ASSESSMENT OF MARGINAL ADAPTATION OF TWO DIFFERENT ROOT CANAL SEALERS USING AN INNOVATIVE IRRIGANT: AN IN VITRO STUDY

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ABSTRACT

(NaOCl) and (EDTA) 17 are the most commonly used irrigants. The active chlorine part in NaOCl that is responsible for its antimicrobial and dissolving activity was greatly reduced when mixed with EDTA. Recently, Dual Rinse HEDP has been proposed as an endodontic irrigant with the ability to remove the smear layer without affecting the antimicrobial and dissolving effect of NaOCl. Epoxy resin and bioceramic sealers are self-adhesive sealers that provided similar acceptable results when tested.

Aim: To compare the efficacy of an innovative additive mixed with (NaOCl) on the marginal adaptation of two root canal sealers to the standard irrigation protocol of using (NaOCl) and 17% EDTA.

Material & Methods: Forty extracted single roots teeth were endodontically accessed and canal enlarged using ProTaper Next up to size X4. Samples were divided into four groups:

Group 1 irrigated with 3% NaOCl and EDTA obturated using Adseal Group 2 irrigated with 3% NaOCl and HEDP; obturated using Adseal Group 3 irrigated with 3% NaOCl and EDTA obturated using Endoseal Group 4 irrigated with 3% NaOCl and HEDP; obturated using Endoseal. The samples were obturated using ProTaper Next gutta percha cone size X4 and subjected to (SEM) evaluation and statistical analysis.

Results: Groups 3&4 obturated using Endoseal showed significantly less marginal gaps in comparison to Groups 1&2 obturated with Adseal sealer regardless of the irrigation protocol used.

Conclusion: It could be concluded that Endoseal MTA provided better adaptation to root canal walls regardless of the irrigation regime. Also Adseal showed better adaptation when NaOCl & HEDP were used.

KEYWORDS: Marginal adaptation, endoseal MTA, dual rinse HEDP

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INTRODUCTION

Endodontic treatment aims to eliminate or minimize the microorganisms present within the canal. However, root canal system is rather a complex structure that has area that could not be reached or cleaned with instrumentation success and survival rates of the endodontic procedure.

The alternative use of Sodium hypochlorite (NaOCl) and Ethylenediaminetetraacetic acid 17% (EDTA) have been advocated by many authors and proved effective at disinfecting the canal system as well as removal of the smear layer from canal walls. NaOCl has an antibacterial ability and able to dissolve the organic substrate from the smear layer through the action of its active chlorine particles that provides the protein-dissolving ability and antibacterial properties of this irrigant. However, NaOCl content of active chlorine particles was greatly reduced when mixed with EDTA, authors recommended that NaOCl and EDTA should not mix in the root canal but rather to be used simultaneously after drying the canals in between the two irrigants. In addition, other authors found that alternating NaOCl & EDTA may result in dentin erosion, they concluded that the sequence, application time, and concentration of the irrigants used affected the final outcome.

Dual Rinse HEDP (Medcem, Weinfelden, Switzerland) has been proposed recently as an endodontic irrigant, it has the ability to remove the inorganic component of the smear layer and it can be mixed with NaOCl without adversely affecting its antimicrobial & dissolving properties, also the manufacturer mentioned that it could prevents and remove the smear layer as it forms during instrumentation. This single combined-solution irrigation concept was suggested by several authors to have a positive effect on the adhesion of various types of sealers to root dentin.

Resin sealers and bioceramic sealers have exhibited acceptable biological; physical; and mechanical properties such as bond and push out strength in addition to high flow. Adseal (MetaBiomed, South Korea) is an epoxy resin-based sealers similar that showed superior flow property, whereas, EndoSeal MTA (Maruchi, Wonju, South Korea) is a bioceramic material based on pozzolan cement that retains the physical and biological properties of MTA, it is available in the form of a premixed, preloaded paste in an air-tight syringe.

