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ORIGINAL ARTICLE

# Effect of Glycolic Acid, Maleic Acid, and EDTA in the Removal of Smear Layer from Root Canal Dentin

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**ABSTRACT**

**Objective:** To assess the efficacy of 5% Glycolic Acid (GA), 17% ethylenediaminetetraacetic acid (EDTA) and 7% maleic acid (MA), in removing the smear layer (SL). **Material and Methods:** For the experiment, forty single-rooted human teeth were selected. To perform the chemo-mechanical preparation, the root canals were instrumented to an apical size of #30, along with simultaneous irrigation of 2.5% NaOCl. The samples were allotted to the experimental groups based on the final irrigating solution (n=10): (1) The GA group: 05%, (2) the EDTA group: 17%, (3) the MA group: 7%, and (4) the control group: Distilled water. The teeth were evaluated for the presence or absence of SL using SEM. **Results:** Comparing the 5% GA, 7% MA and 17% EDTA groups, no statistically significant differences were found at the coronal and middle thirds ($p>0.05$). However, in the apical third, MA had greater efficacy than EDTA ($p=0.002$) and GA ($p=0.041$), with a significant difference in the SL removal, while there was no significant difference between the latter ($p=0.148$). **Conclusion:** Thus, it was concluded that 7% maleic acid as a final irrigating solution is more efficacious than 17% EDTA and 5% glycolic acid in eliminating the smear layer from the apical portion of the root canal.

**Keywords:** Endodontics; Root Canal Therapy; Smear Layer; Edetic Acid.

## Introduction

Endodontic treatment through instrumentation seeks to achieve the objective of mechanically debriding the root canal and facilitating the delivery of disinfecting solutions and, thereby, their action. However, endodontic files, while instrumenting the canal, produce debris comprising of dentin remnants, organic tissue, and bacteria, called the smear layer (SL) [1,2]. This layer interferes with the infiltration of irrigants and medicaments into the tubular structure of dentin. Thereby restricting their effects on bacteria deep-seated within these tubules [3]. It can also resist the bonding of root canal sealers to the dentin walls, increasing the potential of microleakage [4,5].

Ethylenediaminetetraacetic acid (EDTA) and sodium hypochlorite (NaOCl) in combination are often used in irrigation regimens to eradicate the SL [6]. However, EDTA, while an effective chelator, has a downside as its cytotoxic, ineffective in the critical apical third in removing the SL, hinders the bond between resin sealer and root canal dentin and results in the reduction of freely available chlorine ions when used alongside NaOCl [7-9].

Maleic Acid (MA), an organic acid in concentrations of seven percent, has been introduced as an alternative irrigating solution to remove the SL, and was found to be more effective in the apical third as opposed to EDTA [10,11]. Relative to EDTA, it was found to be less cytotoxic and enhanced the bond of resin-based sealer to root dentin [8].

Among the group of alpha hydroxy acids like citric acid and MA, Glycolic Acid (GA) is commonly extracted from sugarcane. It has been suggested as an alternative to EDTA owing to its biodegradability [12,13] and as an alternative etchant for enamel and dentin [14]. Compared to EDTA and Citric Acid, GA had greater antibacterial activity E. faecalis, comparable surface tension, less cytotoxic and comparable efficacy against the SL formed on the root canal [12,13,15,16].

To the extent of our knowledge, the smear layer removal efficacy of both GA and MA has been evaluated separately and has not been compared in the previous literature so far. Thus, this study aimed to assess the efficacy of 5% GA, 7% MA and 17% EDTA as a final irrigating solution on the SL removal following biomechanical preparation of the root canal system.

## Material and Methods

Sample Size Estimation

With a 95% confidence level and 80% power based on previously published literature [13], and standard deviation of 2 and a mean difference of 2.8, a minimum sample size of 10 per group was required.

Sample Preparation

Forty extracted human maxillary incisor teeth with completely developed roots and single, round canals, verified on a radiograph were selected for the study. Teeth showing resorption or filled canals were excluded. The institutional review board granted ethical clearance (IEC-704/2019) for the same. The teeth were kept in a storage solution of 0.2% sodium azide at 4°C until use (Millipore Sigma, St. Louis, MO, USA). The teeth were decoronated with a diamond disk (Horico Dental, Berlin, Germany) to reach a standard root length of 12mm.

