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Evaluating the antibacterial effect of low-powered laser activated irrigation on Enterococcus faecalis infected root canals

[1]Muthanna S. Al-Aqeedi, BDS, HDD, M.Sc.
[2]Salwa H. Mohammed, BSC, M.Sc., Ph.D, Oral Microbiology
[3]Raghad A. Rasheed, BDS, M.Sc.

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ABSTRACT

Enterococcus faecalis is the most resistant microorganisms that can be found within failed root canal treated teeth, it has a high resistant to most intracanal medicaments and irrigants, This in-Vitro study aimed to evaluate the intracanal bacterial reduction using Er,Cr:YSGG laser activation of three irrigants. 42 extracted human mandibular first premolar teeth were inoculated with Enterococcus faecalis strains , incubated for 48 hours then randomly distributed into two groups of 21 root each: **group A (Control group):** conventional irrigation (CI) with syringe using Navi Tip gauge 31 with no activation. **Group B (laser activated group):** irrigated then activated using Er,Cr:YSGG laser (Biolase,USA) for 80 second using radial firing tip (RFT/3) at 0.5 watt. Each group was subdivided into three sub-groups according to the irrigant used (group I = 0.9%NaCl), (group II = 1.5 %NaOCl) and (group III = 3%NaOCl). After treatment, a sample of dentin was taken from each root for colony-forming unit counting (CFU_s). The results showed that Er,Cr:YSGG laser activated irrigation (LAI) group showed a significant reduction in (CFU/ml) comparing to the (CI) protocol. The laser activation of 0.9%NaCl showed (72.15%) reduction, 1.5 %NaOCl showed (97.37 %), while laser activation of 3%NaOCl showed (99.75%) compared to (14.11%) reduction when using 0.9%NaCl with acoustic activation, (68.11%) for 1.5% NaOCl with acoustic activation and (93.1%) eradication for 3%NaOCl. As a conclusion, laser activated irrigation using Er,Cr:YSGG laser is an effective protocol for eradication of Enterococcus faecalis .

Key words: Er,Cr:YSGG, laser activated irrigation, radial firing tip, Enterococcus faecalis.NaOCl.

(1) Senior, Department of Conservative Dentistry, Specialized Dental Center, Ministry of Health, Mosul,Iraq
(2) Lecturer, Department of Basic Dental Science, College of Dentistry, Mosul University.
(3) Ass. Prof., Department of Conservative Dentistry, College of Dentistry, Mosul University.

INTRODUCTION:

The main principles of a successful endodontic treatment based on sound principles of debridement, disinfection, and obturation in order to maintain the dentition [(1)], which requires the combination of physical and chemical agents to remove soft-tissue debris, smear layer [(2,3)], and to eradicate the intracanal bacterial populations or at least reducing them to levels that are compatible with periradicular tissue healing [(4)].

Enterococcus faecalis is frequently isolated from endodontic cases requiring retreatment [(5)]. Numerous studies carried out using Enterococcus faecalis because of its resistance to some medicaments and irrigants including the antimicrobial action of sodium hypochlorite that makes this bacteria able to survive conventional root canal therapy [(6,7).] It is the most frequently recovered microorganisms from refractory periapical periodontitis [(5)].

The antimicrobial irrigant of choice is sodium hypochlorite (NaOCl). It has been shown to kill bacteria and remove debris from the root canal [(8)], However, for its bactericidal effect, sodium hypochlorite relies heavily on the duration of time retained in the canal and the use of copious volumes of the solution since it is the free chlorine which acts as the disinfecting agent and this is used up rapidly. It has been shown that 20-30 minutes is required to clean and debride a canal [(9)]. A small volume used for a short contact time will have a limited effect. Furthermore, there is evidence that hypochlorite is not effective against all pathogenic bacteria specifically Enterococcus faecalis which is associated with recalcitrant canals [(10)], because of its high surface tension that prevents direct contact of this irrigant with the dentinal

walls of the anatomical complexities [11] Paque et al reported that after NaOCl syringe/needle irrigation and instrumentation, 40-60% of the canals still contained cultivable bacteria [12].

