



Research Article

COMPARATIVE EVALUATION OF ANTIMICROBIAL ACTIVITY OF PINEAPPLE EXTRACT (BROMELAIN), 2% CHLORHEXIDINE, 5.25% SODIUM HYPOCHLORITE ALONE AND IN COMBINATION WITH BROMELAIN AS ROOT CANAL IRRIGANTS AGAINST ENTEROCOCCUS FAECALIS ATCC (29212): AN IN VITRO STUDY

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ABSTRACT

Debridement and disinfection of the root canal system is a critical step in endodontic treatment. Most of the irrigants presently used in the endodontic treatment can have an impact on the microbes surviving in the biofilm but none of them are accomplish all of the required tasks. Researches are going on in order to produce an endodontic irrigant having ideal properties.

Aim and Objectives: Successful root canal treatment involves the complete elimination of microorganism from the root canal and the three-dimensional obturation of the canal space. Enterococcus faecalis is the most commonly found bacteria in failed root canal. Chemical irrigation of canals along with biomechanical preparation helps in the elimination of microorganisms. The present study was aimed to evaluate the antimicrobial effect of Pineapple Extract (Bromelain), 2% Chlorhexidine, 5.25% Sodium Hypochlorite alone and in combination with Bromelain.

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INTRODUCTION

Micro-organisms in the root canal are responsible for pulp and periradicular infections.^[1] The aim of the root canal therapy is to eradicate microorganisms from the root canal in order to provide appropriate environment for tissue healing.^[1,2] Enterococcus faecalis generally isolated from failed root canals. It has the ability to invade dentinal tubules and does not depend on the survival of other bacteria.^[1-5] It is an anaerobic gram-positive bacterium responsible for 80–90% of enterococcal infection. It plays an essential role in persistent failure of endodontic therapy. Its virulence is attributed to its resistance to intracanal medicaments^[2] Negative bacterial culture in root canal space helps in healing of periapical areas. Only with mechanical cleaning, all microorganisms cannot be eliminated; hence, appropriate irrigation with intracanal medicament is advised. Root canal irrigation helps in removal of bacteria where instrumentation is inaccessible.^[2,6]

Various chemical root canals irrigants used successfully. Sodium hypochlorite has been the gold standard for irrigation because of its ability to dissolve organic matter and high antimicrobial potential. It is difficult to imagine successful irrigation of the root canal without hypochlorite.¹³

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The high pH of NaOCl influenced by the release of hydroxyl ions, alters the integrity of the cytoplasmic membrane of microorganisms, causes irreversible enzymatic inhibition, biosynthetic alterations in cellular metabolism and phospholipid degradation by lipid peroxidation.¹⁵ However, there are certain major drawbacks associated with the use of sodium hypochlorite i.e irritant to periapical tissues, stains instruments, unpleasant taste, high toxicity, corrosion of instruments, inability to remove smear layer, burning of surrounding tissues and reduction in elastic modulus and flexural strength of dentin. Chlorhexidine (CHX) is a broad spectrum antimicrobial agent that has substantive antimicrobial activity and relatively low toxic effects. However does not dissolve organic tissues. In vitro studies have shown CHX to exhibit sustained antimicrobial activity in the root canal for some time after being used as an endodontic irrigant. Therefore, CHX has been suggested as a root canal irrigant owing to its unique ability to bind to dentin, its effectiveness as an antimicrobial agent, and its substantivity in the root canal system.

Pineapple or ananas comosus belongs to the family of bromeliacea. It has been widely used as a therapeutic plant in several resident cultures and these therapeutic qualities of pineapple are accredited to bromelain, which is an elementary extract from pineapple that contains, along with other compounds, various proteinases. Bromelain has shown to

exhibit various fibrinolytic, antiedematous, antithrombotic, and anti-inflammatory activities both *in vitro* and *in vivo*.⁷ Bromelain is a mixture of Phosphatases, Thiopeptidases, such as ananain and Comosain, Cellulases, Peroxidases, Glucosidases, Glycoproteins, Proteinase inhibitors, such as cystatin. Due to its high medicinal value it is used by people as medicine.^{10,11,12}

METHODOLOGY

Five groups of the study was,

Group 1) Pure pineapple extract (Bromelain)

Group 2) 2% CHX

Group 3) 5.25% NaOCl

Group 4) Combination of bromelain and 2% CHX (1:1)

