Invitro Comparison of Antimicrobial Efficacy of Different Toothpastes

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Abstract:
Aim: To compare different toothpastes for their antimicrobial efficacy.

Background: Anti-microbial agents have been used as a chemotherapeutic agent to improve oral health. This in vitro study was carried out to determine antimicrobial efficacy of different toothpastes and against the oral pathogens. Five different types of toothpastes with different ingredients were taken and tested for its antimicrobial efficacy. The agar plates were made wells by using a 7mm cork borer. 5 wells were made and the dentrifices were loaded into it.

Result: From this study we come to know that the toothpaste formulation A was more effective against the microorganisms which were tested in this study.

INTRODUCTION:
Toothpaste is a paste or gel dentifrice used along with a toothbrush as an accessory to clean and maintain the aesthetics and health of teeth. Toothpaste is used to promote the oral cavity by serving as an abrasive to remove food and dental plaque from the teeth[1]. Toothpastes are considered as one of the most used and common cosmetic and hygienic materials[2]. A very significant proportion of dental problems due to the microbial infection is found in India and developing countries. There are three types of dental problems that include formation of dental plaque, dental caries and periodontal disease[3]. Dental caries is the destruction of enamel, dentin, or cementum of teeth due to bacterial activities, which if left untreated can cause discomfort, considerable pain and costs of the treatment are very high[4]. Caries formation requires four main criteria which includes caries causing bacteria, tooth surface enamel, fermentable carbohydrates such as sucrose etc. and time[5]. Streptococcus mutans is recognized as one of the main opportunistic pathogens of dental caries that can demineralize the enamel[6]. In addition to Streptococcus mutans, other microflora like Escherichia coli and Candida are also associated with active caries lesions[3]. Toothpaste is the most common form of caries control used today[4]. Toothbrushing and flossing are the difficult tasks, and most of the patients are unable to completely remove plaque in all surfaces of the teeth[6], but still toothbrushing is very significant to protect the oral cavity. Periodontal disease is also a bacterial disorder which may result in tooth mobility and loss of tooth by affecting the supporting structures. The traditional periodontal pathogens includes Bacteroides, Streptococci and Spirochetes[9]. Gingivitis is an inflammatory condition of gum, which is the most common form of periodontal disease. The possible pathogens responsible for this disease are Streptococci and Spirochetes[10]. Regular toothpaste formulations mostly contain a combination of fluorides and detergents, mainly sodium dodecyl sulphate to increase the efficacy of brushing and thereby preventing diseases[7]. In commercial toothpastes available today, there are few different main ingredients which have varying effects on the oral flora population. These ingredients include triclosan, sodium lauryl sulfate, chlorhexidine, sodium fluoride, sodium monofluorophosphate, zinc citrate trihydrate, and hexetidine. It is very common for toothpastes to contain different main ingredients, and this leads to an inquiry concerning the efficacy of these various ingredients and the toothpaste product as a whole[8]. This study focuses on the anti microbial efficacy of different toothpastes against S. mutans, C. albicans, E. coli, Lactobacillus, Enterococcus.

MATERIALS AND METHOD:
Microorganisms:
Cultures of Candida albicans, Escherichia coli, Streptococcus mutans, Enterococcus and Lactobacillus were obtained from different clinical samples. Cultures of Candida albicans, E. coli were cultured in nutrient broth (Hi-Media) at 37°C for 24 h while Candida albicans and Streptococcus mutans were cultured in brain heart infusion broth (Hi-Media) at 37°C for 24 h.

INTRODUCTION:
Evaluation of dentrifices:
The research was aimed at knowing the brands of toothpastes that are mostly used. As a result, five toothpastes were selected for assessment of their in vitro antimicrobial activities. They were purchased from local markets in Chennai, Tamilnadu, India. The composition of these toothpastes are given in table 1. The selected dentrifices solutions were taken in a calculated amount of toothpastes (2.0 gms). Nutrient agar and brain heart infusion plates were prepared to assess the antimicrobial activity of dentrifices against the pathogens. All the chemicals and reagents used were of analytical grade.

Antimicrobial essay:
The antimicrobial activity of different concentrations of the dentrifices was determined by modified agar well diffusion method. In this method, nutrient agar plates were seeded with 0.5 mL of 24h broth cultures of each isolate (brain heart infusion agar was used for Streptococcus mutans strain). The plates were allowed to dry for an hour. A sterile 7mm corkborrer was used to cut one central and five wells at equidistance in each of the plates. 0.2g of the toothpaste was introduced into each of the five wells. The plates were incubated at 37 C for 24h. The antimicrobial activity was evaluated by measuring the diameter of the zones of inhibition. All the plates were made five times and all the experiments were repeated five times.

Table 1 – Toothpastes and their ingredients listed on their packages.

