

SMEAR LAYER REMOVAL EFFICACY OF CONVENTIONAL ENDODONTIC IRRIGANTS V/S PHYTOCHEMICAL EXTRACTS-AN IN VITRO STUDY

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ABSTRACT

This study was conducted to evaluate the effectiveness of two natural extracts for the irrigation of root canal.

A descriptive study was conducted at the Department of Operative Dentistry & Department of Oral Biology, Hamdard University Dental Hospital, Karachi, Pakistan. Irrigation solutions were made in Operative Department. 100 single rooted extracted teeth were collected from Oral Surgery Department. These teeth were disinfected and cleaned using ultrasonic tip and stored in distilled water until use. These teeth were decoronated below the cemento-enamel junction to obtain standardized root length of 10mm. Root canals were instrumented using rotary files at working length 1mm short of the apex. Specimens were randomly divided into 4 groups and subjected to each irrigants for 5 minutes according to the irrigation protocol, Group A contained distilled water and was taken as control. Group B contained 17 % EDTA while Group C contained herbal extract of lemon grass and Group D contained herbal extract of Green tea. Specimens were longitudinally sectioned and evaluated under scanning electron microscope for smear layer removal efficacy. Statistical analysis was done by using Kruskal-Wallis test ($\alpha=0.05$).

Group A, Group C and Group D showed the presence of debris and smear layer and they were statistically different from Group B where debris and smear layer were totally removed ($p < 0.05$). Group D showed removal of debris at middle third and ground portion of the tooth. EDS microanalysis showed the presence of Na, P, and Ca elements on the surface.

Among all groups green tea was more effective in removing the smear layer and debris without causing erosion.

Key Words: Endodontic irrigants, lemon grass & green tea.

INTRODUCTION

Endodontics is the branch of dentistry, concerned with the diseases threatening the vitality of a tooth making pulp chamber and root canals containing pulp

tissue vulnerable to infection and necrosis. To rescue this tooth, we perform endodontic therapy or root canal treatment that comprises of chemo-mechanical debridement of canals, subsequent shaping and decontamination with different chemical irrigants.¹The effectiveness of endodontic files, rotary instrumentation, irrigating solutions, and chelating agents to clean, shape, and disinfect root canals underpins the success, longevity, and reliability of modern endodontic treatments.⁴

However, during endodontic therapy an amorphous structure called the smear layer is formed on dentinal walls by the mechanical action of endodontic instruments. McComb and Smith were the first one to find out the existence of smear layer as a part of routine instrumentation.²They proposed that the smear layer is not merely dentin but a mixture of organic pulpal

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material, inorganic dentinal debris, odontoblastic processes, and bacteria⁽²⁾. This layer is a physical barrier to the penetration of endodontic irrigants into the dentinal tubules consequently decreasing their efficacy.^{2,3,4} This layer is composed of two parts i.e. inorganic and organic. Organic substances include fragments of odontoblastic processes, microorganisms and necrotic tissue. For an effective root canal treatment both organic and inorganic parts of this smear layer must be removed. The presence of smear layer, interferes with perfect adaptation of the root canal filling material with the root canal walls. However, some researchers are against smear layer removal; stating that the presence of smear layer prevents bacterial adhesion and colonization of dentinal matrix and inhibits root canal re-infection. According to Love et al. the smear layer acts as a protective barrier and if removed, there would be no physical barrier against bacterial penetration into the dentinal tubules.

Because of the complex anatomy of root canal system, it is impossible to perform complete debridement and disinfection with only mechanical means. Hence irrigation with chemicals exhibiting antibacterial properties is an important step in a successful endodontic treatment. These chemicals easily seep into the hard tissue to reach areas where files can't approach and are able to cleanse dentinal walls effectively.

