Evaluation of the effectiveness of the use of photodynamic therapy (PDT) after cleaning and shaping the root canal: An in vivo study

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ABSTRACT

Objective: The purpose of this study was to assess the effectiveness of photodynamic therapy (PDT) in teeth with periapical lesion seen radiographically. **Methods:** 24 anterior teeth were selected and divided into two groups with 12 teeth each. Three samples were collected in each specimen. In G1, endodontic treatment was performed with a nickel/titanium rotary system, with the first sample being collected at surgical opening; the second, after endodontic treatment; and the third, seven (07) days after canal preparation. In G2, the samples were collected by means of the same procedures used in G1, except for the second sample which was taken after instrumentation and photodynamic therapy (PDT). The third sample was collected seven days (07) after PDT. Sodium hypochlorite at 5.25% was used as irrigating solution, neutralized with 5% sodium thiosulfate at a given time of the study. Photon Laser III was used for 40 seconds, with 0.005% methylene blue dye as photosensitive substance. All samples from both groups were send to laboratory analysis to have potential contamination of the root canal system checked. **Conclusion:** The results revealed no statistically significant difference between groups, but further randomized studies are necessary to demonstrate the effectiveness of PDT.

Keywords: Laser therapy. Enterococcus faecalis. Root canal therapy.

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Introduction

Endodontic treatment poses limitations that can undermine a successful therapy. One often faces hurdles such as anatomical variability, calcifications and atresia when cleaning and shaping the root canal. These difficulties hinder proper cleaning and shaping while favoring bacterial reinfection.

Schilder[13] reported that in cleaning and shaping the root canal one must meet all the requirements necessary to ensure a favorable prognosis. These requirements include conical preparation without changing the position of the apical foramen, removal of bacteria and tissue debris, smoothing of the walls and increase in dentin permeability with the aid of irrigating chemicals which favor microbial decontamination and facilitate access by the filling material. Decontamination and microbial reduction can be more easily achieved in the root canal lumen, although certain bacterial strains can survive in anaerobic environments in the root canal system concealed in accessory, lateral and secondary canals.

Given that the microorganisms reside inside the root canal, they cannot be eliminated by defense mechanisms alone. It is believed that the most likely cause of endodontic failure is the survival of microorganisms in the apical third of the root canal. These microorganisms can also be found in areas of the root canal normally inaccessible to disinfection procedures such as isthmuses, craters, recesses and ramifications of the root canal system, thereby rendering endodontic therapy a daunting challenge.

The microbial species most commonly isolated in root canals when endodontic treatment fails is *Enterococcus faecalis*, a facultative anaerobic microorganism that can thrive with or without oxygen.

Sundqvist et al[18] correlated treatment failure with unfavorable factors or flaws in performing endodontic therapy to treat devitalized teeth with apical periodontitis. They also reported the peculiarities of microbiota found in root canals that had previously undergone unsuccessful endodontic treatment. The failure was due to a selection process that depends on the specific resistance of microorganisms to antimicrobial procedures and drugs used during endodontic therapy, as well as the survivability of certain microbiota in restricted nutritional mediums, where interrelations between bacteria are minimal.

In endodontics, laser has demonstrated its ability to eliminate layers of debris, its capacity to seal the dentinal tubules, vaporize pulp tissue, change dentin permeability and disinfect the root canal system with its bactericidal action.

A number of authors [1,3,4,12,14,15,17,21] reported that several studies have been conducted in Dentistry with the purpose of complementing conventional therapies. Specifically in Endodontics, photodynamic therapy (PDT) plays a promising role as an adjunct in retreating periapical lesions. Studies have been conducted to show intracanal microbial reduction.

This study aimed at assessing the efficacy of PDT in cleaning and shaping the root canal system.

Material and Methods

Twenty-four anterior teeth with radiographically visible periapical lesions were selected. After all patients had signed an informed consent form (Appendix 01), the teeth were divided into two groups (G1 and G2) of 12 teeth each.

The teeth in group 1 (G1) were opened using a round high rotation bur #1015 (KG Sorensen Ltda, São Paulo, SP, Brazil), rubber dam isolation (Madeitex Indústria e Comércio Ltda Latex supplies, São Paulo, São Paulo State, Brazil), disinfected with iodine 0.1% alcohol (Farmax, Minas Gerais State, Brazil), use of Endo Z bur (Dentsply/Maillefer Corp., Bellagues, Switzerland) for convenience form, use of # 2, # 3 and # 4 Gates Glidden drills (Dentsply/Maillefer Corp., Bellagues, Switzerland) for the pre-enlargement phase in finishing the coronal opening. The canal was shaped with ProTaper system (Dentsply/Maillefer Corp., Bellagues, Switzerland) and sanitized with 5.25% sodium hypochlorite (Marcela Dourada Compounding Pharmacy Ltd., Vitória da Conquista, Bahia State, Brazil).