The standard irrigation delivery protocol is by using conventional syringe; unfortunately syringe irrigation showed to be inefficient in the apical third of the root canal**,** or in oval extensions, isthmuses, and anastomoses [13]. The main reason for that is vapour lock that results in trapped air in the apical third of the root canal, preventing the exchange of irrigants and decrease the efficacy of debridement [14].

In the recent years, laser activated irrigation (LAI) or what can call laser energized irrigation (LEI) has been reported as a more powerful and effective protocol for removing intracanal debris than conventional irrigation protocol [15-17], but its antibacterial efficacy has not been fully evaluated. In order to generate LAI technique, radial firing laser tip (Biolase Technology, Inc.) can be used to achieve Better root canal surface coverage, as the tip shape emits the laser energy in a broad cone, allowing better coverage of the root canal walls than end-firing tips. This facilitates entry of the emitted laser energy into the dentinal tubules reaching bacteria that have penetrated deep into the dentin. The present study evaluates the in-Vitro intracanal reduction of Enterococcus faecalis populations promoted by three irrigants (0.9% NaCl, 1.5% NaOCl and 3% NaOCl) using LAI of Er,Cr:YSGG laser using radial firing tip and compare it to conventional syringe irrigation (CI) protocol.

MATERIALS AND METHOD

SELECTION AND PREPARATION OF THE SAMPLES:

42 recently extracted single rooted human mandibular 1st premolar teeth with closed apices were collected. The teeth were decoronated at the level of the cemento-enamel junction (CEJ) using a two-sided diamond coated sectioning disc under copious coolant. The roots were examined by using two views (bucco-palatal and mesio-distal) of the dental radiograph. The length of each root was adjusted to 15 mm from the cervical border of the root to the apical foramen. Setting of working length was 1 mm less than the root canal length (working length 14 mm). Mechanical instrumentation subsequently performed with ProTaper rotary nickel titanium files (ProTaper® Universal, DENTSPLY Maillefer, Ballaigues, Switzerlanden) according to the manufacturer's instructions, the canals were prepared to size F4. A resin based composite restorative material (spectrum universal composite, DENTSPLY ,Germany) shade A2 used to seal the apical foramen (apical 3 mm) of each root .while the external root surface with the apical 3 mm was painted with two layers of nail varnish.

SAMPLES MOUNTING:

A silicone based poly vinyl siloxane impression material mixed and adapted in a stainless steel metal containers on which the sampled roots were mounted vertically about 3mm from the cervical border. Then samples were sterilized by autoclaving at (121 C°/15 pounds/inch² for 15 minutes).

MICROBIAL ISOLATION:

Enterococcus faecalis strains were isolated from canals of teeth with failed endodontic treatment and persisting periapical lesions as described by Pinheiro et al., [18], then Identification and confirmation of the obtained microbes done by a series of laboratory diagnostic tests.

INOCULATION OF THE SAMPLED ROOTS:

Sterile 1-ml insulin syringes were used to fill the sterile roots with 0.1 ml of the prepared broth that contains $8*10^{-7}$ of the isolated Enterococcus faecalis without over flowing. Then these sampled roots were incubated at 37°C for 48 hours according to Lui et al [19].

After 48 hours, each sampled root was irrigated with 1ml of sterile distilled water then dried carefully with sterile paper points to remove the remnants of the culture medium and non-adherent cells the sampled roots were ready for treatment.

TREATMENT GROUPS:

The total samples (n=42) were divided into 2 groups according to the different irrigation protocols into:

1-) group A: (Control group/n=21)

Irrigated by luer-lock hypodermic syringe with Navi tip (Navi Tip; Ultradent products, Salt Lake City, UT, USA) gauge 31 of two sides venting and closed apex at a rate of 5 ml/75 seconds (1ml/15 second) the needle tip was placed 1 mm from the working length. No activation method was used for this group.

2-) Group B: (LAI group/n=21)

Irrigated by luer-lock hypodermic syringe with Navi tip (Navi Tip; Ultradent products, Salt Lake City, UT, USA) gauge 31 of two sides venting and closed apex at a rate of 1.25 ml/10 seconds**,**the tip of the needle placed 1 mm from the working length, then the irrigation solution was activated four times with Er,Cr:YSGG laser (Waterlase i Plus, Biolase Technology, San Clemente, USA) for 5 seconds each, The cycle was repeated for 4 times, for a total of 80 second activation and 5ml of NaOCl with an irrigation rate of 1mm pulling out /second.