Basic purposes of an ideal Irrigant solution includes tissue dissolution, removal of smear layer, lavage of debris, canal disinfection and minimal lubrication. Commonly used irrigating solutions include 5% and 2.5% sodium hypochlorite, 3% hydrogen peroxide, 17% EDTA, MTAD, 50% Citric acid, 0.2% Chlorhexidine.⁵

NaOCl is the most widely recommended endodontic irrigant due to its unique characteristics of organic tissue dissolution.^{6,1} However since the smear layer is composed of an inorganic component as well, calcium chelators are used with NaOCl in root canal treatment to ensure its complete removal. EDTA being the most prominent chelator agent in the history of endodontics.^{3,6} Other chelating agents include 10% citric acid, tanins and maleic acid.⁶ Literature also supports the use of 10ml of 17% EDTA followed by 10ml of 5% sodium hypochlorite as an effective method of smear layer removal.⁶

Although these chemicals are potent in their action but their demerits are also unforgiving. Many studies highlight that these chemical solutions are cytotoxic.^{1,6,7} Their extravasation in the periradicular tissues or leakage into the oral cavity is found to be associated with severe inflammation, hematoma formation, chemical burns, neuronal damages, choking, endophthalmitis, ototoxicity and severe hypersensitivity reactions.^{6,8,9}

Considering the above mentioned side effects world is now moving on towards developing more biocompatible alternatives for the purpose of endodontic irrigation. Research studies are being conducted on various plant extracts to develop better irrigant solutions, including Miswak, Propolis, Turmeric (*curcumin longa*), *Ferula gummosa*, *Punicagranatum Mesocarp*, *Nelumbonucifera* leaf, *Psidiumguajava* leaf and *Coffeacanephora*, *Citrus aurantifolia*, *Sapindusmukorassi* and many others.¹⁰⁻¹²

This study focuses on comparison of smear layer removal efficacy of conventional irrigants i.e. 17% EDTA v/s phytochemical extracts i.e. *Camellia sinensis* (green tea) and *Cymbopogoncitratatus* (lemon grass).

LEMON GRASS

Cymbopogoncitratatus also known as lemon grass is a herb which belongs to family Poaceae. It is well known and utilized for its distinct lemon flavor and citrusy aroma. It is a tall perennial grass which is native to India and tropical regions of Asia. It is a coarse and tufted plant with linear leaves that grows in thick bunches, emerging from a strong base and standing for about 3 meters in height with a meter wide stretch.

In addition to its culinary usage, lemon grass offers a wide array of medicinal benefits and is in extensive demand due to its antipyretic, antidepressant, anti-inflammatory, antiseptic, antibacterial, antimicrobial and anti-carcinogenic properties.¹³ Lemon grass contains substances that are thought to relieve pain, reduce fever and have antioxidant properties.

GREEN TEA

Camellia sinensis also known as green tea is a member of Family Theaceae. It is widely consumed by people in the world due to its relaxing effects on mind and body. Tea mainly comprises of polyphenolic compounds about 60-80% that makes up 30% of the dry weight of flush.

Epigallocatechingallate is the most abundant catechin in most tea brands. There is good evidence that the catechin compounds of green tea are responsible for the observed antibacterial activity and that EGC, EGCg and ECg constitute the most important antibacterial agents (Yam et al. 1997; Hara 2001). Green tea exhibits bacteriostatic and bactericidal activities against both methicillin resistant *Staphylococcus Aureus* (MRSA), *S. Epidermidis* including many other microbes. It has also been found to work against α -hemolytic streptococci, *E. fecalis*, *Lactobacilli* (main etiologic agent of dental caries) and also against *Candida albicans* and *Pseudomonas aeruginosa*.¹² Making tea a deserving candidate to be used in dentistry.¹⁴

