

EVALUATION OF DISINFECTION OF GUTTA PERCHA CONES USING HERBAL IRRIGANTS

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ABSTRACT

Aim: The purpose of this study was to evaluate the effectiveness of chemical agents and herbal alternatives such as Aloe vera, Neem, Ginger and Garlic for rapid decontamination of GP cones. **Materials and Methods:** The reference strains of E. Faecalis were obtained, to know the efficacy of different agents against this microbe. Two experimental groups used in this study comprises of Herbal agents and chemical agents. The extracts of the Herbal agents which included amla, aloe vera, neem were prepared whereas the chemical agents were commercially available. Group 1 – Thulsi, Group 2 – Ginger, Group 3 – Aloe vera, Group 4 – Neem, Group 5 – NaOCl, Group 6 - CHX. The zone of inhibition of the different herbal and chemical agents were checked in two different media. **Results:** The result of the study showed that CHX was effective in disinfection of GP cones for 1 min followed sodium hypochlorite. Whereas Neem was the most effective herbal extract in disinfecting GP followed by Aloe Vera.

Conclusion:

Within the limits of this study, it can be concluded that CHX and Sodium Hypochlorite are efficient chemical disinfectants. Herbal agents also hold a promising future as a medium for disinfection, in which neem gave the best results within a short duration of time.

INTRODUCTION

Microorganisms are major etiological agents in pulpal and periapical disease.[1,2] The major goals of endodontic therapy are the elimination of microorganisms from the root canal system and the prevention of subsequent reinfection. In clinical practice, the dentist is occasionally come across with a problem of infection that occurs after obturation of root canal space. The endodontic treatment involves an initial mechanical phase of the instrumentation of root canal space; the chemical phase which involves antimicrobial solutions as irrigation agents and a final phase of obturation of root canal space with a suitable obturating material. Gutta-percha (GP) at present is the most common obturating material used for filling root canal space. They are biocompatible dimensionally stable radiopaque and thermoplastic and can be removed easily from root canal space.[3] According to some authors as GP cones are produced under the aseptic condition and possess strong antimicrobial properties owing to its zinc oxide component,[4] and they have been used directly from their sterile packages, so the need for decontamination for GP cones is unnecessary. However, the risk of GP cones contamination by glove handling and other physical source during its storage process can also be not ruled out. Unlike most instruments used in the endodontic treatment, GP cones cannot be sterilized by conventional autoclaving or in a hot-air oven; therefore, they require rapid decontamination before use to maintain the aseptic condition required for successful endodontic therapy.[5,6] Various chemical agents have been proposed as GP cones disinfectant, including sodium hypochlorite,[7-12] glutaraldehyde,[9-11] alcohol, iodine compounds, and hydrogen peroxide.

The appropriate disinfecting agent should be effective in killing different bacterial species. The objective of this study is to evaluate the effectiveness of a herbal alternative, Aloe vera and Amla for rapid decontamination of GP cones.

Aloe barbadensis mill, is a short succulent herb, it has strong antibacterial, antifungal, and antiviral properties.[13,14] The antimicrobial effects have been attributed to the plant's natural anthraquinones.[15] Neem has an established antibacterial, antifungal, and antiviral effect.[16] Neem also has anti-inflammatory, antiseptic, astringent, and analgesic properties.[17] Hence, it can be effective in decontaminating GP cones as well.

Therefore, the objective of the present study was to evaluate the antimicrobial properties of three commonly used chemical agents, namely 5% sodium hypochlorite, 2% chlorhexidine (CHX) and four herbal agents, namely Thulsi, ginger, Aloe vera, and Neem, against the most commonly isolated microorganism from the root canal, that is, Enterococcus faecalis.

MATERIALS AND METHODS

E. Faecalis is considered to be the most common Gutta Percha Contaminant, hence in this study the reference strains of E. Faecalis were obtained, to know the efficacy of different agents against this microbe.

Two experimental groups used in this study comprises of Herbal agents and chemical agents. The extracts of the Herbal agents which included thulsi, ginger, aloe vera, neem were prepared whereas the chemical agents were 5% Sodium Hypochlorite and CHX

Group 1 – Thulsi

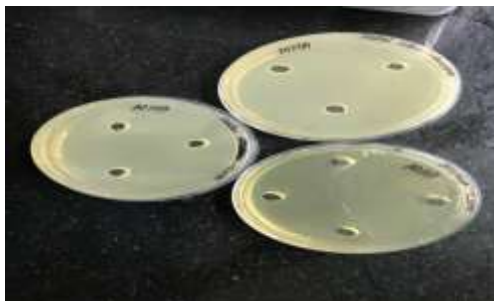
Group 2 – Ginger

Group 3 – Aloe vera

Group 4 – Neem

Group 5 – NaOCl

Group 6 – CHX



Antibacterial activity

The antibacterial activity of the chemical and herbal agents were tested against *E. faecalis* using Mueller-Hinton agar media. The strains of *E. faecalis* were cultured overnight in brain–heart infusion broth, and then, the culture was streaked on a Mueller-Hinton agar media. Three wells of 5 mm × 5 mm measure were made for chemical and herbal agents on the plates of both the media. About 0.1 ml of the agents were delivered into the well using a micropipette. They were then incubated at 37°C for 24 h; these were then monitored for clear zones around the agents. The antibacterial activity was assessed by the diameter of the inhibition zone.

