

بسم الله الرحمن الرحيم



-Call 4000





شبكة المعلومات الجامعية التوثيق الالكتروني والميكروفيلم





جامعة عين شمس

التوثيق الإلكتروني والميكروفيلم

قسم

نقسم بالله العظيم أن المادة التي تم توثيقها وتسجيلها علي هذه الأقراص المدمجة قد أعدت دون أية تغيرات



يجب أن

تحفظ هذه الأقراص المدمجة يعبدا عن الغبار













بالرسالة صفحات لم ترد بالأصل



The Effect of Different Antibiotics Disinfection Protocols on Chemokines Expression in Mature Teeth with Chronic Apical Periodontitis.

(An immunohistochemical animal study

Thesis Submitted to Endodontic Department, Faculty of Dentistry, Ain Shams University in Partial Fulfillment of Doctoral Degree in Endodontics

By

Wael Hamdy El Shater

B.D.S 2004 M.D.S 2012

Supervisors

Prof. Ehab El Sayed Hassanien

Professor and head of Department of Endodontics Faculty of Dentistry, Ain Shams University

Prof. Kariem Mostafa El Batouty

Professor of Endodontics
Faculty of Dentistry, Ain Shams University

Ass. Prof. Mohamed Mokhtar Nagy

Associate Professor at Department of Endodontics Faculty of Dentistry, Ain Shams University

Faculty of Dentistry Ain Shams University 2020

Contents

List of Figures	i
List of Tables	v
List of Abbreviations	vi
Introduction	1
Review of Literature	3
Aim of the Study	32
Material and Methods	33
Results	49
Discussion	71
Summary, Conclusions and Recommendations	84
References	88
Arabic summary	

List of Figures

Figure 1.	Surgical site showing the 3 lower premolar area of dog38
Figure 2.	After flap reflection and initial osteotomy of cortical plate38
Figure 3.	Apicectomy of a single premolar39
<u>Figure 4</u> .	Full apicectomy of all 3 premolars39
	Harvested samples showing embedded apices and surrounding
	tissues (buccal and top views)40
Figure 6.	Sutures in place after the procedure
Figure 7.	One week post-operative showing healing41
Figure 8.	Decalcifier device and fast decalcifying solution42
<u>Figure 9</u> .	ZEN 2 blue edition software interface
Figure 10	. Representative Photomicrographs obtained by fluorescent
	microscope for sampled tissues from different groups
	stained by red flouorochrome for IL-17 and MMP-13 and
	green flourochrome for TGF-β47
Figure 11	. Sample Photomicrographs obtained by fluorescent
	microscope for tissues stained by red flouorochrome
	for IL-1749

Figure 12.	Sample Photomicrographs obtained by fluorescent
	microscope for tissues stained by red flouorochrome
	for MMP-1350
Figure 13.	Sample Photomicrographs obtained by fluorescent
	microscope for tissues stained by red flouorochrome
	for TGF-β51
Figure 14	Day alot segmenting modicy and segmentatives for
Figure 14.	Box plot representing median and range values for
	IL-17 levels in the four groups53
Figure 15.	Box plot representing median and range values for
	TGF-β levels in the four groups55
Figure 16.	Box plot representing median and range values for
	MMP-13 levels in the four groups57
Figure 17.	Scatter diagram representing correlation between
	IL-17 and TGF-β within the control group59
	12 17 with 101 ptime the control group
Figure 18.	Scatter diagram representing correlation between
	IL-17 and TGF-β within the CP group59
Figure 10	Scatter diagram representing correlation between
rigure 19.	Scauer diagram representing correlation between

	IL-17 and TGF- β within the DAP group60
Figure 20.	Scatter diagram representing correlation between
	IL-17 and TGF-β within the TAP group60
Figure 21.	Scatter diagram representing overall correlation between
	IL-17 and TGF-β62
Figure 22.	Scatter diagram representing correlation between
	IL-17 and MMP-13 within the control group63
Figure 23.	Scatter diagram representing correlation between
	IL-17 and MMP-13 within the CP group63
Figure 24.	Scatter diagram representing correlation between
	IL-17 and MMP-13 within the DAP group64
Figure 25.	Scatter diagram representing correlation between
	IL-17 and MMP-13 within the TAP group64

