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# Comparison of smear layer removal ability of different agitation system in the apical third of the root canal using SEM: an *in vitro* study

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## Abstract

**Introduction:** Sodium Hypochlorite (NaOCl) remains gold standard as a result of its antimicrobial effect and tissue dissolution properties, but it has no effect on inorganic portion of smear layer. Thus the combination of NaOCl and EDTA has been proven to have the perfect ability in removal of both organic and inorganic debris. These irrigants when used with conventional syringe irrigation were unable to penetrate the apical portion of the root canal, so new activation devices have come in the market which claims to be effective in delivering the irrigant to the working length. **Objective/Aim:** This study evaluated and compared the efficacy of recently introduced irrigation activation devices EndoActivator, Passive ultrasonic irrigation and Laser on removal of smear layer from the apical third of instrumented root canal using Scanning electron microscope. **Methods:** Forty three single rooted teeth were prepared with the help of protaper files and divided into four groups. Group I: EDTA only, Group II: Endoactivator, Group III: Laser, Group IV: Passive Ultrasonic Irrigation (PUI). Three specimens were not treated with any smear layer removal protocol and were immediately sectioned and sent for SEM examination. The remaining 40 samples from 4 groups after treatment with different activation system were also sectioned and sent for SEM examination. The data obtained were statistically analysed using Friedman's test. **Results:** All the four groups removed smear layer and the laser showed best smear layer removing capability compared to other groups but was significant only with respect to control and group I (EDTA group without any activation) ($p<0.05$). **Conclusions:** Within the limitations of the study, all the activation systems were able to remove the smear layer from the apical third of the root canal with laser showing the best result followed by Endoactivator and then PUI.

**Keywords:** Ethylenediaminetetraacetic acid, Irrigation, Laser, Scanning electron microscopy, Sodium hypochlorite

## INTRODUCTION

Endodontic therapy aims to return the involved teeth to a state of health and function [1]. When dentine is cut using hand or rotary instruments during biomechanical preparation, the mineralized tissues are not shredded or cleaved but shattered to produce considerable quantities of debris called smear layer [2]. Many researchers suggest that this smear layer results in the blockage of dentinal tubules thereby limiting bacterial or toxin penetration by altering dentinal permeability [3, 4]. While some other authors believe that this loosely adherent smear layer must be completely removed from the surface of the root canal wall as it can harbour bacteria and provide an avenue for leakage thereby limiting the disinfection of the dentinal tubules by preventing irrigants and medicaments from penetrating into the tubules] [5, 6]. Because of these toxic and undesired effects of smear layer, the chances of improper disinfection and reinfection increases leading to failure of the endodontic treatment which makes removal of smear layer utmost important in order to achieve the desired result.

Widely accepted conventional irrigation with a syringe has demonstrated the flushing action to remove debris from root canal irregularities, but was not sufficient. It has also been seen in a previous study that when conventional syringe needle was used, the irrigating solution was delivered only 1 mm deeper than the tip of the needle [7]. In a study by Claw it was concluded that effective apical irrigation is directly proportional to the depth of insertion of the needle which at times presents a challenge to the clinician [8].

Studies have shown that instrumentation of root canal, causes formation of the smear layer on root canal walls more significantly in the apical third [9, 10]. To attain maximum efficacy of irrigants an effective irrigation delivery system is required which will delivery it with adequate flow and volume to the working length to be effective in debriding the complete canal system [11, 12].

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Studies have shown that regardless of the pattern of instrumentation and irrigation effectiveness of irrigating solution remains limited in prepared root canals. [12, 13] Hence, improvement in the irrigation protocols have become utmost important during root canal treatment in order to achieve better cleaning efficiency in very complex area] [14].

There are new devices available which agitate the irrigants and increase its penetration and disinfection efficacy in the root canal system, but data to compare these new and emerging devices and methods for disinfection of root canal system are limited in the published literature. Therefore, this study was planned to evaluate and compare in vitro, the efficacy of recently introduced irrigation activation devices EndoActivator, Passive ultrasonic irrigation and Laser on removal of smear layer from the apical third of instrumented root canal using Scanning electron microscope.

**MATERIALS AND METHODS**

The study protocol was approved by the ethical committee of Sri Rajiv Gandhi College of Dental Sciences and Hospital Bangalore. Human mandibular first premolars which were extracted for periodontal or orthodontic purposes, having single straight canals without bi/trifurcation were included. Fractured teeth, carious teeth, teeth with internal or external resorption and teeth with hypoplasia were excluded.

**Specimen preparation**

All the specimens were cleaned of superficial debris, calculus, tissue tags and stored in normal saline They were then decoronated below the cemento-enamel junction using diamond disk to leave a root of 12mm in length and intraoral periapical radiograph (IOPAR) was taken to check for internal anatomy of canals, resorption, caries, fracture and only teeth with straight canals was selected. A size 10 K file was then introduced into the canal until it is visible at the apical foramen and the working length was determined by reducing 1mm from the total root length.

