Comparative evaluation of chlorhexidine and sodium fluoride mouthwashes on streptococcus mutans

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Abstract

Objective: This study was conducted to compare the efficacy of chlorhexidine (0.2%) and sodium fluoride (0.05%) mouthwashes in reduction of Streptococcus mutans count in plaque in seventy-five children of age group 8-14 years.

Methods: Children were equally divided in three groups. Group A and group B were test groups for chlorhexidine and sodium fluoride respectively and group C was control group. The subjects were instructed to rinse the mouth with 10 ml of mouthwash for one minute twice daily for fifteen days. Number of colony of Streptococcus mutans were counted by using mitis salivarius agar plate, at the beginning and at the end of the study period. Inter group and intra group comparisons were done to evaluate the efficacies of mouthwashes by using paired t test and anova test.

Results: There was significant reduction in number of colony of Streptococcus mutans in both test groups. P value was found to be.000 in both whereas in control group, there was no significant reduction in Streptococcus mutans (p=0.100) count. Among the two mouthwashes efficacy of both were found to be statistically equal.

Conclusions: The results of this study suggested that sodium fluoride is a potent antimicrobial agent as chlorhexidine and would be better mouthwash due to its additional remineralization property for prevention of dental caries in children.

Key words: Dental caries, Plaque, Streptococcus mutans, Chlorhexidine, Sodium fluoride

Introduction

Dental caries is the most common disease in children. It is an infectious disease and has multifactorial etiology such as diet, micro flora, host and time. The modern concept of cariogram demonstrates micro-organism as one of the major etiological factors. Among the pathogenic flora, mutans streptococci particularly Streptococcus mutans are considered as main microorganism associated with dental caries1, 2. These bacteria colonize and accumulate in plaque and are highly aciduric and acidogenic. They produce lactic acid and demineralize the tooth structure.

Targeting Streptococcus mutans forms is the most important measure for prevention of dental caries. This can be achieved by various mechanical and chemical aids. Many chemical antiplaque agents in the form of varnishes, dentifrices and mouthwashes have been tried for improvement of oral health. Mouthwashes have been found to be one of the safe and effective delivery system as anti-microbial and antiplaque agent. These mouthwashes are capable of inhibiting bacterial adhesion, colonization and metabolic activity which ultimately affects bacterial growth. A variety of mouthwashes have been examined for ability to control micro-organisms and to affect the plaque formation. Among the various mouthwashes, the most persistent reduction of Streptococcus mutans has been achieved by chlorhexidine mouthwashes3.

Chlorhexidine though, is not toxic, has unpleasant taste and it alters the taste sensation4. It also produces brown staining of teeth and affects mucous membrane and tongue4. This may be related to precipitation of chromogenic dietary factors on to tooth surface and
mucous membrane. Many clinical trials showed that the taste of chlorhexidine mouthwash is not well accepted by the children⁵.

Fluoride mouthwash, commonly used in children is the sodium fluoride mouthwash. Its pH is neutral and is found to be well accepted by the children⁶. It is available in concentration of 0.05% (220ppm) for daily use and 0.2% (900ppm) for weekly use. This mouthwash is recognized as a potent anti-cariogenic agent and is effective in reduction of caries. The anti-caries effect of fluoride on tooth through its remineralization property has been studied and discussed extensively but the effect of fluoride on plaque has taken a backseat. Some recently done studies demonstrated that fluoride mouthwash has potential antimicrobial activities.

The aim of the study was to assess and compare the effect of chlorhexidine and sodium fluoride mouthwashes in reduction of Streptococcus mutans level in plaque in children.

Materials and methods

This study was undertaken in the Department of Pedodontics and Preventive Dentistry, Manipal College of Dental Sciences, Mangalore, India in co-ordination with Department of Microbiology, Kasturba Medical College, Mangalore, India. A double-blind was designed and the study was approved by Ethical Committee, Kasturba Medical College, Mangalore, India. Children with high caries-risk age group, 8-14 (mean age group-12) were recruited from an orphanage in Mangalore. Children with positive medical history, prolonged antibiotic therapy and subjects undergoing orthodontic treatment or with intraoral appliance were excluded. Written consent was obtained from the authorities of the orphanage after the nature of the study and the possible risks were fully explained. Total sample size was 75. Children were randomly divided into 3 groups, each group consisting 25 children as follows:

- **Group A (n=25)** – were given 10ml of 0.2% chlorhexidine gluconate mouthwash (Clohex, DR. Reddy’s) twice daily in the interval of 12 hours for 15 days.

- **Group B (n=25)** – were given 10ml of 0.05% sodium fluoride mouthwash (S-Flo, DR. Reddy’s) twice daily in the interval of 12 hours for 15 days.

- **Group C (n=25)** – were used as control group.

