RESEARCH ARTICLE

Effect of temperature and passive ultrasonic irrigation of EDTA 17% in smear layer removal

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ABSTRACT
Smear layer removal in root canal treatment is required to aid the diffusion of intracanal medicament and help sealer penetrate into dentinal tubules. The need for irrigants is important to facilitate smear layer removal in the root canal. Various methods have been proposed in some articles, but their application in apical third still challenges many clinicians due to the complexity of the root canal. This study aimed to analyze the effect of temperature and passive ultrasonic irrigation (PUI) activation of EDTA 17% solution in smear layer removal. Twenty-four extracted mandibular premolars were decoronated to standard root length of 14 mm. The preparation of root canals was completed with crown-down technique using ProTaper FHU to file F3 (30/.09). The root canal irrigation was done with NaOCl 5.25% and EDTA 17% solution. The samples were divided into four groups, namely 25 ºC of EDTA 17% solution (group 1), 37 ºC of EDTA 17% solution (group 2), 25 ºC of EDTA 17% solution with PUI activation (group 3), and 37 ºC of EDTA 17% solution with PUI activation (group 4). Smear layer removal was observed under scanning electron microscopy (SEM) and scored ordinally from 1-5. Non-parametric Friedman test showed significant results in all the groups (p<0.05). Post hoc Wilcoxon Signed Rank test showed significant results between group 2 and group 3 (p = 0.039), and between group 3 and group 4 (p = 0.038). The combination methods of temperature and PUI activation showed a significant result in smear layer removal.

Keywords: EDTA 17%; PUI activation; root canal irrigation; smear layer; temperature

INTRODUCTION
Root canal treatment is implemented when the pulp is non-vital, to create an aseptic condition in the root canal system.1 This condition aims to avoid the risk of reinfection.1 Root canal treatment is carried out with cavity preparation and guided with triad endodontic (preparation, irrigation, obturation).2,3 During preparation, the file utilized to prepare the root canal produces a smear layer.4 A smear layer is a surface film of debris retained on dentin, consisting of dentin particles, pulp tissues, and bacteria.3 The procedure of smear layer removal is importantly required to aid the diffusion of intracanal medicament and help sealer penetrate into dentinal tubules during obturation. With that being said, the need for irrigants that can provide smear layer removal is necessary.5

The combination of sodium hypochlorite (NaOCl) and ethylenediaminetetraacetic acid (EDTA) solutions is widely used as irrigants in the root canal system.5 The use of NaOCl solution has been approved due to its antibacterial activity, which is able to dissolve organic components. Meanwhile, EDTA solution has been appointed as the golden standard of chelating agent, by forming calcium chelator for smear layer removal.5,6 EDTA solution with a concentration of 17% is often used in dentistry practice for its better ability than EDTA solution with lower concentrations.7

Many factors contribute to EDTA solution effectiveness, among others, time, concentration, temperature, and method.8,9 Importantly, increasing temperature leads irrigants to flow easier in the root canal system owing to the reduction of viscosity.9 It is shown in research that 25 ºC and 37 ºC of EDTA solution remove smear layer better than 4 ºC of EDTA solution in the apical third.10 These temperatures are selected since 25 ºC is the average room temperature, 37 ºC is the average
body temperature, and 4 °C is the average refrigerator temperature.\textsuperscript{10}

The conventional method with syringe and needle is frequently used in root canal irrigation. However, this method does not distribute enough irrigants to reach all the aspects of the root canal system.\textsuperscript{11,12} This reason leads to the development of various irrigation methods, such as ultrasonic activator. Irrigation with activation using ultrasonic tip is based on premise that energy released by the tip enhances the ability of irrigants physically and chemically.\textsuperscript{13,14} Ultrasonic activator is divided into active ultrasonic irrigation (AUI) and passive ultrasonic irrigation (PUI), but it is written in many articles that the use of PUI is more effective than AUI.\textsuperscript{15} The use of AUI has been discharged due to the difficulty of controlling the cut of dentin.\textsuperscript{16} However, the term “passive” in PUI is related to the noncutting action of the ultrasonically activated file, which is potential to create aberrant shapes within the root canal to be reduced to minimum.\textsuperscript{12} The tip of PUI produces sound energy at a frequency of 25-30 kHz, causing the irrigants to flow intensely against the root canal and contributing to the successful disinfection of the root canal system.\textsuperscript{12} It is shown in research that EDTA 17% activated ultrasonically removes smear layer effectively in the root canal system.\textsuperscript{17}

EDTA 17% was purposed as final irrigation. The combination of temperature and ultrasonic activation was proposed in this article to ascertain the effectiveness of EDTA 17% in smear layer removal in the apical third. The application of this combination method in the apical third was observed due to the complexity of the root canal, which still challenges many clinicians. This laboratory experimental study aimed to analyze the effect of temperature and PUI activation of EDTA 17% in smear layer removal. The outcome of this experiment is expected to contribute as a useful reference in endodontics.