All root canals were prepared with rotary Ni-ti instruments (Protaper universal) with a pre-determined working length till size F3. Copious irrigation was done using 3% NaOCl in between instrumentation.

Three samples were sectioned immediately after cleaning and shaping and apical third was evaluated using scanning electron microscope for the presence of smear layer.

**Irrigation Protocol**

The remaining forty specimens were then randomly divided into four experimental groups with 10 samples each and were treated as follows:

**1. Group I : Only EDTA Used**

After preparing the canal as mentioned above, a 27 gauge side vented needle was introduced 2mm short of working length and used to employ final flush with 3ml of 17% EDTA for 1 min using inside-out motion without any physical agitation.

**2. Group II : Endoactivator with EDTA**

Canals were irrigated with 1.5ml of 17% EDTA, and the medium sized tip (# 25 ISO) was attached to the Endoactivator and used 2mm short of the working length with pumping motion for 30 sec for 2 cycles at high speed (10000 cpm). Fresh irrigant was introduced in between cycles.

**3. Group III : Laser with EDTA**

After filling the root canal with EDTA, diode laser (980 nm, 2 W, 200 µm tip) was used for 6 cycles of 10 seconds each. During lasing the laser was held stationary 2mm short of the working length for the first 2 seconds in each lasing period and then withdrawn at the rate of 1mm $s^{-1}$ for the remaining 8 seconds. Total volume of irrigant used was 3ml and after each cycle fresh irrigant was used.

**4. Group IV : Passive ultrasonic irrigation with EDTA**

Passive ultrasonic irrigation device (# 25 size tip) was used and the tip was held stationary 2mm short of the working length for 2 cycles of 30 seconds each with 1.5ml of 17% EDTA in each cycle.

In all groups canals were rinsed with saline to rinse off the residual irrigants and dried with paper points.

**Evaluation**

All samples from the four groups were split longitudinally by first creating grooves on the buccal and lingual side without entering the lumen using a diamond disk and then chisel was used to split the samples into two halves to give a total of 86 halves. One of the two halves in which the apical third was prominently visible was examined under scanning electron microscope for debris and smear layer coverage at 1000X. Photographs were taken from the apical third and were graded from 0 to 3 [22] by blinded observers as follows:

1. No smear layer, open dentinal tubules, smear layer completely removed or melted.
2. Moderate smear layer, outlines of dentinal tubules observable, removed or melting in some areas.
3. Thin smear layer covering the surface outline of the dentinal tubules which were not discernible, and the location of the tubule will be indicated by crack, scattered laser, removed or melting.
4. Heavy smear layer, outlines of tubules obliterated, no visible smear layer removed or melting.

The data was then statistically analyzed using Friedman test which is a non-parametric test for testing the difference between several related samples.

**RESULTS**

The scores given by two blinded observers are given in Table 1 and Table 2:

**Table 1:** Scores given by the blinded Observer 1

| Group I | Group II | Group III | Group IV | Control |
|---|---|---|---|---|
| 2 | 0 | 0 | 2 | 3 |
| 2 | 0 | 0 | 0 | 3 |
| 3 | 3 | 2 | 2 | 3 |
| 1 | 2 | 0 | 2 | |
| 1 | 1 | 0 | 2 | |
| 1 | 0 | 0 | 2 | |
| 3 | 1 | 1 | 1 | |
| 0 | 1 | 1 | 1 | |
| 2 | 2 | 1 | 0 | |
| 1 | 0 | 0 | 0 | |

**Table 2:** Scores given by the blinded Observer 2

| Group I | Group II | Group III | Group IV | Control |
|---|---|---|---|---|
| 3 | 0 | 0 | 2 | 3 |
| 2 | 1 | 0 | 0 | 3 |
| 3 | 2 | 1 | 2 | 2 |

| | | | | |
|---|---|---|---|---|
| 1 | 2 | 0 | 3 | |
| 1 | 2 | 0 | 2 | |
| 2 | 1 | 1 | 1 | |
| 3 | 1 | 0 | 1 | |
| 1 | 1 | 1 | 1 | |
| 3 | 2 | 0 | 0 | |
| 1 | 0 | 1 | 1 | |

The agreement between two observers was evaluated using kappa value. Table 3 shows the kappa value for the four groups and the results revealed substantial agreement (0.61–0.80) in group 1, group 2 and group 4 between the observers, whereas slight agreement (0.01–0.20) was found between the observers in group 3.

Since there was substantial agreement between the value of the both the observers in majority of the groups, either of the two values was selected randomly for the statistical analysis.

