



# Smear Layer Removal Efficacy of EDTA and Citric Acid in Endodontics

Khan A<sup>1\*</sup>, Ahmed R<sup>2</sup>

## AFFILIATION:

- Dr. Atiquzzaman Khan**   
MS in Conservative Dentistry & Endodontics  
Associate Professor & Head,  
Dept. of Conservative Dentistry & Endodontics,  
University Dental College & Hospital, Dhaka, Bangladesh
- Dr. Rubaba Ahmed**   
FCPS in Prosthodontics  
Associate Professor,  
Dept. of Science of Dental Materials,  
University Dental College & Hospital, Dhaka, Bangladesh

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## \* Corresponding Author

**Dr. Atiquzzaman Khan**

MS in Conservative Dentistry & Endodontics

Associate Professor & Head,

Dept. of Conservative Dentistry & Endodontics,

University Dental College & Hospital, Dhaka, Bangladesh

Phone: +88-01716643893

Email: [dratiquzzamankhan@yahoo.com](mailto:dratiquzzamankhan@yahoo.com)

<https://orcid.org/0009-0004-0180-3427>

## ABSTRACT:

**Introduction:** Smear layer is formed by organic and inorganic components generated during endodontic instrumentation. Smear layer has many deleterious effects on the success of root canal treatment. So removal of smear layer is advocated. Sodium Hypochlorite (NaOCl) and a chelating agent like Ethylene Diamine Tetra Acetic Acid (EDTA) or Citric Acid is commonly used alternatively to remove smear layer. But the alternate use increase dentine erosion in root canal surface. This in vitro study attempts to compare the smear layer removal efficacy of 15% EDTA & 10% Citric Acid as a final rinsing solution. **Materials and Methods:** Forty extracted, single rooted and single canal human anterior tooth were used. Endodontic preparation of the root canal was done after grouping and access cavity preparation. Final irrigation was given with 15% EDTA & 10% Citric Acid to the test groups and distilled water control groups and kept for one minute. Photomicrograph of the root canal surface at 3000X magnification at coronal, middle and apical regions were taken with the help of SEM and scoring for smear layer removal was done. Data was analyzed using SPSS version 16 and were compared with the control samples and subjected to statistical analysis by one way ANOVA and Bonferroni multiple comparison tests at 5% level of significance. **Result:** The test solutions removed smear layer but none could be removed completely from all of the three root canal regions. 10% Citric Acid removed smear layer better from the coronal and middle region but in apical region 15% EDTA produced a better result. The difference of smear layer removal efficacy of 15% EDTA & 10% Citric Acid with the Control solution was found statistically significant ( $p < 0.001$ ) but in between 15% EDTA & 10% Citric Acid it was not significant in all three root canal regions. **Conclusion:** Regarding the smear layer removal efficacy of 10% Citric Acid was found better as a final rinse solution.

**KEYWORDS:** Smear layer, EDTA, Sodium Hypochlorite (NaOCl), Scanning Electron Microscope (SEM)

## INTRODUCTION:

Smear layer is one of the most disputable issues in endodontics nowadays. The term "Smear Layer" refers to a morphologically discrete layer of 'debris' produced iatrogenically following instrument manipulation of enamel, dentine, cementum and even calculus<sup>1</sup>. McComb and Smith suggested that the smear layer associated with the root canal treatment consisted of not only dentine as in coronal smear layer, but also remnants of odontoblastic processes, pulp tissue and bacteria.<sup>2</sup> Hence, it may contain organic and inorganic material. The morphology and composition of smear layer was fully determined only after the invention of Scanning Electron Microscope (SEM). When viewed under SEM the smear layer has an amorphous, irregular and granular appearance. This appearance may be formed by translocating and burnishing the superficial components of the dentine walls during endodontic instrumentation<sup>2</sup>. In course of time, smear layer has become the subject of many controversies and discussions for the question whether smear layer should be removed or not from the canal walls. Intact smear layer that was formed after instrumentation can act in two ways. It can prevent bacteria and /or fluids from penetrating into dentinal tubules following endodontic treatment, and at the same time stop microorganisms, which have already leaked into the tubules, to return back in canal and periapical tissues if obturation was incorrect<sup>3</sup>.

