

**Comparative evaluation of smear layer removal with ultrasonic activation using two different irrigants- An Invitro Sem Study.**

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**Citation of this Article:** : Dr Pradeep P.R, Dr. Rishi Rajan, Dr Ananthakrishna S, Dr Aksa Mariam Varghese, “Comparative evaluation of smear layer removal with ultrasonic activation using two different irrigants- An Invitro Sem Study”, IJDSIR- July - 2021, Vol. – 4, Issue - 4, P. No. 221 – 228.

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**Type of Publication:** Original Research Article

**Conflicts of Interest:** Nil

**Abstract**

**Introduction:** Smear layer is a submicroscopic granular structure made up of inorganic debris trapped in an organic matrix. It extends 1-40 µm deep inside the dentinal tubules. Since it is a loosely adherent structure which acts as a substrate for microbial growth, its removal by various irrigants like NaOCl, EDTA and activation devices like ultrasonic, endoactivators etc. is necessary for ensuring a predictable endodontic outcome, however this is controversial. Purpose of this study is to compare the efficiency of ultrasonic activation in smear layer removal from the apical 1/3rd of root canals using EDTA and an innovative irrigant OXUM as final irrigating solution.

**Methodology:** Thirty extracted single rooted human mandibular premolars were decoronated and standardized to a length of 16mm. The root canals are cleaned and

shaped using universal ProTaper rotary system and irrigated with 5ml of 3% NaOCl after each instrument change. The final irrigation sequences were as follows, Group I- 0.9% saline(control), Group II – OXUM(superoxidised water), Group III- 17%EDTA. Each group is further subdivided in to 2 subgroups. Subgroup A- without ultrasonic activation of irrigant & Subgroup B- with ultrasonic activation. All the samples were irrigated with 4ml of respective irrigants for two minutes. Then samples will be irrigated with 4ml of distilled water to remove any precipitate. Each tooth sample will be split longitudinally and apical third smear layer removal was studied using scanning electron microscopy.

**Results:** EDTA was found to have the highest smear layer removal ability, regardless of the irrigation method (with

or without ultrasonic activation) ( $p < 0.05$ ). In addition, the use of ultrasonic activation did not significantly influence smear layer removal at the apical third when the same solution was used.

**Conclusion:** Oxum showed smear layer removal significantly higher than saline, and similar to EDTA. There is no significant differences were identified in the apical region of the different groups, with and without ultrasonic activation of irrigants.

### Introduction

The discipline of endodontics is governed by paradigms like clinical protocol, quality of instrumentation, effective irrigation, disinfection and obturation of the entire pulp space to achieve a three dimensional seal. The anatomical complexity and variations within the root canal systems enhances bacterial invasion and also makes the cleaning and shaping procedure task oriented.<sup>1</sup> Endodontic instrumentation using both hand and rotary instruments produces organic and inorganic debris that are embedded within a layer of amorphous tissue referred to as the 'smear layer'. Presence of smear layer has proven to be deleterious because it prevents the penetration of irrigants, intracanal medicaments and also the filling materials into the dentinal tubules.<sup>2</sup> Irrigation solutions are used as lubricants and disinfection agents during chemo-mechanical endodontic treatment which improve the permeability of the canal and the elimination of the contaminated dentin. Currently, Sodium hypochlorite (NaOCl) (0.5–6%) and ethylenediaminetetraacetic acid (EDTA) (15–17%) are the commonly used intracanal irrigants in endodontic practice. NaOCl acts as an organic material solvent and an antimicrobial agent, while EDTA serves as an inorganic solvent or smear layer removal agent. Thus, the consecutive use of these two solutions represents an optimal irrigation protocol.<sup>3</sup> One such widely investigated irrigant is super-oxidized water.

It is one of most powerful antimicrobial agent available for use in both medical and dental field <sup>4</sup>.

The greatest difficulty in endodontic instrumentation involves the apical third of the canal. Studies demonstrating the removal of the smear layer in this area showed remaining debris with both conventional and activated irrigation techniques.<sup>5</sup>

The purpose of this study was to evaluate the effectiveness of different final irrigants with or without ultrasonic agitation technique on smear layer removal at the apical third of the root canals.

### Materials and methodology

Thirty extracted single rooted human mandibular premolars were decoronated and standardized to a length of 16mm. The root canals are cleaned and shaped using universal ProTaper rotary system and irrigated with 5ml of 3% NaOCl after each instrument change. The final irrigation sequence will be as follows,

Group I- 0.9% saline (control)

Group II – OXUM (superoxidised water)

Group III- 17%EDTA.