The working length was determined using a #10 K file (Mani Inc- Tochigi Ken, Japan) inserted until seen at the apical foramen, observed under the 3.2x dental loupes. 1mm was decreased from this to achieve the desired working length. Sticky wax was used to seal the apices against extrusion. The canals were instrumented with a rotary file (Protaper Gold Dentsply, Maillefer, Ballaigues, Switzerland) up to size F3 using an endomotor (CanalPro 2; Coltène/Whaledent AG, Altstätten, Switzerland). Irrigation was performed intermittently between each instrument with 5ml of 2.5% NaOCl for 1 minute, using a 30 gauge side-vented needle (Vista Dental Inc. USA) placed 2mm short of the working length. Finally, to remove any remaining debris or formed residue, the canals were irrigated with 5ml of distilled water.

Irrigation Regimen

The samples were allocated into four experimental groups at random based on the final irrigating solution to be used (n=10):

- Group 1: 5% GA (Tokyo Chemical Industry Pvt Ltd, India);
- Group 2: 17% EDTA (Vista APEX, Racine, WI, USA);
- Group 3: 7% MA (Sigma-Aldrich Corp., St. Louis, MO, USA);
- Group 4: Distilled water (Control).

Irrigation was conducted with 5ml of the test solution for 1 minute with the irrigation needle placed 2mm short of the working length and a final rinse of 5ml distilled water for 1 minute. Paper points were used to obtain a dry canal (Dentsply Maillefer, Switzerland). On both the facial and lingual aspects, two longitudinal grooves were prepared with a diamond disc (Horico Dental, Germany), ensuring the preparation would not penetrate the canal. Using a chisel, the roots were split into halves. For each sample, the half with the more evident portion of the apical third was evaluated. Three external markings were made on this half with a fine-tip pen on the external root surface, perpendicular to the long axis, to divide it into cervical, middle, and apical thirds measuring 4mm in length from the root tip. The markings served as references to make 3 grooves in the canal wall, delimiting the root into thirds (cervical, middle, and apical).

Scanning Electron Microscopic Analysis

Increasing concentrations of ethyl alcohol (25%, 50%, 75%, and 100%) were used to dehydrate the specimens for 15 minutes. The specimens were finally mounted and sputtered using a gold ion sputter and evaluated through the Scanning Electron Microscope (SEM) (Carl Zeiss VO MA18 with Oxford EDS(X-act). Images obtained at a magnification of 1000x. The presence or absence of the SL was assessed at coronal, middle and apical thirds of the canal using the criteria given by Torabinejad et al. [17]:

1. No SL: No SL on the surface of the root canals; all tubules were clean and open.
2. Moderate SL: No SL on the surface of the root canal, but tubules contained debris.
3. Heavy SL: The SL covered the root canal surface and the tubules.

These were evaluated by two independent examiners unaware of the experimental groups to which the samples belonged.

Data Analysis

Chi-Square and Fisher Exact tests were used for the analysis of the data. A 5% level of statistical significance was adopted. The software SPSS was used for data analysis (IBM Corp., Armonk, NY, USA).

## Results

For the interpretation of scores, the inter-examiner agreement was evaluated by the coefficient of Kappa test. In the coronal third, the Kappa scores were 0.81, 0.49 and 0.36 for MA, GA and EDTA, respectively; in the middle third, it was 0.10, 0.62 and 0.16 and in the apical third, the scores were 0.07, 0.40 and 0.58. However, in certain areas, the agreement between examiners was inadequate, wherein a third independent observer scored the images.

The percentages of SL distribution after the use of the different irrigating agents amongst the different thirds of the canal are represented in Figure 1. In coronal and middle thirds, there were no statistically significant differences between GA, MA and EDTA (p>0.05). Though, there was a significant difference among the test solutions in the apical third, in which MA was significantly more effective in removing the SL when compared to EDTA (p=0.002) and GA (p=0.041). Between GA and EDTA, there was no statistically significant difference (p=0.148). In the control (distilled water) group, all the specimens had heavy SL in all three sections of the canal.