The subjects were provided non-fluoridated toothpaste (Dabur Red, Hindustan Limited) and were told to brush their teeth twice daily, once in the morning and once in the evening. Hence the action of fluoride on plaque and bacteria from other sources was not possible and the antimicrobial action of mouthwashes was evaluated without any interference.

At the beginning of the study, a small amount of plaque was collected from an individual to record number of Streptococcus mutans. After initial examination, the subjects were instructed to rinse the mouth with 10 ml of specific mouthwash for one minute twice daily for fifteen days. One doctor was allotted to deliver the mouthwash to the children. A plastic measuring cup was used to measure 10ml of mouthwash and it was given in disposable glass. The supervision was done by same person while children were taking mouthwash and instruction was given not to rinse the mouth for half an hour. Final data was recorded for number of Streptococcus mutans level at the end of 15 days.

Clinical assessments were performed in the clinic by a single examiner using mouth mirror and probe. A dental prophylaxis was not performed so that the study could begin with normal, existing level of plaque deposits of the subjects.

Prior to the examination, the children were asked to rinse the mouth with water in order to remove the food debris. A small amount of plaque was collected from the tooth surface with help of sterilized toothpick. Sample of dental plaques were prepared for culture process in mitis salivarius agar (MSA). The plates were then incubated at 37°c in anaerobic condition for 48 hours, followed by incubation at 4°c. After 12 hours, number of colonies were counted using colony counter.

The collected data was subjected to statistical analysis. For intra–group and inter group comparison of plaque and Streptococcus mutans, Paired t-test and One way Anova test were applied.

Results

**Intra group comparison**

Comparison of differences in number of colony of bacteria between before intervention and after was done with paired t-test.

Table 1 shows number of bacteria before and after intervention in group A (chlorhexidine)

Table 2 shows number of bacteria before and after intervention in group B (Sodium fluoride)

Table 3 shows intra group comparison of group C (Control Group)
**Inter-group Comparison**

Intergroup comparison was performed in between group A and group B to compare the efficacy of chlorhexidine mouthwash and sodium fluoride by using one way Anova test.

On comparison, the result showed there was no significant difference in and number of colony of Streptococcius mutans between two experimental groups before the interventions and after the interventions as depicted in Graph No. 1.

Hence in the experimental groups (group A using chlorhexidine mouthwash and group B using sodium fluoride mouthwash), number of colony of Streptococcus mutans at the end of the study were significantly lower than the scores recorded at the baseline. In the control group (group C, no mouthwash), at the end of the study period, there was no statistically significant reduction in number of colony of Streptococcus mutans. Amongst the two experimental groups, at the end 15 days, almost comparable reduction was found in group A and group B. The differences in efficacy of these two mouthwashes were highly non-significant.

**Table No. 1**

<table>
<thead>
<tr>
<th>Pair 1</th>
<th>Number of bacteria before intervention</th>
<th>Mean</th>
<th>N</th>
<th>Std. deviation</th>
<th>Std. Error Mean</th>
<th>P value</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Number of bacteria after intervention</td>
<td>88.80</td>
<td>25</td>
<td>106.00</td>
<td>21.200</td>
<td>.000</td>
</tr>
</tbody>
</table>

**Table No. 2**

<table>
<thead>
<tr>
<th>Pair 1</th>
<th>No. of bacteria before intervention</th>
<th>Mean</th>
<th>N</th>
<th>Std deviation</th>
<th>Std error mean</th>
<th>P value</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>No. of bacteria after the intervention</td>
<td>699.20</td>
<td>25</td>
<td>322.890</td>
<td>64.578</td>
<td>.000</td>
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**Table No. 3**

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<tr>
<th>Pair 1</th>
<th>No. of bacteria before intervention</th>
<th>Mean</th>
<th>N</th>
<th>Std Deviation</th>
<th>Std. error mean</th>
<th>P value</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>No. of bacteria after intervention</td>
<td>410.64</td>
<td>25</td>
<td>397.932</td>
<td>79.586</td>
<td>0.100</td>
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**Table No. 4**

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<th>Df</th>
<th>Mean square</th>
<th>F</th>
<th>Significance p value</th>
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<tbody>
<tr>
<td>No. of bacteria</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>before intervention</td>
<td>Between groups</td>
<td>1</td>
<td>157922.000</td>
<td>1.36</td>
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<tr>
<td></td>
<td>Within groups</td>
<td>48</td>
<td>115531.833</td>
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<tr>
<td></td>
<td>Total</td>
<td>49</td>
<td></td>
<td></td>
</tr>
<tr>
<td>No. of bacteria</td>
<td>Between groups</td>
<td>1</td>
<td>24420.500</td>
<td>2.641</td>
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<tr>
<td>after intervention</td>
<td>Within groups</td>
<td>48</td>
<td>9247.167</td>
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<tr>
<td></td>
<td>Total</td>
<td>49</td>
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</tbody>
</table>