Laser showed the minimum smear layer remaining or maximal removal of smear layer in the apical third of the root canal followed by Endoactivator, PUI and EDTA alone. Maximum remaining smear layer was found in the control group.(table 3)

On pairwise comparison laser was found to be the best in removal of smear layer compared to Endoactivator and passive ultrasonic irrigation but the value was not statistically significant. Control group showed maximum remaining smear layer which was found to be statistically significant when compared to all other groups ($p < 0.05$). There was also significant difference in the remaining smear layer between the EDTA group and laser group ($p < 0.05$).

**Table 3:** Comparison of mean values between different groups

| Group | N | Mean | SD | Min. | Max. | Median | ChiSquare^ | 'p' value |
|---|---|---|---|---|---|---|---|---|
| Group I | 3 | 2.33 | .577 | 2 | 3 | 2.00 | | |
| Group II | 3 | 1.00 | 1.732 | 0 | 3 | 0.00 | | |
| Group III | 3 | 0.67 | 1.155 | 0 | 2 | 0.00 | 5.3500 | *0.021* |
| Group IV | 3 | 1.33 | 1.155 | 0 | 2 | 2.00 | | |
| Control | 3 | 3.00 | .000 | 3 | 3 | 3.00 | | |

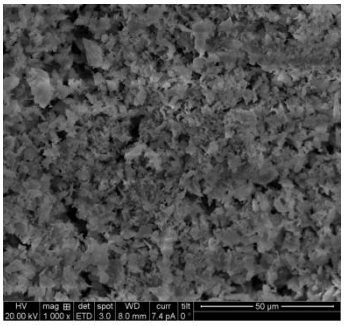

**Figure 1:** SEM image showing remaining smear layer in control group

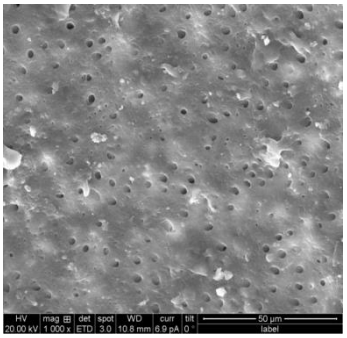

**Figure 2:** SEM image showing remaining smear layer in EDTA group (Group 1)

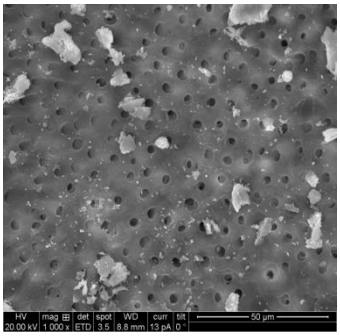

**Figure 3:** SEM image showing remaining smear layer Endoactivator group (Group 2)

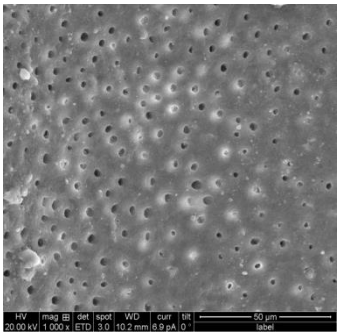

**Figure 4:** SEM image showing remaining smear layer Laser group (Group 3)

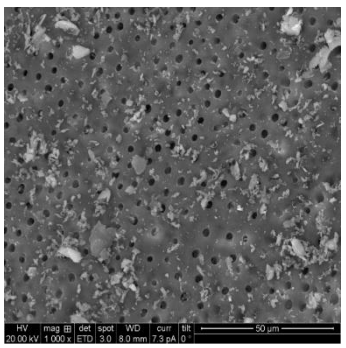

**Figure 5:** SEM image showing remaining smear layer PUI group (Group 4)

**DISCUSSION**

This in vitro study compared and evaluated the efficacy of different irrigation systems in removal of smear layer from the apical third of the root canal system using scanning electron microscope. The study was done under strict and standardized condition to minimize the bias and confounding factors. In all the groups samples were enlarged till size F3 to keep the apical enlargement uniform for all the samples and the time and quantity of the irrigant used during and after biomechanical preparation was standardized to 1min and 3ml respectively and the tip of the agitation system was kept 2mm short of apex for all the samples.

The reason because of which the apical part of the root canal was analysed was that significant amount of smear layer formation in that part and inability of the irrigant to be delivered by conventional needle delivery system to go 1mm beyond the tip. Therefore only apical third was evaluated in the study as studies show removal of smear layer is lot easier in coronal and middle third due tobetter delivery of irrigants [9, 11, 12].

