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Effect of Indian Medicinal Herbal Extracts on Surface Characteristics of Gutta Percha Cones

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Abstarct

Aim: The aim of the study was to evaluate the surface changes of Gutta Percha (GP) cones after disinfection using three different Indian herbal extracts.

Objective: To evaluate the surface changes of gutta percha cones after disinfection with 5.25 % Sodium hypochlorite, Vetiver extract, Karpooravalli extract and Citronella essential extract using Scanning electron microscope (SEM).

Methods: 104 gutta percha cones were considered in this vitro study and the samples were divided into four different groups such as Group I, II, III, and IV and were treated with 5.25% Sodium hypochlorite, Karpooravalli extract, Citronella extract and Vetiver extract respectively and for two different time intervals (1 min and 5 mins). The surface topography analysis of the samples were performed using SEM.

Results: The results of the study showed that Sodium hypochlorite had severe surface changes upto 77% of the GP for 5 minutes of time interval when compared with Karpoorvalli extract, Citronella extract and Vetiver extract. Among these Indian herbal extract used it was proven that vetiver extract showed the least changes on surface of GP cones both for 1 minute and 5 minutes.

Conclusion: According to the result of our study, GP cones that was immersed in Sodium hypochlorite showed higher surface irregularity than the GP cones immersed in Indian herbal extract.

Keywords: Gutta Percha; Karpooravalli Oil; Vetiver Oil; Citronella Oil

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Introduction

The ultimate success of root canal therapy mainly depends on removal of micro-organisms from the root canal system and to prevent reinfection, which will be done by thorough disinfection and obturation [1]. Gutta percha (G-P) is the best and an inert obturating material for root canal as it is dimensionally stable, biocompatible, radiopaque and thermoplastic with antimicrobial properties. GP cones which are available in sealed boxes and packaging are unsterilized and they can easily get contaminated during storage, handling, and even by exposure in chair side procedures resulting in breach of aseptic chain [2]. It is often seen that even in endodontic procedures performed under aseptic condition reinfection may occur. Number of chemical agents are available as GP disinfectants that is used in chair side practice such as Sodium hypochlorite, Glutaraldehyde, Alcohol, Iodine compounds, and hydrogen peroxide [3]. Sodium hypochlorite and chlorhexidine has oxidizing, hydrolyzing, and proteolytic properties which is the reason for its anti-bacterial and anti-viral effect. Sodium hypochlorite is most commonly used to sterilize GP cones. 5.25% sodium hypochlorite solution is recognized as broad spectrum antimicrobial agent who is efficient, inexpensive and reliable. It has been said that during GP disinfection, sodium hypochlorite shows some deposits on the surface of GP and because of strong oxidizing power it changes the structure of GP cones. Extreme topographic alterations and aggressive deterioration in the cones can be seen, which results in decrease of adhesion or bond strength of GP cones to endodontic sealers [5]. Though chemicals used for disinfection helps in eliminating microorganisms to an extent, it leads to some drawbacks such as surface changes in the GP cones. Different time periods and concentration have varying effect on microorganisms. Therefore an effective, but safer disinfectant is desirable for disinfecting GP cones. Now days number of articles shows that various herbs have antimicrobial, antiseptic, antiviral, antifungal, and immune modulatory properties. It is also noted that, they are safely used in food and pharmaceutical industries with little or no side effects [7,8]. Herbal agents are eco-friendly but have not been explored as an alternative pre-operative disinfection medicament for GP cones and whether it has any impact on the properties of GP.

Vetiver extract is popularly known as Khas Khas, Khas or Khus grass in India. Vetiveria zizanoids is profoundly used in Ayurvedic medicine. The chemical constituents present in the plant are vetiverol, vetivone, khusimone, khusimol, vetivene, khositone, trepenes, benzoic acid, tripene-4-ol, vetivenyl vetivenate, isonkhusimol, vetivazulene. Ayurvedic literatures mentioned that this plant has antimicrobial efficacy [9]. Studies indicates that vetiver extract was as effective as chlorhexidine mouthwash in its ability in reducing probing depth, clinical attachment loss and bleeding on probing [10].

