In Vitro Evaluation of The Antibacterial Effect of Three Endodontic Sealers

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Elimination of microorganisms from the root canal system has always been an integral part of successful endodontic therapy and a primary objective to be reached during root canal treatment since bacteria or their byproducts are considered to be the main etiological agents of pulpal necrosis, periapical lesions and failure of well treated teeth resulting from persisting microorganisms in the apical portion of the root canal system due to the anatomical complexities of many root canals, such as dentinal tubules, ramifications, deltas, and fins which cannot be sufficiently cleaned.

Current strategies of biomechanical instrumentation, irrigation, and intra-canal medication may reduce significantly the population of microorganisms in an infected root canal, but hardly achieve complete elimination of biofilms from endodontic sites, leaving 40%–60% of the root canals still positive for bacterial presence [(1)].

By eliminating or reducing the bacterial concentration within a canal system the prognosis of endodontic therapy is significantly influenced; aiming to lower the critical concentration of microbes and disrupting biofilms will give the best chance for a positive host response.

It had been advocated that an ideal root canal sealer should be biocompatible, dimensionally stable, it should seal well and have a strong antimicrobial effect upon contact with microbes and biofilms and ideally over time be able to maintain this effect.

Enterococcus species constitute only a small proportion of the initial flora in untreated root canal; however this genus is the most commonly recovered one from teeth with failed root treatment about 38% [(2)] and even up to 77% [(3)] in recent studies. Hence, the choice of a sealer with high antimicrobial activity may be advantageous in decreasing or avoiding growth of these remaining microorganisms particularly in clinical situations of persistent or recurrent infection.

The agar diffusion test was the most commonly used technique to assess antibacterial activity of sealers. But it has many limitations as it is depends on physical properties and diffusion of tested materials. Direct contact test was developed by Weiss et al. [(4)] where the antibacterial activity of the endodontic sealers can be evaluated by measuring the kinetics of bacterial growth. Even insoluble materials can be tested with this quantitative assay.

Bioceramic based materials have been recently introduced in endodontics, mainly as repair cement and as root canal sealer.

Bioceramics are the result of the combination between calcium silicate and calcium phosphate that are applicable for biomedical and dental use. According to manufacturers, bioceramic materials show alkaline pH, antibacterial activity, radiopacity, and biocompatibility.

Based on such evidence, the antibacterial effect of three different based sealers currently available in the market was thought to be of value.

*A- **Microbiology of Dental Infection***

Rocas et al [5] investigated the prevalence of E. faecalis in endodontic infections by taking samples from cases of untreated teeth with asymptomatic chronic periradicular lesions, acute apical periodontitis, or acute periradicular abscesses and root filled teeth associated with asymptomatic chronic periradicular lesions. DNA was extracted from collected samples, then a 16S rDNA-based nested polymerase chain reaction assay was done. They found that E. faecalis is significantly more associated with asymptomatic cases of primary endodontic infections than with symptomatic one; furthermore E. faecalis was much more likely to be found in cases of failed endodontic therapy than in primary infections.

Gomes et al [6] investigated the presence of Enterococcus faecalis in endodontic infections by culture and polymerase chain reaction analyses (PCR). Microbial samples were obtained from 100 teeth, 50 with untreated necrotic pulps (primary infection) and 50 with failing endodontic treatment (secondary infection). E. faecalis detection was done by culture techniques including serial dilution, plating, incubation, and biochemical identification. For PCR detection, samples were analyzed using a species-specific primer of the 16S rDNA and the downstream intergenic spacer region. They found that in culture the test species were detected in

23 of 100 and in PCR E. faecalis was detected in 79 of 100 teeth. E faecalis was cultured from 2 (4%) of 50 necrotic canals and from 21 (42%) of 50 root treated canals. PCR detection identified the target species in 41 (82%) and 38 (76%) of 50 primary and secondary infections respectively. They concluded that E faecalis was detected as frequently in teeth with necrotic pulp as in teeth with failing endodontic treatment when a PCR analysis was used.

Zoletti et al [7] investigated the prevalence of Entercoccus faecalis in root filled teeth with or without periradicular lesions using species specific 16S rRNA gene based polymerase chain reaction (PCR) or conventional culture procedures for identification of this species. They found that of 27 root filled teeth with no periradicular lesions, E. faecalis was found in 22 cases (81.5%) by PCR and in five cases (18.5%) by culture and of 23 root filled teeth with periradicular lesions, E. faecalis was identified in 18 cases (78%) by PCR and in three cases (13%) by culture. They concluded that PCR was significantly more effective than culture in detecting this bacterial species and regardless of the identification technique used, no significant difference was observed when comparing the occurrence of E. faecalis in root filled teeth with and without periradicular lesions these findings put into question the status of E. faecalis as the main species causing endodontic treatment failure.

Gomes et al [8] investigated the presence of nine bacterial species in 45 canal samples of root filled teeth associated with periapical lesions. Microbial detection was carried out by a PCR assay using species specific primers of 16S rDNA and the downstream intergenic spacer region. They found that Enterococcus faecalis was the most prevalent species 77.8% of the study teeth. They concluded that E. faecalis was the most frequently identified test species by PCR in teeth with failing endodontic treatment.

Murad et al [9] conducted a study to investigate the composition of the root canal microbiota in endodontic failures in order to identify and quantify these microorganisms. Microbiological samples were taken from 36 root canals with persistent endodontic infection. The presence, levels, and proportions of 79 bacterial species were determined by checkerboard DNA-DNA hybridization. The Pearson correlation coefficient was used to investigate the relations between bacterial counts and clinical conditions. They found that Enterococcus faecium (36%), Streptococcus epidermidis (36%), Enterococcus faecalis (28%), Eubacterium saburreum (28%), Parvimonas micra (28%),Streptococcus sanguis (28%), Capnocytophaga sputigena (28%), Leptotrichia buccalis (28%), and Staphylococcus warneri (28%) were the most prevalent species;

and there was a low prevalence of Treponema socranskii(3%), Fusobacterium periodonticum (3%), Capnocytophaga gingivalis (3%), and Spiroplasma ixodetis (3%). The highest mean levels were found for the following species: E. faecium, Dialister pneumosintes, Staphylococcus epidermidis and Helicobacter pylori. A positive correlation was found between the area of the periapical lesion and the levels of gram negative and rod species. They concluded that the microbiota from teeth with persistent apical periodontitis presents a mixed and complex profile, hosting E. faecium (36%) and S. epidermidis (36%) as the most highly prevalent species. No correlation was found between any of the species tested and clinical findings; however periapical lesions with the largest areas presented higher counts of gram-negative and rod species.

Zhang et al [10] did a systematic review to compare the prevalence of Enterococcus faecalis in primary and persistent intraradicular infections. An exhaustive literature search combined with specified inclusion criteria was performed to collect all studies comparing the prevalence of E. faecalis in root canals with primary and persistent intraradicular infections. The systematic review included 10 studies covering 972 teeth after descriptive statistics were applied and excluding studies with uncertain forms of pulpal and periradicular lesions in their