An investigation into the potential use of Dennkur™ as an intra-canal irrigant and medicament in Endodontics

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Abstract
Achieving complete asepsis within the root canals during Endodontic therapy is an elusive goal of every dental practitioner. While many commercial root canal irrigants and medicaments are employed to achieve the same, they are not without undesirable side effects. A fair amount of scientific interest is currently being generated by the potential of traditional herbal agents and their use in achieving asepsis in the root canal. Research in nearly all branches of medicine exists documenting the potential of Turmeric as an antimicrobial agent. Curcumin, the active ingredient in Turmeric, has shown fair results in its antimicrobial action in Periodontics. Our in vitro study explores the antimicrobial potential of a curcumin-containing product Dennkur™ against endodontic pathogens E. faecalis and C. albicans by the Agar cup method.

Keywords: Curcumin, Dennkur, E. faecalis, C. albicans

Introduction
The success of endodontic treatment depends on the eradication of microorganisms from the root-canal system and prevention of reinfection. The root canal is shaped with hand and rotary instruments under constant irrigation to remove inflamed and necrotic tissue, microbes/biofilms, and other debris from the root-canal space. Several studies using advanced techniques such as microcomputed tomography (CT) scanning have demonstrated that proportionally large areas of the main root-canal wall remain untouched by the instruments, emphasizing the importance of chemical means of cleaning and disinfecting all areas of the root canal. Presently the most reliably and universally used root canal irrigants in endodontic practice are Sodium hypochlorite and Chlorhexidine gluconate. Calcium hydroxide is considered as a gold standard for use as an intracanal medicament in endodontic cases with suspected infection.

Due to the clinical drawbacks encountered with the use of the above irrigants and medicaments, research has shifted in the direction of traditional medicine in search for an ideal irrigant and medicament. The rhizome Turmeric (Curcuma longa) is one such traditional Asian herb that has sparked a great deal of interest among researchers. The active ingredient in Turmeric is Curcumin. Curcumin’s potent anti-inflammatory properties have lead to active research on its use for a variety of inflammatory conditions, including postoperative inflammation, arthritis, uveitis, inflammatory pseudotumors, dyspepsia, irritable bowel syndrome, inflammatory bowel disease, pancreatitis, and Helicobacter pylori infection. Most studies are promising and further exploration of curcumin’s therapeutic value for inflammatory conditions is warranted (Jurenka, 2009).

Dennkur™ is a mouth-dissolving pastille that contains Curcumin with its natural antiseptic, antibacterial and anti-inflammatory properties. It slowly dissolves in mouth giving a longer contact time for the local action on the floor of mouth. The natural antibacterial action of curcumin reduces the bacterial load in the oral cavity. Each soft gelatine pastille (Lozenge) contains Curcuma Longa (Curcuma) -100mg, excipients and flavouring agents.

Dennkur™ is believed to have the same antibacterial action as chlorhexidine and hence an investigation into its potential use as an endodontic irrigant and intracanal medicament was carried out. The aim of this study was to test the antibacterial action of Dennkur™ against endodontic pathogens Enterococcus faecalis and Candida albicans.

Materials and Method
Group 1 was Sterile Nutrient Agar (for Enterococcus Faecalis).
Group 2 was Sterile Sabourauds Agar (for Candida Albicans)
The following five samples were compared in the following in vitro study
1. 3% Sodium Hypochlorite (Prime Dental Products)
2. 2% Chlorhexidine (Stedman Pharmaceuticals Pvt Ltd)
3. Calcium Hydroxide (Prime Dental Products)
4. Curcumin (Dennkur™ Lozenges(I tablet in 10 mL distilled water
5. Normal saline

Means used were:
1. Sterile Nutrient Agar (for Enterococcus Faecalis)
2. Sterile Sabourauds Agar (For Candida Albicans)
Microbial cultures used were:
1. Enterococcus faecalis (NCIM no 5025)
2. Candida albicans (ATCC No. 10231)

**Armamentarium used:**
1. Petri plate
2. Agar
3. Cork boner

**Procedure:** The method employed to carry out the following in–vitro study was Agar Cup Method. The equipment was sterilized and a sterile environment in order to prevent cross contamination. Preparation of the culture mediums, i.e. Sterile Nutrient Agar (for Enterococcus Faecalis) and Sterile Sabourauds Agar (for Candida Albicans) was carried out and it was kept for boiling to maintain asepsis. After appropriate temperature was reached they were allowed to cool down to lukewarm. The culture medium (Sterile Nutrient Agar) was removed by the use of pipette and was placed between two Bunsen burners, this was done to maintain a zone of absolute sterilization. 25 ml of sterile culture medium with an addition of 1 mL of cultural strain (Group I containing Enterococcus Faecalis) was poured in a petri plate and allowed to solidify to room temperature.