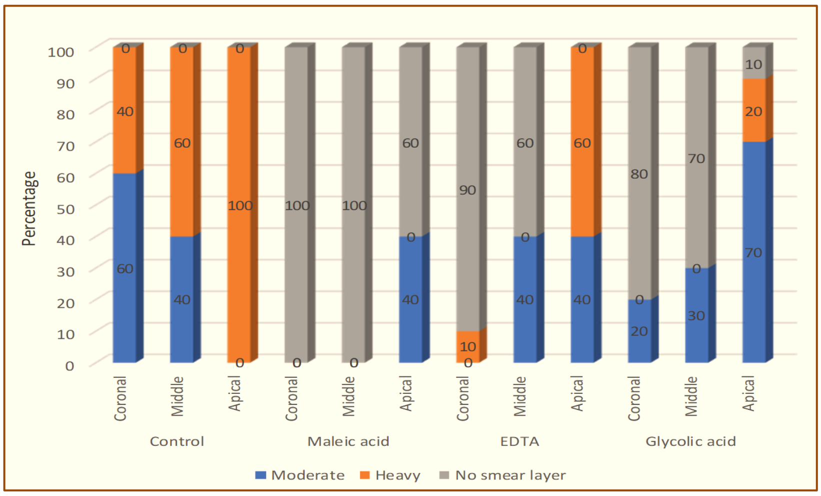


**Figure 1. Comparison of the percentage of smear layer removal among the test irrigants at coronal, middle, and apical thirds of the root canal system.**

Figure 2 represents the images obtained by SEM of the canal walls treated with the various experimental solutions.

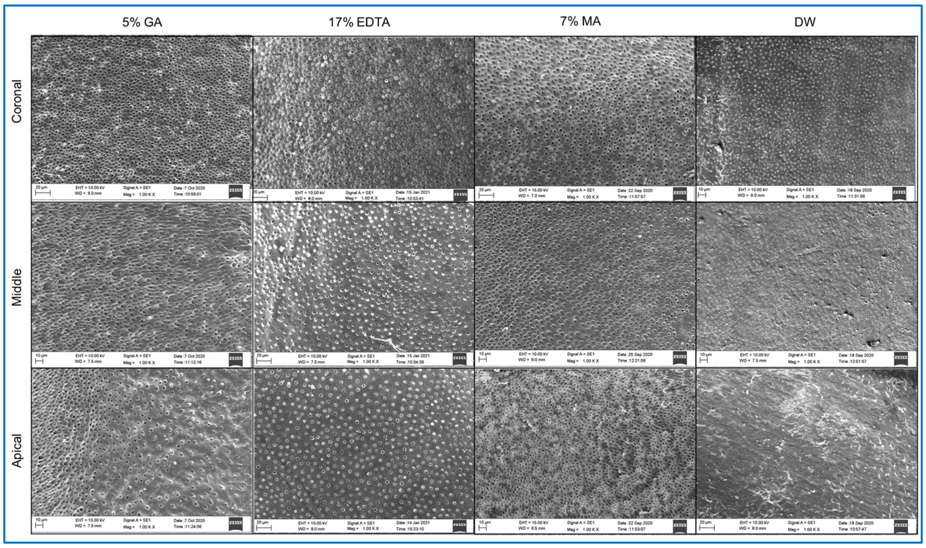


**Figure 2. Photomicrographs of root canal walls instrumented with the test irrigants in the coronal, middle and apical thirds.**

**Discussion**

The effectiveness of 17% EDTA, 7% MA and 5% GA as the final irrigating solution in eliminating the SL from the canal walls at the coronal, middle and apical thirds was assessed in this study. The outcomes showed that 7% MA was more effective in the removal of SL when contrasted with 17% EDTA and 5% GA in the apical third of the root canal. All the tested irrigating solutions were effective, with no significant differences between them, in the middle and coronal third. The finding is as per published literature that have revealed EDTA to be less successful in the apical third compared with the coronal and middle thirds for the removal of SL [10,17-19]. Since EDTA is a chelator, its capacity to decalcify the root canal dentin is free from the high concentration of hydrogen ions and is found to be effective at a pH that is neutral. The fall in the pH results from a trade by hydrogen ions for calcium from dentin. This decrease in pH over a period of time causes the ensuing reduction in the efficacy of EDTA [20]. It has been shown that the dentinal tubules progressively sclerose in the apical third [21,22]. Thus, the activity of EDTA may not be as effective in the apical third.