AIM OF THE STUDY

To compare the efficacy of an innovative additive to be mixed with sodium hypochlorite that is regularly used for root canal irrigation on the marginal adaptation of two different root canal sealers to the standard irrigation protocol of using sodium hypochlorite followed by a final flush with 17% EDTA solution.

MATERIALS AND METHODS

Sample size calculation:
To detect the difference of means between the different cross-sections within each group with 80% power and 0.05 error; it was decided that only 2 specimens was needed at each interval (cross-section), however a total of 10 (n=10) were used in similarity to Remy et al.

Sample Selection:
Forty extracted straight single rooted maxillary or mandibular bicuspid teeth with mature apices and patent canals were collected for this study; extraction was done for orthodontic; periodontal; or prosthetic reasons based on clinical evaluation, no humans were involved. The samples were examined under magnification for cracks, resorption or any defects. They were also examined radiographically to ensure they conceded with Vertucci Type I canal classification, the single canal configuration was confirmed through high magnification; bucco-lingual and mesio-distal radiographs. Exclusion criteria included roots with cracks, decay, defects, or had more than one canal. The specimens had
their external surfaces cleaned using Ultrasonic scaler to ensure removal of any debris or remnants attached to the external tooth surface. Finally the teeth were stored in distilled water and 2.5% NaOCl in proportion of 10:1 till the time of the experiment.

**Sample Preparation**

All Specimens were endodontically accessed, the Working length was determined by inserting a size #15 K-type file (DENTSPLY Maillefer, Ballaigues, Switzerland) inside the root canal until visible at the apex and then 1mm was subtracted. The crowns of all teeth were then adjusted to a standardized working length of 18 mm. The root canals were then cleaned and shaped using ProTaper Next system (PTN)-(DENTSPLY Maillefer, Ballaigues, Switzerland); the master Rotary file was set at size # X4.

The specimens (n=40) were randomly divided into four equal groups each having ten specimens (n=10) as follows:

**Group I:** The canals will be irrigated with 1 mL of 2.5% NaOCl following each instrument for the duration of 30 seconds each and irrigation with 1 mL of 17% EDTA (tg cleanser 17%, Technical & General Ltd. UK) was also done as a final flush. The canals will then be properly dried with sterile paper points (Dentsply Maillefer, Ballaigues, Switzerland) and obturated using the single cone Obturation technique with ProTaper Next gutta percha cone size #X4 (Dentsply Maillefer, Ballaigues, Switzerland) and ADSEAL resin sealer (MetaBiomed, South Korea).

**Group II:** The canals will be irrigated with a fresh mixture of Dual Rinse HEDP (Medcem GmbH, Austria) mixed with 2.5% NaOCl according to the manufacturer’s instructions immediately before the start of the experiment. The canals will then be properly dried with sterile paper points (Dentsply Maillefer, Ballaigues, Switzerland) and obturated using the single cone Obturation technique with ProTaper Next gutta percha cone size #X4 (Dentsply Maillefer, Ballaigues, Switzerland) and ADSEAL resin sealer (MetaBiomed, South Korea).

**Group III:** The canals will be irrigated with 1 mL of 2.5% NaOCl following each instrument for the duration of 30 seconds each and irrigation with 1 mL of 17% EDTA (tg cleanser 17%, Technical & General Ltd. UK) was also done as a final flush. The canals will then be properly dried with sterile paper points (Dentsply Maillefer, Ballaigues, Switzerland) and obturated using the single cone Obturation technique with ProTaper Next gutta percha cone size #X4 (Dentsply Maillefer, Ballaigues, Switzerland) and EndoSeal MTA (Maruchi, Wonju, South Korea).