<table>
<thead>
<tr>
<th>Toothpastes</th>
<th>Ingredients as listed on packages</th>
</tr>
</thead>
<tbody>
<tr>
<td>A</td>
<td>Sorbitol, Water, Hydrated silica, Sodium lauryl sulfate, PEG-32, Flavor, Cellulose gum, Sodium fluoride, Sodium saccharin, Cl - 17200</td>
</tr>
<tr>
<td>B</td>
<td>Fluoride, Silica, SD Alcohol, Zinc Citrate Trihydrate, Cellulose gum, Titanium dioxide, Sodium saccharin, Sodium lauryl Sulfate, Flavor, Sorbitol.</td>
</tr>
<tr>
<td>C</td>
<td>Calcium Carbonate, Herbal extract(Piper nigrum, Piper longum, Zanthoxylum alatum, Zingber officinale), Red ochre, Flavor, Sorbitol, Sodium lauryl sulfate, Methyl paraben, Propylparaben, Formalin.</td>
</tr>
<tr>
<td>D</td>
<td>Miswak, Neem, Pomegranet, Triphala(Emblica officinalis, Terminalia chebula, Terminalia bellirica),Tumpara, Babool, False black pepper and leaved chaste tree</td>
</tr>
<tr>
<td>E</td>
<td>Glycerine, Water, Dicalcium phosphate, Extracts of Ginger, Black pepper, Long pepper, Terminalia Chebula, Gooseberry,Licorice, Camphor, Menthol and Clove oil, Sodium carboxymethyl cellulose,Sodium Benzoate Sodium saccharin, Red ochre, Calcium carbonate,</td>
</tr>
</tbody>
</table>
The microbes responsible for the formation of plaque are S. mutans, when compared to S. mutants and C. albicans, Lactobacillus, when compared to E. coli. Maintenance of good oral hygiene is the main factor to prevent the dental diseases. To maintain the oral health, it is necessary to brush everyday. The major problem of the dental problems is the formation of dental plaque. The dental plaque slowly causes the destruction of enamel. The microbes responsible for the formation of plaque are S. mutants, E. coli and C. albicans.

This study focuses the in vitro comparison of antimicrobial activity of different toothpastes. From the data collected, among all the toothpastes investigated the toothpaste formulation A appears to be most effective against all the five microorganisms. This is due to the ingredients present in the toothpaste formulation A. Moreover the active ingredient in the toothpaste formulation A is sodium monofluorophosphate (0.76%). The toothpaste formulation B was next to toothpaste formulation A, it showed good antimicrobial activity against all the microorganisms. Here the active ingredient is Sodium fluoride (0.24%). The fluoridated products toothpaste formulations A and B showed antimicrobial efficacy against all the microorganisms. The effectiveness of the fluoridated toothpaste varies with its concentration. In this study the toothpaste formulation A has high concentration than B. In a previous study, Jenkins had stated that fluoridated toothpaste formulation have shown 30 – 70% reduce caries when compared to no fluoride therapy. The toothpaste formulations C, D and E showed no antimicrobial activity against E. coli. The toothpaste formulation E is a herbal based toothpaste, and it exhibited good antimicrobial activity against S. mutans and C. albicans, but showed less efficiency against Enterococcus and Lactobacillus. The reason behind its efficacy against the microbes is the ingredients present in the toothpaste formulation E which are mostly herbal products and the usage of chemical substances were reduced. The toothpaste formulation D showed the least efficiency against all the microorganisms. This may be due to the ingredients present in that toothpaste formulation which mostly consisted of fruits and natural products.

In this study the herbal formulations were equally efficient to the fluoride formulations but not greater than that. The usage of natural substances for treatment of the diseases increased nowadays. This is because the contribution of the herbal products is comparatively more than modern products. The effective antimicrobial activity of herbs is due to the presence of secondary metabolites such as flavonoids, polyphenols, alkaloids and lectins. Nowadays Fluorides are used in the toothpastes to prevent dental caries.

**RESULTS:**

Table 2 - Antimicrobial activity of dentrifices against the test organisms

<table>
<thead>
<tr>
<th>Toothpaste</th>
<th>E. coli</th>
<th>S. mutans</th>
<th>C. albicans</th>
<th>Enterococcus</th>
<th>Lactobacillus</th>
</tr>
</thead>
<tbody>
<tr>
<td>A</td>
<td>24</td>
<td>24</td>
<td>28</td>
<td>27</td>
<td>28</td>
</tr>
<tr>
<td>B</td>
<td>20</td>
<td>21</td>
<td>26</td>
<td>23</td>
<td>26</td>
</tr>
<tr>
<td>C</td>
<td>0</td>
<td>20</td>
<td>25</td>
<td>19</td>
<td>0</td>
</tr>
<tr>
<td>D</td>
<td>0</td>
<td>18</td>
<td>23</td>
<td>18</td>
<td>15</td>
</tr>
<tr>
<td>E</td>
<td>0</td>
<td>24</td>
<td>26</td>
<td>22</td>
<td>17</td>
</tr>
</tbody>
</table>

From the results of the above investigation, the toothpaste formulation A had maximum zones of inhibition against all test organisms, when compared to other toothpaste formulations. The toothpaste formulation A showed greater zone of inhibition against Candida albicans, Enterococcus, Lactobacillus, when compared to S. mutants and E. coli.

**DISCUSSION:**

Maintenance of good oral hygiene is the main factor to prevent the dental diseases. To maintain the oral health, it is necessary to brush everyday. The major problem of the dental problems is the formation of dental plaque. The dental plaque slowly causes the destruction of enamel. The microbes responsible for the formation of plaque are S. mutants, E. coli and C. albicans.

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In this study the herbal formulations were equally efficient to the fluoride formulations but not greater than that. The usage of natural substances for treatment of the diseases increased nowadays. This is because the contribution of the herbal products is comparatively more than modern products. The effective antimicrobial activity of herbs is due to the presence of secondary metabolites such as flavonoids, polyphenols, alkaloids and lectins. Nowadays Fluorides are used in the toothpastes to prevent dental caries.

**CONCLUSION:**

From this study we conclude that, the toothpaste formulation A is more effective against all the microorganisms tested and helps in maintaining oral hygiene when compared to the other toothpaste formulations. The effectiveness of toothpaste formulation A was due to the presence of the active ingredient Sodium monofluorophosphate. Hence we conclude that the presence of this ingredient in toothpaste increases the effectiveness of that toothpaste formulation.

**REFERENCES:**

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