sex was determined accurately in 26 of 30 teeth (86%). Among the remaining 4 teeth, no amplified band of Y chromosome specific sequence was observed in 3 teeth, although the samples were obtained from males and in 1 male sample, neither X nor Y specific amplification bands appeared. In these teeth therefore, decay due to bacteria was considered to have progressed markedly by infiltration of drainage water in to the pulp cavity and DNA of pulp tissue was considered to have been degraded to such a degree that amplification of the Y chromosome specific sequence by PCR became impossible. In these three samples, the amplified band of the X chromosome specific sequence was detected although that of the Y chromosome specific sequence was not, possibly because the amplified fragment of the X chromosome specific sequence, which is shorter than that of Y chromosome-specific sequence, was less liable to be effected by DNA degradation due to decay and because the number of repetitions of the X chromosome specific sequence in a single genome is several tens of times greater than that of the Y chromosome specific sequence (Murakami et al., 2000). In the remaining one tooth, it was considered that autolysis and DNA fragmentation reached such an extent that even detection of X chromosome specific sequence became impossible. Chances of PCR inhibition by substances in drainage water also was not ruled out.

The findings of our study were in accordance with the findings of Murakami et al. (2000) who mentioned that ‘in wet state, pulp tissue gradually lyses due to autolysis and decay and its sampling becomes difficult’. This can be further accelerated in drainage water which is rich in various types of bacteria and chemical substances making DNA sampling difficult to near impossible.

The results of the present study indicate that sex determination of teeth by means of PCR is considered to be extremely useful for identification of markedly decayed or skeletonized bodies, which has been difficult using the conventional morphological methods contributing in identification of victims of various incidents and disasters including child abuse.

Sex determination is an important step in personal identification in forensic sciences. Surrounding environment has a definite role in preservation of pulp tissue; primary teeth can be regarded as an effective tool in preservation of pulp tissue which can provide DNA for sex determination and other DNA analysis procedures.

REFERENCES

Dragan P (Unpublished). Identification of human remains from mass graves found in Croatia and Bosnia and Herzegovina.


Plaque removal efficacy of a novel oral care device: A microbiological assessment

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Accepted 1 July, 2013

In adults with inflammatory problems, self-performed mechanical plaque removal is insufficiently effective and should be improved. The aim of this study was to determine the biofilm removal efficacy of a new oral care device, the digital brush (Enacare, Micerium), a disposable gauze product soaked in 0.12\% chlorhexidine. Changes in supragingival microbiota were investigated in 30 Caucasian patients (14 males and 16 females) aged 8 to 90 years. All subjects provided written informed consent. Pre-treatment (pre-T) and post-treatment (post-T) samples of supragingival plaque were taken from the right vestibular and lingual mucosa in 15 subjects and from the buccal aspect of the anterior sextant in 15 subjects using sterile swabs flocked with sterile nylon fibers. The samples were analyzed to determine the presence of Candida albicans, Candida species, Staphylococcus aureus, Staphylococcus epidermidis, Staphylococcus species, oral streptococci, and Enterobacter species. Groups were compared using Pearson’s chi-squared test. The following bacteria were detected: C. albicans (8 pre-T and 3 post-T), Candida spp. (3 pre-T and 0 post-T), Enterobacter spp. (2 pre-T and 2 post-T), S. aureus (12 pre-T and 4 post-T), S. epidermidis (2 pre-T and 1 post-T), Staphylococcus spp. (29 pre-T and 22 post-T), and Streptococcus viridans (29 pre-T and 22 post-T). Microbiota differed between sampling sites. Within the limits of this preliminary clinical and microbiological evaluation of biofilm reduction in a small sample, the digital brush appears to be an effective plaque removal device. Mechanical cleaning with this tool appears to be more effective on hard surfaces than on mucous membranes.

Key words: Plaque removal, home care, gauze, digital brush, chlorhexidine, bacteria, brush.

INTRODUCTION

Routine toothbrushing is the principal method used by individuals to remove biofilm and control plaque-related diseases, such as periodontitis and caries (Creeth et al., 2009; Lucchese et al., 2012a). However, in some adults, especially those with inflammatory problems, self-performed mechanical plaque removal is insufficiently effective and should be improved (van der Weijden and Hioe, 2005).

To improve dental health care, professional recommendations should always fit patients’ specific needs (Silverman and Wilder, 2006). Given the strong adhesion of biofilms grown from whole saliva (Verkaik et al., 2010), a mechanical plaque removal strategy must be implemented to achieve satisfactory oral health. The introduction of a novel device may improve patients’ compliance (Chongcharoen et al., 2012; Sicilia et al., 2003).

The aim of this study was to determine the biofilm elimination capability of a new oral care device, the digital brush (Enacare, Micerium S.p.A., Genoa, Italy), a disposable gauze product containing 0.12\% chlorhexidine. This device can be used as an alternative to conventional oral hygiene, when performing the latter is difficult or as additional device to improve the quality of self-performed mechanical plaque removal.

