Efficacy of caries removal of Papain and Bromelain based caries removal
gel decorated with silver nanoparticles- Patent

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Abstract: Introduction
Conventionally caries has been removed by drilling using dental burs. But drilling causes anxiety, pain in patients. Another method of caries removal is Chemicomechanical method using gel. Papain has been used for this purpose and studied in literature. This study is about novel patented caries removal gel and its efficacy.

Aim
To compare the efficacy of caries removal time taken for caries removal and surface changes of patent group after caries removal with that of drilling and commercial caries removal gel under Scanning electron microscope.

Materials and methods
• Papain and bromelain based gel decorated with silver nanoparticles has been prepared. Characterisation has been done based on EDX, FTIR and SEM.
• Three groups were taken- patented gel, drilling bur and commercially available caries removal gel.
• Caries removal was done on extracted molar with class 1 caries. Time taken for caries removal is recorded. After caries removal surface changes of tooth was checked under SEM.

Results
Time taken for caries removal was less in drilling group compared to that of other two groups. Efficacy of caries removal of equal in all groups. SEM results favored patented caries removal gel.

Conclusion
Patented caries removal gel can be considered as a method for caries removal as surface changes and marginal gap is more in drilling group.

Keywords: caries, drilling, papain, bromelain, drilling.

1. Introduction
Dental caries is one of the most common diseases affecting the oral cavity in the population of the world. The removal of caries has been done conventionally using dental burs (1). This drilling method of caries removal though is fast, it definitely has certain adverse effects. It can cause anxiety, pain, fear especially in children (2). Not just that there is every possibility that pulpal damage can occur.

Different other methods of caries removal have been tried in the past. The mechanical rotary or non-rotary instruments, chemo- mechanical caries removal system and lasers (3). The non-rotary, non-invasive techniques include air-abrasion, air-polishing, ultrasonic and sonic abrasion. The chemo-mechanical caries removal system, based on the principle of minimal invasive dentistry involves the application of substances like Caridex and Carisolv which is referred to as the Chemico mechanical method (4). This method involves the usage of caries removal gels for the purpose of removing caries. The Chemo-mechanical caries removal technique involves the application of chemical agents, to cause a selective softening of the carious dentine and facilitate removal by
gentle excavation(5). Many caries removal gels are available in the market. The important ingredient in these gels is papain. Papain is an enzyme derived from the fruit of papaya. It is a proteolytic enzyme causing breakdown of complex proteins into simpler groups(6). This concept was applied for removal of caries. Papacarie was introduced as a new method for CMCR. This product contains active ingredients, including papain and chloramines, with bactericidal, bacteriostatic, and anti-inflammatory properties. Papain can specifically digest infected or dead tissues because their collagen molecules are partially degraded and they lack α1-anti-trypsin, which normally inhibits protein digestion in healthy tissues(7).

The present study is about a novel self made caries removal gel which is based on papain, Bromelain decorated with silver nanoparticles used for removal of caries. Bromelain is an enzyme derived from the plant of pineapple(8). It is said to be a potent proteolytic enzyme compared to papain. Silver nanoparticles were biosynthesised using papain and Bromelain and formulated as a gel. Bromelain contains thiol endopeptidases and other components, such as, peroxidases, cellulases, phosphates, glucosidases, glycoproteins, and carbohydrates(9). Bromelain is stable at pH 3.0 to 6.5. Bromelain when applied topically helps in the removal of burnt debris and accelerates healing(10). Silver nanoparticles are well known for their antimicrobial action and increasing the efficacy of the prepared formulation(11).

The aim of this study was to compare the efficacy and time taken for caries removal of self prepared caries removal gel with that of drilling bur based caries removal and commercially available caries removal gel.

2. Materials And Methods

Preparation of caries removal gel:
0.5 gram of Papain and bromelain powder which was readily available in the market was taken. To this 100 ml of distilled water was added. To this solution, 0.1gram of silver nitrate powder was added and placed in the shaker for 24 hours. After 24 hours, a change in color of the solution was seen indicating that biosynthesis of silver particles had occurred. The supernatant was removed and residue was further centrifuged to obtain papain and bromelain based silver pellets.

The silver pellets were added to a gel preparation which contained carbopol, carboxymethyl cellulose and sodium benzoate to obtain the final caries removal gel.
Characterization of prepared gel:

**Energy Dispersive X ray Spectroscopy (EDX):** This is a method to detect the elemental composition of different compounds. It is important to determine the elements present in the prepared gel. This method was used for the same purpose. The elements detected were carbon, oxygen and silver particles majorly. The percentage of oxygen present in the gel was 43.1%, carbon was 38% and silver particles were 19%. This acted as a proof for the presence of silver particles in the prepared gel.

**Fourier Transform Infrared Spectroscopy:** also known as FTIR Analysis or FTIR Spectroscopy. FTIR analysis helps to understand and characterize materials and products. FTIR offers quantitative and qualitative analysis for organic, inorganic samples and polymers. Fourier Transform Infrared Spectroscopy (FTIR) identifies chemical bonds in a molecule by producing an infrared absorption spectrum. The spectra produce a profile of the sample, a distinctive molecular fingerprint that can be used to screen and scan samples for many different components. FTIR is an effective analytical instrument for detecting functional groups and characterizing covalent bonding information. The majority of elements detected by this method in the prepared gel was carbon, oxygen, silver, polymers and nitrogen.

