

Clinical Strategies for Management of Cases with Refractory Apical Periodontitis

Thesis, submitted to

Faculty of Dentistry, Ain Shams University

In partial fulfillment of the requirements of the Doctor
Degree in Endodontics

By

Bassem Nasr Eldin Hassan Elhady
(B.D.S. – M.Sc.)

Ain Shams University

Faculty of Dentistry
Ain Shams University

2016

بِسْمِ اللَّهِ الرَّحْمَنِ الرَّحِيمِ

﴿ قَالُوا سُبْحَانَكَ لَا عِلْمَ لَنَا إِلَّا مَا عَلَّمْتَنَا

﴿ إِنَّكَ أَنْتَ الْعَلِيمُ الْحَكِيمُ

صَدَقَ اللَّهُ الْعَظِيمُ
الْآيَةُ (32) سُورَةُ الْبَقَرَةِ

SUPERVISORS

Prof. Salma Hassan El-Ashry

Professor of Endodontics
Faculty of Dentistry
Ain Shams University

Dr. Shehab El-Din Mohamed Saber

Associate Professor of Endodontics
Faculty of Dentistry
Ain Shams University

Dr. Marwa Saad Mohammed Fathi

Associate Professor of Medical
Microbiology & Immunology
Faculty of Medicine
Ain Shams University

Dedication

This work is dedicated to:

*My parents for their endless love and care,
My wife for her continuous encouragement,
And my beloved children Kareem and Nada.*

Acknowledgment

First of all, I would truly thank God for giving me the power to complete this work and for helping me to pass every difficulty throughout my whole life.

I wish to express my deepest gratitude and appreciation to Prof. Salma Hassan El Ashry for her unlimited guidance, encouragement and valuable advice since the undergraduate part of my life. Without her motherly support this work would not have come out to light.

I would also like to express my deep thanks and gratitude to Dr. Shehab Eldin Mohamed Saber for his continuous support, stimulating guidance and his generosity with time and effort. It was a real pleasure working under his supervision.

I would also take the opportunity to thank Dr. Marwa Saad Fathi for her help, knowledgeable advice and professional guidance in the microbiologic part of this work.

Many thanks are due to Dr. Khaled Keraa for his help in accomplishing the statistical work for this study.

I would like to express my thanks to all staff members, department of Endodontics, Ain Shams University for their support and for pushing me ahead to accomplish this work.

Contents

	Page
Contents.....	I
List of Tables.....	III
List of Figures.....	V
Introduction.....	1
Review of literature.....	3
Apical periodontitis	3
Refractory Apical periodontitis.....	4
Pathogenesis of Apical periodontitis.....	5
Microbiology of the root canal system.....	8
Elimination of Endodontic infection.....	13
Enterococcus faecalis.....	16
Actinomyces.....	25
Endodontic treatment failure.....	30
Endodontic surgery.....	33
Aim of the study.....	39
Subjects and methods.....	40
Results.....	81

Discussion.....	107
Summary and conclusions.....	118
References.....	121
Appendices.....	144
Arabic summary.....	149

List of Tables

Table (1): Bacterial identification & counts in the 1st culture.

Table (2): Antibiotic susceptibility of bacterial samples of the first culture collected from part I cases.

Table (3): Enterococci counts in 1st & 2nd culture in terms of Log₁₀ CFU (descriptive statistics).

Table (4): Mean, standard deviation (SD) values and results of Wilcoxon signed-rank test for the changes in Log₁₀ CFU of Enterococci counts after the use of antibiotics.

Table (5): Actinomyces counts in 1st & 2nd cultures in terms of Log₁₀ CFU (descriptive statistics).

Table (6): Mean, standard deviation (SD) values and results of Wilcoxon signed-rank test for the changes in Log₁₀ CFU of Actinomyces counts after the use of antibiotics.

Table (7): Descriptive statistics of radio-density.

Table (8): Mean, standard deviation (SD) values and results of repeated measures-ANOVA and Tukey's tests for the changes in radio-density by time.

Table (9): Mean, standard deviation (SD) values and results of repeated measures-ANOVA for the comparison between radio-density of granulation tissue and apical cyst.

List of figures

Figure (1): Amies transport medium.

Figure (2): Eppendorf tubes containing transport media: left-broth, right-amies.

Figure (3): Microbiologic sampling.

Figure (4): Temperature preserving container used for sample transportation.