Numerous experimental and clinical studies have confirmed that smear layer and occlusion of dentinal tubules significantly decreasing the effects of intracanal medicaments in endodontic therapy<sup>3-5</sup>. Another significant clinical implication of smear layer on root canal walls is related to obturation. Researchers have shown that adhesion

and bonding strength of obturation materials were highly improved after smear layer had been removed<sup>3,5</sup>. The main argument for the greater number of scientists recommending removal of the smear layer is the fact that this layer obturates dentinal tubules in root canal and effects of canal medication are blocked, as well as the efficacy of disinfecting agents during endodontic treatment<sup>3,6</sup>. Another important consideration in endodontics is the ultimate seal of root canals in order to prevent possible micro leakage which may be the cause of the future failure of the root filling. The prepared dentine surfaces should be very clean to increase sealing efficiency of obturation. Smear layer on root canal walls acts as an intermediate physical barrier and may interfere with adhesion and penetration of sealers into dentinal tubules<sup>2</sup>. Despite controversies on the impact that smear layer may have on quality of instrumentation, medication and obturation and the fact that it may contain microorganisms, it is reasonable and justified to recommend its removal. For removal of smear layer chemical agents like EDTA, NaOCl, citric acid, polyacrylic acid, antibiotics; ultrasonics and laser techniques have been attempted<sup>3</sup>. The rationality for using the above-mentioned chemicals lies in the fact that studying the smear layer by electron microscopy, it has shown that it contains both organic and inorganic substances<sup>7</sup>.

NaOCl is the most widely accepted and regular root canal irrigant. It has wide spectrum, non specific killing efficacy on all microbes. It has sporicidal and virucidal and greater tissue dissolving effects. Further more NaOCl is cheap, easily available and demonstrate good shelf life. But NaOCl alone cannot remove smear layer completely, because smear layer consists of both organic and inorganic components and NaOCl can remove only the organic part. So scientists have decided that in addition to NaOCl some chelating agents must be used, which will help to remove the inorganic component.

EDTA is widely used as a chelator in endodontic therapy. For effective removal of both organic and inorganic components of the smear layer, it is generally recommended to use EDTA followed by NaOCl. Baumgartner and Mader(1987) reported that the combination of EDTA and NaOCl caused a progressive dissolution of dentine at the expense of peritubular and intertubular areas, and they suggested that this effect may have resulted from the alternating action of NaOCl, which dissolved the organic component of the dentine, and EDTA which demineralized the inorganic component.<sup>8</sup> In a study conducted by Calt and Serper (2002) compared the smear layer removal capability and the structural effects of EDTA on root dentine with respect to duration of application and concluded that excessive erosive effects were observed with increased time of exposure and recommended to keep the application time as short as possible<sup>9</sup>.

The citric acid is recommended recently for the root canal irrigation, in the aim of the smear layer removal. The citric acid is weak organic acid, which, the same as EDTA, belongs to the chelate agents. Citric acid is highly biocompatible. Like EDTA, this demineralizing agent has been recommended as an adjuvant in root canal therapy. Citric acid has also been recommended for use as an endodontic irrigant because of its low pH, which causes dentine dissolution and thereby produced similar results to EDTA. Moreover, when used in concentrations of 10%, 25%, and 50%, citric acid has been shown to remove the smear layer associated with instrumentation of the canal system<sup>10</sup>.

Most of the studied for smear layer removal suggested that alternate use of at least two irrigation solution, most commonly these are

NaOCl and EDTA or NaOCl and citric acid. NaOCl removes the organic components and the EDTA or citric acid removes the inorganic components. But as NaOCl is very commonly used irrigating solution during root canal preparation. The complementary effect of two irrigating solutions increases the erosion of the dentine. No study compared the smear layer removal potential of EDTA and Citric acid as a final rinse. That is why this study was designed to evaluate the smear layer removal capacity of EDTA and Citric Acid when used as final rinse solution.