Group 5) Combination of Bromelain and 5.25% NaOCl (1:1)

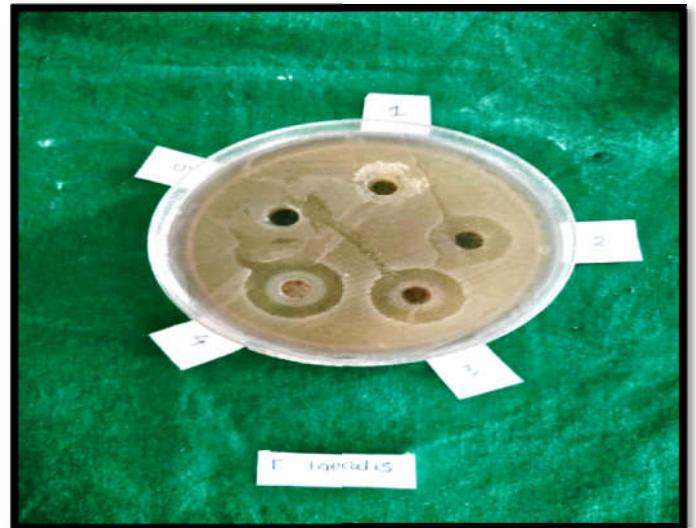


Preparation of Pineapple Extract (Bromelain)

The crude extract of pineapple was prepared. 200 grams of Stem was separated from outer fruit portion and cut into smaller pieces. 20 ml of distilled water added into that and Grind into the grinder. The obtained filterd was collected and then filtered through whatmans filter paper. The obtained filter was then centrifuged in centrifugation machine. The supernant was collected and stored at 4°C before use.

Agar disc diffusion test

E. faecalis (ATCC 29212) species were obtained from GMC Indore, India. The culture was grown overnight in brain heart infusion (BHI) broth at 37°C and inoculated in Mueller–Hinton agar plates and adjusted to 0.5 turbidity reading on McFarland scale (1.5×10^8 bacteria/ml). Five uniform wells of size 6mm were prepared on the *E. faecalis* culture plates. 200µl of each experimental solution were added to the respective wells on each plate. Total 5 plates were prepared and 5 wells in each plate prepared. These plates were incubated for 24 hours at 37°C in an incubator. After incubation period, plates were checked for zones of inhibition of bacterial growth and diameters of the zones achieved by each group against *E. faecalis*. Zones were recorded in centimetre (cm)



DISCUSSION

E. faecalis was chosen in the present study because of its high prevalence in secondary endodontic infection. Herbal irrigants are gaining importance because of their therapeutic properties. The present study evaluated antibacterial efficacy of Pure pineapple extract (Bromelain), 2% CHX, 5.25% NaOCl, Combination of bromelain and 2% CHX (1:1), Combination of Bromelain and 5.25% NaOCl (1:1)

Since its introduction NaOCl has been considered as an irrigant of choice for root canal irrigation because of its antimicrobial activity and tissue dissolving capacity. High pH of NaOCl interferes with the cytoplasmic membrane integrity and cause biosynthetic alterations in cellular metabolism attributing to its antimicrobial nature. Tissue dissolving action and dissolution rate of NaOCl is directly proportional to its concentration. But not only its actions like antimicrobial activity, tissue dissolving capacity and smear layer removing ability but also the caustic potential and toxicity of NaOCl also increases with the increase in concentration.

Chlorhexidine gluconate (2%) is a good disinfecting agent with a property of substantivity contributing to its prolonged time of action. On comparison with NaOCl, the irrigant is having less toxicity and foul taste. Chlorhexidine is proposed to be an alternative to NaOCl in open apex cases and NaOCl allergic patients. But the major disadvantage persisting is its inferior tissue dissolving action as a primary endodontic irrigant.

RESULTS

Results were tabulated and the statistical analysis was performed using SPSS software (Statistical Package for Social Sciences) version 16.0. Analysis of variance (ANOVA) was used followed by Post Hoc test. (P value < 0.05 considered as significant difference). Bromelain had showed antibacterial efficacy against *E. faecalis* but the combination of Bromelain and 5.25% Sodium Hypochlorite shown higher antibacterial property than other groups comparatively against *E. faecalis*.

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