Photoactivated Disinfection of Root Canal System with FotoSan

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Abstract: The aim of this in vitro study was to determine the antibacterial activity of photoactivated disinfection with FotoSan on an experimental bacterial biofilm in the root canal system consisting of Candida albicans and Enterococcus faecalis. Material and methods: The study was performed on 22 single root extracted teeth. The selected teeth were prepared with rotary nickel-titanium endodontic files and divided in three groups – two test groups (n=20) and one control group (n=2). According to the root canal disinfection method, the test samples (n=20) were randomized in two sub-groups: 1^{st} group - passive irrigation, 2^{nd} group – photoactivated disinfection with FotoSan. Results: After microbiological analysis no Candida albicans colonies were observed in test samples treated withFotoSanphotoactivated disinfection and the percentage of Enterococcus faecalis was reduced to 99,9%.

Keywords: FotoSan, photoactivated disinfection, Candida albicans, Enterococcus faecalis.

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I. Introduction

The main objective of endodontic treatment is to eliminate the pathogenic microorganisms and their byproducts from the root canal system by shaping, cleaning and disinfecting an infected root canal space. However, chemo-mechanical procedures may fail to clean each area of the root canal system¹. The most common microbial species associated with endodontic failure are: *Enterococcus faecalis, Streptococci, Staphylococci, Actinomyces, Pseudomonas, Candida albicans, Prevotella*etc². Nowadays there are new methods such as sonic, ultrasonic irrigation, various lasers and photodynamic therapy, which provide extra disinfection of root canal system^{3, 4,5, 6}.

Photoactivated disinfection (PAD) has been proposed as an appropriate antimicrobial treatment to maximize root canal disinfection. Light of an appropriate wavelength activates a photosensitizing molecule attached to the bacterial/fungal membrane and eventually leads to production of highly reactive oxygen species able to kill microorganisms⁷.

The aim of this *in vitro* study was to determine the antibacterial activity of PAD with FotoSan (CMS Dental, Denmark)on an experimental *Candida albicans*(CA) and *Enterococcus faecalis* (EF) biofilm in the root canal system of single root extracted teeth.

II. Material And Methods

The study was performed on 22 single root freshly extracted teeth. The root canals were prepared with rotary nickel-titanium endodontic files (Pro Taper, Dentsply, Maillefer) to final apical size F3 (30/09). For chemical disinfection during the root canal preparation a2.5% NaOCl was used. After the final irrigation the root canals of all extracted teeth were dried and infiltrated with experimental biofilm containing: 200 mL of LBG growth media, 1 cm³ CA and 1 cm³ EF. Teeth were placed in vials with 5 mL LBG and were incubated at 37°C for 7-10 days. As control group were used 2 teeth (n=2). After the contamination, the control samples weren't subjected to any disinfection. According to the root canal disinfection method, the test samples (n=20) were randomized in two sub-groups: 1st group (n=10): passive irrigation with 2.5% NaOCl for 1 min, distilled water, 17% ethylenediaminetetraacetic acid (EDTA) for 1 min and drying with 95° alcohol and paper points; 2ndgroup (n=10) - PAD with FotoSan (Fig. 1). The protocol for the second test group included: irrigation with 2.5% NaOCl for 1 min, distilled water and 17% EDTA. After drying with 95° alcohol and paper points, a photo sensitizer (PS) was applied into the root canal (Toluidine Blue) and activated with LED light source (628 nm,

2000mW/cm²) for 1 min (Fig. 2). After the activation the PS was removed from the root canal by irrigation with distilled water and drying with paper points.





Dentinal debris for subsequent microbiological analysis was taken from each root canal of the test groups (n=20). The material was collected with sterile H-files and stored insterile containers.

In the microbiological laboratory it was made ten-fold dilutions from control group. From each dilution a material was taken and seeded into 3 petri dishes with the following media: EVA for EF and Sabouraud dextrose chloramphenicol agar for CA. The samples were thermostated for 24 ± 3 h at 37° C (for the bacteria) or 24-48 h at $32.5\pm2.5^{\circ}$ C (for yeast) until single colonies appeared. The number of grown colonies was determined.

