

The effect of Grapefruit-seed extract as an endodontic irrigant on bonding to root canal dentin (An In Vitro Study)

Thesis

Submitted to Endodontics Department
Faculty of Dentistry, Ain Shams University
for partial fulfillment of the Requirements of Master Degree in

Endodontics

By

Maisa Ahmed Hussein

B.D.S Faculty of Dentistry, Ain Shams University (2004)

> Faculty of Dentistry Ain Shams University 2012

SUPERVISORS

Dr. Ahmed Abd El Rahman Hashem

Associate Professor of Endodontics,

Faculty of Dentistry - Ain Shams University

Dr. Abeer Abdulhakeem Mahmoud

Associate Professor of Endodontics,

Faculty of Dentistry - Ain Shams University



سورة التوبة (105)

Acknowledgement

All praise and thanks be to **Allah**, the most Compassionate, the most Merciful.

I am proud to be supervised by Doctor Ahmed Abd El Rahman Hashem, Associate Professor of Endodontics, Faculty of Dentistry, Ain Shams University, whom I have a spontaneous appreciation for his valuable instructing of the theme of my study which was originally suggested by him.

Words can never express my sincere gratitude to **Doctor Abeer Abdulhakeem Mahmoud**, Associate Professor of Endodontics, Faculty of Dentistry, Ain Shams University, for her great efforts, grace and cooperative remarks during this study.

Dedication

To my father who encouraged me throughout my way.

He is the most important person in my life whom I could not have reached this level without him.

To my mother who supported me in each step of my life.

She gave me everything I needed or dreamt of.

LIST OF CONTENTS

	Page
LIST OF TABLES	ii
LIST OF FIGURES	iii
INTRODUCTION	1
REVIEW OF LITERATURE	3
1-Smear layer removal using different irrigation regimens	3
2-Bond strength of resin based root canal filling material to root canal dentin	27
3-Grapefruit-seed Extract	41
AIM OF THE STUDY	45
MATERIALS AND METHODS	46
Materials	46
Methods	47
RESULTS	62
DISCUSSION	82
SUMMARY AND CONCLUSIONS	91
REFERENCES	94
ARABIC SUMMARY	

LIST OF TABLES

Table No.	Title	Page
1	Smear layer scoring according to Guerisoli et al 2002	51
2	Mean ± standard deviation (SD), of the overall smear laye scores and results of comparison between different irrigation regimens	
3	Comparison between mean ± standard deviation (SD) o smear layer score of different irrigation regimens at the three segments	
4	Comparison between mean ± standard deviation (SD) o smear layer score at different segments within the same irrigation regimen	e
5	Mean ± standard deviation (SD), values (in MPa), and result of comparison between push-out bond strength of the five groups at the whole root surface area	e
6	Mean ± standard deviation (SD), values (in MPa), and results of comparison between push-out bond strength of the different irrigation regimens at the same root canal level	e
7	Mean ± standard deviation (SD), values (in MPa), and results of comparison between push-out bond strength a different root canal level within the same irrigation regimen.	ıt

LIST OF FIGURES

Fig. No.	Title	Page
1	Scanning electron micrograph X1500 representing score 4	52
2	Scanning electron micrograph X1500 representing score 3	52
3	Scanning electron micrograph X1500 representing score 2	53
4	Scanning electron micrograph X1500 representing score 1	53
5	(1)Coronal,(2) middle, and(3) apical root sections were cut	
	after setting of the acrylic resin	
6	The loading fixture	58
	A. The upper part accurately fitted in the lower part. B. The upper and lower parts of the loading fixture.	
7	Schematic diagram for the loading fixture used in the push-	
	out test	
8	Different plungers used in the push out test	59
	A. The plunger used for the coronal segment (1mm) B. The plunger used for the middle segment (0.75mm) C. The plunger used for the apical segment (0.5mm)	
9	Loading of the tested sample using Instron testing machine	60
10	Bar chart representing the overall mean smear layer scores of the different irrigation regimens	63
11	Bar chart representing mean smear layer score of the three segments with different irrigation regimens	66
12	Bar chart representing mean smear layer score of the five groups at the three root canal segments	69
13	Scanning electron micrograph of samples irrigated using 2.5% NaOCl (Magnification X1500); A, coronal. B, middle. C, apical	70
14	Scanning electron micrograph of samples irrigated using 2.5% NaOCl+17% EDTA (Magnification X1500); A, coronal. B, middle. C, apical	71
15	Scanning electron micrograph of samples irrigated using 2.5% NaOCl+GSE (Magnification X1500); A, coronal. B, middle. C, apical	72
16	Scanning electron micrograph of samples irrigated using GSE (Magnification X1500); A, coronal. B, middle. C, apical	73
	(iii)	

