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ANTIBACTERIAL EFFICACY OF QMIX 2 IN 1 AND 908 NM DIODE LASER IN E.FAECALIS INFECTED ROOT CANALS: AN IN-VITRO STUDY.

Dental Science

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ABSTRACT

Aim: To compare antimicrobial efficacy of QMix 2 in 1, 908 nm diode laser and their combination against E.faecalis.

Methodology: 49 single rooted extracted premolars were divided into five groups - positive control group (n=12), Group I (QMix 2 in 1) (n=12), Group II (908 nm laser) (n=12), Group III (QMix 2in 1+Laser (n=12), negative control group (n=1). Root canal was performed. Teeth were autoclaved and inoculated with E.faecalis infected nutrient broth except for negative control group. All groups were treated with respective antimicrobial solutions except for positive control group. Dentin shavings were collected with paper points and CFU/mL was counted for each group.

Results: Group I was found to be significant than Group II. Group I and Group III showed non-significant results and highest antimicrobial activity was observed with combination.

Conclusions: QMix 2 in 1 with or without use of laser has the ability to disinfect root canal.

KEYWORDS

Diode Laser, E.faecalis, Q Mix 2in 1.

INTRODUCTION:

The primary aim of any root canal treatment is to eliminate the pathogenic bacteria from the infected root canals and ensure periapical healing. The complex structure of root canal system does not facilitate the complete disinfection of microorganisms and possess the threat of re-infection. Biofilm is a slime layer which develops naturally where bacteria attach to a solid support such as dentin and contains extracellular polymeric substrate and other organic materials that acts as a natural glue to immobilize the cells. Bacterial biofilms are difficult to eliminate as they have increased resistance to wide range of antimicrobial agents. There are bacteria that form biofilm such as E.faecalis which provides a safe habitat for them against the antibacterials. E.faecalis is a gram positive, facultative anaerobic cocci (Denotti et al., 2009) and possess several defense mechanisms that allow it to survive the effect of intracanal irrigants and medication (Stuart et al.)(1-3). It is one of the most resistant bacteria in the endodontic infections and its presence is related to higher probability of failures of endodontic treatments. Despite of thorough mechanical instrumentation of a root canal system, microorganisms are readily found in the intricacies of root canal. Hence, different irrigating solutions are recommended to completely disinfect the root canal space. Irrigating solution like sodium hypochlorite (NaOCl) used extensively for irrigation is insufficient to eliminate bacteria found in isthmus, fins, ramifications and lateral canals and anatomical structures that are difficult to access for mechanical instrumentation. Also irrigation with sodium hypochlorite does not completely remove the smear layer. This smear layer is produced due to root canal instrumentation containing organic and inorganic materials along with bacteria and their byproducts. The smear layer hinders the penetration of intracanal medications and irrigating solutions into dentinal tubules and it becomes difficult to achieve complete disinfection of root canal system. Another root canal irrigant is Chlorhexidine, which is a synthetic bisbiguanide. At low concentration (0.12%), its action is bacteriostatic but at high concentration (2%), its action is bactericidal. Its main characteristics are the wide-spectrum antimicrobial effect, substantivity, and relative low toxicity (Sassone et al., 2008). As well as NaOCl, CHX has been used in several concentrations providing bacteriostatical or bactericidal effect depending on the microorganism sensitivity (Mohammed & Abott). (4-6).

It is being found that the antibacterial effectiveness of sodium hypochlorite and chlorhexidine depends on the location of bacteria. If the bacteria are found on the root canal surface, both the irrigants are capable of eliminating E.faecalis but in deeper layers, E.faecalis is more resistant to the antimicrobial effect of sodium hypochlorite and chlorhexidine because of the presence of smear layer. Another root canal disinfectant is 17% EDTA which is a chelating agent and efficiently removes the smear layer from the root canals and facilitates the penetration of irrigants in the deep layers of dentin. A newer irrigating solution, QMix 2 in 1(Dentsply) is a single step irrigant with combination of bisbiguanide antimicrobial agent (CHX) with calcium chelating agent (17% EDTA), saline and a surfactant. It has a been reported from studies, that QMix is as effective as 17% EDTA in eliminating the smear layer but still theories are lacking regarding its effectiveness against E.faecalis.(7) Despite of using various chemical irrigants, the problems associated with the smear layer and biofilm still persists. Newer methods to disinfect root canals such as lasers have been introduced. Various studies have proved that diode laser has good antibacterial properties in the disinfection of root canal system. Hence, in the present in vitro study, antibacterial efficacy of QMix 2 in

1(Dentsply) and 908 nm diode laser (DoctorSmile) along with their combined efficacy was evaluated against E.faecalis.

