RESEARCH PAPER

"Comparative Evaluation Of Different Root Canal Disinfection Techniques - An In Vivo Study"

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ABSTRACT-
BACKGROUND- Root canal disinfection is one of the most important step of Root canal treatment which also determines the success of this treatment. Till date many chemical, herbal agents have come up which claim to provide disinfection. So this study has been conducted in order to assess the efficacy of different agents and when they are combined, what effect they provide on the disinfection process.
AIM-- To assess the effectiveness of different ways of root canal disinfection.
MATERIALS AND METHOD- 60 patients were selected and divided them into 3 groups who were clinically or/and radiographically diagnosed cases of acute irreversible pulpitis. Percentage of bacterial growth reduction was assessed in blood agar. Percentage of bacterial reduction was analyzed with the Mann Whitney U test table.
RESULT- The result showed that there is significant reduction of bacterial count in the group C, sodium hypochlorite along with laser combination than group B and group A. CONCLUSION- The study showed that sodium hypochlorite and diode laser had antimicrobial effect. However, combined effect of diode laser with NaOCl found to be more significant in bacterial colony count reduction.

Keywords- Diode LASER, Disinfection, Root Canal, Sodium Hypochlorite
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Introduction-
The successful endodontic treatment depends on elimination/eradication of microorganism from the root canal system before obturation. The chemical debridement helps in removal of
residual tissue and bacterial biofilm, mainly from the non-instrumented areas of root canal system.[1] The major cause of endodontic failure is the survival of microorganisms in the apical portion of root canal treated teeth, of which, E.faecalis is considered one of the primary organism in patients with post-treatment endodontic infection.
E.faecalis has the ability to establish monoinfections in medicated root canals. The organism has the ability to acquire, accumulate and share extra-chromosomal elements, encoding virulence raits, which help to colonize, compete with other bacteria, resist host defense mechanisms and produce pathological changes directly through the production of toxins or indirectly through the induction of inflammation.[2] Several irrigating solutions have been used to reduce microorganisms, necrotic tissues and residual debries. With the introduction of Lasers to the field of conservative dentistry and endodontic treatment has been enriched by a multitude of new treatment methods that improved the chances for a successful treatment outcome.
Laser energy can eliminate microorganisms existing in main canal, lateral canals and dentinal tubules which may cause pulp and peri-apical infection.[3]
Field of antibacterial chemotherapy is a constant challenge. The current problem of bacterial drug resistance perhaps best illustrates the continuing requirement both for new agents and new approaches

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to eliminate infection from root canal system.

Materials and Method-

This present study included patients of age 20-50 years who visited for root canal treatment at the hospital. Total 60 patients after radiographic examination were selected. The pulp vitality was performed using either electric pulp test or cold test. Patients were first explained about the procedure and after their approval written consent was taken before treatment. A detailed medical and dental history was taken. The inclusion criteria included permanent anterior maxillary teeth with acute irreversible pulpitis, teeth with vertucci's type I root canal configuration, completely developed single root with closed apex. Exclusion criteria comprised of pregnant women and lactating mothers, patients with systemic conditions, patients allergic to any components of materials being used in study, patients who are on antibiotic treatment, teeth with root resorption and calcified canals, teeth having dental anomalies, nonvital teeth, patients who are having any sort of periodontal disease with clinically and radiographic changes suggestive of periodontal/combined lesion, teeth which have been previously endodontically treated. Method-Povidine iodine solution 5% was used for disinfecting tooth and surrounding area. Local anaesthesia was administered, and isolation was done with rubber dam. A high-speed handpiece and sterilized round bur was used for access opening. Debridement was done using saline. Working length was determined radiographically and confirmed by apex locator which was kept 0.5mm short of the apex. The root canal patency was checked by size10K-file. The presterilized paper points were used for collection of initial pre-treatment root canal samples. One paper point was placed in the canal for 60 seconds and then transferred into pre sterilized tube. This was designed as the first sample(Sample1).All the samples were transferred directly in thioglycolate broth and cultured in blood agar and were kept in an incubator at 37°C for 24 hours. Colonies were counted with colony counter manual method. Chemicomechanical preparation was done using Hyflex EDM rotary file system at 450 rounds per minute at a torque of upto 2.5Ncm was used for cleaning and shaping except the glidepath files which was used at 300 rpm and at a torque of upto1.8 Ncm and then patients were randomly divided into three groups–

GroupA-Saline along with diode laser combination was used on 20 patients

GroupB-5.25%NaOCl was used alone on 20 patients.