Tooth length measurement was performed with a foramen locator (Root ZX, J Morita Corporation, Ltda. Japan) seeking apical foramen patency.

Three samples of the material were collected in the following order:

In G1, the first collection was made after surgical opening; the second, after canal preparation and the third seven days after cleaning and shaping the root canal. Canal preparation was performed by means of the crown-down technique using spherical high rotation burs #1015 (KG Sorensen Ltda, São Paulo, São Paulo

State, Brazil). In the pre-enlargement phase, Gates Glidden #2, 3 and 4 burs (Maillefer Corp., Bellagues, Switzerland) and F3, F4 and F5 rotary files Dentsply ProTaper system (Maillefer Corp., Bellagues, Switzerland) were used. Five ml of sodium hypochlorite at 5.25% (Macela Dourada compounding pharmacy Ltda., Vitória da Conquista, Bahia State, Brazil) were used for irrigation every time instruments were replaced. Prior to collection, the sodium hypochlorite was neutralized with sodium thiosulfate at 5% so that the hypochlorite would not interfere with the results of the samples. Throughout the study intracanal medication was not employed between sessions. Balls of sterile cotton were inserted into the specimens, and then closed with a temporary sealer (Coltosol by Vigodent ind. e Com. Ltda, Rio de Janeiro, Brazil). The material was collected from the root canal system with the aid of absorbent paper points #20 or 25 (Dentsply/Dentsply Ind. And Com. Ltda., Petrópolis, Brazil), placed into a transport medium and sent to a clinical analysis laboratory (Hermes Pardini Ltda., Belo Horizonte, MG, Brazil) to ensure the growth of a culture of anaerobes, thereby disclosing whether or not contamination had occurred in the root canal system.

Teeth in G2 underwent the same procedures as G1, but with the addition of Photodynamic Therapy (PDT) after cleaning and shaping the root canal system.

The device used in the research was a Photon Laser III (DMC Equipmentos Ltda, São Carlos, SP, Brazil) with red laser irradiation and optical fibers of 660nm, 100mw, 140j/cm² for 40 seconds. The dye used in the study was methylene blue at 0.005%, which was left for 5 minutes for pre-irrigation inside the canal. The material was then collected with an absorbent paper point #20 or 25 (Dentsply / Dentsply Ind. e Com. Ltda., Petrópolis, Brazil), and taken to the clinical analysis laboratory (Laboratório Hermes Pardini Ltda, Belo Horizonte, MG, Brazil) to check whether or not bacterial growth culture had occurred, thereby demonstrating — or not — the effectiveness of Photodynamic Therapy (PDT) as an adjunctive therapy in reducing contamination of microorganisms in the root canal system.

The results were analyzed with a view to proving or disproving the effectiveness of PDT in reducing contamination of microorganisms in the root canal system.

The data were initially analyzed by contingency tables and correlated Chi-square tests by comparing the response variable distribution according to the levels of the factor "treatment" at each stage. Thereafter, analysis of variance was applied utilizing a model suitable to experiments with two factors and interaction. In this analysis, the existence of repeated measurements was noted, since the same volunteers were evaluated at both stages. The analyses were calculated using SAS (SAS Institute Inc., The SAS System, Release 9.2. SAS Institute Inc., Cary NC. 2008). The contingency tables and Chi-square tests were calculated by FREQ procedure, and the analysis of variance using GLIMMIX procedure.

Results

For the purposes of statistical analysis, it became clear that all samples in the first collection (in both G1 and G2) were contaminated and, therefore, only the second and third samples were analyzed.

For clarification purposes, the term "BEFORE" was used to refer to the first sample, "DURING" to the second, and "AFTER" to the third sample. In the present study the significance level was set at 5%.

The analysis began with the contingency table, which compares the frequencies and ratios in the levels of the treatment factor in the "DURING" phase (Table 1).

Likelihood ratio chi-square test (G2) provides no evidence ($p>0.10$) of an association between treatment and outcome.

Whereas in the treatment with a rotary instrument there were 02 positive cases and 10 negative cases, treatment with PDT yielded very similar quantities, with 03 positive cases and 09 negative cases. Clearly, this difference can be incidental, since it refers to a single event in the PDT group which happened to migrate from negative to positive.

Hence the absence of significant differences, which occurs when there is evidence to assert that such difference is not incidental, which would not be reasonable in this case.

Table 1. Frequency (percentage) of result in treatments during experiment.

Treatment	Results		χ^2 Likelihood ratio Chi-square
	Negative	*Positive*	
Rotary (G1)	10 (83.33)	2 (16.67)	G2: 0.2540 – GL: 1 Valor-p: 0.6143
PDT (G2)	9 (75.00)	3 (25.00)	
Total	19 (79.17)	5 (20.83)	

The next step was to evaluate the result ratios in the "AFTER" phase (third sample) displayed in Table 2.