#### MATERIALS AND METHODS:

Freshly extracted completely formed single rooted human maxillary and mandibular anterior teeth with single root canal which were extracted after proper diagnosis for either orthodontic purposes or due to loss of periodontium (excessive mobility) were collected for this study. Teeth with resorptive defects, broken down crown and developmental defects like excessive curve root or dilacerated tooth were excluded. The sample size of this study was forty. The selected teeth were carefully cleaned to eliminate calculus, stain and remaining tissue with the help of ultrasonic and hand scaler. Then the specimens were immersed in distilled water. The specimens were randomly divided into three groups. Group A was the control group consisting of ten teeth. The canals would be irrigated with 5.25% NaOCl and normal saline alternatively after each instrumentation during preparation and final irrigation would be given with distilled water. Group-B consisted of 15 teeth, the canals would be irrigated with NaOCl 5.25% and normal saline alternatively after each instrumentation during preparation and final irrigation would be given with 15% EDTA. Group-C consisted of 15 teeth, the canals would be irrigated with NaOCl 5.25% and normal saline alternatively after each instrumentation during preparation and final irrigation would be given with 10% Citric acid solution.

Coronal access cavity was prepared using a round diamond bur in high speed contra-angle handpiece with water spray coolant system. The canal orifice was enlarged with diamond fissure bur. A size 10 K-type root canal file was introduced into each root canal to establish patency. For determination of working length a size 10 K-type file was inserted in the canal until its tip just appeared through the apical foramen. Now the silicon rubber stop pre fitted to the shaft of the file was adjusted to a coronal reference point, the file was withdrawn and the length from file tip to silicon stop was measured. Finally individual working length for each tooth was calculated by subtracting 1mm from the measured length.

The root canal preparation was performed by step-back technique using K-files. The files were used sequentially to clean and shape each canal until a size #40 K-file at the working length. During preparation of the canals the canals were irrigated with NaOCl 5.25% and normal saline alternatively for all groups after each instrumentation through out the whole root canal preparation. The canals were then step back up to #60 K-file at one millimeter for each number of instruments and the coronal part was flared by gates glidden bur #2 and #3. During step back procedure, after each instrumentation, the master apical file was placed up to the working length to maintain the patency and to prevent blocking of the canal by collection of dentine chips and debris.

The final irrigation was done by leaving 1 ml of the tested solution in the canal for 1 minute, after which the canals were flushed with 4 ml of the same solution. Then the each sample was irrigated with 10 ml

of distilled water and then dried with absorbent paper points. The irrigation solution was delivered with a hypodermic syringe and the needle was placed as apically as possible without binding. At the time of irrigation each specimen were embedded in a putty silicone material to simulate the hydraulic counter-pressure produced physiologically by periapical tissues.

After irrigation and drying the root canal orifices were blocked with small piece of cotton. Longitudinal grooves were cut on the buccal and lingual surfaces with the aid of a slow speed hand piece and diamond coated disc without penetrating into the root canal space. The root was then splitted longitudinally into two with cutting pliers. After splitting of the samples the one halves of the roots were examined in the Scanning Electron Microscope (Hitachi S 3400N, Japan, Scanning Electron Microscope). Photo micrographs were taken at the coronal middle and apical third of the root canal of each sample at a magnification of 3000x.

The images were then analyzed for the amount of smear layer and degree of erosion of dentinal wall as per predetermined evaluation criteria. Qualitative analysis of canal cleanliness was based on the following rating system developed by Rome et al.<sup>11</sup>

- 0-No smear layer with opened tubules free of debris
- 1-Smear layer present only in the apertures of the dentinal tubules (minimum smear layer)
- 2-Thin smear layer covering the root canal surface and dentinal tubular apertures (moderate smear layer).
- 3-Heavy smear layer masking dentinal tubular apertures