The plant Karpooravalli which is also called as Plectranthus amboinicus, Coleus amboinicus, Coleus aromaticus commonly known as Country borage, Indian borage, is a dicotyledonous plant belonging to the family Lamiacea [11,12]. It is a large succulent aromatic perennial herb. The Phytochemical study reveals the presence of various Áavonoids like quercetin, apigenin, luteolin, salvigenin, genkwanin and volatile extract in the leaves [12]. The plant is also known to contain the constituents responsible for cytotoxicity and anti-bacterial activity [13,14]. In Literature leaf juice extracted from karpooravalli plant were used for healing chapped lips and cracked mouth [15].

Citronella essential extract is extracted from an herbaceous grass like tropical plant, Cymbopogon nardus, through steam distillation. Though it contains more than 22 compounds, geraniol, trans-citral, cis-citral, geranyl acetate, citronellal (6-octenal, 3, 7-dimethyl) and citronellol are the major constituents. Citronellal alone constitutes about 29.6% of Citronella essential extract [16]. Its use started long back as mosquito repellent [17]. Citronella essential extract had immunomodulatory effect [18]. It has also been reported to have antifungal and antimicrobial properties. Studies demonstrated that citronella based mouthwash presented higher antbiofilm effect and showed lowest cytotoxic effect than the commercial mouthwash used [19].

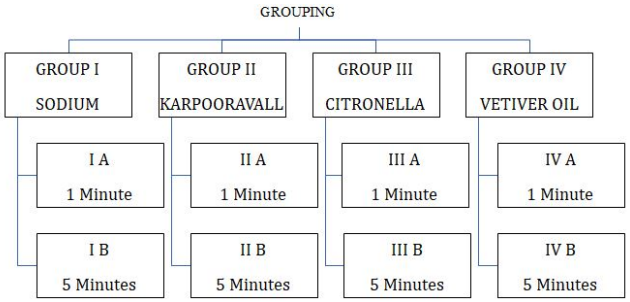
On analysing the literature there were no studies which compares the effect of Sodium hypochlorite and herbal agents such as Vetiver extract, Karpooravalli extract and Citronella extract as GP disinfectants. Thus, this present invitro study is undertaken to the compare the surface changes in GP cones disinfected using herbal extract (Vetiv-

er extract, Karpooravalli extract, Citronella essential extract) and 5.25% Sodium hypochlorite at different time intervals by scanning electron microscope (SEM).

Materials and Methods

From a sealed packet, 104 GP cones of size F3 protaper (dentsply sirona,USA) were selected and distributed into groups. Each group consist of two subgroups containing 13 GP cones in each.

The following were the groups.



Each GP cones were directly taken from the box and examined under scanning electron microscope (Pre-SEM).

Disinfection of Gp Cones

After taking PRE-SEM, all the GP cones were washed using distilled water to remove the dirt and deposits which was done to obtain a better results. The GP cones were air dried using sterile gauge pad for 30 minutes in a laminar flow chamber to remove the moisture content on the surface of GP cones. These 104 GP cones were then divided into four groups with 13 cones in each subgroup and were immersed in beakers containing respective extracts. The cones that were immersed in groups IA, IIA, IIIA and IVA were treated for 1 minute and the cones immersed in groups IB,IIB,IIIB and IVB were treated for 5 minutes.

Topographic Examination of Gp Cones

Scanning electron microscope (FEI QUANTA FEG 200 SCANNING ELECTRON MICROSCOPE) at 500x magnification was used for the topographical examination of GP cones. After disinfection cones were arranged in a stub (stainless steel block) for sputter coating which was done to prevent charging of specimen and to withstand the high vacuum which was done using magnetron sputtering. After sputtering process stub was placed in sample chamber of the microscope and the topographical examination was done.

Statistical analysis: Statistical analysis has been performed using SPSS software VERSION 24 and carried out using Chi- square test to compare the changes seen on the surface of GP cones.

Results

13 GP cones where selected from each group and the percentage was calculated by the number of cones that showed changes divided by the total number of cones. The results were tabulated as shown in table 1.

