Evaluation of Antimicrobial Activity of Herbal Mouthwash on Streptococcus Mutans – An In vitro Study

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ABSTRACT

The presence of *Streptococcus mutans* has been consistently linked with the etiology of many oral problems such as the dental caries due to their mechanisms of plaque biofilm development and accumulation of acids. Several antimicrobial chemicals, incorporated into mouthwash, can be beneficial in the prevention of dental caries. Inhibition of each step in this process of caries formation contributes to the prevention of dental caries. Several medicinal plants have been recently found to have antimicrobial properties. Hence the extracts of these plants have been used to improve dental health and oral hygiene. The aim of the study is to evaluate the antibacterial activity of herbal mouth rinse on streptococcus mutans level – an in vitro study. The Agar well diffusion technique was used for determining the inhibitory effects of two commercially available herbal mouthwashes when compared with control 0.2% chlorhexidine. The result shows that herbal mouthwash 2 has got antimicrobial activity equal to the activity of conventional chlorhexidine mouth wash than the herbal mouthwash 1. Hence, use of herbal mouthwashes may reduce the streptococcus count due to their antimicrobial activity and could help lower the prevalence of dental problems and maintain oral hygiene in the population.

Keywords: *Streptococcus mutans*, Dental caries, Herbal mouthwash, Antibacterial.

INTRODUCTION

Dental caries is the most prevalent infectious oral disease that is caused by the bacterial breakdown of the hard tissues of the teeth (enamel, dentin and cementum). The caries process is initiated by activity of bacteria within the biofilm and dental plaque. Certain bacteria in the biofilm produce acid that attaches on to the tooth surface and destroys tooth enamel and results in tooth decay. The mouth contains a wide variety of oral bacteria, but only a few species of bacteria are believed to play an important role in causing dental caries; *Mutans streptococcus* and *Lactobacilli*. The elimination of cariogenic bacteria from the oral cavity using antibacterial agents is one of primary strategies for prevention of dental caries. Mouthwashes are widely used as adjuncts to oral hygiene and in the delivery of active agents to the teeth and gums. These mouthwashes have the ability to influence plaque formation and to alter the course of gingival formation due to their antibacterial, anti-inflammatory and cytostatic effects. Herbs are being widely explored to discover alternatives to synthetic antibacterial agents. Several antimicrobial chemicals incorporated into these herbal extracts help to improve dental health and oral hygiene without side effects. As it contains natural herbs that have natural cleansing and healing property to teeth and gums. In this article, the research was done with herbal mouthwash which has the following ingredients: i) pomegranate (*Punica granatum*) and Miswak and ii) papaya (*Carica papaya*) and pineapple (*Ananas comosus*).

Pomegranate (*Punica granatum*)

Punica granatum L, a commonly found fruit, is found to be beneficial in maintaining oral health. Pomegranates contain polyphenols, tannins, ellagic acid and anthocyanins. These compounds are powerful antioxidants that are effective against oral bacteria and have an antimicrobial activity.

Miswak

Miswak is a teeth cleaning twig made from the Salvadora persica tree. It prevents dental plaque formations and associated with multiple medicinal benefits and uses such as it prevents plaque and gingivitis, Gums strengthening, Bad breath, Strengthens teeth enamel, Tooth whitening, Anti-cariogenic, Prevents tooth decay.

Papaya (*Carica papaya*)

The bark and leaf extracts of papaya are effective in relieving gums and teeth problems.

Pineapple (*Ananas comosus*)

Pineapples can help whiten teeth by acting as a natural stain remover. Bromelain also helps break up plaque, the sticky film of bacteria that accumulates on your teeth. The bacteria in plaque produce acids that can erode the enamel of your teeth and lead to dental caries.
MATERIALS AND METHODS
In this in vitro study, isolated colonies of Streptococcus mutans of two commercially available herbal mouthwashes i) papaya and pineapple and ii) miswak and pomegranate is compared with control 0.2% chlorhexidine and saline was prepared for an antimicrobial mouth rinse test. The Agar well diffusion technique was used for determining the Minimum inhibitory concentrations (MIC). The minimum inhibitory concentration (MIC) was used to evaluate the inhibitory effects of herbal mouthwash.

Screening of antibacterial activity [Agar well diffusion technique]
Broth culture of the test organism compared to Mac Farland’s standard was prepared. Lawn culture of the test organisms were made on the Muller Hinton agar [MHA-Hi media M1084] plates using sterile cotton swab and the plates were dried for 15 minutes. Well measuring 4mm depth was made on the agar with sterile cork borer. Different concentrations of the mouthwashes were added to the wells. The plates were incubated at 37°C overnight and the zone of inhibition of growth was measured in millimetres. (2)

RESULTS
The antibacterial activity of the mouthwashes at different concentrations was screened by Agar well diffusion technique and the zone of inhibition was measured in mm diameter. The results are given in the table 1. The results of this study has shown that the herbal mouthwash 2 (miswak & pomegranate) is more significant with a zone of inhibition of 23mm at concentration 100μl than the herbal mouthwash 1 (papaya & pineapple) which had a zone of inhibition of 21mmmat 100μl concentration when compared with standard chlorhexidine 0.2% mouthwash.

Table 1: Zone of Inhibition in mm diameter

<table>
<thead>
<tr>
<th>Mouthwash</th>
<th>25μl</th>
<th>50μl</th>
<th>100μl</th>
</tr>
</thead>
<tbody>
<tr>
<td>Herbal Mouthwash 1</td>
<td>9</td>
<td>17</td>
<td>21</td>
</tr>
<tr>
<td>Herbal Mouthwash 2</td>
<td>12</td>
<td>18</td>
<td>23</td>
</tr>
<tr>
<td>Chlorhexidine 0.2%</td>
<td>18</td>
<td>23</td>
<td>28</td>
</tr>
</tbody>
</table>

DISCUSSION
According to the results of the present study, showed that herbal mouthwash can cause inhibition of bacterial growth. Bacterial plaques have been proven to have a role in the etiology of dental caries and periodontal diseases. The use of mouthwashes as disinfectants can help mechanical methods to reduce plaques (2). The association between oral diseases and the oral microbiota is well established. (8) The level of pathogenic organisms in the oral microbiota is one of the etiological factors for dental caries and other periodontal diseases. (9) According to many studies that have been conducted on the effects of mouthwashes on oral microorganisms, chlorhexidine mouth rinse provides better results in its antimicrobial efficacy against Streptococcus mutans than the herbal mouth rinses. (10) In the present study herbal mouthwash 2 had better antibacterial activity in comparison with herbal mouthwash 1 and is found significant than the chlorhexidine mouthwash.

CONCLUSION
The efficacy of herbal mouthwash was equally effective in reducing plaque and gingivitis as compared to chlorhexidine mouthwash and may be considered as a good alternative. The chlorhexidine mouthwash was reported with many side effects such as discoloration of teeth, altered taste sensation, mucosal irritation, parotid swelling, and enhanced supra-gingival calculus formation which limits its acceptability and long-term use, whereas the presently tested herbal mouthwash has no side effects apart from mild burning sensation. (12)

REFERENCES
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