**Group IV:** The canals will be irrigated with a fresh mixture of Dual Rinse HEDP (Medcem GmbH, Austria) mixed with 2.5% NaOCl according to the manufacturer’s instructions immediately before the start of the experiment. The canals will then be properly dried with sterile paper points (Dentsply Maillefer, Ballaigues, Switzerland) and obturated using the single cone Obturation technique with ProTaper Next gutta percha cone size #X4 (Dentsply Maillefer, Ballaigues, Switzerland) and EndoSeal MTA (Maruchi, Wonju, South Korea).

**TABLE (1): Summary of Sealers & irrigation regime**

<table>
<thead>
<tr>
<th>Groups</th>
<th>Preparation system</th>
<th>Sealer</th>
<th>Irrigation regime</th>
</tr>
</thead>
<tbody>
<tr>
<td>Group I</td>
<td>ProTaper Next up to size X4</td>
<td>Adseal Resin sealer</td>
<td>NaOCl &amp; EDTA final flush</td>
</tr>
<tr>
<td>Group II</td>
<td>ProTaper Next up to size X4</td>
<td>Adseal Resin sealer</td>
<td>NaOCl &amp; HEDP mixture</td>
</tr>
<tr>
<td>Group III</td>
<td>ProTaper Next up to size X4</td>
<td>Endoseal MTA sealer</td>
<td>NaOCl &amp; EDTA final flush</td>
</tr>
<tr>
<td>Group IV</td>
<td>ProTaper Next up to size X4</td>
<td>Endoseal MTA sealer</td>
<td>NaOCl &amp; HEDP mixture</td>
</tr>
</tbody>
</table>
The specimens were grooved with a diamond disk (4217, DFS, Riedenburg, Germany) and split longitudinally using chisel and mallet. One half of each tooth was randomly chosen and placed using carbon tape in a circular metal stub measuring 10 mm in diameter and 5 mm in height. Then the samples were coated with gold for SEM evaluation (JSM 6460 LV; JEOL, Tokyo, Japan). The images were all performed without knowledge of the group tested. One point was selected in each section to be evaluated at the canal cervical, middle and apical thirds under 100x magnifications and the maximum width of the marginal gap at each tooth section on all three levels will be recorded.

The evaluation was scored in a blind manner by two separate well experienced observers and the result Data was tabulated and statistically analyzed.

**Statistical Analysis**

Statistical analysis was performed using SPSS in general (version 20), while Microsoft office Excel was used for data handling and graphical presentation.

Shapiro-Wilk test of normality was used to test normality hypothesis of all quantitative variables for further choice of appropriate parametric and non-parametric tests. Mostly the variables were found non-normally distributed allowing the use of non-parametric tests. Kruskal–Wallis analysis of variance was used to compare the marginal adaptation between the three different groups and for individual comparisons Mann–Whitney U-test was used; p<0.05 with a hypothesized value of 0.11.

**RESULTS**

This study evaluated 40 specimens for different root canal sealers and different irrigation regimes and made the following observations:

