
**Mechanical Reduction of Enterococcus Faecalis
following Root Canal Enlargement Using Four
Rotary Nickel-Titanium Systems**

[An In Vitro Study]

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Introduction

The cleaning of dentin within the root canal system and the removal of inflamed and/or necrotic tissue remains as one of the most important steps in endodontic therapy. It has been demonstrated that eradication of endodontic infection enhances the success rate of the endodontic therapy. In infected root canals reducing the bacterial count is accomplished by a combination of mechanical instrumentation, various irrigation solutions, and antimicrobial agents placed into the canal.

Enterococci are part of the human normal flora. They are recognized as potential human pathogens causing 12% of the nosocomial infections. Development of multiple resistances to various antibiotics in *enterococci* poses serious therapeutic problems. *E. faecalis* accounts for around 80% of all infections caused by *enterococci*. In endodontics, *E. faecalis* is rarely present in primary apical periodontitis, but it is the dominant microorganism in root-filled teeth presenting with the post-treatment apical periodontitis. It is often isolated from the root canal in pure culture, but it can also be found together with some other bacteria or yeasts. When present in mixed infections, *E. faecalis* is typically the dominant isolate.

Reduction of bacterial counts by mechanical means depends on the type of root canal enlarging instruments and the method of instrumentation. The introduction of rotary (NiTi) enlarging instruments has revolutionized the old method of canal instrumentation. These recent systems offer an easy, efficient and

safe method for canal enlargement. However, differences in geometrical designs between these different systems could have a direct impact on their cleaning efficiency.

Aim of the Study

The aim of the present study was to assess the efficiency of four rotary NiTi root canal enlarging systems in cleaning root canals infected by *Enterococcus faecalis* biofilm.

Review of Literature

1. Intracanal Microbes in Relation to Pulp and Periapical Pathosis

The role of microorganisms in the initiation and propagation of pulp and/or periapical diseases have been clearly demonstrated since the classic study by *Kakehashi* ⁽¹⁾. The bacterial population inside infected root canals shows wide variety of aerobic and anaerobic species. Different studies have reported on the incidence of different micro-organisms.

Kannangara et al ⁽²⁾ studied the microbiologic and therapeutic aspects of sixty-one cases of pyogenic dental infection through the use of modern anaerobic culture methods. Forty-five (74 percent) patients had anaerobic infections. Among them, eighteen (29.5 percent) had *Bacteroides fragilis*, of which six were resistant to penicillin at 16 microgram/ml. but all were susceptible to clindamycin at less than 2 microgram/ml. Of twenty-five patients treated with 4 to 20 million units of penicillin per day, twenty were cured and did not suffer relapse. The five patients in whom treatment failed had mandibular fractures infected with *B. fragilis*. Of ten patients treated with clindamycin (600 mg. intravenously every 6 hours), which included five patients with *B. fragilis* infections, all were cured. The presence of *B. fragilis* in dental infections has not been recognized. Dental infections associated with mandibular fracture that fail to respond to conventional penicillin therapy should be routinely cultured for *B. fragilis*.

Williams et al ⁽³⁾ cultured aspirates from 10 dental abscesses of endodontic origin, all of which had penetrated beyond the bony alveolus to produce fluctuant swelling. Sampling was done by syringe aspiration under strict anaerobic techniques, including the use of an anaerobic chamber. Randomly selected

colonies (100) from each culture were purified, characterized, and identified. Results showed that seventy percent of the bacterial isolates were either strict anaerobes or microaerophilic. One abscess yielded a pure culture of a viridans streptococcus, *Streptococcus milleri*. *Streptococcus intermedius* dominated the flora in a second abscess. The common oral streptococcus, *Streptococcus sanguis*, constituted only 2% of the isolates from one additional infection. *Fusobacterium nucleatum*, *Bacteroides melaninogenicus*, other *Bacteroides* including *B. oralis* and *B. ruminicola*, anaerobic diphtheroids, *Peptostreptococcus micros*, and *Staphylococcus epidermis* were other predominant isolates.