Each of group (A, B) sub-divided into 3 sub-groups as following:

Sub-group I: (0.9% NaCl) (n=7) irrigated with 0.9% sterile saline.

Sub-group II: (1.5%NaOCl) (n=7) irrigated with 1.5%NaOCl (Parcan™; Septodont, Saint –Maur-des-Fossés, France).

Sub-group III: **(3%NaOCl) (n=7)** irrigated with 3%NaOCl (Parcan™; Septodont, Saint –Maur-des-Fossés, France). At the end of the laser activation, sub-group (2) and (3) were neutralized with 1.2ml 2.4ml respectively of 5% sodium thiosulphate for 60 second respectively. All the irrigants in both groups used were at room temperature during irrigation.

LASER PARAMETERS:

Er,Cr:YSGG dental laser (Waterlase i Plus, Biolase Technology, San Clemente, USA) of 2,780 nm was used at panel setting of 0.5 Watt average power and a pulse repetition rate of 20 Hz . The laser beam was conducted by a tapered quartz tip RFT 3 (Endolase, Biolase Technology, Irvine, USA) with (400µm diameter/ length of 25mm /52° angled tip) in the presence of air spray sets at 10%. No water spray was activated in this study.

DETERMINATION OF BACTERIAL VIABILITY AND BACTERIAL COUNTING:

After 1 minute of the treatments, a 10 µL of a sterile ringer solution was placed in all root canals according to Pedullà et al [20] to wash out the canal contents. A sample of dentin shaves from (coronal, middle, and cervical third) was taken from each treated root by inserting a sterile file size F5 into the root canal. The file was rotated 360° clockwise direction within the canal, reaching the full working length to engage and remove dentin from the coronal, middle and apical third of the root . The file with the dentin stuck was transferred to a sterile screw-capped vial of brain heart infusion broth, dispersed for 30 second with a vortex mixture.10 folds serial dilution in brain heart infusion broth was carried until 10^{-7} .0.1 ml of the final dilution of each root specimen were cultured on enterococci selective agar. Plates were incubated at 37°C for 48 hours to determine the number of remaining viable bacteria after treatment.

RESULTS

The findings of this study showed that the root canal treated by LAI exhibits the least viable count bacteria 1 minute after management (Table 1). LAI group showed a significant reduction in (CFU/ml) comparing to the (CI) protocol. The laser activation of 0.9%NaCl showed (72.15%) reduction, 1.5 %NaOCl showed (97.37 %), while laser activation of 3%NaOCl showed (99.75%) %) compared to (14.11%) reduction when using 0.9%NaCl with (CI) protocol, (68.11%) for 1.5% NaOCl with (CI) protocol and (93.1%) eradication for 3%NaOCl.

Table (1): Eradication rate of each irrigant according to the irrigation protocol

Groups		Eradication rate of E. faecalis
Laser activated irrigation (LAI) with Er,Cr:YSGG	3% NaOCl	99.75%
	1.5% NaOCl	97.37%
	0.9% NaCl	72.15%
Conventional irrigation (CI) with conventional syringe	3% NaOCl	93.1%
	1.5% NaOCl	68.11%
	0.9% NaCl	14.11%

STATISTICAL ANALYSIS:

Duncan's multiple range test for CI (Table 2) shows that the value of using 0.9%NaCl with CI is significantly lower than that with both 1.5% and 3% NaOCl ($P \leq 0.05$), and for 1.5%NaOCl is lower than that of 3%NaOCl ($P \leq 0.05$).

Table (2): Duncan's multiple range tests for CI with (0.9%NaCl, 1.5%NaOCl, and 3%NaOCl) irrigants.

Irrigant used in the study	N	Subset for alpha = 0.05		
		1	2	3
3%NaOCl	7	55.2857		
1.5%NaOCl	7		255.1429	
0.9%NaCl	7			687.1429
Sig.		1.00	1.00	1.00

Duncan's multiple range test for LAI (Table 3) shows that the value of using 0.9%NaCl with LAI is significantly lower than that with both 1.5% and 3% NaOCl ($P \leq 0.05$), and for 1.5%NaOCl is lower than that of 3%NaOCl ($P \leq 0.05$).