![Graph showing before and after intervention comparison between chlorhexidine and sodium fluoride](attachment://graph.png)
Discussion

Chlorhexidine is considered to be the "gold standard" antiplaque mouthwash due to its prolonged broad spectrum antimicrobial property\(^1\). It is active against a wide range of gram positive and gram negative organisms, fungi, facultative anaerobes and aerobes\(^6\). Gram positive cocci especially Streptococcus mutans seems to be sensitive to chlorhexidine\(^7\) which acts by binding to bacterial cell wall and affects its function\(^8\). In this present study, the children who used 0.2% chlorhexidine mouthwash showed significant reduction in number of colony of Streptococcus mutans at the end of 15 days. Complete inhibition of bacterial accumulation by chlorhexidine mouthwash has been reported by Schiott (1973)\(^9\). In this present study, reduction in number of colony of Streptococcus mutans was found to be statistically significant (p value = 0.000). This result well corroborates with the result obtained in previous study done by Clark and Guest (1994)\(^10\) who found greater reduction of Streptococcus mutans in the subjects who used 0.12% chlorhexidine. Similar studies done by H. Koo (2003)\(^11\), Kulkarni (2003)\(^12\) and Menendez (2005)\(^13\) found greater reduction in number of colony of Streptococcus mutans.

Sodium fluoride is the most commonly used mouthwash in children for the prevention of dental caries. Laske et al\(^14\), laske, Ripa and Green\(^15\) have reported that long term use (2-4 year) of 0.2% sodium fluoride resulted lower DMFT index. They found that DMFT index was 20.4% lower in children who rinsed the mouth with sodium fluoride. The caries preventive action of fluoride is reported to be due to its effects on the teeth, bacteria and plaque. Fluoride alters the physiochemical properties of teeth by making them more resistant to acid dissolution due to the formation of fluorapatite or fluorhydroxyapatite\(^16\). It increases the post eruptive maturation, enhances remineralization and inhibits demineralization. It also has antimicrobial action on dental plaque. Fluoride affects the potential cariogenicity of plaque in many ways. It will reduce acid production and lead to eliminate sensitive bacterial population. It also interferes with the formation of cellular polysaccharide that is required for adhesion. It has been known for a long time that fluoride inhibits glycolytic enzyme enolase which could directly reduce acid production\(^17\). Inhibition of enolase causes depletion of phosphoenolpyruvate that would reduce sugar transport which would, in turn, reduce acid reduction and glycogen synthesis. Reduced glycogen synthesis would adversely affect the ability of bacteria to survive and ultimately reduce bacterial population in the plaque\(^18\).

The result of this study demonstrated that number of colony forming units of Streptococcus mutans were reduced in 15 days on twice daily use of 0.05% sodium fluoride mouthwash. This reduction is statistically highly significant. P values in case of reduction of colony of Streptococcus mutans were 0.000 indicating that fluoride mouthwash is potent antimicrobial agent. A previous study, semi-quantitatively measuring salivary Streptococcus mutans using Dentocult S. M. Kit, reported that children using fluoride mouthwash had a significantly lower level of salivary Streptococcus mutans than those using no particular preventive measures (Yoshihara et al, 2001)\(^19\). Similar study carried out by Yoshihara and Noboru (2005)\(^20\) to evaluate long term use (2years) of sodium fluoride mouthwash on salivary level of Streptococcus mutans in school children of age 9-10 year old revealed that children who used fluoride mouthwash had significant lower level of salivary Streptococcus mutans. Similarly Kulkarni and Damle (2003)\(^12\), Sharma U (2004)\(^21\) and S.E. Jabbarifar (2005)\(^22\) found significant reduction of Streptococcus mutans and plaque with sodium fluoride mouthwash.

When comparison was done between chlorhexidine and sodium fluoride mouthwashes, no statistically significant difference was found regarding their antimicrobial activities. Kulkarni and Damle(2003)\(^12\) reported that sodium fluoride had reduced Streptococcus mutans count significantly but was less effective than chlorhexidine. Similarly, more reduction in plaque was found by 0.2% chlorhexidine mouthwash than 0.2% sodium fluoride mouthwash in the study done by Sharma U (2004)\(^21\). However in this present study, no statistically significant difference was found between these two mouthwashes.

Conclusion

Hence, it can be concluded that mechanical oral hygiene practice aids in the removal of accumulated plaque, whereas the chemical agents reduce Streptococcus mutans. Chemical agents thus can be used as adjunct to mechanical cleansing for effective oral hygiene maintenance. To conclude, the results of present study, sodium fluoride mouthwash would be a better mouthwash due to antimicrobial property besides its additional remineralization ability since Chlorhexidine is not preferred by children due to its bitter taste.

References