Likelihood ratio chi-square test (G2) provides scarce evidence ($p>0.10$) of an association between treatment and outcome.

In the "AFTER" phase, a different behavior is observed in the positive treatments, in which most of the results were positive (58.33%) and treatment with PDT remained at precisely the same ratio as in the "DURING" phase, in which most of the results were negative.

Treatment with a rotary instrument showed a change, whereas PDT treatment results remained stable in this case. It can be concluded that there were no statistical differences between the two types of treatment used in this study.

Discussion

According to Nair et al,[7] Peciuliene et al,[8] and Sundqvist et al,[18] the presence of microorganisms in the root canal system favors the appearance of periapical lesions as a result of microbial aggression. Thus, the key objective of endodontic treatment is to eliminate infection, given that allowing a viable microbiota to survive within the dentinal tubules may — depending on their virulence factor — enable the pathogenic process to reestablish itself and consequently undermine therapy. Therefore, disinfection of the root canal system is a decisive factor for therapeutic success.

According to Wilson,[22] the use of low power lasers causes denaturation of microorganisms due to structural damage (bacterial wall) or metabolites produced by oxygen when the photosensitizer is activated by laser light, which causes cell death.

Table 2. Frequencies in treatment results in the "AFTER" phase (third study sample).

Treatment	Results		χ^2 Likelihood ratio Chi-square
	Negative	*Positive*	
Rotary (G1)	5 (41.67)	7 (58.33)	G2: 2,8046 – GL: 1 Valor-p: 0.0940
PDT (G2)	9 (75.00)	3 (25.00)	
Total	14 (58.33)	10 (41.67)	

According to Love[6] and Sirien et al,[16] the microbiota found in the canal system are dynamic and diverse.

According to Walsh,[20] temperature variation is less critical due to low power being used during the procedure, but professionals should be aware of this fact. A previous study showed an increase of 3.9° C in the mean temperature, which indicates biocompatibility.

Ferrari,[2] Peciuliene[8] and Pinheiro et al[9] argue that *Enterococcus faecalis* is the bacterium predominantly found in cases of refractory apical periodontitis.

According to Radaelli,[11] in cases of necrotic pulp, the bacterial flora is rich in substrate, and metabolic changes tend to occur in the specimens.

Garcez et al[4] and Walsh et al[19] demonstrated that methylene blue is the best photosensitizer given that it is easy to handle and widely available on the market. Additionally, it features an absorption coefficient compatible with the wavelength of low laser power (PDT).

Zanin[23] found that the older the biofilm, the less sensitive it appeared to be to photodynamic therapy. He also noted that photosensitizing occurred in the outer layers of the biofilm. Bacteria were, therefore, allowed to exist in the innermost regions of the biofilm due to poor penetration of the dye or difficulties penetrating the biofilm.

Garcez et al[4] showed that in conventional endodontic treatment of infected root canals a reduction in bacteria count is accomplished by a combination of mechanical instrumentation, various irrigation and medication solutions or antimicrobial dressings placed within the canal. Photodynamic therapy is a treatment which can be considered an adjunct to conventional endodontic therapy for producing a significant bacterial reduction.

Pinheiro et al[9] demonstrated the use of low-power lasers in various dental specialties such as Pediatric Dentistry, Periodontics, Endodontics and Implantology in Pediatrics.

The present study used 24 teeth with periapical lesions radiographically evidenced and divided into two groups of 12 teeth each. In G1, canal preparation was performed using rotary instrumentation and the ProTaper system. In G2, in addition to the rotary instrumentation, photodynamic therapy (PDT) was employed shortly after endodontic instrumentation.

In order to standardize the methodology used in this study, three samples of each specimen were collected from both groups and sent to a clinical analysis laboratory to determine whether or not PDT was effective.

Furthermore, a single device (Photon Laser III, DMC Equipamentos Ltda, São Carlos, Brazil) was used, as well as methylene blue dye as photosensitizer.

All patients were treated by a single operator working in the same environment to ensure research standardization. Recent studies have proved PDT to be highly effective in reducing the presence of microbes in the root canal system. Several studies have reported the use of photodynamic therapy as an adjunct in endodontic treatment.

Conclusions

Based on the results of this study it is reasonable to conclude that:

» No statistically significant differences were found among the results of the samples.

» Additional studies are warranted to further investigate this topic.

» Photodynamic therapy can be used as an adjunct treatment in addressing root canal system issues.