Formula for Calculating the Percentage of GP Cones Undergone Changes

% of GP undergoing changes = GP cones undergone changes ×1020066002667000 / Total no of GP cones

In group 1 (Sodium hypochlorite) for one minute showed severe changes of 77% and moderate changes of 23 %. In group II (Karpooravalli extract) for one minute showed moderate changes of 54 % and mild changes of 46 %. In group IV (Vetiver extract) for one minute showed moderate changes of 30.7%, mild changes of 53.8% and no changes of 15.3%. In group III (Citronella extract) for one minute showed severe changes of 23%, moderate changes of 46% and mild changes of 30.7%. After 5 minutes, group I (Sodium hypochlorite) showed severe changes of 92.3% and moderate changes of 7.6%. In group II (Karpooravalli extract) showed severe changes of 23%, moderate changes of 38. 5% and mild changes of 38.5 %. In group III (Citronella extract) showed severe changes of 23% moderate changes of 38.4% and mild changes of 30.7%. In group IV (Vetiver ex-

tract) showed moderate changes of 46 % and mild changes of 46 % and no changes of 7.6%. The images of SEM obtained is shown in the Figure 1.

Table 1: Representing the percentage of GP cones undergone changes

GROUPS	MILD%	MODERATE %	SEVERE%	NO CHANGES%
SODIUM HYPOCHLORITE 1 MIN	0	23	77	0
SODIUM HYPOCHLORITE 5 MINT	0	7.6	92.3	0
KARPOORAVALLI EXTRACT 1 MINT	46	54	0	0
KARPORAVALLI EXTRACT 5 MINTS	38.5	38.5	23	0
CITRONELLA EXTRACT1 MINT	30.7	46	23	0
CITRONELLA EXTRACT 5 MINTS	30.7	38.4	23	0
VETIVER EXTRACT 1 MINTS	53.8	30.7	0	15.3
VETIVER Extract 5 MINTS	46 %	46 %	0	7.6

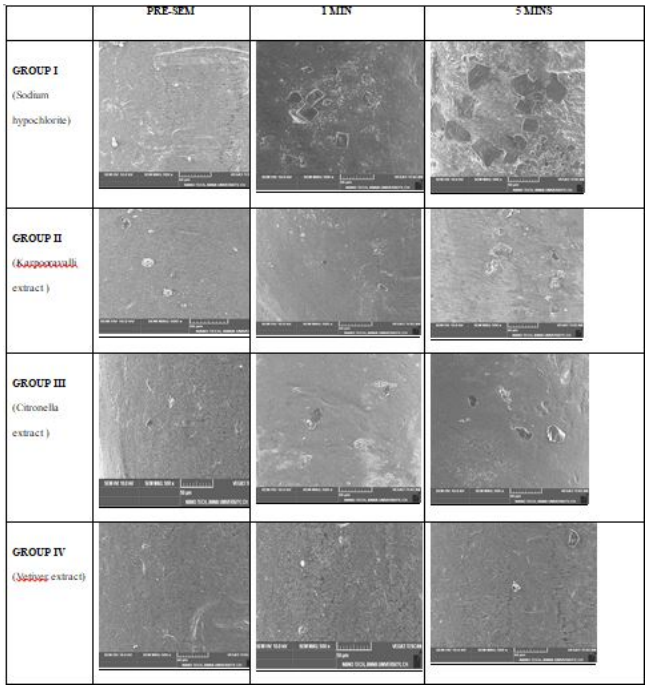


Figure 1: Shows the images of SEM

Discussion

GP cones are the most commonly used root canal filling material which is famous for its thermoplastic and viscoelastic nature. Though the GP cones manufacturer's claim that the production is under aseptic conditions, several studies have proven the presence of microorganisms even in opened fresh boxes [20]. As root canal filling materials need to be in the root canal for lifetime, they must be sterile. In endodontics, 5.25% Sodium hypochlorite solution is the most commonly used disinfection agent, which may be attributed to the fact that it has bactericidal properties, organic material dissolution and low in cost. In Sodium hypochlorite due to the presence of hypochlorous acid, acts against bacteria due to which is routinely used as an irrigating solution and 5.25% Sodium hypochlorite used as GP disinfectant [21]. But Sodium hypochlorite is known to be a strong oxidizing agent. Which generally reduce the chemical stability of chain polymer in resin and waxes of GP cones. Because of this chemical instability the mechanical properties