**Photo micrographs of tooth samples of three groups:**

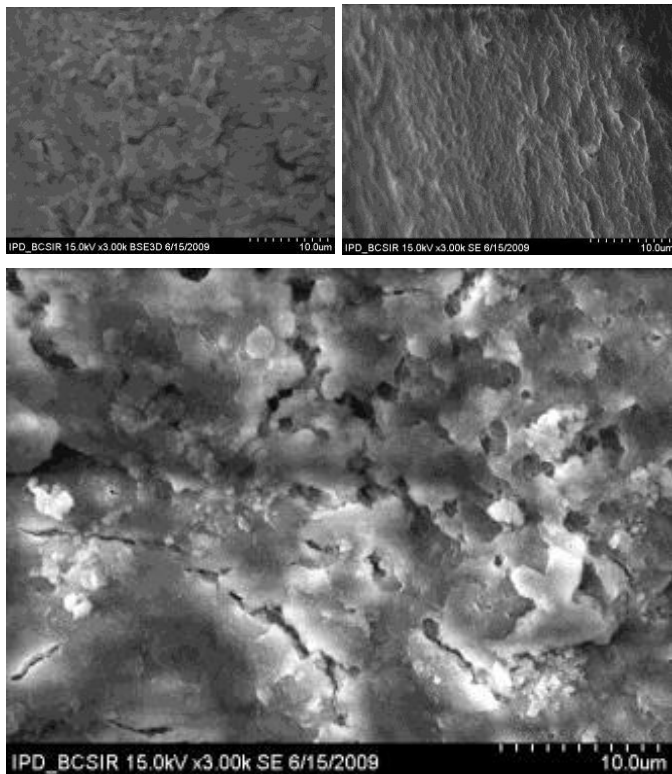


Figure No: 1: Group-A (Distilled Water): Coronal then middle (left to right Upper photo micrograph) and apical (below)

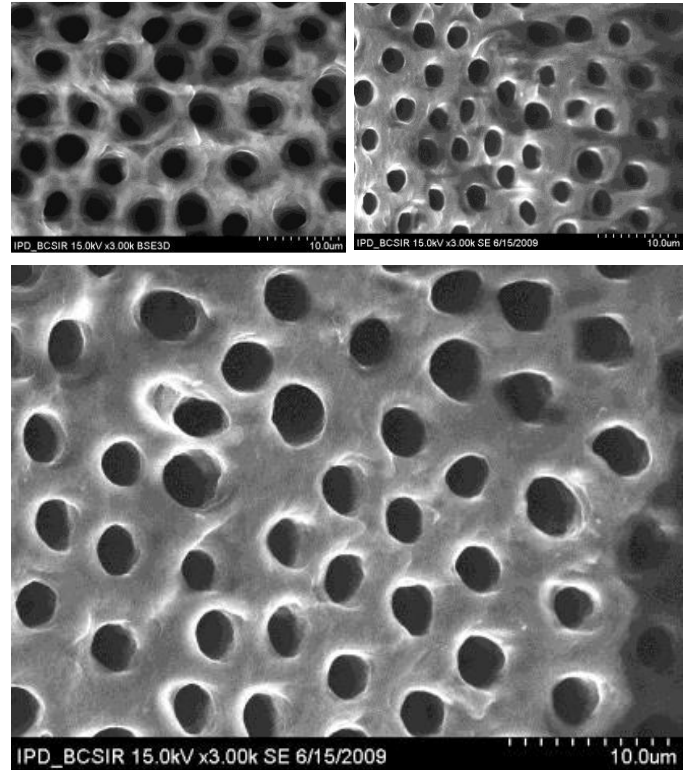


Figure No: 2: Group- B (15% EDTA): Coronal then middle (left to right Upper photo micrograph) and apical (below)