Kipioti et al ⁽⁴⁾ examined the flora from the root canals and periodontal pockets of teeth with advanced periodontal disease in order to compare the predominant cultivable microflora from the canals with those found in the adjacent periodontal pockets. Three samples were collected from each of sixteen patients, one from the root canal and two from the adjacent periodontal pocket in two different sites. Criteria for inclusion in the survey were pocket depth of greater than or equal to 7 mm, intact clinical crowns, and lack of periapical lesions. The study has demonstrated that the microorganisms present in the root canals of caries-free teeth with advanced periodontitis generally resembled those found in the adjacent periodontal pockets. The similarity in the recovery of organisms in the current study suggests that the pocket could be the source of the root canal infections.

Ranta et al ⁽⁵⁾ studied the microbiology and treatment of apical periodontitis in 62 patients, followed-up for 1 year. The clinical treatment and the radiological technique were standardized. One third of the patients received no systemic antibiotics, one third received phenoxymethylpenicillin for 1 week, and one third for 3 months. The results showed that on the average, 0–2 facultatively anaerobic

and 4–6 anaerobic bacteria were isolated from periapical infections. The genera *Bacteroides* and *Fusobacterium* in addition to anaerobic gram-positive cocci were found most frequently. Minimum inhibitory concentrations of benzylpenicillin against 269 strains were tested. 12 strains were resistant at the concentration 2.4 µg/mi, including 4 strains of *Fusobacterium* sp. All patients were free of symptoms 1 month after the admission. Only 1 patient failed to show any progress in radiological healing. No differences between the groups in relation to penicillin therapy were found.

Baumgartner ⁽⁶⁾ observed that oral microorganisms, either directly or indirectly, produce the majority of cases of pulpal and periapical disease. Because microorganisms play such an important role in the pathogenesis of pulpal and periapical disease, a fundamental understanding of the disease process is important. Culture studies have identified infections of endodontic origin as being polymicrobial and predominantly anaerobic. Although several species of bacteria have been associated with infections of endodontic origin, there is no absolute correlation with signs and symptoms.

Sundqvist ⁽⁷⁾ investigated the existence of commensal or antagonistic relationships between microorganisms in the root canals of teeth with apical periodontitis. Samples were taken from 65 infected human root canals and were analysed according to species, frequency of occurrence and proportion of the total isolated flora. The most frequent species were *Fusobacterium nucleatum*, *Prevotella intermedia*, *Peptostreptococcus micros*, *Peptostreptococcus anaerobius*, *Eubacterium alactolyticum*, *Eubacterium lentum* and *Wolinella recta*. An odds ratio system was used to calculate positive or negative associations between the isolated bacteria. Strong positive associations were found between *F. nucleatum* and *P. micros*, *Porphyromonas endodontalis*, *Selenomonas*

sputigena and W. recta. There was also a positive association between P. intermedia and P. micros, P. anaerobius and the eubacteria. The author concluded that the results are consistent with the concept of a special and selective environment occurring in the root canal that is due, in part, to the cooperative as well as antagonistic nature of the relationships between bacteria in the root canal.

In a second study *Sundqvist* ⁽⁸⁾ reported that the root canal represents a special environment in which selective pressures result in the establishment of a restricted group of the oral flora. Population shifts occur over time with obligate anaerobes ultimately dominating the bacterial mix. Bacterial interrelationships and the nutritional supply are key factors in determining the outcome of the infection. Endodontic treatment, apart from directly eliminating bacteria, can completely disrupt the delicate ecology and deprive persisting bacteria of their nutritional source.

Trowbridge and Stevens ⁽⁹⁾ reported that the greatest cause of endodontic and periapical pathosis is microbial infection of the pulp. Most odontogenic infections are of a polymicrobial nature. With advances in anaerobic isolation and culturing techniques, much has been learned about the presence of pathogenic organisms such as Porphyromonas and Prevotella species (formerly classified as black-pigmented Bacteroides species) in infected root canals.