Fig. No.	Title	Page
17	Scanning electron micrograph of samples irrigated using GSE+ 17% EDTA (Magnification X1500); A, coronal. B, middle. C, apical	74
18	Bar chart representing mean push-out bond strength of the five groups at the whole root surface area	76
19	Bar chart representing mean push-out bond strength of the three segments with different irrigation regimens	79
20	Bar chart representing mean push-out bond strength of the five groups at the three segments	81

The main objectives of root canal therapy are cleaning and shaping and then obturating the root canal system in three dimensions to prevent reinfection. Mechanical instrumentation cannot sufficiently disinfect root canal. Irrigation solutions are required to eradicate microorganisms, dissolves necrotic tissue, lubricates the canal and removes the smear layer.

The smear layer consists of organic and inorganic substances, including fragments of odontoblastic processes, microorganisms, and necrotic materials. Presence of smear layer prevents penetration of intracanal medication into the irregularities of the root canal system and the dentinal tubules and prevents complete adaptation of obturation materials. Several endodontic irrigating solutions have been used to remove such preparation smears with varying degrees of success. They have ranged from acids and chelates, to those intended to dissolve the organic debris.

Sodium hypochlorite (NaOCl) is used as an irrigant because it combines important properties such as tissue dissolving capability and microbicidal activity but it cannot remove the smear layer. The most widely used chelating irrigant is ethylenediamine tetra-acetic acid (EDTA) which is effective in eliminating the smear layer. However, the chelating action results

in wide dentinal tubules in the root canal walls, sometimes also completely destroying the intertubular dentin.

Grapefruit-seed extract (GSE) is a commercially available substance that has received some attention for having antimicrobial properties. GSE is made by first converting grapefruit seeds and pulp into an acidic liquid. This liquid is loaded with polyphenolic compounds which are unstable and are chemically converted into more stable substances that belong to a diverse class of products called quaternary ammonium compounds. Recent researches report GSE to be effective against multiple bacteria strains, so this substance may have a potential as an endodontic irrigant but its effect on the smear layer or on the bonding to root dentin is still unclear.

1- Smear layer removal using different irrigation regimens

The smear layer is a 1-5 mm thick layer ⁽¹⁾ of denaturated cutting debris produced on instrumented cavity surfaces, and is composed of dentine, odontoblastic processes, non-specific inorganic contaminants and microorganisms ⁽²⁾. Its removal provides better sealing ability of the endodontic filling material to dentin, thereby avoiding leakage of microorganisms into the oral tissues ⁽³⁾.

Sodium hypochlorite (NaOCl) in a 1-5.25% concentration is an irrigant solution used widely in root canal treatment because of its bactericidal properties and ability to dissolve organic tissues (4,5); but NaOCl has not been shown to be effective in removing the smear layer (6). Decalcifying solutions such as phosphoric acid, citric acid and EDTA have been reported as suitable for removing the smear layer (7). So removal of the smear layer requires a combination of NaOCl (an organic solvent) and acids such as: citric acid, tannic, poly acrylic acid, or chelating agents such as EDTA or REDTA for the removal of the inorganic part (3).