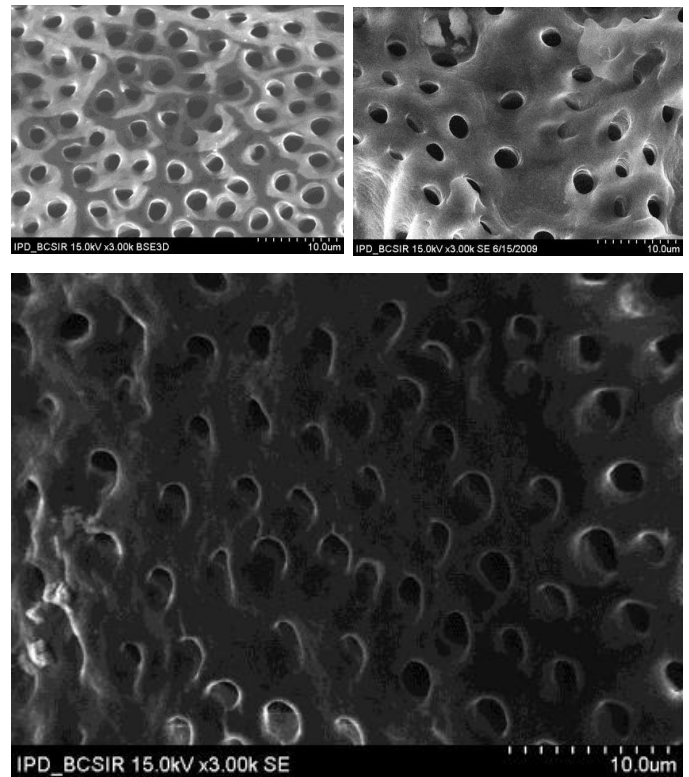


Figure No: 2: Group- C (10% Citric Acid): Coronal then middle (left to right Upper photo micrograph) and apical (below)

**RESULTS:**

Smear layer was removed in varying degrees ranging from complete removal from the root canal surface with clear visibility of most of the dentinal tubular openings to partial removal with smear layer removed from the root canal surface irregularly with blocked tubular openings or the root canal surface completely covered with smear layer with no tubular opening visible in three thirds (viz. coronal, middle and apical) by both the test solutions, 15% solution of EDTA and 10% solution of Citric Acid.

The walls of the root canal in control samples in all of the three thirds (viz. coronal, middle and apical) were covered with large quantity of smear layer, which blocked the openings of dentinal tubules and they produced the highest score (3) for smear layer on the root canal surface.

So it appears that none of the two test solutions used for final irrigation was capable to remove smear layer completely from all three levels of root canal surface.

**Table I: Mean distribution of scores for smear layer removal of different region of three groups of tooth samples.**

Regions	Group –A (Distilled water) (mean±SD)	Group –B (15%EDTA) (mean±SD)	Group –C (10%Citric acid) (mean±SD)	P value
Coronal	3.00±0.00	0.27±0.458	0.00±0.00	0.001 <sup>s</sup>
Middle	3.00±0.00	1.00±0.925	0.47±0.516	0.001 <sup>s</sup>
Apical	3.00±0.00	0.87±0.834	1.60±0.507	0.001 <sup>s</sup>

s= significant Group A = Distilled water, Group B = 15%EDTA, Group C = 10% Citric acid

For removal of smear layer 10% solution of Citric acid produced more effective result compared to 15% EDTA solution in coronal and middle region. However in apical region 15% EDTA removed smear layer more effectively than 10% Citric acid solution. Significant mean coronal, middle and apical area differences were found among three sample specimen groups in ANOVA test.

**Table II: Comparison of scores for smear layer removal of coronal area among three groups:**

Region	Groups	Mean difference	P value
Coronal area	Group A Vs Group B	2.73	0.001 <sup>s</sup>
	Group A Vs Group C	3.00	0.001 <sup>s</sup>
	Group B Vs Group C	0.26	0.065 <sup>ns</sup>

**Table III: Comparison of scores for smear layer removal of middle area among three groups**

Region	Groups	Mean Difference	P value
Middle area	Group A Vs Group B	2.00	0.001 <sup>s</sup>
	Group A Vs Group C	2.53	0.001 <sup>s</sup>
	Group B Vs Group C	0.53	0.136 <sup>ns</sup>

**Table IV: Comparison of scores for smear layer removal of apical area among three groups**

Region	Groups	Mean difference	P value
Apical area	Group A Vs Group B	2.13	0.001 <sup>s</sup>
	Group A Vs Group C	1.40	0.001 <sup>s</sup>
	Group B Vs Group C	-0.73	0.012 <sup>s</sup>