MATERIALS AND METHODS:

The study was performed on 49 single rooted, single canal intact premolars. Crowns of all the teeth were cut off at CEJ using a high speed handpiece and the root lengths were standardized to a 15 mm length. All the root canals were prepared using the Protaper rotary system till F3. The smear layer was removed with the help of 17% EDTA and the root canals of all samples were disinfected with 3% sodium hypochlorite. The apical foramen was sealed using self-cure GIC and the external surfaces of the teeth were covered with 2 layers of nail varnish to prevent liquid penetration. All the teeth were mounted on the self-cure acrylic blocks.

FIG.1. Decoronation of samples

FIG. 2. Mounting of samples on the acrylic blocks

The samples were autoclaved oat 121º C and 15 psi pressure for 30 mins. One tooth was randomly selected as the negative control and was placed in an incubator for 2 weeks. Nutrient broth was inoculated with E.faecalis strain (ATCC 29212) and was kept for overnight incubation.

FIG. 3. Inoculation of E.facealis infected Nutreint Broth

Next day, the turbidity of E.faecalis broth was compared with 0.5 Macfarland standard. The samples were randomly divided into four groups, namely, positive control group, QMix (Group I), Laser (Group II) and QMix + Laser (Group III), keeping 12 teeth in each group (n=12). The E.faecalis broth was inoculated in the root canal of each sample and then the root canal opening of each sample were sealed with sterile bone wax. All the samples were kept for overnight incubation. In the positive control group, all the samples were irrigated with normal saline. Samples of Group I (QMix) were first irrigated with normal saline, followed by 2 mL/1-2 min. of QMix and finally by normal saline. Samples of Group II (Laser) were irradiated with a 908 nm diode laser with the output power of 2.5 w, continuous wave mode, 10 m/sec. pulse duration into the canals 1 mm shorter than the actual root length, using an optical fibre. The handpiece was used in a circular motion at the time of lasing. The samples in Group III (QMix+Laser) were irrigated with normal saline followed by QMix (2 mL/ 1-2 min) and was irradiated with a 908 nm diode laser with the output power of 2.5 w, continuous wave mode, 10 m/sec. pulse duration into the canals 1 mm shorter than the actual root length, using an optical fibre.

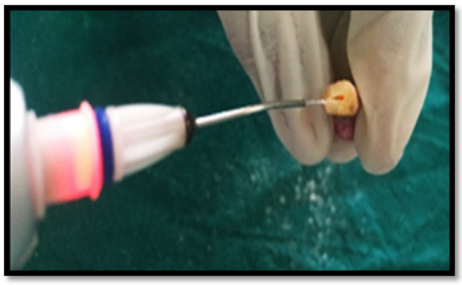


FIG. 4. Irradiation of Laser in the samples of group II and group III

The root canals of all the samples were dried by sterile paper points and then the canals were inoculated with sterile nutrient broth and kept for overnight incubation.

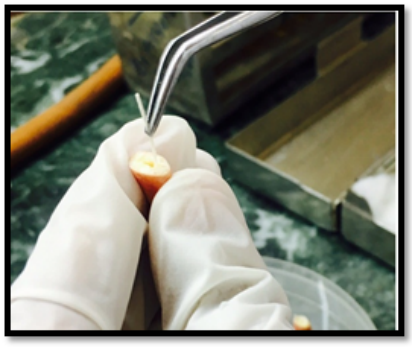


FIG. 5 collection of dentinal shavings with sterile paper point

For the root canal sampling, the samples were filled with normal saline and a 25 K file was used to generate the sample from the root canal. The sterile absorbent paper points were used to collect the sample from the root canal and were immersed into the sterile nutrient broth for overnight incubation. Next day, inoculated nutrient broth were cultured on the nutrient agar plates at 37°C for 24hours and the colonies were counted in CFU/mL.

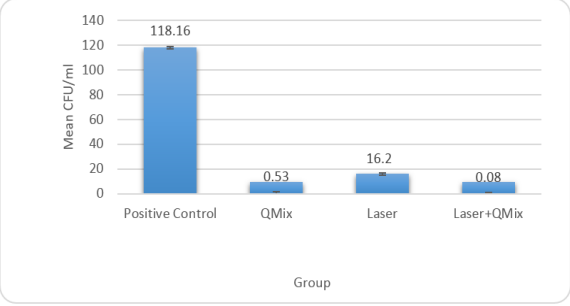
FIG. 6 Inoculation of samples collected from root canals on the nutrient agar plate

Statistical Analysis and results:-

CFU/mL in four groups were compared using one way ANOVA and Multiple Comparison: Tukey Test and software used in the analysis were SPSS 17.0 version and EPI-INFO 6.0 version and $p<0.05$ is considered as level of significance. Mean CFU/ml in positive control group was 118.16±10.99, in QMix group (Group I) it was 0.53±0.33, in Laser group(Group II) it was 16.20±2.29 and in Laser+QMix (Group III) it was 0.08±0.04. By using one way ANOVA statistically significant variation was found in mean CFU/ml in four groups($F=1225.50$,p-value= 0.0001). On comparing mean CFU/ml level in four groups statistically significant difference was found in between all the groups ($p<0.05$) except in between QMix (Group I) and Laser+QMix (Group III) which shows statistically no significant difference ($p=0.997$).