METHODS AND MATERIALS

100 human single rooted teeth that were extracted due to orthodontic and periodontal problems were selected. Sample size was calculated on Open-Epi, version 3, with 95% Confidence interval & power of study was calculated at 80%. Teeth with straight anatomy of roots and type 1 canal anatomy were included in this in vitro study, which left us with 94 teeth as the rest of the teeth showed a different configuration of canal. The teeth extracted were then cleaned by ultrasonic scaler, sterilized and stored in distilled water until use. The teeth were decoronated at CEJ using flexible diamond disc to standardize the root length to 10mm. Samples obtained were then divided randomly into 4 experimental groups. The patency and the working length of the canal was determined by inserting #10 K file (Mani Inc., Tochigi Ken, Japan) and the working length was calculated by subtracting 1mm from this length. Size 15 and 20 K files (DentsplyMaillefer) were used at the working length. The root canals were cleaned and shaped using Universal Protaper Rotary System (Dentsply-Maillefer, Switzerland) as per manufacturer's protocol up to F3. Irrigation was performed with 1ml of 2.5% of NaOCl (Ups Hygenies, Mumbai, India) solution after each instrument change.

All the irrigating solutions were introduced into the canal using stainless steel 27-G beveled needle. The needle was placed within 1 to 2mm of the working length in each canal. Thereafter, the root canals were irrigated with 5ml of distilled water to remove any residue. The canals were then blot dried with sterile paper points and a sterile cotton pellet was placed and the access cavity was sealed.

PREPARATION OF HERBAL EXTRACT

PREPARATION OF HERBAL EXTRACT (LEMON GRASS)

For the purpose of decoction of lemon grass leaves, the leaves were placed in distilled boiling water for 5 minutes, at solid:liquid ratio 1:1. Later the vessel was covered and removed from the heat and allowed to cool for 5 minutes. The herbal material and liquid were then strained through cheese cloth and the resulting decoction placed into 100ml reagent bottles and stored at 4°C.

PREPARATION OF HERBAL EXTRACT (GREEN TEA)

10 gm fresh green tea leaves were grounded and extracted by soaking for two days using 100 ml of distilled water in 250ml of sterile conical flask. The extracts were filtered using Whatmann filter paper no.1, the filtrates were then concentrated by rotavapour and refrigerated stored at 4°C prior to use.

SEM PREPARATION

Using a diamond disc at slow speed, longitudinal grooves were made on the buccal and lingual surfaces of each root without penetrating the canal. The roots were then gently split into two halves using a chisel and were stored in deionized water at 37 °C in an incubator until SEM analysis. The specimens were dehydrated using 100% ethyl alcohol and were placed in a furnace at 600C for 24 hours. The samples were then mounted on metallic stubs, gold sputtered using an ion sputter, and examined under scanning electron microscope (LEO 440i, Carl Zeiss, Tokyo, Japan) for the presence or absence of the smear layer. Photomicrographs of the surface morphology at magnification of x300, x3000, x5000 were performed. The images were scored according to the criteria given by Torabinejad et al.^{4,6,10}

Score 1 = No smear layer, No smear layer on the surface of the root canal; all tubules were clean and open.

Score 2 = Moderate smear layer, No smear layer on the surface of the root canal, but tubules contained debris.

Score 3 = Heavy smear layer. Smear layer covered root canal surfaces and tubules.

In addition, the degree of erosion of dentinal tubules was scored as follows:

Score 1 = No erosion. All tubules looked normal in appearance and size.

Score 2 = Moderate erosion. Peri-tubular dentin was eroded.

Score 3 = Severe erosion. Intertubular dentin was eroded and tubules were connected with each other.

These areas were evaluated by two independent evaluators who were unaware of the experimental groups to which the samples belonged.

STATISTICAL ANALYSIS

The non-parametric analysis of variance (KruskalWallis's test) was applied to independently evaluate the effect of the two dependent variables, smear layer removal and degree of erosion. These analysis were performed using SPSS version 22.