References

1. Cavalheiro F M. Avaliação da redução microbiana em condutos radiculares contaminados comparando três técnicas de irradiação com laser de baixa potência associado a fotossensibilizador [dissertação]. São Paulo (SP): Universidade de São Paulo; 2007.
2. Ferrari PHP. Effect of endodontic procedures on enterococci enteric bacteria and yeasts in primary endodontic infections. I. Int Endod J. 2005;38(6):372-80.
3. Fonseca MB, Tessare Júnior PO, Pallota RC, Ferreira Filho H, Denardin OV, Rapoport A, et al. Photodynamic therapy for root canals infected with E. faecalis. Photomed Laser Surg. 2008;26(3):209-13.
4. Garcez AS. Laser em baixa intensidade associado à fotossensibilizador para redução bacteriana intracanal comparado ao controle químico [dissertação]. São Paulo (SP): Universidade de São Paulo; 2002.
5. Garcez AS, Nuñez SC, Hamblin MR, Ribeiro MS. Antimicrobial efect in photodynamic therapy on patients with necrotic pulps and periapical lesion. J Endod. ;34(2):138-42.
6. Love RM. Enterococcus faecalis: a mechanism for its role in endodontic failure. Int Endod J. 2001;34(5):399-405.
7. Nair PN, Sjögren U, Krey G, Kahnberg KE, Sundqvist G. Intrarradicular bacteria and fungi in root-filled, asymptomatic human teeth therapy-resistant periapical lesions: a long-term light and electron microscopic follow-up study. J Endod. 1990;16(12):580-8.
8. Peciuliene V, Balciuniene I, Eriksen HM, Haapasalo M. Isolation of Enterococcus faecalis in previously root-filled canals in a lithuanian population. J Endod. 2000;26(10):593-5.
9. Pinheiro ET, Gomes BP, Ferraz CC, Teixeira FB, Zaia AA, Souza Filho FJ. Evaluation of root canal microorganisms isolated from teeth with endodontic failure and their antimicrobial susceptibility. Oral Microbiol Immunol. 2003;18(2):100-3.
10. Pinheiro SL, Schenka AA, Alves Neto A, de Souza CP, Rodriguez HM, Ribeiro MC. Photodynamic therapy in endodontic treatment of deciduous teeth. Lasers Med Sci. 2009;24(4):521-6.
11. Radaelli CARM. Avaliação da redução bacteriana em conduto radicular infectado e irradiado com laser de diodo estudo in vitro [dissertação] São Paulo (SP): Instituto de Pesquisas Energéticas e Nucleares; 2002.
12. Ng R, Singh F, Papamanou DA, Song X, Patel C, Holewa C, et al. Endodontic photodynamic therapy Ex vivo. J Endod. 2011;37(2):217-22.
13. Schilder H. Cleaning and shaping the root canal. Dent Clin North Am. 1974;18(2):269-96.
14. Seal GJ, Ng YL, Spratt D, Bhatti M, Gulabivala K. An in vitro comparison of the bactericidal efficacy of lethal photosensitization or sodium hyphochlorite irrigation on Streptococcus intermedius biofilms in root canals. Int Endod J. 2002;35(3):268-74.
15. Silbert T, Bird OS, Milburn GJ, Walsh LJ. Disinfection of root canals by laser dye photosensitization. J Dent Res. 2000;79:569.
16. Siren EK, Haapasalo MP, Ranta K, Salmi P, Kerosuo EN. Microbiological findings and clinical treatment procedures in endodontic cases selected for microbiological investigation. Int Endod J. 1997;30(2):91-5.
17. Sousa L, Brito P, Siqueira Jr JF. Terapia fotodinâmica com dois fotossensibilizadores diferentes como suplemento na promoção intracanal de redução de Enterococcus faecalis. J Endod. 2010;36:292-6.
18. Sundqvist G, Figdor D, Persson S, Sjögren U. Microbiology analyses of teeth with endodontic treatment and the outcomes of conservative retreatment. Oral Surg Oral Med Oral Pathol Oral Radiol Endod. 1998;85(1):86-93.
19. Walsh LJ. The current status of low level laser therapy in dentistry. Part 2. Hard tissue applications. Aust Dent J. 1997;42(5):302-6.
20. Walsh LJ. The current status of low level laser therapy in dentistry. Aust Dent J. 2003;48(3):146-55.
21. Williams JA, Pearson GJ, Colles MJ, Wilson M. The effect of variable energy input from a novel light source on the photoactivated bactericidal action of toluidine blue O on Streptococcus Mutans. Caries Res. 2003;37(3):190-3.
22. Wilson M. Bacteria in supragengival plaque samples can be killed by-power laser light in the presence of a photosensitizer. J Appl Bacteriol. 1995;78(5):569-74.
23. Zanin ICJ. Susceptibility of Streptococcus mutans biofilms to photodynamic therapy: an in vitro study. J Antimicrob Chemother. 2005;56(2):324-30.