Each group is further subdivided in to 2 subgroups.

Subgroup IA-0.9% saline without ultrasonic activation of irrigant

Subgroup IB- 0.9% saline with ultrasonic activation.

Subgroup IIA- OXUM without ultrasonic activation of irrigant

Subgroup IIB - OXUM with ultrasonic activation.

Subgroup IIIA - 17%EDTA without ultrasonic activation of irrigant

Subgroup IIIB - 17%EDTA with ultrasonic activation.

All the samples will be irrigated with 4ml of respective irrigants for two minute. Then samples will be irrigated with 4ml of distilled water to remove any precipitate. Each tooth sample will be split longitudinally and apical third

smear layer removal was studied using scanning electron microscopy.

### Conventional Irrigation Group

The canal was flushed with 4mL of irrigants, the solution was left in place for two minutes with no agitation.

### Ultrasonic Group

All the samples of both the groups were irrigated with 4 ml of irrigating solution for 2 minutes using conventional irrigation method and ultrasonic irrigation method .The irrigation time in the group was divided as follows:

- i) 30 sec conventional syringe irrigation
- ii) 20 sec passive ultrasonic irrigation
- iii) 20 sec conventional syringe irrigation
- iv) 20 sec passive ultrasonic irrigation
- v) 30 sec syringe irrigation.

1.5mL of irrigant was used in the first 30 s of irrigation, 1 mL in between the two periods of passive ultrasonic irrigation and then 1.5 mL of irrigant in the final stage of irrigation. The irrigation needle size used is 30 gauge with closed end and side vented.

Finally, the specimens were irrigated with 5 mL sterile distilled water, dried, temporarily sealed, and stored separately in labeled bottles<sup>6</sup>.

### Scanning Electron Microscopy Examination

The canals were dried with absorbent paper points, and the entrance to each of the canals was protected with a cotton pellet. Using carborundun discs, deep grooves were cut on the buccal and palatal surfaces of the roots, without perforating the root canal. The roots were then split with a chisel and a hammer. One half of each tooth was selected and prepared for SEM examination. After assembly on coded stubs, the specimens were placed in a vacuum chamber and sputter-coated with a 300 Å gold layer (Bal-Tec SCD 005; Bal-Tec Co., Balzers, Liechtenstein). The specimens were then analysed using a Zeiss EVO SEM XL 30 (Carl Zeiss Microscopy). The dentinal wall of the

apical thirds of roots were observed at magnifications of up to X1000 for the presence or absence of smear layer and the visualization of the entrance to dentinal tubules. The smear layer was scored according to the criteria given by Hulssman et al .

Score 1: No debris.

Score 2: Clumps of debris covering

Score 3: Clumps of debris covering 25–50% of the canal wall.

Score 4: Clumps of debris covering more than 50–75% of the canal wall.

Score 5: More than 75% of canal wall covered by debris.

### Statistical Analysis

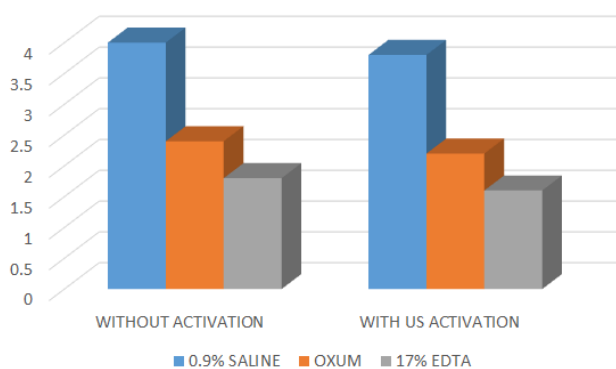
Statistical Package for Social Sciences [SPSS] for Windows, Version 22.0. Released in 2013. Armonk, NY: IBM Corp., was used to perform statistical analyses. Descriptive analysis includes expression of smear layer removal score in terms of mean & standard deviation (SD).One-way ANOVA test was used to compare the mean smear layer removal score between 03 irrigants for each irrigation technique. Independent Student t Test was used to compare the mean smear layer removal score between the irrigating techniques under each irrigant .The level of significance [P-Value] was set at P<0.05.