RESULTS

Out of 100 teeth, 94 teeth were included in the study as the rest of the 6 teeth had a different canal configuration. Group A was assigned with 25 teeth, while rest of the groups (B,C,D) were assigned with 23 teeth in each group. Group A which consisted of distilled water (control), Group C which was based on herbal extracts of lemon grass and Group D which was based on herbal extracts of green tea showed the presence of debris and

TABLE 1: GROUPS WITH DIFFERENT IRRIGANTS

GROUPS	IRRIGANTS USED
GROUP A	DISTILLED WATER (CONTROL)
GROUP B	17%EDTA(pulpdent pulpdent corporation, MA, USA)
GROUP C	HERBAL EXTRACTS 1(LEMON GRASS)
GROUP D	HERBAL EXTRACT 2 (GREEN TEA)

TABLE 2: STATISTICAL ANALYSIS OF DEGREE OF EROSION AND SMEAR LAYER REMOVAL OF DIFFERENT EXPERIMENTAL GROUPS

Experimen- tal Groups	Mean Score for Smear Layer			Mean Score for Erosion		
	Coronal 3 rd	Middle 3 rd	Apical 3 rd	Coronal 3 rd	Middle 3 rd	Apical 3 rd
Group A	3.00	3.00	3.00	1.00	1.00	1.00
Group B	1.33	1.53	1.83	2.83	2.63	1.93
Group C	2.33	2.53	2.13	1.00	1.00	1.00
Group D	3.00	1.43	1.63	1.00	1.00	1.00

smear layer and they were statistically different from Group B which was based on 17% EDTA solution where debris and smear layer were totally removed ($p < 0.05$). Group B showed complete removal of smear layer in all thirds of the tooth with significant erosion. Group C showed partial removal in coronal and apical portions of the root whereas middle portion seems to be having heavy smear layer around. Group D showed moderate removal of debris at middle third and ground portion of the root (Table 2). No erosion was seen in group C (Extract of Lemon grass) and D (Extract of Green tea) (Table 2) while Group B (17% EDTA) showed maximum erosion in middle third. EDS microanalysis showed the presence of Na, P, and Ca elements on the surface.

DISCUSSION

The ultimate goal of root canal preparation is canal debridement to promote apical healing.³ Thickness of smear layer is normally around 1-2 μm . It consists of two layers: a surface smear layer and a subsurface smear layer that is packed into the dentinal tubules. It is present into the tubules to a depth of about 40 μm . Smear layer serves as a good substrate for bacteria hence it should be eliminated as early as possible from the root canal wall. There seems to be a long list of commercially available irrigating solutions but still there is an ongoing search for an ideal root canal irrigant as the microbiota of smear layer are stubborn and resistant. Most popular name in the removal of smear layer is a combination of 17% EDTA with 5.25% NaOCl but the main disadvantage of EDTA is dentinal erosion with limited antibacterial activity. NaOCl reacts with EDTA to release chlorine gas at relatively low levels.⁶

Our data suggests that irrigation with the extract of lemon grass & green tea are effective in removing

smear layers but lemon grass extract was more effective as it achieved better scores compared to green tea extract (table 2). These inferences are in conjugation with the work of Rathakrishnan M et al & Chhabra N et al. Our data also suggests that these herbal extracts does not causes erosion of the dentinal wall (Table 2), thus minimizing the short coming of NaOCl & EDTA. Recent research shows an excessively aggressive effect of this combination on the root canal walls causing too much erosion and degradation of the dentin.⁶ Although its effect on the long-term viability of endodontic treatment remains controversial, erosion could result in an alteration of mechanical properties of dentin with insufficient adaptation of the root filling materials to the canal walls.⁶ Other advantage of these herbal extracts is the low cost of treatment as the newer alternatives are costlier & have other short comings.

The only short coming we felt was the preparation of fresh extracts of these herbal products as it is difficult to store these solutions in activated form. Further studies are required to observe the long term effect of using these extracts.

CONCLUSION

Among all groups Lemon grass extract was more effective in removing the smear layer and debris without causing significant erosion. More studies are necessary to further investigate the properties of these two novel irrigants.

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CONTRIBUTIONS BY AUTHORS

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|---------------------------|--|
| 1 Syed Abrar Ali: | Wrote manuscript, Collected data & analyzed. |
| 2 Mehmood Hussain: | Assisted in writing Discussion. |
| 3 Haroon Shah: | Reviewed the literature cited in article. |
| 4 Hina Khan: | Helped in data collection. |