The null hypothesis of this study was that the presence
of microbiota (representing the cleansing effect) before and after the use of a medicated gauze product on the mucosa and teeth would not differ.

MATERIALS AND METHODS

A disposable gauze product containing 0.12% chlorhexidine can serve as an alternative device for oral hygiene, even outdoors, or as an additional device for individuals with special care needs, bedridden patients, and caregivers.

Patients

The study group comprised 30 Caucasian patients (14 males and 16 females) with a mean age of 48.3 (range, 8 to 90) years. All patients provided written informed consent.

Sampling

At baseline, pre-treatment (pre-T) supragingival plaque samples were taken from the right vestibular and lingual mucosa in 15 subjects (group 1) and from the buccal aspect of the anterior sextant in 15 subjects (group 2) using sterile swabs.

The subjects were instructed in proper oral hygiene and the use of the digital brush as a cleansing device (Figures 1 and 2), using a rolling motion technique (Figure 3A and B) for ~2 min. Post-treatment (post-T) microbiological sampling was performed immediately after cleaning.

Culture protocol

Saliva samples were collected with flocked swabs (Copan Italia S.p.A., Brescia, Italy) designed for biological sample collection that contained a transport medium specific to aerobic and anaerobic bacteria. Samples subjected to delayed (>24 h after collection) microbiological evaluation were transferred to cryovials and stored at ~80°C to ensure preservation.

Bacterial culture was performed as follows. Using a disposable sterile loop, 10-μl samples were streaked onto the following plates (Vacutest; Kima [ARZERGRANDE, Pd, Italy]): horse blood agar (for non-selective growth of streptococci groups A to C, pneumococci, and staphylococci), azide agar (for selective growth and isolation of streptococci, Enterococcus species, and Enterococcus species, including Enterococcus species, Herellea agar (for selective growth and isolation of Gram-negative bacteria), and CHROMagar Candida (for Candida identification). The plates were incubated at 37°C for 24 h, then examined to distinguish colonies on the basis of morphology, pigmentation, and macroscopic shape. In cases of positive growth, standard identification procedures were applied to selected colonies.

The isolated colonies were identified using the VITEK® automatic system (bioMérieux, Inc, Hazelwood, Mo). For colony counts, samples were serially diluted to 1:10^6. The number of colony-forming units (CFUs)/ml in the original sample was determined by multiplying the number of colonies (30 to 300) per plate by the dilution factor.

Statistical analysis

The presence or absence of microorganisms (including Candida species, Staphylococcus aureus, Staphylococcus epidermidis, Streptococcus viridans, Enterobacter species) was determined before and after cleaning with the digital brush. Pearson’s Chi-squared test was used to analyze bacterial concentrations and compare data from groups 1 and 2.

RESULTS

The results of microbiological evaluation are reported in Table 1. The following bacteria were detected: Candida albicans (8 pre-T and 3 post-T), Candida spp. (3 pre-T and 0 post-T), Enterobacter spp. (2 pre-T and 2 post-T), S. aureus (12 pre-T and 4 post-T), S. epidermidis (2 pre-T and 1 post-T), Staphylococcus species (29 pre-T and 22 post-T), and S. viridans (29 pre-T and 22 post-T). Microbiota differed between sampling sites.

No significant difference in the presence of bacteria was detected between groups 1 and 2. The mean post-T reduction in bacterial concentration was 2.36 log_{10}. Using this cut-off value, data from groups 1 and 2 were compared by Pearson’s chi-squared test. Although more reduction was visible in group 2 samples (from the buccal aspect of the anterior sextant), no significant difference was found.

DISCUSSION

The oral cavity can serve as a reservoir of pathogens that can cause systemic infection. C. albicans was the most prevalent yeast found in the periodontal pockets (76.2%) and oral cavities (63.0%) of patients with periodontal disease (Cuesta et al., 2010).

Many studies have demonstrated the essential etiological role of pathogenic dental biofilm in the development of gingivitis, additionally finding that most people fail to maintain sufficient mechanical plaque control to prevent disease (van der Weijden and Hioe, 2005; Barnett, 2006).

In adults, professional mechanical plaque removal (PMPR) in combination with oral hygiene instruction (OHI) may be more effective than no treatment, but patient compliance in combination with repeated OHI may have an effect similar to that of PMPR (Needleman et al., 2005).

Oral health care professionals generally recommend that individuals brush their teeth for at least 2 min using an appropriate technique; however, adequate interdental cleaning requires 4 min or more (Chongcharoen et al., 2012; Gjermo and Flotra, 1970). Patients’ failure to comply with the correct use of cleaning devices for an adequate period of time can be a problem. The average brushing time in the general population is ~45 to 50 s, only 10% of which is spent cleaning the lingual tooth surfaces (Olaydon, 2008).