### Results

Experimental Group	Score for Smear Layer at apical third (MEAN ± SD)			
	Without ultrasonic activation	ultrasonic	With ultrasonic activation	ultrasonic
Group I 0.9% saline	4 ± 0		3.8 ± 0.4	
Group II Oxum	2.4 ± 0.55		2.2 ± 0.83	
Group III 17% EDTA	1.8 ± 0.83		1.6 ± 0.24	

EDTA was found to have the highest smear layer removal ability, regardless of the irrigation method (with or without ultrasonic activation) ( $p < 0.05$ ). In addition, the use of ultrasonic activation did not significantly influence smear layer removal at the apical third when the same solution was used. In this study Oxum, showed smear layer removal significantly higher than saline, and similar to EDTA. According to this study, even though no significant differences were identified in the apical region of the different groups, with and without ultrasonic activation of irrigants, the smear layer removal scores were higher in the conventional group than ultrasonic activated group.

COMPARISON OF DIFFERENT IRRIGANTS



## Discussion

Successful endodontic treatment depends upon thorough cleaning, shaping, disinfecting and obturation of root canals so as to achieve three-dimensional hermetic sealing of the pulp spaces. During mechanical preparation or whenever dentin is cut using hand or rotary instruments, the mineralized tissues are scattered producing considerable amount of debris called “smear layer”.

American Association of Endodontists defined smear layer as a “surface film of debris retained on dentin or other surfaces after instrumentation with either rotary instruments or endodontic files; consists of dentin particles, remnants of vital or necrotic pulp tissue, bacterial components & retained irrigants” The thickness

of smear layer depends on the type and sharpness of cutting instrument and also whether the cutting was carried out on dry or wet dentin. The complexity of root canal system usually limits the efficacy of thorough cleaning the pulp spaces. The deviated root canal anatomy may pose difficulties in instrumentation, subsequently leading to unevenly prepared zones on root canal walls. These uneven zones may contribute to produce more of smear layer. When dentin chips are accumulated in flutes of the instrument, working effect is impaired and friction between instrument and canal walls is increased. The cutting effect is decreased; consequently larger amount of smear layer is formed. It has been established that the amount of smear layer produced during rotary instrumentation is far greater as compared to hand filling. It is established that sonic and ultrasonic preparation of root canal are the most effective methods leading to only minor formation of smear layer. Massive irrigation directed towards working part on the instrument facilitates removal of dentin and prevents debris binding on the root canal walls. Vent needles enable detachment of smear layer from root canal walls under pressure thus making debridement more efficient.<sup>7</sup>

White et al. (1987) found that PHEMA, silicone and Roth 801 and AH26 sealers extended into tubules consistently when smear layer was removed.<sup>8</sup> Genc,og˘lu et al. found removing the smear layer enhanced the adaptation of guttapercha in both cold laterally compacted and thermoplastic root fillings without sealer.<sup>9</sup> Gutmann also showed that after removing the smear layer, thermoplastic gutta-percha adapted with or without sealer.<sup>10</sup>

Root canal irrigants plays an important role in removal of smear layer partially or completely.<sup>11</sup> Most commonly used irrigating solutions are EDTA (ethylenediaminetetraacetic), Sodium Hypochlorite(0.5-

6%) , citric acid , MTAD , Etridonic acid , chlorhexidine , saline , distilled water etc.

NaOCl is a widely used irrigant in endodontic treatment because of its solvent action on organic pulp tissue and its bactericidal and cleansing properties. It does not, however, remove the smear layer produced during instrumentation of the root canal<sup>12</sup>

Neutral ethylenediaminetetraacetic acid (EDTA) solutions, in a 15–17% concentration, are effective in demineralizing the dentine , and can be used to remove the smear layer. EDTA which reacts with the calcium ions in dentine and forms soluble calcium chelates. Nygaard-O'stby reported that EDTA decalcified dentine to a depth of 20–30  $\mu$ m in 5 min ;<sup>13</sup> however, Fraser stated that the chelating effect was almost negligible in the apical third of root canals. However, as it does not dissolve organic matter, EDTA has been used with sodium hypochlorite (NaOCl) solution which in addition to acting on pulp tissue remnants it has antimicrobial properties.<sup>14</sup>

Super-oxidized water is a powerful anti-microbial agent against bacteria, fungi, protozoa and viruses. It is rich in reactive oxygen with a neutral pH and studies show its ability to remove the smear layer. Super-oxidised water, commercially available as Oxum, is stable & has longer shelf life.<sup>15</sup>

In this study smear layer removing ability of different irrigating solutions were evaluated at the apical third of the root canal. According to Takeda et al, the apical third of the root canal is the most difficult portion to clean possibly because of its narrower dimensions, which can prevent effective penetration of irrigants, resulting in limited contact of solutions with root canal surfaces<sup>16</sup>. Mechanical debridement efficacy of an irrigation system depends on its ability to deliver the irrigating solution to the apical and non-instrumented areas of the root canal space and flush out debris away from the canal walls.<sup>17</sup>