GroupC- 5.25%% NaOCl and diode laser on 20 patients.

Post treatment sample collection was done after disinfection, according to selected protocol and the canals were treated according to the groups given below.

GroupA- Irrigation was done with 5ml of saline and then canal irradiated with Diode laser wavelength of 980nm was used at 1.5 watt power, frequencies 15 Hz, energy 21.2 j , average power-0.7watt, then, the fiber optic tip 200 micrometer was placed 1 mm short of the working length and reccess in circular movement with speed of 1 mm/sec and repeated six times at intervals of 10 seconds between each one. After completion of irradiation samples was taken with sterile paperpoint and designated as (SampleA2).

GroupB- The Canal was irrigated using 5ml of 5.25% NAOCL irrigating solution with 30 gauge side vented needle which was kept 1mm short of the apex. Then canal was flushed with saline and sample was taken with sterile paper point and designated as (SampleB2).

GroupC- First the canal was irrigated with 5.25% sodium hypochlorite and then irradiated with diode laser wavelength of 980nm was used with 1.5 watt power, frequencies 15 Hz, energy 21.2 j ,average power- 0.7watt, then the fiber optic tip 200 micrometer was placed 1 mm short of the working length, in circular movements with speed 1 mm/sec, and repeated six times at intervals of 10 seconds. And then sample collection was done with sterile paper point and designated as (SampleC2)

All the samples were transferred directly in thioglycolate broth and cultured in blood agar and were kept in an incubator at 37°C for 24 hours.

The lab procedure included microbiological culturing of the collected samples(paper point),inoculated in nutrient broth and Thioglycolate broth were vortexed and then incubated at 37degree Celsius for 24 hours to 48 hours to observe the turbidity. Calculation was done for counts per ml of diluted broth with multiplication by dilution factor.

Sample was plated on MacConkey agar and blood agar plates (HiMedia Lab Pvt. Ltd. India)with a calibrated loop. Incubation of agar plate was done for 24-48hours at 37 degree celsius

. and counted for the growth of bacteria and followed with manual colony counting for observing the efficacy of A, B, and C solutions. The similar procedure was performed in both the groups pretreatment and post treatment samples in order to figure out the effectiveness of the solution against the bacteria.(Fig 1a to 1h)

Observation and Result-

Statistical analysis was carried out and following results were observed- Mean colony count manually in Saline alongwith diode laser group was710890±752689; in 5.25%NaoOCL alone group it was 478755±424803 and in 5.25%NaOCL laser group it was 81766.75±87856.27. By using Krushkal Wallis Chi square test statistically significant variation was found in mean colony count manually in three groups of patients(χ2-value=8.04,p-value=0.001) (Table 1a).

On comparing mean colony count manually among

three groups by using Mann Whitney U Test, statistically significant difference was found between saline along with diode laser group
and 5.25% NaOCL and diode laser group(p=0.001) and between 5.25% NaOCL alone and 5.25% NaOCL and diode laser group(p=0.040) and no significant difference was found between saline along with diode laser and5.25%NaOCL group(p=0.316) (Table 1b). Whereas reduction in bacterial colonies in saline along with diode laser group was 320449.50±270677, in 5.25% NaoOCL alone group it was 239735±263889 and in 5.25% NaOCL Laser
group it was 9984±15823.56. With the help of Krushkal Wallis Chi square test statistically significant variation was found in mean reduction in bacterial colonies in three groups of patients (χ2-value=10.87,p-value=0.0001) (Table 2a).
On comparison of mean reduction in bacterial colonies amongst three groups by using Mann Whitney U Test statistically significant difference was observed between saline along with diode laser group , 5.25% NaOCL and diode laser group(p=0.0001) and between 5.25% NaOCL alone and 5.25% NaOCL and diode laser group(p=0.004) and no significant difference was found between saline along with diode laser and5.25%NaOCL along group(p=0.477) (Table 2b).