**Scanning Electron Microscope (SEM) analysis:** Scanning electron microscopic view of the particles of the gel showed that the silver particles synthesized were more or less spherical with size ranging from 10 to 100nm.

**Removal of caries:** Fifteen extracted mandibular permanent molar teeth with class 1 caries were taken and randomly divided into three groups. The three groups included were drilling burs, self made patented caries removal gel and commercially available caries removal gel. Caries from the extracted teeth were removed and the time taken for caries removal was recorded in each of the groups. The efficacy of caries removal was evaluated by two trained endodontists and cross checked by a third operator. The teeth were then filled with composite restorative material. The restored teeth were mounted on acrylic and sectioned to a thickness of 0.5mm using hard tissue microtome. The sections were viewed under scanning electron microscope, analyzed and evaluated further.

3. Results

**Results of EDX:**

EDX microanalysis is a technique of elemental analysis that is based on the generation of characteristic X Rays in atoms of the specimen by the incident beam electrons. The EDX analysis of self prepared caries removal gel
showed the presence of oxygen as 43.1%, Carbon was 38% and silver was 19%. This shows the presence of silver particles in the compound.

Results of FTIR:

![Graph showing FTIR analysis of caries removal gel](image)

The most common use is in the identification of unknown materials and confirmation of production materials. The above graph shows the presence of carbon, silver as major ingredients in the prepared gel.

Time taken for caries removal: Time taken for caries removal in the drilling group was 1.5 minutes.

<table>
<thead>
<tr>
<th>S No.</th>
<th>Group</th>
<th>Mean Time taken in minutes(S.D)</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>Patent Gel</td>
<td>3(+/-.0.495)</td>
</tr>
<tr>
<td>2</td>
<td>Commercial</td>
<td>3(+/-.0.495)</td>
</tr>
<tr>
<td>3</td>
<td>Drilling</td>
<td>1.5(+/-.0.395)</td>
</tr>
</tbody>
</table>

Table 1: Table showing time taken for caries removal

Efficacy of caries removal:
A trained operator performed all caries removal and restorations. The efficacy of caries removal was evaluated by tactile and visual inspections performed by another trained dentist, who was blinded to the group status. For tactile criteria, complete caries removal was defined as a smooth passage of a blunt explorer and the absence of a catch or a “tug-back” sensation (12).

SEM analysis of the sections:

![SEM view of patent gel](image)

![SEM view of patent gel](image)
4. Discussion

Dental caries is a disease that affects the majority of the global population (13). The removal of caries was traditionally done by drilling, using dental burs. But this can cause fear in patients, pain and anxiety which makes it very difficult for the operator to complete a restorative procedure. In order to avoid and overcome this, different methods of caries removal have been tried and evaluated (14). One such method is the Chemico mechanical method where in caries removal gel is used for the purpose.

Different caries removal gels are available in the market but majority of them do not show much success (15). The key ingredient in such gels is majorly papain, a proteolytic enzyme obtained from the plant of papaya. In the present study a novel caries removal gel has been formulated which is based on papain (16), Bromelain which is a newer and more potent, effective proteolytic enzyme. To this, silver nitrate has been added for biosynthesis of silver nanoparticles. Silver nanoparticles are known for their antimicrobial efficacy and to enhance the efficacy of caries removal of papain and Bromelain (17). Biosynthesis of nanoparticles, especially silver nanomaterials from plant extracts or organic sources has been receiving huge interest because of their plentiful abilities and a wide range of bioactive reducing metabolites (18). Plants are known as highly preferable sources for synthesizing nanoparticles (14). Compared with bacteria and algae, plants are more resilient to metal toxicity, thereby offering a green alternative for synthesizing of silver nanoparticles (19).

The characterisation of the gel has been done and caries removal efficacy, time taken for caries removal was compared with that of drilling burs and commercially available caries removal gel. The time taken for caries removal was more in commercially available gel and patented caries removal gel group compared to drilling burs group. However there was no significant difference between the patented group and commercially available group.

The efficacy of caries removal was equal in all the three groups with no significant difference between the groups. The study by Kemporn et al, aimed to evaluate the performance of Papacarie in comparison with the drilling method. According to the author no significant difference was seen between the two groups (7). A study by Vamsi et al, aimed to evaluate and compare bromelain with papain as the chemomechanical caries removal agent in relation to their efficacy. According to the author, that bromelain was more effective in amount of caries removal than papain. The amount of remaining demineralized dentin was found to be lower in bromelain group than the papain group and time taken for the carious dentin removal was almost found similar in both bromelain...
and papain groups(20). Pallvi Goomer et al. conducted a clinical trial to compare different methods of caries removal in terms of efficacy, time taken and pain during caries removal. Chemical Mechanical removal of caries with Carisolv was found to be effective measure of caries removal and could be considered as viable alternatives to painful procedures like airrotor in management of dental caries especially in children(21).

The teeth were restored with composite restorative material and further sectioned to view under scanning electron microscope. The SEM analysis results showed that the gap between restoration and tooth structure in self prepared gel group was around 16.97μm-17.89μm, in the commercially available gel it was,43.8μm- 55.6μm where as in the drilling group it ranged between 100.1μm-256μm. Also the presence of microcracks were evident and prominent in the drilling group compared with that of self made caries removal gel.

5. Conclusion
Efficacy of caries removal is equal in all the three groups. Time taken for caries removal is though higher for patent group and commercial gel, the surface changes and marginal gap is more in drilling group. Although it is difficult to employ any one single method to achieve clinical excellence, patented gel shows patient friendly and promising results.

References

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