Table (3): Duncan's multiple range tests for LAI with (0.9%NaCl, 1.5%NaOCl, and 3%NaOCl) irrigants.

Irrigant used in the study	N	Subset for alpha = 0.05	
		1	2
3%NaOCl	7	2.000	
1.5%NaOCl	7	21.000	
0.9%NaCl	7		222.8571
Sig.		0.133	1.00

DISCUSSION

During the last years, attention was drawn toward reducing the concentration of chemicals and irrigants introduced into the root canal to reduce their disadvantages and toxic effects, this reduction must be balanced by another method that activates and increases the efficiency of these irrigants against resistance microorganisms, especially with the increased antimicrobial resistance of bacteria. As The efficacy of LAI on the bactericidal property of irrigants has not been fully evaluated, the present In-Vitro study was conducted to compare the antibacterial efficacy of two activation protocols (CI) protocol and LAI protocol using Er,Cr:YSGG laser in reducing intracanal E. faecalis populations.

Tay et al [(13)] stated that (CI) protocol generates a '' dead water zone'' at the apical 0.5-1.0 mm. causing ''apical vapour lock ''; inhibiting accurate bacterial eradication. Many authors stated that entrapped air bubbles cannot be removed with (CI); these results are in agreement with our results which showed the eradication rate of 0.9% NaCl with conventional irrigation protocol showed the least eradication rate of Enterococcus faecalis among all other treatment groups, the eradication rate was only (14.11%) which can be related to the limited mechanical flushing action of the manual irrigation protocol. Using hand irrigation with conventional syringe has no activation effect on the irrigants and the eradication effect can be only related to the irrigant itself. These results are in agreement with Seet et al.[(21)]

LAI of 0.9%NaCl showed an improved reduction in CFU (72.15%) when compared to CI that proves the effectiveness of photo acoustic streaming in decontamination of root canal by creating rapid implosion of vapor bubbles as reported by Matsumoto et al. [(22)], which helps in decomposing the biofilm and detaches some of the colonies as stated by Seet et al. (2012). Lasers have been used to produce cavitation of liquids, thereby increasing the cleaning ability of the liquid. George and Walsh [(15)] reported the first in-vitro study to examine the capacity of lasers to activate irrigants inside root canal systems to increase their action on the smear layer. Di Vito et al.[(23)] stated that laser energized water increases smear layer removal, as water alone has no effect on organic or inorganic components of the smear layer.

When laser pulses are focused into a limited volume of liquid, plasma is generated. Plasma formation can lead to rapid heating of the material followed by an explosive expansion and the emission of a shock wave (Di Vito et al. [(23)]). This technique is referred to as laser activated irrigation or laser energized irrigation. The results of our study are in agreement with Seet et al. [(21)], Pedullà et al. [(20)], and Christo [(24)].

ROOT CANAL IRRIGATION WITH 1.5% NAOCL:

The apical lock generates either by entrapped air bubbles by the advancing irrigant interface during irrigant delivery in a dry root canal as reported by Peeters and Gutknecht [(25)], or by gas bubbles produced in situ during NaOCl reaction with the organic materials represented by the remnant of pulp tissue or presented within the composition of the dentin (Vera et al.[(26)]) causing debris accumulation and fluid stagnation that prevents adequate irrigant replacement and inhibits accurate bacterial eradication that needs a forceful current which can be created by sonic or ultrasonic devices. Many authors stated that entrapped air bubbles cannot be removed with CI (De Gregorio et al. [(27)]; Gu et al. [(28)]; Tay et al. [(14)]).