Discussion-

Ideal property of an irrigant is that it should be bactericidal, germicidal, and fungicidal; ability to serve as a lubricant during instrumentation; also dissolve organic and inorganic dentinal tissues(pulp tissue, collagen, and biofilm);non-irritating to periapical tissues; prolonged and sustainable antibacterial activity after use; activity in an environment in which blood, serum, and
tissue protein products are present and it should remove the smear layer completely.[4]
Sodium hypochlorite and hydrogen peroxide, or the combined use of both are the most frequently used irrigation solutions. Their benefits include good tissue dissolving and
disinfecting capability whereas NaOCl reacts with organic tissue, resulting in saponification, amino acid neutralization, and chloramine reactions. Owing to its solvent effect on necrotic
tissues,[5] NaOCl has become the most widely used irrigation solution in endodontics.
The concentration of the irrigants is still a matter of debate and remains controversial; many authors recommend a 5.25% concentration of sodium hypochlorite, others prefer a lower concentration of 3% or even 0.5%. [6] Sodium hypochlorite is highly effective antimicrobial agent, but it has some drawbacks also.
As adjunct to currently used disinfection methods in root canal disinfection various LASER systems have been examined to improve the efficacy of dynamic irrigation techniques.
Since the development of the ruby laser by Maimanin1960and the application of the laser for endodontics by Weichman in 1971, a variety of papers on potential applications for lasers in endodontics have been published.[7]
The first laser use in endodontics was reported by Weichman & Johnson.[8] The impact of the laser light depends on the interaction of the light quanta and the molecules and the molecular formations in the target material. Working of the laser and its effect on biological tissue is determined by interaction of laser radiation parameters, such as: wavelength, physical characteristics of the illuminated tissue, energy radiation, continuous or pulsed mode, diameter of the laser beam, and the exposure time.

Hyflex EDM (COLTENE)

rotary is a one file system, which has controlled memory, greater flexibility and extreme fracture resistance, retain their shape in curved canals and donot possess the spring back action, thereby avoiding any perforation. The conventional NiTi files have another drawback of not giving a warning sign before breakage (unlike stainless steel files, which show such signs in the form of unwinding of flutes, or presence of a shiny spot on the file, indicating that the file should be discarded).[9] 5.25% NaOCl is significantly more efficient in eliminating microorganisms and in particular the resistant Enterococcus faecalis as compared to the antimicrobial efficacy of 1.3% NaOCl (Khoand Baumgartner, 2006).[6]
Study done by Moritz et al . showed that treatment of root canals with 2W diode laser(810nm), when the irradiation was repeated 5 times at each laser treatment, each time for a period of 5sec with short breaks in-between, a maximum of two irradiations resulted in nearlycomplete elimination of E.faecalis. Gutknecht et al. also demonstrated that diode laser can eradicate microbes that have migrated deep into the dentine and more specifically Enterococcus faecalis(Gutknecht et al.,2004).[10]
An early study by Gutknecht et al. (1996) reported that 83% of infected cases were treated successfully, after been unsuccessfully treated by conventional chemomechanical method.
Also NjwanF. Sehaba et al done a study and showed that 10 sec (6 cycles) exposure time to laser, was more effective than 5 sec (6 cycles) at each output power, and no significant difference between 2W and 2.5W at 5 sec (6 cycles).[11] These results indicated that time plays an important role in root canal disinfection when using laser. So in this present study we have used 1.5WATTat10 seconds(6cycles).
Mashalkaretal. and Agrawal AA et al. also concluded from the in-vivo comparative study that conventional method of root canal disinfection using sodium hypochlorite and hydrogen peroxide as irrigating solutions were highly effective, however lasers when used can also reduce the bacterial load of the infected root canal.[2]
Limitation in laser application is rise in temperature which eventually damages the periapical area, especially when the roots are close to proximity of mental foramen, canal of the inferior
alveolar nerve or maxillary sinus. Passage of laser