Better results of MA in the removal of SL from apical portion of the canal as opposed to EDTA is in concurrence with previously published literature [7,11]. This might be ascribed to the possibly greater depth of demineralization of MA in the apical third, caused due to its lower pH of 1.3, which results in a greater decalcifying effect in a shorter time [23]. Trevelin et al. [24] discovered that when MA was utilized at a similar concentration for etching dentin, the penetration was more than GA, notwithstanding the low molecular weight of GA. Since sclerosis occurs in the dentinal tubules in the apical section of the canal [21,22], the demineralization impact of MA might be more pronounced, yielding the outcomes found in the current study. Further investigations to confirm this mechanism are warranted.

The capability of GA as an irrigating solution was assessed utilizing various concentrations ranging from 5 to 17%, in which 5% was found as effective as higher concentrations in eliminating the SL and caused less decrease in the dentin microhardness [12]. Additionally, a recent study revealed that 5% GA had lesser cytotoxicity when compared with 17% EDTA [25]. Consequently, the concentration of 5% GA was chosen in this study. The volume of EDTA recommended to eliminate the SL goes from 3 to 20 ml for each canal [26,27]. In any case, the transport of a huge volume of the solution through a fine needle requires additional time and results in fatigue to the clinician. In this study, 5 ml of final irrigating solution was utilized. Mello et al. [28] showed that 5 ml was as effectual as 10 or 15 ml of EDTA in eliminating the SL as a final rinse.

In this study, 2.5% NaOCl was utilized for the chemo-mechanical arrangement between each instrument, which could eliminate the organic substance of SL. This is in accordance with a study that expressed that weakened NaOCl is just as effective as a greater concentration of NaOCl for the removal of the organic part of SL [17]. In the current experiment, the apex was prepared up to ISO size no. 30. Khademi et al. [26] found that the minimum size of canal instrumentation for the infiltration of irrigating solutions into the apical part of the canal is #30. They suggest it for viably eliminating the SL and debris while avoiding the dangers of iatrogenic errors, particularly in narrow and curved canals. Presently, there are no standard recommendations for the ideal duration of the use of EDTA. A 1-minute time interval; was decided on in the current experiment, similar to previously published investigations [27,29,30]. Furthermore, on exposing the dentinal tubules for more than 1 minute, EDTA causes erosion, accordingly diminishing the microhardness of dentin and causing fragility of the root [31,32]. Distilled water which is utilized as a control, was observed to be ineffective in eliminating the SL, with results like past studies [33].

Other than traditional SEM, different modalities that might be utilized to assess the presence of SL are digital image analysis, micro-computed tomography, atomic force microscopy, environmental SEM, and co-site optical microscopy [34]. However, in this study, SEM was selected as it is a conveniently accessible instrument. Results observed in this study might be bound to just teeth with single roots having a practically straight root canal configuration. Posterior teeth with curved canals and complex root canal anatomy can be more challenging for effective cleaning. Subsequently, further studies should be performed using these experimental irrigating solutions to judge their efficacy in the removal of SL in teeth where the canal anatomy is more complex.

## Conclusion

Within the constraints of this research, it can be concluded that 7% MA as a final irrigating solution is more effective than 17% EDTA and 5% GA in eliminating the SL in the apical portion of the human root canal.

### Authors' Contributions


| | | |
|---|---|---|
| TM | https://orcid.org/0009-0002-9866-8553 | Conceptualization, Formal Analysis, Investigation and Writing - Original Draft. |
| NVB | https://orcid.org/0000-0002-2208-9443 | Methodology, Writing - Review and Editing and Supervision. |

All authors declare that they contributed to critical review of intellectual content and approval of the final version to be published.


### Financial Support

None.

### Conflict of Interest

The authors declare no conflicts of interest.

### Data Availability

The data used to support the findings of this study can be made available upon request to the corresponding author.

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