Multiple Comparison (Bonferroni 't') test was done as the test of significance

s= significant, ns = not significant, Group A = Distilled water, Group C = 15%EDTA, Group D = 10% Citric acid

Coronal areas were found significant ( $p < 0.001$ ) differences between group A with group B and group A with group C, however almost similar between group B with group C. Middle areas were found significant ( $p < 0.001$ ) difference between group A with group B and group A with group C, however almost similar between group B with group C. Apical areas were found significant ( $p < 0.001$ ) differences in all three sample groups.

**Table V: Comparison of scores for smear layer removal among coronal, middle and apical regions in two groups of tooth samples.**

Groups	Regions			P value
	Coronal (mean±SD)	Middle (mean±SD)	Apical (mean±SD)	
Group B	0.27±0.458	1.00±0.925	0.87±0.834	0.001 <sup>s</sup>
Group C	0.00±0.00	0.47±0.516	1.60±0.507	0.001 <sup>s</sup>

s= Significant

Group B = 15%EDTA solution

Group C = 10% Citric solution

Comparing the individual effect of 15% EDTA and 10% Citric acid solution for smear layer removal in coronal, middle and apical region were found significantly different in ANOVA test.

**Table VI: Comparison of scores for smear layer removal efficacy of EDTA (15%) solution among three areas**

Group	Regions	Mean Difference	P value
Group B	Coronal Vs Middle	-0.73	0.037 <sup>s</sup>
	Coronal Vs Apical	-0.60	0.114 <sup>ns</sup>
	Middle Vs Apical	0.13	1.000 <sup>ns</sup>

**Table VII: Comparison of scores for smear layer removal efficacy of Citric acid (10%) solution among three areas**

Group	Regions	Mean difference	P value
Group C	Coronal Vs Middle	-0.46	0.012 <sup>s</sup>
	Coronal Vs Apical	-1.60	0.001 <sup>s</sup>
	Middle Vs Apical	-1.13	0.001 <sup>s</sup>

Multiple Comparison (Bonferroni 't') test was done as the test of significance

s= Significant, ns = not significant Group B = 15%EDTA solution, Group C = 10% Citric Acid solution

EDTA (15%) solution was found significant ( $p < 0.001$ ) difference between coronal and middle area, however, almost similar between coronal with apical and middle with apical.

Citric acid (10%) solution was found significant ( $p < 0.001$ ) difference in all three regions.

#### DISCUSSION:

This study evaluated the effectiveness of smear layer removal from the root canal surface by a final irrigation with 15% EDTA solution and 10% Citric acid solution. The result demonstrates that 10% solution of citric acid was found more effective than 15% solution of EDTA for removing smear layer from coronal and middle third of root canal surface whereas in apical third 15% EDTA was more effective.

Zivcovic (2005) found similar result in their study that, samples which were irrigated with 10% solution of citric acid a little bit better cleaning was noticed comparing to the samples irrigated with 17% solution of EDTA. They concluded that the 10% solution of citric acid is efficient in smear layer removal from the root canal walls and in complete cleaning of the canal system and that it can be used as a final irrigant during the endodontic treatment<sup>12</sup>.

Similarly the results of Takeda and associates also showed the superior cleaning of the root canal with final irrigation using 6% citric acid solution comparing to 17% EDTA solution. The authors also pointed out that, although it was very effective in the middle third of the root canal, citric acid did not remove the smear layer in the apical third of the root canal<sup>13</sup>.

Di Lenarda and associates (2000) also evaluated the efficiency of EDTA and citric acid in removal of the smear layer from the walls of the root canal, but in the function of different techniques of instrumentation. Their results showed the similar efficiency of the 19% solution of citric acid ( $1 \text{ mol L}^{-1}$ ) and 15% solution EDTA. The authors also emphasized that in the group of mechanically instrumented samples, which had been done in a shorter time interval, citric acid was more efficient. On the basis of results achieved the authors summarized that the efficiency of the tested means depended on duration of application and that the canal cleaning could be realized quicker by citric acid implementation<sup>14</sup>.