During the conventional needle irrigation, replenishment and fluid exchange do not extend much beyond the tip of the irrigating needle.<sup>18</sup> The trapped air in the apical third of root canals causing a vapour lock effect may obstruct the exchange of irrigants and hamper the debridement effectiveness of irrigants.<sup>19</sup>. Ahmad et al<sup>20</sup> reported that the use of ultrasonics has been suggested to improve irrigation in the root canal. Less debris and smear layer have been observed in the apical region of the canal than its coronal aspects with the use of ultrasonics; this effect is thought to be generated by acoustic streaming. Thus EMS Ultrasonic irrigation system was used in this present study to allow the flow of irrigants in the apical third of the root canals. In this study 4 ml of each irrigating solutions are irrigated for about 2 minutes under with ultrasonic irrigation. This is in accordance with Bhuva et al<sup>21</sup> which states that an irrigation time of 2 min shown to eradicate the biofilm reliably within this time period along with the removal of smear layer.

All irrigation protocols were done using 30 gauge needle (close-ended single side vented) as it allows the clinician to place the needle as apical as clinically possible without canal binding amongst all the endodontic needle gauges according to Gopikrishna et al.<sup>22</sup>

Cunningham et al investigated a continuous flow of NaOCl activated by an ultrasonic delivery system was used for the preparation and irrigation of canals.<sup>23</sup> Cameron et al found smear-free canal surfaces while using this method.<sup>24</sup>

In this study, a comparison of the efficacy of smear layer removal by 17% EDTA , 5.25% and a commercially available super-oxidized water (Oxum) as a final irrigant was done. Oxum, is a solution with less dentinal erosion and more antimicrobial activity used as one of the irrigants, showed smear layer removal significantly higher than saline, and similar to EDTA .According to this study,

even though no significant differences were identified in the apical region of the different groups, with and without ultrasonic activation of irrigants, the smear layer removal scores were higher in the conventional group than ultrasonic activated group.

Vasiliadis et al., reported that dentin in the apical third is sclerosed and that EDTA may not have such a pronounced effect on the apical third as compared to middle or coronal third of the dentine.<sup>25</sup> Michael O Connell et al., compared EDTA of various concentrations and pH and concluded that at high pH, excess number of hydroxyl ion prevented the dissolution of hydroxyapatite crystals thus limiting the number of calcium ions for chelation. Thus at neutral or low pH, the calcium ions from dentine becomes more readily available for chelation due to dissociation of hydroxyapatite crystals<sup>26</sup>. Hulsman et al. proposed that the ideal concentration of EDTA was from 15–17% with neutral or low pH<sup>27</sup>. At neutral pH, EDTA showed lesser degree of decalcification in the apical third of root dentine because the content of non-collagenous proteins decreases in the apical third<sup>28</sup>

Super-oxidized water is a powerful anti-microbial agent against bacteria, fungi, protozoa and viruses. It is rich in reactive oxygen with a neutral pH. The main advantage of this super-oxidized water is that it is stable and has a longer shelf life. It mainly contains oxidized solution (H<sub>2</sub>O), sodium hypochlorite, hypochlorous acid, hydrogen peroxide, ozone, chlorine dioxide, sodium hydroxide, sodium carbonate and sodium chloride. The molecules are broken into ions and free radicals, which rapidly react and denature protein of bacterial cell wall. It produces an environment of unbalanced osmolarity that damages the cell wall of single cell organisms. The low pH in oxum may sensitize the outer membrane of bacterial cell, thereby enabling oxygen anion radicals to attack the bacterial cell more efficiently<sup>29</sup>. The damage is due to the

difference in osmolarity between the concentrations of ions in solution versus the concentration of same ions in the cell<sup>30</sup>. Multicellular organisms are not prone to such changes so host tissues are spared. For these reasons it is referred to as a well suited alternative irrigating agent. Based on the present study, it has shown that oxum when used as an irrigant, cleans the root canal surfaces in a clinically significant manner and removed the smear layer in large areas leaving the collagen fibers intact and completely exposed with less erosion.

### Conclusion

Within the limitation of the present study, oxum the commercially available super-oxidized water proved to be significantly equal in smear layer removal with less significant erosion when compared to EDTA. However it may be worthwhile to investigate further, the effect of oxum alone as a root canal irrigant to evaluate its effect on smear layer and on dentine.

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**Legend Figure**

**Sem Images of Saline, Oxum, EDTA When Used As Irrigants with and Without Ultrasonic Irrigation**