These facts were confirmed by the results of our study when we used CI of 1.5% NaOCl, the results were (68.11%) eradication rate of Enterococcus faecalis. This rate stills low and not accepted for root canal obturation in clinical practice. Because CI methods offers no additional activation for the irrigants as mentioned before.in addition, Senia et al.[29] demonstrated that NaOCl did not extend any closer than 3 mm apically from working length when delivered with CI protocol. This might be attributed to the fact that NaOCl reacts with organic material in the root canal and quickly forms micro gas bubbles at the apical termination that coalesce into an apical vapor lock with subsequent instrumentation. Because the apical vapor lock cannot be displaced within a clinically relevant time frame through simple mechanical actions, it prevents further irrigants from flowing into the apical region. Disinfection agents such as NaOCl require direct contact with the bacteria what is often impossible in peripheral areas of the root canal such as anastomoses, fins and the most apical part of the main root canal (Haapasalo et al. [30]). Caron [31] stated that sonic activation systems can provide deeper penetration of an irrigant to all areas of endodontic space in comparison to CI, and to effectively dislodge the biofilm from root canal wall.

Meire et al [32] proved that NaOCl has a high absorption rate of laser wave length of 2500nm and above which indicates that Er family lasers (2780nm, 2940nm) are the best for LAI. Lasers have been used to produce cavitations of liquids. Thereby increasing the cleaning ability of the liquid [17].when laser pulses are focused into a limited volume of liquid, plasma is generated. Plasma formation can lead to rapid heating of the material followed by an explosive expansion and the emission of a shock wave. LAI of 1.5%NaOCl showed (97.37%) reduction in viable bacteria, with a better antibacterial result when compared to CI 1.5%NaOCl, because laser activation results a stronger modulation in the reaction rate of NaOCl, significantly increasing the production and consumption of available chlorine and oxygen ions as stated by Macedo et al. [33]. Incomplete dissolution and residual biofilm appears to be common under clinical conditions following full-strength NaOCl irrigation. A previous study by Muller et al. [34] shown that the LAI technique was effective in disrupting plaque-derived biofilm in the absence of antimicrobials. Biofilm disruption can change the bacteria to their planktonic form, making them more susceptible to antimicrobial agents.

ROOT CANAL IRRIGATION WITH 3% NAOCL:

Using of 3% NaOCl in this study was of clinical importance, Hafez et al. [35] showed that 3% NaOCl was biocompatible solution, because pulps treated with this concentration demonstrated no evidence of pulpal necrosis after 7 and 27 days. They also showed that 3% NaOCl was biocompatible as a hemorrhage control agent, because pulps treated with this concentration demonstrated no evidence of pulpal necrosis after 7 and 27 days.

Our study showed that CI with 3% NaOCl gave a (93.1%) eradication of Enterococcus faecalis. This can be related to the antibacterial effect of the 3% concentration of NaOCl. To prevent recurrence of bacterial growth in infected root canals, the depth of irrigation penetration is more important than the concentration. Which means that the depth of penetration (apically) towards the full apical depth and the depth of penetration (laterally) within the dentinal tubules. Using CI is limited (apically) by the vapor lock phenomena that prevent full canal sterilization, and it is limited (laterally) by the depth of about 100-120 micron. Debbie et al. [36] stated that CI of 3%NaOCl can penetrates deep to only 100-300 micron within dentinal tubules. Clegg et al. [37] evaluated the effectiveness of three concentrations of NaOCl (6%, 3%, and 1%), on apical dentine biofilms in-vitro. Their findings indicated that 6% NaOCl was the only irrigant capable of both rendering bacteria nonviable and physically removing the biofilm.

For LAI group, the results of our study showed that LAI of NaOCl effectively increases the eradication rate of Enterococcus faecalis; it was the highest eradication rate among all other study groups as 3% NaOCl with CI gave 93.1% eradication, while LAI of 3% NaOCl gave 99.75% eradication. This improves that LAI is a valuable protocol for increasing the efficiency of even low concentrations of NaOCl to reduce the complications related to high concentrations. This fact has been proved by Seet et al. (2012) who used 0.25W Er,Cr:YSGG laser with 4% NaOCl and with Christo (2012) who used 0.5 W with 4% NaOCl.

Using of low power laser is important to overcome the problems associated with heat generated with high power laser, as the amount of heat delivered may have undesirable effects such as charring and cratering of dentin (Depraet et al., [38]) or possible thermal injury to the periodontal ligament, resulting in root resorption, ankylosis or peri radicular necrosis (Bahcall et al., [39]).