TABLE 1: comparison of mean CFU/mL of samples of each group

Group	N	Mean	Std. Deviation	Std. Error	95% Confidence Interval for Mean		Minimum	Maximum
					Lower Bound	Upper Bound		
Positive Control	12	118.16	10.99	3.17	111.18	125.15	102.00	140.00
QMix	12	0.53	0.33	0.09	0.31	0.74	0.18	1.16
Laser	12	16.20	2.29	0.66	14.75	17.66	12.00	19.00
Laser+ QMix	12	0.08	0.04	0.01	0.05	0.11	0.01	0.16



GRAPH 1: Comparison of mean CFU/mL of QMix, Laser and combination of both in four groups

DISCUSSION:

For the successful endodontic treatment outcome, it is necessary that the bacteria and their toxins are eliminated from the root canal space. Most of the endodontic failures and retreatment is due to the presence of E.faecalis. This gram positive cocci is highly resistant to many endodontic irrigants and antibacterial agents. QMix 2 in 1 is a new endodontic irrigant that consists of an antibacterial agent i.e. Chlorhexidine, calcium chelating agent such as EDTA and a surface active agent. Recent studies demonstrated that QMix2 in 1 is effective against Enterococcus faecalis. On the other hand, various studies have showed good antibacterial properties of laser in the disinfection of root canal space.

In the present study, QMix 2 in 1 which is a new irrigating solution has been compared with a 908 nm diode laser. Also the combined efficacy of both, laser as well as QMix 2 in 1 is evaluated against E.faecalis. The methodology used in this study is as same that used by **Khosrow et al**. He evaluated the disinfection ability of 980-nm diode laser in comparison with sodium hypochlorite (NaOCl) as a common root canal irrigant in canals infected with Enterococcus faecalis (E. faecalis). In the current study, QMix (CFU/mL 0.53±0.33) showed statistically significant antimicrobial property as compared to laser (CFU/mL 16.20±2.29).

Stojicic et al reported that QMix effectively killed E. faecalis biofilms grown on collagen-coated hydroxyapatite discs in vitro, and was superior to CHX and MTAD. The efficacy of QMix is related to its surface active agent which increases the wettability and allows deeper penetration of the antibacterial agent.

According to **Moritz et al**., complete eradication of bacteria was achieved only through high-power irradiation of diode laser that raises the temperature on the root surface. Diode lasers and Nd: YAG lasers have strong water sorption ability. The laser energy is absorbed in superficial dentinal tubules filled with water and hence the superficial layers receive the most antibacterial effect than the deeper layers and bacteria present in the deep layers are not eluted from the root canal. This could be the reason of lower antibacterial efficacy of laser as compared to QMix 2 in 1 in this study. (4, 8)

Also, in this study, Laser+ QMix (CFU/mL 0.08±0.04) has shown statistically significant antimicrobial property than Laser (CFU/mL16.20±2.29). In the study conducted by **Rahimi et al**., it was reported that laser is less effective in root canal disinfection compared to combined use of laser and NaOCl; hence, using laser in combination with root canal irrigants was recommended.

In the present study, a combination therapy of QMix 2 in 1 along with diode laser was used to disinfect E.faecalis from the root canals and this was found to be the most significant. (9) The additive effect of QMix and laser is attributed to its individual properties. As the surface active agent increases the wettability and provides the larger surface area for the calcium chelating agent to remove the smear layer, the antibacterial property of chlorhexidine is enhanced due to this and the application of diode laser penetrates deeper into the dentinal tubules producing the highest rate of disinfection.

Mehrvarzfar et al. suggested the combination therapy including chemical irrigation and laser irradiation as an effective treatment option for elimination of E.faecalis from the root canal system. (10) In a similar study, by **Thomas et al,** when 908 nm diode laser was used in conjunction with NaOCl and EDTA, a complete elimination of E.faecalis was demonstrated when analyzed using culture methods. (11)

In the present study, QMix (CFU/mL 0.53±0.33) and Laser+QMix (CFU/mL 0.08±0.04) has shown statistically non-significant antimicrobial properties. This might be due to the surface active agent present in the QMix which decreases the surface tension, increases the wettability and allows deeper penetration of the antibacterial agent and laser.

To summarize, QMix 2 in 1 and its combination with diode laser showed significantly better antibacterial efficacy against E.faecalis as compared to diode laser. When QMix 2 in 1 was compared with its combination with diode laser, QMix 2 in 1 showed non-significant results but latter showed higher antimicrobial activity. Hence, QMix 2 in 1 and its combination with diode laser can be considered as important adjunct to the biomechanical preparation for disinfecting the root canal system.

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