Considering the cleaning efficacy of citric acid in the present study we found that it produced the most cleaning result in the coronal region and least at the apical, which was consistent with some of the previous results. Khademi et al. 2004 found in the similar way that 7% citric acid cleaned the cervical and middle region better than apical region. On the other hand Khademi et al. 2004 in the same study compared the effect of EDTA and citric acid for smear layer removal in the mesial canals of mandibular first molar and found that 17% EDTA was more effective than 7% citric acid. They also noted that the superiority of EDTA over citric acid was specially observed in middle and apical regions which have similarity with the present study for the apical region<sup>15</sup>. Yamada et al. reported that 17% EDTA is more effective than 25% citric acid and they found the apical region as the cleanest region which is very much similar with the present study, where hand instrumentation and gates glidden drilling were used in canal preparation<sup>15</sup>. Scelza et al. reported no difference between EDTA-T (a combination of 17% EDTA and Tergentol) and 10% citric acid. The study was done on straight single rooted teeth and the canals were prepared only by hand instrumentation<sup>15</sup>.

For evaluation of the effect of EDTA, it was observed that it produced highest smear layer removing effect in the apical region then coronal

and least effect in the middle third. This result differs from many of the previous studies. Ciucchi, B and associates in 1989 used 15% as a final irrigation and found least effect of removing smear layer in apical region comparing with coronal and middle third. They explained this decline of efficiency along the apical part could be attributed, to limited distribution of the irrigant, the limited size and pronounced curvature of the canal being obstacles to the optimal apical flooding of the irrigation solution. Specially because they have taken only the moderately curved canals on the basis of the Schneider classification. (i.e. the mesial roots of molars)<sup>16</sup>. Similar result was also found by Prabhu S. G. et al. 2003. They summarized that EDTA is efficient in removing smear layer mainly from the middle third; its action in apical third is very much impaired<sup>17</sup>. Teixeira et al. 2005 found notably clean canal surface in the cervical and middle third than the apical third<sup>18</sup>. On the other hand, Garberoglio and Becce 1998 using EDTA for 30 sec. reported good cleaning of the apical third.<sup>19</sup> Lopes et al. 1996 upon irrigating the canals for 5 minutes reported that the mechanical stirring of EDTA for 2min using a Lentulo spiral allowed for the near complete removal of the dentinal smear layer from the apical third.<sup>20</sup> The authors explained that, on account of the reduced dimension of the root canal, air bubbles frequently remain trapped and prevent total filling with the irrigant. Mechanical stirring with a Lentulo spiral removes the air bubbles, favors improved contact of EDTA with the canal walls, and takes the solution to areas that are not reached by the irrigating needle<sup>18</sup>.

In the present study the possible reasons for maximum cleaning of smear layer in apical third by EDTA may be due to only the straight rooted single canal anterior teeth were included in this study and they were enlarged up to file #40 up to the apex which were large enough to receive the needle tip of the hypodermic syringe used for irrigation, the irrigation was applied by placing the needle as much apical as it could be with out binding. So it was the apical portion of the canal that received the irrigation first then the middle and coronal portion. Finally the tip of the needle pushed the irrigation in a forward flow motion in to the apical part of the canal. Moreover the flow dynamics and wettability of EDTA and Citric acid may be different which could play a role for the difference in the apical region. These two factors should be considered in the further studies.

#### CONCLUSION:

Within the observation of the present study it can be concluded that both 15% EDTA and 10% Citric Acid removed smear layer but none of them did it completely. Comparing the efficacy in three root canal regions 10% Citric acid removed smear layer from coronal part completely and in middle region the efficacy of 10% Citric Acid was better than 15% EDTA but in apical region 15% EDTA showed better result. So we can conclude that regarding the smear layer removal efficacy, 10% Citric acid was found better as a final rinse solution.

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**DATA AVAILABILITY STATEMENT:** The data presented in this study are available on reasonable request from the corresponding author.

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