Figure 26.	Scatter diagram representing overall correlation between
	IL-17 and MMP-1365
Figure 27.	Scatter diagram representing correlation between
	TGF- β and MMP-13 within the control group67
Figure 28.	Scatter diagram representing correlation between
	TGF-β and MMP-13 within the CP group67
Figure 29.	Scatter diagram representing correlation between
	TGF-β and MMP-13 within the DAP group68
Figure 30.	Scatter diagram representing correlation between
	TGF-β and MMP-13 within the TAP group68
Figure 31.	Scatter diagram representing collective correlation between
	TGF-β and MMP-1370

List of Tables

Table 1:	Steps of immunofluorescence processing technique and substances used for the samples' preparation44
Table 2:	Descriptive statistics and results of Kruskal-Wallis test for comparison between IL-17 levels in the four groups53
Table 3:	Descriptive statistics and results of Kruskal-Wallis test for comparison between TGF- β levels in the four groups54
Table 4:	Descriptive statistics and results of Kruskal-Wallis test for comparison between MMP-13 levels in the four groups 56
<u>Table 5</u> :	Results of Spearman's correlation coefficient and P-values for the correlation between IL-17 and TGF-β61
<u>Table 6</u> :	Results of Spearman's correlation coefficient and P-values for the correlation between IL-17 and MMP-1365
<u>Table 7</u> :	Results of Spearman's correlation coefficient and P-values for the correlation between TGF-β and MMP-1369

List of Abbreviations

BSA: Bovine Serum Antigen

CFU: Colony Forming Unit

CH: Calcium Hydroxide

CP: Ciprofloxacin

DAP: Double Antibiotic Paste

ECM: Extracellular matrix

ELISA: Enzyme linked immunosorbant assay.

EMMPRIN Extracellular metalloproteinase inducer

Foxp3 Forkhead box p3 Transcription factor

HDPF Human dental pulp fibroblast

IFNγ Interferon Gamma

IgG Immunoglobulin G

IL- Interlukin-

LSTR Lesion sterilization and tissue repair.

MCP Monocyte chemotactic protein

MIP Macrophage inflammatory protein

MMP- Matrix metalloproteinase-

PCR Polymerase chain reaction

PBS Phosphate-buffered saline

qRT-PCR Quantitative real time polymerase chain reaction

RANKL Receptor activator for nuclear factor kappa-B ligand

REP Regenerative endodontic procedures

RORγt RAR (Retinoic acid related)-orphan receptor gamma

RT-PCR Real time polymerase chain reaction

SCAP Stem cells of the apical papilla

T regulatory cells

TAP Triple antibiotic paste

TGF- β Transforming growth factor beta.

Th1/2/17 T helper 1/2/17

TIE TGF-β Inhibitory element

TIMP Tissue inhibitors of metalloproteinases

TNF-α Tumor necrosis factor alpha

Introduction

With the continuous advancements in tissue engineering and stem cell technology, root canal treatment concepts have been shifted to attempted tissue regeneration; that is to regenerate pulp like or more ideally actual pulp tissue inside the canal space, while maintaining the basic concepts of proper disinfection and proper canal seal. This will result not only in preserving the tooth as a structure, but also restoring the tooth functionally and histologically with almost normal architecture. In long standing cases however, where the pulp becomes necrotic, bacterial infection affects the periapical tissues resulting in alveolar bone destruction and degradation of several extracellular matrix components, manifested clinically as apical periodontitis and later as a periapical lesion. The host immune response involved in this process is complex and involves the recruitment of inflammatory cells and the production of variable cytokines, enzymes and other bioactive substances, whose monitoring may give indications about the degree of progression or regression of this lesion. Examples of these cytokines include interleukin 17 (IL-17) and transforming growth factor (TGFβ) which are major contributors to the inflammatory process. Other substances of interest on the extracellular level include the enzyme matrix metalloproteinase13 (MMP-13) which degrades extracellular matrix.

These periapical reactions also pose a challenge for the regenerative endodontic procedures (REP) due to the necessity to properly disinfect the canal space and minimize or eradicate the unfavorable immunological reaction to make way for the