STATISTICAL ANALYSIS

The data were summarized as mean ± standard deviation. The groups were compared by two-way analysis of variance and the significance of mean difference within (inter), and between (intra) the groups was done by Tukey's honestly significant difference post hoc test after ascertaining normality by Shapiro–Wilk's test and homogeneity of variance by Levene's test. A two-tailed ($\alpha = 2$) $P < 0.05$ ($P < 0.05$) was considered statistically significant.

RESULT

The present study compares the different disinfecting agents for GP cones. Total 30 samples treated under Muller-Hinton agar (MHA) medium with thulsi (Group 1), Ginger (Group 2), Aloe vera (Group 3), Neem (Group 4), NaOCl (Group 5), and CHX (Group 6) each with five samples. The outcome measure of the study was zone of inhibition against *E. faecalis* measured in millimeter (mm).

DISCUSSION

Disinfection of GP cones before to use as an obturating material is so important for a successful root canal treatment. Obturation is the final and one of the important steps in the root canal treatment. Hence, the materials used for obturation should also be properly disinfected to prevent the persistence of microbes. These cones can be easily get contaminated due to its improper storage or even when they exposed in clinics as well as during handling. Even though the fact that, some authors have said that there is no need to use disinfectant solutions on cones because it may change the properties of GP. In above study, cones contaminated with well-defined microbial populations were sterilized in different antimicrobial solutions. Due to its thermoplastic properties of cones, it become impossible to sterilize them by heat as it would result in their deformation. Therefore, the cones were sterilized with ethylene oxide. Several other chemical solutions have been proposed for a rapid decontamination of GP cones.

E. faecalis is found to be the main cause of endodontic failures as this is one of the most resistant bacteria which greatly adapt with the endodontic system. Hence, this bacteria is chosen in this study including sodium hypochlorite, CHX as chemical disinfectant; Thulsi, Ginger, Neem, Aloevera as herbal disinfectant.

Sodium hypochlorite is used at varying concentration, and it is most commonly used disinfectant. It has antibacterial as well as sporicidal effect, the mechanism of action involve the liberation of active chlorine which is an oxidizing agent and help to inactivate the bacterial enzyme.[18,19] Gomes et al. found that the time required for sodium hypochlorite to eliminate the microbes was inversely proportional to concentration of the solution used.[20] The sodium hypochlorite (NaOCl) of 5.25%, can easily eliminate *Candida albicans* and *E. faecalis* within 45 s, whereas 2.5% NaOCl takes 10 min for the complete elimination of these microbes. That's why, NaOCl can be used effectively for the disinfection of GP cones. It is very important to wipe off GP with gauze piece after disinfecting with sodium hypochlorite because at concentrations of 5.25% NaOCl can be a reason of having a large quantity of chloride crystals on the GP cone and also might cause GP deterioration as well as loss of its elasticity, which could hamper the obturation quality and can also impair the seal of the root canal. In case of having lower concentration of NaOCl can take more time to disinfect the GP.[21]

CHX is a cationic bisbiguanide with broad-spectrum antibacterial properties. The CHX molecule reacts on the bacterial cell surface (with negatively charged groups), causing an irreversible loss of cytoplasmic components. Gomes et al. stated that 2% CHX liquid took <30 s to completely eliminate *E. faecalis* from contaminated GP cones. Whereas, in other study, 2% CHX solution was found more effective from 15 s to 2 h in direct contact with infected GP cones.[20]

Recently, some herbal alternatives are also being suggested as disinfectants for GP cones. In this study, we have used Amla, Thulsi, Ginger, and Neem as a herbal alternative for GP cones disinfection.

Aloe vera has a popular about its antimicrobial activity credited to many compounds which are now precisely acknowledged as p-Coumaric acid, ascorbic acid, pyrocatechol, and cinnamic acid. It has been used from previous years for the treatment of a many type of diseases ranging from peptic ulcers to its use in cosmetic purpose. Aloe vera gel has been found to be effective in decontamination of GP cones within 1 min.

In other study, the extracts of Neem leaf showed significant antibacterial activity against. However, the extracts of Neem leaf shows inhibitory activities of were both organism – as well as solvent-dependent. Its extracts reduced the growth of Gram-positive as well as Gram-negative bacterial species. It also shows an anti-adherence property by modifying bacterial adhesion, and it became more helpful to limit the colonization by microorganisms within the root canals.[22]

Ginger and Thulsi also found to have antimicrobial properties, so in this it has been tested against other herbal irrigants.

For the evaluation of the efficacy of these disinfecting solutions, these solutions were cultured in Muller-Hinton media for 24 h at 37°C. The antimicrobial efficacy was assessed by the presence of zones of inhibition. *E. faecalis* showed maximum zone of inhibition with CHX in chemical agents and neem among the herbal agents.

The result of the study showed that CHX was effective in disinfection of GP cones for 1 min followed by sodium hypochlorite. Whereas, Neem was the most effective herbal extract in disinfecting GP followed by Aloe vera, ginger and thulsi.

CONCLUSION

Within the limits of this study, it can be concluded that CHX and Sodium Hypochlorite are efficient chemical disinfectants. Herbal agents also hold a promising future as a medium for disinfection, in which neem gave the best results within a short duration of time.

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