Takeda et al ⁽⁸⁾ evaluated the effects of endodontic irrigants and two types of laser on a smear layer created by hand instrumentation. Sixty human mature extracted mandibular

premolar teeth with a single root canal were classified into five groups (12 each). Root canals were instrumented to a size 60 master apical file with a step-back technique; canals were irrigated with 3ml of 5.25% NaOCl and 3% H₂O₂, alternately, between each file size. Final flush was 17% EDTA (group 1), 6% phosphoric acid (group 2), 6% citric acid (group 3), group 4 the root canals were irradiated with a carbon dioxide (CO₂) laser, and group 5 were irradiated using an Er: YAG laser. The teeth were split longitudinally and prepared for examination by SEM. Irrigation with 17%EDTA, 6%phosphoric acid and 6% citric acid did not remove all smear layers from the root-canal system and demineralized the intertubular dentin with enlargement of tubular openings. The CO₂ laser was useful in removing and melting the smear layer on the instrumented root-canal walls and the Er: YAG laser was the most effective in removing the smear layer from the root-canal wall.

Peters and Barbakow ⁽⁹⁾ evaluated debris and smear layer in canals prepared with either Lightspeed (LS) or ProFile (PF) rotary instruments. Irrigants used were tap water (group A) or alternating 5.25% NaOCl and 17% EDTA (group B). The roots were split longitudinally and examined at apical, middle and coronal levels for debris and the smear layer using a 5-step scale. Using only water, mean debris and smear layer scores were similar for LS and PF. In contrast, with EDTA/NaOCl, LS and PF

prepared canals had similar debris scores at the apical and coronal levels, but there was a significant difference at the middle level. Also with NaOCl and EDTA, mean smear layer scores were significantly different at the apical and middle levels, but not at the coronal level. Neither technique was superior in removing debris, but with LS instruments there was a more effective removal of the smear layer in the EDTA-NaOCl group.

Scelza et al (10) determined the degree of removal of pulpal remnants and smear layer from root canals after final irrigation with three different solutions. Thirty extracted single-rooted human teeth were randomly distributed into 3 groups (n=10) for the final 4-min irrigation. During instrumentation the step-back preparation and 1% NaOCl were used. Solutions used in the final irrigation for group1 were 10 ml of 1% NaOCl, followed by 10 ml of 10% citric acid, and finally 10 ml of distilled water; for group 2: 15 ml of 0.5% NaOCl, followed by 15 ml of EDTA-T (17% EDTA + sodium lauryl ether sulphate); for group 3: 10 ml 5.25% NaOCl, followed by 10 ml of 3% hydrogen peroxide, and finally 10 ml of 5.25% NaOCl. The teeth were split longitudinally for SEM examination at different thirds. The largest number of visible tubules in the three groups was in the cervical thirds, followed by the middle and apical thirds. There was no statistically significant difference between groups 1 and 2 at different thirds; however, groups 1 and 2 had significantly more visible dentinal tubules than group 3.

Semra and Ahmet (11) compared the effects of EGTA and EDTA on removal of the smear layer using SEM. Fifteen extracted single-rooted teeth were instrumented to size 60 using 10 ml of 5% NaOCl irrigation. Three teeth served as the control group, and the remaining teeth were randomly assigned into two experimental groups; samples of the first group were irrigated with 10 ml of 17% EDTA, followed by 10 ml of 5% NaOCl. The second group was irrigated with 10 ml of 17% EGTA, followed by 10 ml of 5% NaOCl. The irrigation process in all groups was continued for 2 minutes. All the roots were sectioned longitudinally and processed for SEM evaluation. Results showed that the smear layer was completely removed by EDTA, but it caused erosion of the tubuli. EGTA was somewhat effective in removing the smear layer without inducing erosion.

O'connell et al ⁽¹²⁾ compared smear layer removal using three solution of EDTA: 15% concentration of the alkaline salt, 15% concentration of the acid salt, and 25% concentration of the alkaline salt to smear layer removal in root canal system. All solutions were adjusted to pH 7.1 using either NaOH or HCI. When the EDTA solutions were alternately used for root canal irrigation with 5.25% NaOCl, they completely removed the smear layer in the middle and coronal thirds of canal preparations, but were less effective in the apical third. None of the EDTA solutions by themselves were effective at completely removing the smear