Figure (5): Vortex device used to resuspend bacteria in media.

Figure (6): Enterococci colonies on blood agar.

Figure (7): Blackening Effect of Enterococci colonies on Bile Aesculin.

Figure (8): Bleaching (reduction) of litmus milk by Enterococci.

Figure (9): Microscopic confirmation of Enterococci.

Figure (10): AnaeroGen atmosphere generating system.

Figure(11): Characteristic Actinomyces colonies on blood agar.

Figure (12): PCR work set-up.

Figure (13): Heat block used for bacterial DNA extraction.

Figure (14): Centrifuge used for bacterial DNA extraction.

Figure (15): Thermal cycler used for bacterial DNA extraction.

Figure (16): Laminar flow for mastermix preparation.

Figure (17): Electrophoresis apparatus.

Figure (18): Gel documentation system.

Figure (19): Enterococcus fecalis multiplex PCR.

Figure (20): Enterococcus Real Time PCR.

Figure (21): Zones of inhibition around antimicrobial disks on blood agar.

Figure (22): Resistance of a microbial sample to all tested antibiotics.

Figure (23): Ultrasonic device and retrotip.

Figure (24a): Surgical site of one case after apicectomy and retrograde filling.

Figure (24b): Surgical site of another case after apicectomy and retrograde filling.

Figure (25): Digital Radiographs for a case of Part II. a) one week after surgery. b) 3 months. c) 6 months.

Figure (26): Lesion in formalin before being sent to histopathology.

Figure (27): Histopathologic section for one of the samples showing granulation tissue X400.

Figure (28): Histopathologic section for one of the samples showing inflammatory cyst X200.

Figure (29): Bioaggregate retrograde filling material.

Figure (30): The paralleling device with aiming ring.

Figure (31): The aiming device with putty index.

Figure (32): Measuring Radiodensity using Digora for Windows ver 1.51.

Figure (33): Digital Radiographs of one of the cases in which signs and symptoms subsided after antibiotic application. a) preoperative, b) 3 months after obturation, c) 6 months after obturation.

Figure (34): Digital Radiographs of another case in which signs and symptoms subsided after antibiotic application. a) preoperative, b) 3 months after obturation, c) 6 months after obturation.

Figure (35): *Enterococcus fecalis* gel electrophoresis from real time PCR.

Figure (36): *Actinomyces* PCR.

Figure (37): Bacterial resistance to all tested antibiotics.

Figure (38): Bacterial sensitivity to Amoxicillin + clavulanate, intermediate sensitivity to Ciprofloxacin and resistance to Clindamycin.

Figure (39): Bacterial sensitivity to both Amoxicillin + clavulanate and Ciprofloxacin, intermediate sensitivity to Clindamycin is also evident.

Figure (40): Bar chart representing mean Log10 CFU of *Enterococci* counts.

Figure (41): Bar chart representing mean Log10 CFU of *Actinomyces* counts.

Figure (42): Digital radiographs of the sample with minimum radiodensity in the "1 week" observation period. a) 1 week postoperatively. b) 3 months. c) 6 months.

Figure (43): Digital radiographs of the sample with maximum radiodensity in all postoperative periods. a) 1 week postoperatively. b) 3 months. c) 6 months.

Figure (44): Digital radiographs of the sample that showed minimum radiodensity in both "3&6 months" postoperative periods. a) 1 week postoperatively. b) 3 months. c) 6 months.

Figure (45): Digital Radiographs illustrating minimal change in radiodensity overtime in a case of part II. a) 1 week postoperatively. b) 3 months. c) 6 months.

Figure (46): Digital Radiographs illustrating minimal change in radiodensity overtime in another case of part II. a) 1 week postoperatively. b) 3 months. c) 6 months.

Figure (47): Bar chart representing mean radio-density at different follow-up periods.

Figure (48): Histopathologic section showing Granulation tissue with inflammatory cell infiltration and bone fragments (from curettage) X200.

Figure (49): Histopathologic section showing Granulation tissue and foreign body giant cell infiltrate X400.

Figure (50): Histopathologic section showing inflammatory cyst epithelial lining X200.

Figure (51): Histopathologic section showing heavy inflammatory cell infiltration with Histiocytes most prominent X400.

Figure (52): Pie chart representing Histopathological findings.

Figure (53): Bar chart representing mean radio-density of granulomas and apical cysts.

INTRODUCTION