THE ANTIMICROBIAL ACTION OF THREE DIFFERENT INTRA-CANAL MEDICAMENTS AGAINST ENTEROCOCCUS FAECALIS

Thesis
Submitted to Endodontic Department
Faculty of Dentistry
Ain Shams University

For Partial Fulfillment of the Requirements of
the Master Degree In Endodontics

By
Soha Ahmed Ismail Gaber
B.D.S., Faculty of Dentistry
Ain Shams University, (2004)
Endodontic Department
Faculty of Dentistry
Ain Shams University
2011

Supervisors

Prof. Dr. Hossam Mohamed Tawfik
Professor of Endodontics
Chairman of Endodontic Department
Faculty of Dentistry,
Ain Shams University

Dr. Abeer Abd El-Hakim M. Mohamed
Lecturer of Endodontics
Faculty of Dentistry,
Ain Shams University

Dr. Soha Abd El Rahman El-Hady
Ass. Professor of Bacteriology and Immunology
Faculty of Medicine,
Ain Shams University

المقاومة البكتيرية لثلاث أنواع مختلفة من عقارات القنوات العصبية تجاه المكورة المعوية

رسالة

مقدمة إلى كلية طب الأسنان جامعة عين شمس
توطئة للحصول على درجة الماجستير فى
علاج الجذور

مقدمة من

الطبيبة/ سها أحمد إسماعيل جابر
بكالوريوس طب وجراحة الفم والأسنان
جامعة عين شمس

كلية طب الأسنان
جامعة عين شمس
٢٠١١

المشرفون

أ.د./حسام محمد عصام الدين توفيق
أستاذ ورئيس قسم علاج الجذور
كلية طب الأسنان - جامعة عين شمس

د./ عبير عبد الحكيم محمود محمد
مدرس علاج الجذور
كلية طب الأسنان - جامعة عين شمس

د./ سهـا عبـد الرحمـن الهادى
أستاذ مساعد البكتريا والمناعة الطبية
كلية الطب - جامعة عين شمس

كلية طب الأسنان
جامعة عين شمس
٢٠١١

Contents

List Of Tables

List Of Figures

List Of Figures *(Cont..)*

List Of Figures *(Cont..)*

INTRODUCTION

The main goal of root canal treatment is to eliminate bacteria and their by products from root canals before filling. The majority of bacteria found in the root canal may be simply eliminated by the mechanical action of endodontic instruments. Nevertheless, because of the anatomical complexities of many root canals, organic residues and bacteria located in the dentinal tubules cannot be sufficiently cleaned, even after meticulous mechanical procedures. Therefore, various irrigation and medication have been used during and immediately after root canal preparation to remove debris and necrotic pulp tissue and to eliminate microorganisms from the root canal [(1)].

As a facultative organism, *Enterococcus faecalis* can tolerate a wide variety of growth conditions, including temperatures of 10°C to 45°C, and hypotonic, hypertonic, acidic, or alkaline environments. Several studies have shown that *E.faecalis* resist various intracanal treatment procedures. This is attributed to their ability to penetrate dentinal tubules, withstand high pH values, possession of virulence factors and biofilm formation[(2)].

Bacterial biofilms is a "polysaccharide matrix enclosed bacterial populations adherent to each other and/or to surfaces

or interfaces". Biofilms are highly organized structures consisting of mushroom-shaped clumps of bacteria bound together by a carbohydrate matrix and surrounded by water channels that deliver nutrients and remove wastes. Bacteria sequestered in biofilms are shielded and are often much harder to kill than their free-floating or "planktonic" counterparts[3].

Although systemic antibiotics appear to be an effective adjunct in certain surgical and nonsurgical endodontic procedures, their administration is not without the potential risk of adverse systemic effects, such as allergic reactions. Local application of antibiotics within the root canal system has been advocated. Recently a mixture of ciprofloxacin, metronidazole, and doxcycline has been shown to be very effective in eliminating endodontic pathogens in vitro and in situ[4]. However, the efficiency of this triple antibiotic paste toward *E.fecalis* biofilm is still unclear and need further researches.

REVIEW OF LITERATURE

An infection of the pulp can result in microbial colonization of the entire root canal system, together with the dentinal tubules adjacent to the canal. Instrumentation and antibacterial irrigation will render 50% to 70% of infected canals free of microorganisms while the remaining canals contain vital bacteria. These microorganisms and their toxic metabolic products are responsible for the development and persistence of apical periodontitis of endodontal origin.