III. Result

After the microbiological analysis, the following numbers of isolated microorganisms were found:

- control group: $CA - 4x10^4$ chm/cm³, EF - 6.1x10⁵ chm/cm³; - 1st group (Fig. 2): petri dish 1: CA - 4 chm/cm³, EF - 28 chm/cm³; petri dish 2: CA - 30 chm/cm³, EF - 82 chm/cm³; petri dish 3: CA - <1 chm/cm³, EF - 82 chm/cm³; petri dish 4: CA - 15 chm/cm³, EF - 36 chm/cm³; petri dish 5: CA - 47 chm/cm³, EF - 18 chm/cm³; petri dish 6: CA - 6 chm/cm³, EF - 18 chm/cm³; petri dish 7: CA - <1 chm/cm³, EF - 37 chm/cm³; petri dish 8: CA - 11 chm/cm³, EF - 39 chm/cm³; petri dish 9: CA - <1 chm/cm³, EF - 20 chm/cm³; petri dish 10: CA - <1 chm/cm³, EF - 74 chm/cm³;





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- 2^{nd} group (Fig. 3):
petri dish 1: CA - <1 chm/cm<sup>3</sup>, EF - 2 chm/cm<sup>3</sup>;
petri dish 2: CA - <1 chm/cm<sup>3</sup>, EF - 5 chm/cm<sup>3</sup>;
petri dish 3: CA - <1 chm/cm<sup>3</sup>, EF - 6 chm/cm<sup>3</sup>;
petri dish 4: CA - <1 chm/cm<sup>3</sup>, EF - 10 chm/cm<sup>3</sup>;
petri dish 5: CA - <1 chm/cm<sup>3</sup>, EF - 8 chm/cm<sup>3</sup>;
petri dish 6: CA - <1 chm/cm<sup>3</sup>, EF - 15 chm/cm<sup>3</sup>;
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petri dish 7: CA - <1 chm/cm<sup>3</sup>, EF - 7 chm/cm<sup>3</sup>;
petri dish 8: CA - <1 chm/cm<sup>3</sup>, EF - 3 chm/cm<sup>3</sup>;
petri dish 9: CA - <1 chm/cm<sup>3</sup>, EF - 20 chm/cm<sup>3</sup>;
petri dish 10: CA - <1 chm/cm<sup>3</sup>, EF - 12 chm/cm<sup>3</sup>;
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The study found that no CA colonies were observed in FotoSan PAD, and the percentage of EF was reduced to 99,9%.

IV. Discussion

Treatment failure and persistent periapical pathology can result from microbialin fections because of inadequatedisinfection of therootcanals. Root canals function as an incubator since they are closed spaces with low oxygen concentration and therefore they promote the growth of microorganism. As a result of limited access to different regions in the root canal system, preparing a canal completely devoid of microorganisms in an infected tooth is extremely difficult or even impossible despite proper instrumentation and irrigation. Resistant microorganisms such as EF, gram-negative facultative anaerobic bacilli, CA can have a major role in the persistence of infection in the root canal system⁸.

In the last few years new irrigating solutions or additional techniques for endodontic disinfection have been proposed and tested. A modern device with the aim to minimize or eliminate the bacterial population in the root canal is PAD^{9, 10, 11, 12}. PAD is not only effective against bacteria, but also against other microorganisms including viruses, fungi, and protozoa^{7, 13, 14, 15, 16, 17}. Many studies have shown its efficacy in endodontic bacteria (Fusobacteriumnucleatum, Peptostreptococcus micros, Prevotellaintermediaand Streptococcus intermedius) in planktonic suspension and in root canals, and concluded that PAD kills endodontic bacteria at statistically significant levels¹⁸. Soukos et al investigated the effects of PAD on endodontic pathogens in planktonic phase as well as on EF biofilms in experimentally infected root canals of extracted teeth⁹. In 2010 Schlafer et al tested the antimicrobial effect of FotoSan on endodontic suspension and after inoculation of S. intermedius into prepared root canals of extracted molars. In the test they verified a strong reduction of the number of viable endodontic pathogens both in planktonic suspension and in root canals⁷.

One of the advantages of photosensitization compared to traditional antimicrobials is that as the interaction of highly reactive oxygen with organic molecules is not specific, any macromolecule within microbial cell may become a potential target, thus hindering the development of mechanisms of microbial resistance. Furthermore, the procedure can be repeated several times, as it there are no reports of cumulative effects^{19, 20}.

PAD has other attributes that make it an excellent tool in intracanal bacterial reduction, such as: safety for human tissues, ability to eradicate pathogens in biofilms, easy to apply and painless²¹.

V. Conclusion

FotoSan may be considered an adjunctive procedure to kill residual bacteria in the root canal system after standard endodontic treatment.

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