**MATERIALS AND METHODS**

This study used a post-test only control group design. The samples in this study were determined by purposive sampling, based on predetermined criteria. The samples were obtained by Federer formula.

Twenty-four samples of extracted mandibular premolars decontaminated in formalin 10% for a week were decoronated to standard root length of 14 mm with a diamond disc burr. The preparation of the root canals was completed with a crown-down technique using ProTaper FHU to file F3. The working length was set at 13.5 mm. The root canal irrigation was done with NaOCl 5.25% and EDTA 17% solutions. The samples were divided into four groups, namely 25 °C of EDTA 17% (group 1), 37 °C of EDTA 17% (group 2), 25 °C of EDTA 17% with PUI activation (group 3), and 37 °C of EDTA 17% with PUI activation (group 4).

Increasing the temperature of EDTA 17% in this study was conducted by thermostat to reach the temperature of 37 °C. EDTA 17% was poured into a beaker glass soaked in a container of water, with the convective heat transfer from the thermostat. The temperature of 25 °C of EDTA 17% was obtained by placing EDTA 17% in an icebox due to incongruity of room temperature, indicating a temperature of 27 °C. The setting of temperature was controlled using a digital thermometer.

During irrigation, 30-G needle and syringe were used. The volume of irrigants was precisely set at 1 ml for each application in the root canals and completed for 1 minute. In this study, ultrasonically activated groups were completed by PUI (Ultra X, Eighteenth, China) with a silver tip.

Smear layer removal was observed by placing the samples that had been cut longitudinally under scanning electron microscopy (SEM) with a magnification of 1000x, 3000x, and 5000x. The observation focused on the apical third. The sample scoring was performed by 3 observers based on ordinal scoring.\textsuperscript{18} Score 1 was assigned when no smear layer was found, score 2 was assigned when thin smear layer was found in less than 25% of the area and the dentin tubule opening was visible, score 3 was assigned when the distribution of smear layer was up to 50% of the area and the dentin tubule opening was visible, score 4 was assigned when homogenous smear layer covered
the canal wall, and score 5 was assigned when inhomogenous smear layer covered the canal wall. Interclass correlation coefficient (ICC) test was established to measure the agreement among the observers.

**RESULTS**

This study was held in September – November 2020 in Radiology Installation of Islamic Hospital Dental Sultan Agung; Pre-Clinical Dentistry Laboratory, Faculty of Medicine, Diponegoro University; and Integrated Laboratory, Diponegoro University, Semarang. Ethical clearance was obtained from the Research Ethic Committee of the Faculty of Medicine, Diponegoro University. Figure 1 shows the results of the smear layer removal in the apical third point under SEM.

Using non-parametric Friedman test, this study indicated that there was a significant difference in all the groups (Table 1). Post hoc Wilcoxon Signed Rank test continually revealed the difference between the groups (Table 2).

**DISCUSSION**

Smear layer removal aims to help the diffusion of intracanal medicament and achieve the success of obturation in root canal treatment. Smear layer removal increases the apical seal and the success of long-term endodontic treatment. In this study, the low score of mean rank indicated a small area covered by the smear layer, signifying that there was an improvement of smear layer removal. The differences in the smear layer removal in all the groups showed a significant result. Consequently, the null hypothesis was rejected.

Post hoc test between EDTA 17% solution at different temperatures, namely 25 °C and 37 °C, resulted in an insignificant difference (p = 0.063). The result of this study is in line with Çiçek, et al (2015) that there was no significant difference between 25 °C and 37 °C of EDTA 17% solutions.