Different scoring systems were used in the previous studies which vary from simple criteria, such as 'debris present or absent' [15,16] to arbitrary three-, four-, five-, or seven-point scoring systems [17-20]. Scores may be expressed in terms of amount of debris or smear layer per root level or canal, or alternatively, as percentage area of root surface occupied [21]. Given the subjective nature of the scoring, some form of reproducibility tests should be performed but are rarely reported. In the present study SEM photographs so obtained were scored by two blinded observers using the scoring criteria given by Takeda *et al* in 1998 [22] because of its simplicity, clarity and ease of scoring after which data was collected and subjected to statistical analysis [22]. Two blinded observers were used in the study to reduce the systematic errors and bias and also to eliminate inter-observer variability.

Removal of the smear layer during or after root canal instrumentation requires the use of irrigants that can dissolve both organic and inorganic components. The most commonly used irrigant 1-7% of NaOCl is effective in removing the organic portion of the infected root canal However, its capacity to remove smear layer from the instrumented root canal walls has been found to be lacking [23]. The conclusion reached by many authors is that the use of NaOCl during or after instrumentation produces superficially clean canal walls with the smear layer present [11,24]. In the present study this control group showed the highest mean remaining smear layer on the canal walls and when compared to other groups it was found to be statistically significant which is in accordance with the previous studies.

Only EDTA was used as the smear layer removal protocol in group I with the help of 27 gauge needle with in and out motion without any agitation. Analysis of the apical third of root canal revealed very high remaining smear layer on the root canal (mean - 2.33) with most of the dentinal tubules obliterated. When compared to the control group it showed statistically less remaining smear layer but when compared to the remaining three groups it showed high remaining smear layer but was significant only when compared to group III (Laser). This can be explained based on the fact that irrigation with needle without any agitation removes debris by its flushing action and penetrate only 1mm beyond the tip of the needle keeping the apical third almost untouched by the irrigant [29].

Researchers advocate the use of different agitation techniques to make the irrigants reach the apical third of the root canal to render their action.

The EndoActivator System uses sonic energy in which activator tips are used in conjunction with the hand piece driver to provide the energy for tip oscillation and vibration which produces cavitation and acoustic streaming thereby improving debridement and the disruption of the smear layer and biofilm.

The result of the present study showed that Endoactivator removed smear layer effectively in the apical third of the root canal with the mean remaining smear layer score of 1. Group I (Needle Irrigation), Group IV (PUI) and control group showed more remaining smear layer on the canal walls when compared with Endoactivator but was significant only with respect to control group. A possible explanation for irrigant activation giving cleaner canals is that higher frequency resulting in higher flow velocity which helps in dislodging the debris more efficiently [26]. The results are in accordance with the study done by Mathew *et al*., to evaluate antimicrobial efficacy where EndoActivator showed better results than conventional needle irrigation and also by the study done by Manuele Macini *et al* in which Endoactivator removed the smear layer effectively in the apical third of the root canal when compared to PUI [27,28].

The recently introduced laser technique and devices have gained utmost importance due to its compactness and low cost. The added advantage because of which it is recommended for endodontic treatment because its wavelength is within the infrared range, and thin and flexible fibres can be used. Previous studies have shown the bactericidal effects of 810-nm wavelength and 980-nm wavelength diode lasers [29]. Because of the above mentioned advantages, we have used diode laser with 200 nm fibre tip for lasing the root canals to agitate the irrigant in our study.

In the present study the diode laser parameters used were based on the known threshold laser settings required to induce agitation, cavitation and shockwaves [30]. The findings of the present study demonstrated that Laser effectively removed the smear layer in the apical third of the root canal with the least remaining smear layer score (mean 0.67). The result is in accordance with the previous study done by Manfred Langemann *et al* in which diode laser effectively removed the smear layer [31]. This result can be attributed to warming of the irrigant solution (although not to the point of decomposition) as well as to physical agitation of the fluid, which enhances the debridement through shear forces and hydraulic stresses agitating the solution [32].

The mechanism of action of PUI is by transmission of acoustic energy from an oscillating file or smooth wire to an irrigant in the root canal. The energy gets transmitted via ultrasonic waves which induces acustic streaming and cavitation of the irrigant [33]. Results of the present study showed effective smear layer removal by PUI when compared to non-agitated EDTA group (Group I) and control which was found to be significant with respect to the later. Whereas laser and Endoactivator showed better cleaning efficacy in terms of smear layer removal compared to PUI. The result is in accordance with the majority of the studies done in the past where laser was found to be effective in removal of the smear layer from the root canal, the reason being attributed to cavitation and subablative settings of the laser that do not damage the canal wall [33, 34].

**CONCLUSION**

Within the limitations of the study it can be inferred that Laser showed the best result in removing smear layer from the apical part of root canal and on pairwise comparison, it showed significant smear layer removing capability when compared to EDTA and control group but was found to be non-significant when compared with other agitation systems.

**Conflict of Interest**

The author reports no conflicts of interest.

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