Significantly lower dental plaque scores have been recorded immediately after an oral self-care demonstration; a mean of 27.4% plaque removal was observed after the demonstration (Yuen et al., 2009), compared with 40 to 55% plaque removal after 1 min of manual toothbrushing in the general (young and middle-aged) healthy, non-
disabled population, as reported in a meta-analysis (van der Weijden GA, Hioe, 2005). Consistently, no more than 60% of the overall plaque is removed during each episode of cleaning (Claydon, 2008). Less plaque was removed from mandibular teeth and lingual tooth surfaces than on the maxillary teeth and buccal surfaces (Claydon, 2008; Yuen et al., 2009).

A previous dental review (van der Weijden and Hioe, 2005) proved that self-performed mechanical plaque removal is insufficiently effective and should be improved. Treatment procedures should always include customized patient education and OHI. In some instances, such instruction can be used as appropriate to reduce, eliminate, or change the nature of microbial pathogens and to remove bacterial plaque, although only from the supragingival regions.

User skill is a more important factor than the design of the toothbrush for the efficacy of cleaning (Yuen et al., 2009). Thoroughness may be improved by the use of tactile receptors in the fingers to guide a device, such as the digital brush, the novel home care device used in this study. The use of the digital brush with a wiping motion enables an individual to reach frequently neglected dental surfaces.

Studies of the oral microbial environment have demonstrated that oral mucosal tissues act as reservoirs of the bacteria that colonize tooth surfaces (Silverman and Wilder, 2006; Verkaik et al., 2010; Needleman et al., 2005; Gjermo and Flotra, 1970; Claydon, 2008). This finding supports the incorporation of an effective antimicrobial mouth rinse into the daily oral hygiene regimen to complement mechanical plaque control (Silverman and Wilder, 2006; Verkaik et al., 2010; Yuen et al., 2009; West and Moran, 2008; Gunsolley, 2006).

Chlorhexidine remains the gold standard of antiplaque
agents (Silverman and Wilder, 2006). According to one meta-analysis, seven studies have documented the strong antiplaque, anti-gingivitis effects of mouth rinses with 0.12% chlorhexidine (Verkaik et al., 2010). The gingival index has also been used to demonstrate the significant anti-gingivitis effects of these mouth rinses (Raul, 2008). Twice-daily oral care with 0.12% chlorhexidine gluconate may hold promise for the prevention of nosocomial infection (Bopp et al., 2006).

The persistence of staining on natural dentition after the use of chlorhexidine gluconate mouth rinse is a well-known side effect of this antimicrobial agent that counter indicates long-term use (Bagis et al., 2011). This staining effect should be expected to be most pronounced in the first few days of use. Other reported side effects of chlorhexidine use include pain, burning sensation, pruritus, xerostomia, taste disturbance, mucosal irritation, and discoloration of tooth and tongue surfaces (Gürgan et al., 2006).

In the present study, a greater reduction in microbial concentration occurred in group 2 (samples taken from the buccal aspect of the anterior sextant) than in group 1.

Figure 3. The digital brush is wrapped around the index finger and utilized with a sweeping motion in an apico-oclusal direction from the oral mucosa to the tooth surfaces, similar to the roll brushing technique. Finger tactile receptors can guide cleaning movements to better reach frequently neglected dental surfaces in the posterior lingual/palatal areas. This device may improve the thoroughness and efficiency of cleaning.
Table 1. Microbiological data.

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(samples taken from the right vestibular and lingual mucosa), although this difference was not significant. Mechanical cleansing with the digital brush tended to be more effective on hard surfaces than on the mucous membranes.

The lack of significant findings may be due to the small sample size. Further research may support our findings by detecting significant differences.

Conclusions

Within the limits of this clinical and microbiological evaluation of a small sample, the digital brush seems to be an effective plaque removal device. Its use as an alternative tool when conventional oral hygiene is difficult to implement or as a supplementary device to improve the quality of self-performed mechanical plaque removal can be recommended (Lucchese et al., 2012b). Further studies with larger samples are necessary to more fully evaluate the cleansing effectiveness of this novel device.

REFERENCES


UPCOMING CONFERENCES

5th International Conference on Food Engineering and Biotechnology, Penang, Malaysia, 12 Mar 2014

17th International Conference on Biopesticides: Current Status and Future Prospects, Alexandria, Egypt, 1 Apr 2014
Conferences and Advert

December 2013
IEEE International Conference on Bioinformatics and Biomedicine (BIBM), Shanghai, China, 18 Dec 2013

February 2014
5th International Conference on Legal Medicine, Medical Negligence and Litigation in Medical Practice & 5th International Conference on Current Trends in Forensic Sciences, Forensic Medicine & Toxicology (IAMLE 2014), Goa, India, 25 Feb 2014
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