of GP cones are adversely affected. It is also reported that Sodium hypochlorite reduces the polymeric component of GP cones which in turn reduces the tensile strength [21]. In order to avoid the disadvantage of Sodium hypochlorite plant based medicine can be considered as alternative for various medical health practices throughout the globe because of their safety, availability and cost effectiveness. Plant derived extract and extracts is used for various disease and helps in maintaining good health too. So in this study we have included three different Indian herbal extract such as Vetiver essential extract, Citronella essential extract and karpooravalli extract.

Vetiver was considered as an alexiteric agent in several folk medicines, e.g.,as a preservative against poisons and venoms. The volatile extract which is taken from roots of vetiver is a viscous liquid, with different colors from pale-yellow to dark brown, as well as a deep, smoky, earthy, and woody odor [21]. Vetiver extract is known for its odor which can be used as a flavor adding agent in the food industry. It has its special aroma, it has been proven that Vetiver extract also possesses various biological properties, such as antioxidant, antibacterial and anti-inflammatory properties. Vetiver extract is known by its botanical name *Chrysopogon zizanioides.*

Citronella or *Cymbopogon nardus* is one of the *Cymbopogon* species with its essential extract widely used in the production of citronella essential extract, food preservatives, and pharmaceutical products. Citronella extract is said to have phyto therapeutic potential for antibacterial and antifungal activity. The physical qualities of the essential extract, which are natural products, include painkillers, anticonvulsants and anxiolytics [14]. The fragrance, cosmetic, medicinal and flavoring industries employ the steam extracted volatile essential extract from its leaves.

Karpooravalli essential extract is known by its botanical name *Plectranthus amboinicus sprengel.* Phytochemicals in Karpooravalli extract, possess antimicrobial activity against a wide range of bacteria, yeast and mould. Essential extract of Karpooravalli was reported to have a synergistic effect on the antibiotic toxicity toward resistant bacterial strains when combined with aminoglycosides. The essential extract in Karpooravalli has antioxidant properties [22].

In the present study, three herbal extract Vetiver extract, Karpooravalli extract and Citronella extract were used to evaluate the surface changes when compared with sodium hypochlorite which is commonly used disinfectant.

In the present study, SEM was used to scan the surface at a magnification of 500x. Sodium hypochlorite treated GP cone had shown highest surface alteration when compared with other three herbal extract (Karpooravalli, Vetiver, Citronella) which showed less surface changes. It was reviewed that Sodium hypochlorite leaves multiple pitting and deposition on the surface of GP cones, which reduces the bond strength or adhesion of GP cones to endodontic sealers. Hence, it is said that these effects of sealing ability and reinforcement in the root canal may be the main cause of endodontic failure. It is evident that Sodium hypochlorite increases the elasticity of GP cones due to which the resin bonds break and reduce the bonding capacity and it also forms crystals of Sodium hypochlorite which may precipitate and remain on the surface. whereas herbal extracts has no effect on the elasticity of GP cones and it forms less crystals when compared with Sodium hypochlorite which may precipitate and remain on the surface [23,24]. Hence there was less changes seen in herbal extracts, among these three herbal extracts Vetiver extract showed least changes on the surface of GP cones. Therefore Vetiver extract can be used as an alternative for disinfecting GP cones.

Conclusion

Within the limitations of this study, Sodium hypochlorite (5.25%) showed greater deterioration on the surface of GP cones, whereas lesser deterioration was observed with Karpooravalli extract, Vetiver extract and Citronella extract. The least changes were seen in GP treated cones with Vetiver extract. Further research in this area should be conducted to investigate the surface energy of this extract for using it as GP disinfectants.

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