Haapasalo ⁽¹⁰⁾ in a review article suggested that necrotic dental root canal infections are polymicrobial infections dominated by anaerobic bacteria. The number of different species in one canal is usually low, approx. 4-7 species. The species isolated most frequently belong to the genera Prevotella, Porphyromonas, Fusobacterium, Peptostreptococcus, Eubacterium and Streptococcus. The frequency of isolation of black-pigmented Gram-negative anaerobes in endodontic infections varies from 25% to > 50%. Pr.

intermedia is the most commonly found pigmented species, followed by *Pr. denticola* and two *Porphyromonas* species, *P. gingivalis* and *P. endodontalis*.

Sundqvist ⁽¹¹⁾ demonstrated that bacteria present in infected root canals include a restricted group of species compared with the total flora of the oral cavity. Conditions exist in the root canal that permit the growth of anaerobic bacteria capable of fermenting amino acids and peptides, whereas bacteria that mainly obtain energy by fermenting carbohydrates are restricted by lack of available nutrients. During the course of infection interrelationships develop between microbial species and population shifts are produced as a result of these interactions. Strong associations occur between certain species which are most likely based on nutritional demands and nutritional relationships. The pathogenicity of the polymicrobial root canal flora is dependent on bacterial synergy.

Weiger et al ⁽¹²⁾ examined the occurrence of bacteria in 12 endodontically induced periodontal lesions associated with sinus tracts. The microbial flora encountered in the sinus tract was compared with that of the root canal of the involved teeth which had not experienced any prior endodontic therapy. All microbiological samples taken from the sinus tract and from the root canal system contained bacteria. Seventy-one strains were detected in the extraradicular lesions. Of the anaerobic species, *Fusobacterium nucleatum* (7 strains), *Prevotella intermedia* (4 strains) and *P. oralis* (4 strains) were most frequently found. In the group of the facultative anaerobes *Streptococcus* spp. were predominant. Ninety-four strains were isolated from the root canal system of the 12 teeth. *P. intermedia* (6 strains), *P. buccae* (5 strains), *F. nucleatum* (5 strains) and *Lactobacillus plantarum* (5 strains) were most common. In 9 cases, species present in the root canal could be revealed in the extraradicular lesions. It was

concluded that a variety of microorganisms were capable of colonizing endodontically induced, extraradicular lesions clinically characterized by sinus tracts.

Chaudhry et al ⁽¹³⁾ evaluated microbiological and clinical data from 56 patients with endodontic infections. Samples were collected using autoclaved paper points. Specimens were processed for isolation of aerobic and anaerobic bacteria. Antimicrobial sensitivity and resistance profiles of the recovered isolates was also performed. Forty nine positive cultures (87.5%) were obtained from the 56 consecutive necrotic root canal systems which were sampled. A total of 69 aerobic bacteria and 21 anaerobic bacteria were recovered. Aerobic bacteria were isolated from 35 patients (72%), anaerobic bacteria from 3 (6%) and mixed aerobic and anaerobic bacteria from 11 patients (22%). The most common aerobic isolate was *Klebsiella pneumoniae*. The predominant anaerobic isolate was *Bacteroides* species. One isolate was recovered from 25 patients (51%) whereas in the remaining 24 patients (49%) more than 1 isolate were recovered.

Iren et al ⁽¹⁴⁾ investigated the relationship between bacteriological findings and clinical treatment procedures in root canal treatment cases. Two groups of teeth were selected based on the type of infection present in the root canal system. The 'enteric bacteria' group consisted of 40 sequential cases where *Enterococcus faecalis* and/or other facultative enteric bacteria or *Pseudomonas* sp. were found in the samples in pure culture (35%) or together with other types of bacteria. The group 'non-enteric bacteria' consisted of 40 sequential cases where only non-enteric bacteria were found. Depending on the clinical handlings of cases it was found that if the root canals had been unsealed at some point during the treatment, enteric bacteria were found more frequently than in canals with an adequate seal between the appointments. Of cases with enteric bacteria 55% had been open