Blanken et al. [40] reported that Er,Cr:YSGG laser can develops cavitation effect even with a low laser output as with 0.25W/20Hz. This property is highly correlated to the Er family lasers as these lasers are highly absorbed by water. Wang et al. [41] reported that using Er,Cr:YSGG laser alone for direct laser irradiation gave only 77%-96% eradication rate of E. faecalis when using 1W and 1.5W. Yavari et al. [42]stated that using Er,Cr:YSGG laser for direct laser irradiation at 2W and 3W resulted in 97.6% and 98.47% reduction in E. faecalis, both studies gave lower eradication rates than the results of LAI protocol used in this study. Using LAI to activate 3% NaOCl with only 0.5W energy gave better antibacterial effect than the high direct irradiation of (1, 1.5, 2, and 3W) was using lower laser energy to get a higher antibacterial effect which means that LAI is a valuable, effective, and safe disinfecting tool for root canal therapy.

LAI performs better than CI with a deeper penetration depth. In a study done by Gordon et al. [43]; they stated that it was possible to achieve expansion and collapse of intra tubular water as deep as 1,000 µm or more. This micro pulse-induced absorption was capable of producing acoustic waves strong enough to disrupt and kill intra tubular bacteria within the dentinal tubules. These findings are very important in clinical field as bacteria have been identified to reach the depths of 1,000 µm, with E. faecalis at depths of 800 µm (Haapasalo and Orstavik [44]). De Souza et al. [45] found that with progressive decrease in diameter of the deep dentinal tubules, the penetration of irrigants is restricted. However, laser irradiation with its inherent properties of light scattering, local intensity enhancement and attenuation allows light penetration deeper in the dentin tubules contributing to a superior antimicrobial efficacy.

Using LAI offers the advantages of using sub ablative laser energy to generate a 3-dimentionally activated movement of the irrigant, resulting in a better debridement and a high bacterial eradication with a lower NaOCl concentration, reducing the concentration will reduce the drawbacks of high concentration that includes the toxic effect of high concentrations, the reduction of flexural strength, elastic modulus, tensile strength and micro hardness (Patil and Uppin,[46]) and the adverse effect on dentin bonding with resin based materials as lower concentrations of NaOCl has lower adverse effects on dentin (Erdemir et al., [47]).

When using LAI, there is a little concern about adverse thermal effect. Schoop et al. [48] measured temperature rise of the external root canal surface during dry laser irradiation of the root canal using 2W and 3W with 20 Hz and RFT. They found that temperature changes were only 1.3°C and 1.6°C respectively. The geometry of the laser tip plays a major role in determining the quality of laser-induced vapor bubbles formation, secondary cavitation effect, beam shape and laser-induced heat distribution. The selection of RFT was necessary to develop the photo-acoustic streaming, as the shape of the laser beam in this tip is conical which encourages cavitation development. Using RFT allowed for a more homogenous laser irradiation of root canal walls.

Gu et al. [28] who stated that the combination of LAI effect and NaOCl increases the antibacterial effect of NaOCl by many ways as cavitations developed by LAI produces a transient weakening of the bacterial cell wall and cytoplasm, making the bacteria more susceptible to the antibacterial effect of NaOCl even with a lower concentration. This cavitation (primary and secondary cavitation effect) removed the need to insert the laser tip close or at the working length as the cavitation effect was efficient for cleaning the apical part as well the middle and coronal parts. To explain the effect of LAI protocol, the interactions between the Er,Cr:YSGG laser and the NaOCl can be summarized as following:

1-) Photo-physical interaction, where the Er,Cr:YSGG laser reduces surface tension of NaOCl to facilitates contact with the target tissues which is necessary for effective dissolution of the tissue as heat application reduces surface tension .

2-) Photo-chemical interaction, Macedo et al [33] stated that laser activation increase significantly the reactivity of NaOCl. As LAI activates the NaOCl dissociation into its active components (Hypochlorous acid $HOCl^-$, hypochlorite ions OCl^-).The chloramination reaction between chlorine and the amino group (NH) forms chloramines that interfere in cell metabolism. Chlorine (a strong oxidant) has an antimicrobial action, inhibiting bacterial enzymes and leading to an irreversible oxidation of SH groups (sulphydryl group) of essential bacterial enzymes.