The same procedure was followed for second culture medium (Sterile Sabouraud’s Agar) which was mixed with Group II strain (containing Candida Albicans) and poured into sterile petriplates and was allowed to cool at room temperature. The cork boner was placed in the aseptic zone between two Bunsen burners and 5 wells of 8 mm diameter were punched. After the wells were made, 0.1 mL of undiluted sample was introduced in the respective well.

Saline was used as a control. After the incubation period of 24hrs, the plates were removed and zones of inhibition were recorded. The experiment was repeated 3 times (each time after a gap of 48hrs) and zones of inhibition were recorded using Vernier Calipers in mm (millimeters).

**Fig. 1:** Action of Dennkur, calcium hydroxide, chlorhexidine sodium hypochlorite on E.faecalis (Group 1)

**Fig. 2:** Zones of inhibition demonstrated on action with E.faecalis (Group 1)

**Fig. 3:** Action of Dennkur, calcium hydroxide, sodium hypochlorite, chlorhexidine on Candida Albicans (Group 2)

**Fig. 4:** Zones of inhibition demonstrated on action with C.Albicans (Group 2)
Results

Table 1: Enterococcus Faecalis on Nutrient Agar Plate (Group 1) - Zone of Inhibition (mm)

<table>
<thead>
<tr>
<th>Materials</th>
<th>Undiluted</th>
<th>1:2 dilution</th>
<th>1:10 dilution</th>
</tr>
</thead>
<tbody>
<tr>
<td>Sodium, Hypochlorite 3%</td>
<td>13mm</td>
<td>11.5mm</td>
<td>8mm</td>
</tr>
<tr>
<td>Calcium Hydroxide</td>
<td>13mm</td>
<td>12mm</td>
<td>0</td>
</tr>
<tr>
<td>Chlorhexidine 2%</td>
<td>21mm</td>
<td>18mm</td>
<td>18mm</td>
</tr>
<tr>
<td>Dennkur™</td>
<td>0</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>Saline</td>
<td>0</td>
<td>0</td>
<td>0</td>
</tr>
</tbody>
</table>

Table 2: Candida Albicans on a Sabouraud’s Agar plate (Group 2) Zone of Inhibition (mm)

<table>
<thead>
<tr>
<th>Materials</th>
<th>Undiluted</th>
<th>1:2 dilution</th>
<th>1:10 dilution</th>
</tr>
</thead>
<tbody>
<tr>
<td>Sodium, Hypochlorite 3%</td>
<td>16mm</td>
<td>13mm</td>
<td>0</td>
</tr>
<tr>
<td>Calcium Hydroxide</td>
<td>14mm</td>
<td>12mm</td>
<td>0</td>
</tr>
<tr>
<td>Chlorhexidine 2%</td>
<td>15mm</td>
<td>12mm</td>
<td>12mm</td>
</tr>
<tr>
<td>Dennkur™</td>
<td>0</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>Saline</td>
<td>0</td>
<td>0</td>
<td>0</td>
</tr>
</tbody>
</table>

Discussion

By virtue of their mechanism of action, present day root canal irrigants and medicaments nearly effectively clean the root canal. They are able to disinfect and penetrate dentin and its tubules, offer long-term antibacterial effect (substantivity), remove the smear layer, and should be non-antigenic, nontoxic and non-carcinogenic. In addition, they require to have no adverse effects on dentin or the sealing ability of filling materials. Furthermore, they should be relatively inexpensive, convenient to apply and cause no tooth discoloration. The irrigants currently used that are considered gold standard are Sodium Hypochlorite & Chlorhexidine. The universally used and accepted intra-canal medicament is Calcium Hydroxide.

The most effective method for eliminating *E. faecalis* from the root canal space and dentinal tubules is by the use of Sodium hypochlorite (NaOCl) and Chlorhexidine in a gel or liquid concentration form. But the disadvantages of sodium hypochlorite are its unpleasant taste, toxicity, and its use is suspected in weakening of the tooth structure by decreasing the hardness and structural integrity of the dentin within the root canal.\(^{(5)}\)

Chlorhexidine gluconate is a cationic biguanide that seems to act by adsorbing onto the cell wall of the microorganism and causing leakage of intercellular components. At low concentration, it has a bacteriostatic effect and at higher concentrations it has a bactericidal effects. It has a lower cytotoxicity as compared to sodium hypochlorite. However, unlike NaOCl, it cannot dissolve organic substances and necrotic tissues present in the root canal system. In addition, it is unable to kill all bacteria and is not effective in removing the smear layer as well. No adverse effects have been published regarding Chlorhexidine being used as an irrigant or intra-canal medicament. However, the direct effect in an *in vitro* test on human stem cells of apical papilla showed lack of viable cells after its use to remove the smear layer. The disadvantages of hypochlorite and calcium hydroxide have motivated researchers to explore the potential of different chemicals and herbs as root canal irrigants.\(^{(6)}\)