The same result was obtained with the post hoc test between 25 °C of EDTA 17% and 37 °C solution without PUI and with PUI activation, resulting in insignificant differences. The level of effectiveness of PUI activated irrigation in smear layer removal signified the best result in the cervical third, followed with the middle third, and the apical third. Kato, et al (2016) clarified that PUI tip with a size of #20 (as used in this study) was the average size in endodontic treatment, while the root canal preparation with a crown-down technique reached file F3 (similar to file #30). As stated by Kato, the comparison between PUI tip size and prepared root canals was inadequate enough. A previous study by Ahmad, et al (1987) mentioned that, for PUI activation to be effective, the prepared root canal must be operated within a space three times

**Table 1.** Non-parametric Friedman test

<table>
<thead>
<tr>
<th>Group</th>
<th>Mean Rank</th>
<th>p-value</th>
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<tbody>
<tr>
<td>1 Smear layer removal</td>
<td>3.17</td>
<td></td>
</tr>
<tr>
<td>2 Smear layer removal</td>
<td>1.75</td>
<td>0.017*</td>
</tr>
<tr>
<td>3 Smear layer removal</td>
<td>3.33</td>
<td></td>
</tr>
<tr>
<td>4 Smear layer removal</td>
<td>1.75</td>
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*Significance level p<0.05

**Table 2.** Post hoc Wilcoxon Signed Rank test

<table>
<thead>
<tr>
<th>Group</th>
<th>1 and 2</th>
<th>1 and 3</th>
<th>1 and 4</th>
<th>2 and 3</th>
<th>2 and 4</th>
<th>3 and 4</th>
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<tr>
<td></td>
<td>-1.857</td>
<td>0.000</td>
<td>-1.857</td>
<td>-2.060</td>
<td>0.000</td>
<td>-2.070</td>
</tr>
<tr>
<td>Asymp. Sig. (2-tailed)</td>
<td>0.063</td>
<td>1.000</td>
<td>0.063</td>
<td>0.039*</td>
<td>1.000</td>
<td>0.038*</td>
</tr>
</tbody>
</table>

*Significance level p<0.05

**Table 1.** Non-parametric Friedman test

**Table 2.** Post hoc Wilcoxon Signed Rank test
Figure 1. Smear layer removal in apical third point under SEM in group of EDTA 17% in 25 ºC at 1000x (A), 3000x (B), 5000x (C)

Figure 2. Smear layer removal in apical third point under SEM in group of EDTA 17% in 37 ºC at 1000x (D), 3000x (E), 5000x (F)

Figure 3. Smear layer removal in apical third point under SEM in group of EDTA 17% in 37 ºC + PUI at 1000x (J), 3000x (K), 5000x (L)

Figure 4. Smear layer removal in apical third point under SEM in group of EDTA 17% in 37 ºC + PUI at 1000x (J), 3000x (K), 5000x (L)
greater than the PUI tip size. This possibility might result in the insignificant differences in these groups.

The mean rank showed 37 °C of EDTA 17% solution with PUI activation was better than 25 °C of EDTA 17% solution. However, the post hoc test resulted in an insignificant difference. Possibly, this result was caused by the sufficient wettability of EDTA at a temperature of 37 °C, which was better than that at a room temperature. This condition caused a decrease in the surface tension of EDTA 17%. At the sufficient wettability, the surface tension of irrigants reduces and facilitates the irrigants to diffuse easier in dentinal tubules. According to Paragliola, et al (2010), the use of PUI activation increases the diffusion of irrigants in a root canal system.

EDTA 17% solution at a temperature of 37 °C significantly removed smear layer better than EDTA 17% solution at a temperature of 25 °C with PUI activation. EDTA 17% solution at 37 °C had suitable wettability. The wettability of EDTA solution at a temperature of 25 °C was supposed to be less suitable. The combination of room temperature and PUI activation was less significant, as clarified by Amin, et al (2016) that PUI activation in apical third showed an insignificant result. Reasonably, it was because of the narrowing of the apical third diameter which obstructed the energy produced in the tip of PUI.

The smear layer removal in the apical third in EDTA 17% solution with PUI activation at 25 °C was significantly different with that at 37 °C as shown in the results of group 3 (of EDTA 17% solution with PUI activation) and group 4 (p = 0.038). The combination of these methods successfully serves as an alternative in improving the effectiveness of irrigant. EDTA 17% solution with PUI activation at 37 °C revealed a better result rather than PUI activation at 25 °C. Similar to the previous explanation, it might be due to the fact that the wettability of EDTA at a temperature of 37 °C was more appropriate to remove smear layer compared to that at 25 °C, despite PUI activation. Further studies are required to evaluate the wettability of EDTA 17% at a temperature of 37 °C with PUI activation.

The limitation of this study is that the PUI tip size used in this study was selected based on the standard size. The PUI tip size of #10 needs to be developed to achieve optimum result in PUI activation.

CONCLUSION
A single method of increasing temperature and PUI activation insignificantly affects EDTA 17% in smear layer removal in apical third. However, the combination method can be considered as a solution during root canal disinfection in clinical practices to remove smear layer, particularly in apical third.

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