3-) Photo-mechanical interaction, where the shock waves generated from cavitation formation by the interaction between aqueous medium and Er,Cr:YSGG laser can reach a speed of up to 100 km/hr [15] . The cavitation effects disrupt the bacterial cell wall. Leaving the bacteria more susceptible to the irrigant effect, even with a lower concentration of NaOCl as stated by Muller et al,[34] who described the mechanism by which Er,Cr:YSGG lasers has the disinfection effect as the radiation has a high absorption rate by water in the smear layer and debris causing a rapid temperature rise and therefor a kind of explosive removal that disrupt bacterial biofilm and cell wall, Biofilm disruption can change the bacteria to their planktonic form, making them more susceptible to antimicrobial agents **.**

4-)Photo-thermal interaction, heating NaOCl provides an opportunity for a lower concentration solution to be used to achieve the desired clinical result similar to the higher concentrations.

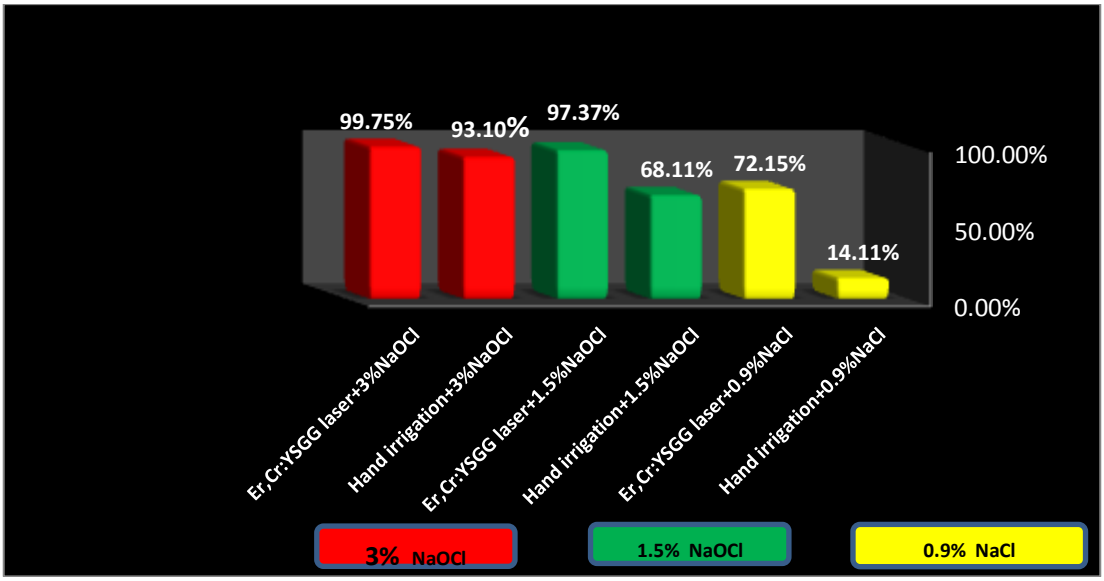
5-)Photo-static interaction, As the Er,Cr:YSGG laser has high affinity to water, absorption by water or NaOCl creates bubbles which are 1,600 times the original volume, when these bubbles present in a spaced root canal, a pressure gradient will arise that will act as a fluid pump. When the vapour bubble implodes the process works opposite, making the irrigant moves and producing turbulence within the irrigant that delivers the irrigant to areas of complex anatomy that cannot be reached with conventional methods of irrigation.

According to the results of our In-Vitro experiments, LAI of NaCl did not appear to be an effective protocol for reduction of bacterial colonies, while LAI of NaOCl showed superior antibacterial results when compared to CI method

of NaOCl, these findings improves that LAI is an effective protocol when used for activation of NaOCl. This finding is in agreement with Seet et al.[21], Pedullà et al.[20], and Christo [23].

CONCLUSIONS:

Under the conditions of the current study, LAI with Er,Cr:YSGG laser was superior to hand irrigation with (CI) protocol. LAI of 3% NaOCl were the best irrigation protocol for root canal disinfection.



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