Calcium hydroxide was originally introduced to the field of endodontics by Herman in 1920 as a pulp-capping agent, it was found effective as intra-canal medicament.\(^{(7)}\) The antimicrobial activity of Ca(OH)\(_2\) is related to the release of hydroxyl ions in an aqueous environment (Siqueira 2001). Hydroxyl ions are highly oxidant free radicals that show extreme reactivity with several biomolecule. The lethal effects of hydroxyl ions on bacterial cells are probably due to (Siqueira & Lopes 1999) damage to the bacterial cytoplasmic membrane, protein denaturation and damage to the DNA.

The major disadvantage of Calcium hydroxide is its ineffectiveness in completely eliminating microorganisms. Various studies done by DiFiore et al. (1983), Haapasalo & Orstavik (1987), Safavi et al. (1990) have shown the ineffectiveness of Calcium hydroxide against *E. Faecalis* and *C. Albicans*.

Thus, looking at the various disadvantages and adverse effects of the mentioned irrigants and medicaments, there is a need for alternatives that would endeavor to eliminate residual microorganisms without causing any adverse effects. Various Indian herbal and ayurvedic products such as extracts from Neem (Azadiracta Indica), Ashwaganda (Withania Somnifera) and Curcumin (Curcuma Longa) are being investigated as alternatives.

Turmeric or Curcumin with a chemical formula (1,7-bis(4-hydroxy-3-methoxyphenyl)-1,6-heptadiene-3,5-dione) and other curcuminoids constitute the main phytochemicals of curcuma longa L. (Zingiberaceae family) rhizome with common name of turmeric. Several Studies have reported the broad spectrum antimicrobial activity for curcumin including antibacterial, antiviral, antifungal and antimalarial activities.\(^{(2,3,4,6,7,8)}\) An in vitro study evaluated the efficacy of three intracanal medicaments 10% and 20% of turmeric, calcium hydroxide, 1% chlorhexidine gel and 1% chlorhexidinegel with 10% calcium hydroxide against *E. Faecalis* in necrotic primary teeth and concluded that chlorhexidine with or without calcium...
hydroxide was more effective than calcium hydroxide alone (Kumar, Hemanshi, 2013).

Neelakantan et al. found that curcumin had significant antibacterial activity against E. faecalis. (9) Haukvik et al. studied the phototoxic effects of curcumin against gram positive bacteria like E. faecalis, streptococcus intermedius and gram negative bacteria E. coli were investigated in aqueous preparations. (10)

Prasanna et al. (2011) conducted an in vitro study to evaluate the antimicrobial efficacy of curcumin against E. faecalis, C. Albicans considering sodium hypochlorite (3%) as reference for comparison. The result of his study revealed that curcumin had significant antibacterial activity against E. faecalis he concluded that the antibacterial activity of curcumin was similar to sodium hypochlorite and thus herbal medicine can be used in endodontics for root canal failure. (11)

Lívia N. Dovigo, Ana Cláudia Pavarina, Ana Paula D. Ribeiro (2011) also carried out an in vitro investigation and found that Curcumin was effective against Candida Albicans. (12)

But Hegde and Kesaria (13) found that Turmeric showed no antibacterial action against E. Faecalis in an in vitro study by agar cup method testing antibacterial action of several new agents for use as root canal irrigants.

In our study, we observed that the antibacterial action of the test sample was nil in all dilutions, the readings for the test sample, sodium hypochlorite, chlorhexidine, calcium hydroxide were tabulated and statistically analysed by ANOVA showed the results highly significance (p>0.05).

The active component of Dennkur™ (Curcumin) was claimed to have antiseptic as well an anti-inflammatory activity. It was also suggested to have endodontic use and claimed to be as effective as chlorhexidine. Since no specific study was done to show the effectiveness of the product (Dennkur™) against endodontic flora, the present study was carried out to investigate the antibacterial activity of this product against E. Faecalis and C. Albicans. In our study, Dennkur™ showed zero zone of inhibition against E. Faecalis & C. Albicans as compared with the gold standard irrigants and medicament, which showed zones of inhibition ranging from 8-16mm (as shown in Table 1 and Table 2).

Conclusion
Curcumin has shown a good potential in terms of antibacterial activity to warrant further research. However the commercial product Dennkur™ in its present formulation appears to be ineffective in showing the similar antibacterial activity. While Dennkur™ shows use as oral lozenges, its activity against endodontic pathogens seems to indicate that this commercial product in its present formulation does not have any endodontic application.

Disclaimer
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References