beam through the apex of the roots can damage this anatomic region. Laser-assisted or ultrasonic irrigant agitation techniques proved to optimize irrigation through earlier research on dentinal debris removal capabilities.[12] There is a lack of evidence comparing these methods of cleaning efficacy of biofilm-infected dentin. Endodontic biofilms are therapeutically significant as they are regarded as one of the basic survival methods employed by bacteria in times of starvation and are resistant to clinical antimicrobial therapy.[13] The superior smear layer removing the ability of 18% etidronic acid activated with ultrasonics was also
highlighted in the study by Awati *et al*. that was based on the confocal imaging technique.[14] Laser-assisted or ultrasonic irrigant agitation techniques proved to optimize irrigation through earlier research on dentinal debris removal capabilities.[15] The pulse energy of the erbium group of lasers is strongly absorbed by water and NaOCl, resulting in vaporization and creation of vapor bubbles inducing a secondary cavitation phenomenon.[16]

Conclusion-

The chemomechanical preparation forms an integral part of rootcanal treatment. The eradication of persisting microorganisms in distant areas of the tubular system is a major challenge in the present-day treatment regimen. Findings of this study also indicate that sodium hypochlorite and diode laser had antimicrobial effect. However, combined effect of diode laser with NaOCl found to be more significant in bacterial colony count reduction. This study can prove to be helpful for efficient endodontic treatment in future although Laser is not much cost effective and hence has its limitation.

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Table 1a: Comparison of colony count manually in three groups Descriptive Statistics

GROUP	N	Mean	Std. Deviation	Std.Error	95% ConfidenceIntervalforMean		Minimum	Maximum
					LowerBound	UpperBound		
A. (Saline alongwith diodelaser)	20	710890	752689	168306	358620.53	1063200	48200.00	3250000
B.(5.25% NaOClalone)	20	478755	424803	94988.83	279941.07	677568.92	32400.00	1209600
C.(5.25% NaOCl anddiodelaser)	20	81766.75	87856.27	19645.26	40648.74	122884.75	3240.00	330800

Table 1b-Mann Whitney U Test

Group		Mean Difference(I-J)	Std. Error	p-value	95%ConfidenceInterval	
					Lower Bound	Upper Bound
Saline along with diode laser	5.25%NaOCl alone	232135	158610	0.316,NS	-149547.90	613817.90
	5.25%NaOCl and diode laser	629123	158610	0.001,S	247440.34	1.0108E6
5.25%NaOCl Alone	5.25%NaOCl and diode laser	396988	158610	0.040,S	15305.34	778671.15

Table 2a: Comparison of Reduction in Bacterial Colonies in three groups Descriptive Statistics

Group	N	Mean	Std. Deviation	Std.Error	95% Confidence Interval for Mean		Minimum	Maximum
					Lower Bound	Upper Bound		
Saline along with diode laser	20	320449.50	270677	60525.14	193768.91	447130.08	20610	826800
5.25%NaOCl Alone	20	239735.00	263889	59007.30	116231.28	363238.71	9800	865700
5.25%NaOCl And diode laser	20	9984.00	15823.56	3538.25	2578.34	17389.65	0.00	54310

Table 2b- Mann Whitney U Test

Group		Mean Difference(I-J)	Std. Error	p-value	95%ConfidenceInterval	
					Lower Bound	Upper Bound
Saline along with diode laser	5.25%NaOCl alone	80714.50	69078.09	0.477,NS	-85516.28	246945.28
	5.25%NaOCl and diode laser	3466	69078.09	0.0001,S	144234.71	476696.28

5.25%NaOCl Alone	5.25%NaOCl and diode laser	2751	69078.09	0.004,S	63520.217	395981.78