- ***Endodontic microbial flora***

Baumgartner and Falkler[5], tested the presence of bacteria in the apical 5 mm of infected root canals. Ten freshly extracted teeth which had carious pulpal exposures and periapical lesions contiguous with the root apex were placed inside an anaerobic chamber and the apical 5mm of the root canals cultured. In addition to anaerobic incubation, duplicate cultures were incubated aerobically. Fifty strains of bacteria were isolated and identified. The most prominent bacteria cultured were *Actinomyces, Lactobacillus, black-pigmented Bacteroides, Peptostreptococcus, nonpigmented Bacteroides, Veillonella, Enterococcus faecalis, Fusobacterium nucleatum*, and *Streptococcus mutans.* Of the 50 bacterial isolates, 34 (68%) were strict anaerobes. This study demonstrates the

presence of predominantly anaerobic bacteria in the apical 5mm of infected root canals in teeth with carious pulpal exposures and periapical lesions.

Molander et al.[6], examined the microbiological status of 100 root-filled teeth with radiographically verified apical periodontitis (the pathology (P) group)and of 20 teeth without signs of periapical pathosis(the technical (T) group). In the P group 117 strains of bacteria were recovered in 68 teeth. Facultative anaerobic species predominated among these isolates. Growth was classified as 'sparse' or 'very sparse' in 53%, and as 'heavy' or 'very heavy' in 42%. *Enterococci* were the most frequently isolated genera, showing 'heavy' or 'very heavy' growth in 25 out of 32 cases (78%). In 11 teeth of the T group no bacteria were recovered, whilst the remaining nine yielded 13 microbial strains. Eight of these grew 'very sparsely'. It is concluded that the microflora of the obturated canal differs from that found normally in the untreated necrotic dental pulp, quantitatively as well as qualitatively.

Peciuliene et al.[7], studied the occurrence of *Enterococcus faecalis* in root canals of previously root filled teeth that requiring retreatment. Twenty-five asymptomatic teeth were included in the study. Microbiological samples were taken from the canals before and after preparation and irrigation with sodium hypochlorite and EDTA. Microbes were isolated from 20 of 25 teeth. *E.faecalis* was isolated from 14 of those 20

culture positive teeth usually in pure culture or as a major component of the flora. Second samples taken after preparation revealed growth in 7 of the 20 teeth. Five of the seven cases were *E.faecalis* in pure culture. Isolation of *E.faecalis* was not related to the use of any particular root filling material in the original root filling. The results indicate that, rather than previous chemical treatment, it is the ecological conditions present in the incompletely filled root canal that are important for the presence of *E faecalis* in these teeth.

Love[8], explained how *E. faecalis* could survive and grow within dentinal tubules and reinfect an obturated root canal. Cells of *Streptococcus gordonii*, *Streptococcus mutans*, or *E.faecalis* were grown in brain heart infusion broth containing various amounts of human serum for 56 days. The ability of the three species to invade dentine and bind to immobilized type 1 collagen in the presence of human serum was assessed by dentine invasion and microtitre. Results show that**,** cells of all three bacteria were able to invade dentine and bind to immobilized collagen. Both of these properties were inhibited by the presence of collagen in the cell solution. Human serum inhibited dentine invasion and collagen adhesion by *S. gordonii* and *S. mutans*, whilst dentine invasion by *E. faecalis* was reduced in the presence of serum, but not inhibited, and binding to collagen was enhanced.

John et al.[3], tested the hypothesis that *Enterococcus faecalis* resists common intracanal medications by forming biofilms . *E. faecalis* colonization of 46 extracted, medicated roots was observed with scanning electron microscopy (SEM) and scanning confocal laser microscopy**.** SEM analysis showed bacterial colonization of root canals medicated with calcium hydroxide points within 2 days. While biofilms in canals medicated with calcium hydroxide paste were detected in an average of 77 days. Scanning confocal laser microscopy analysis of two calcium hydroxide paste medicated roots showed viable colonies in a root canal infected for 86 days, whereas in a canal infected for 160 days, a mushroom-shape typical of a biofilm was observed.

Mário et al.[9], evaluated the presence of bacterial biofilm on the external surface of the root apex in teeth with pulp necrosis, with and without radiographically visible periapical lesions, and in vital teeth. Twenty-one teeth were extracted, eight with pulp necrosis and periapical lesions, eight with pulp necrosis without radiographically visible periapical lesions, and five with vital pulp. The roots were sectioned, and the root apices were processed for scanning electron microscope evaluation. The surface of the apical root was evaluated for the presence of microorganisms, root resorption, and biofilm. Results showed that there were no microorganisms on the apical root surface of either teeth with