**TABLE (2): Summary of irrigation protocol after each instrumentation**

<table>
<thead>
<tr>
<th>PTN</th>
<th>Group I</th>
<th>Group II</th>
<th>Group III</th>
<th>Group IV</th>
</tr>
</thead>
<tbody>
<tr>
<td>X1</td>
<td>1 mL 3 % NaOCl \ 30 sec</td>
<td>1 mL 3 % NaOCl + HEDP mix \ 30 sec</td>
<td>1 mL 3 % NaOCl \ 30 sec</td>
<td>1 mL 3 % NaOCl + HEDP mix \ 30 sec</td>
</tr>
<tr>
<td>X2</td>
<td>1 mL 3 % NaOCl \ 30 sec</td>
<td>1 mL 3 % NaOCl + HEDP mix \ 30 sec</td>
<td>1 mL 3 % NaOCl \ 30 sec</td>
<td>1 mL 3 % NaOCl + HEDP mix \ 30 sec</td>
</tr>
<tr>
<td>X3</td>
<td>1 mL 3 % NaOCl \ 30 sec</td>
<td>1 mL 3 % NaOCl + HEDP mix \ 30 sec</td>
<td>1 mL 3 % NaOCl \ 30 sec</td>
<td>1 mL 3 % NaOCl + HEDP mix \ 30 sec</td>
</tr>
<tr>
<td>X4</td>
<td>1 mL 3 % NaOCl \ 30 sec</td>
<td>1 mL 3 % NaOCl + HEDP mix \ 30 sec</td>
<td>1 mL 3 % NaOCl \ 30 sec</td>
<td>1 mL 3 % NaOCl + HEDP mix \ 30 sec</td>
</tr>
<tr>
<td>Total</td>
<td>4 ml 3 % NaOCl \ 30 sec and 1 mL 17 &amp; EDTA \ 30 sec</td>
<td>4 ml 3 % NaOCl + HEDP mix \ 30 sec</td>
<td>4 ml 3 % NaOCl \ 30 sec and 1 mL 17 &amp; EDTA \ 30 sec</td>
<td>4 ml 3 % NaOCl + HEDP mix \ 30 sec</td>
</tr>
</tbody>
</table>

**TABLE (3): Mean difference and significance using Bonferroni method group I (Adseal + NaOCL\EDTA)**

<table>
<thead>
<tr>
<th>group I (Adseal + NaOCL\EDTA)</th>
<th>Mean</th>
<th>P-Value (P&lt;0.05)*</th>
</tr>
</thead>
<tbody>
<tr>
<td>Coronal Middle</td>
<td>0.17</td>
<td>0.05</td>
</tr>
<tr>
<td>Coronal Apical</td>
<td>8.84</td>
<td>0.001*</td>
</tr>
<tr>
<td>Middle Apical</td>
<td>8.67</td>
<td>0.001*</td>
</tr>
</tbody>
</table>
TABLE (4): Non-Parametric Mann-Whitney test for comparing 2 segments in group I (Adseal + NaOCl\EDTA)

<table>
<thead>
<tr>
<th>Segment</th>
<th>Mean</th>
<th>Mann-Whitney U</th>
<th>P-Value (P&lt;0.05)*</th>
</tr>
</thead>
<tbody>
<tr>
<td>Coronal</td>
<td>0.17</td>
<td>36.50</td>
<td>0.31500</td>
</tr>
<tr>
<td>Middle</td>
<td>8.84</td>
<td>1.00</td>
<td>0.00000*</td>
</tr>
<tr>
<td>Apical</td>
<td>8.67</td>
<td>0.00</td>
<td>0.00000*</td>
</tr>
</tbody>
</table>

TABLE (5): Non parametric Kruskal-Wallies test for comparing more than 2 segments in group I (Adseal + NaOCl\EDTA)

<table>
<thead>
<tr>
<th>Segment</th>
<th>Mean</th>
<th>SD</th>
<th>P-Value (P&lt;0.05)*</th>
</tr>
</thead>
<tbody>
<tr>
<td>Coronal</td>
<td>32.77</td>
<td>2.96</td>
<td>0.00006*</td>
</tr>
<tr>
<td>Middle</td>
<td>32.60</td>
<td>1.14</td>
<td></td>
</tr>
<tr>
<td>Apical</td>
<td>23.93</td>
<td>2.75</td>
<td></td>
</tr>
</tbody>
</table>

Assessment of marginal adaptation of different segments within each group: results showed that group I (Adseal + NaOCl\EDTA) showed presence of significant marginal gaps (P > 0.05) between apical & middle (P < 0.001); Apical & middle (P < 0.001), while marginal gaps difference between coronal and middle was non-significant (P > 0.05). Group II (Adseal + NaOCl\HEDP); group III (Endoseal + NaOCl\EDTA); and group IV (Endoseal + NaOCl\HEDP) showed uniform adaptation without any marginal gaps regardless of the irrigation regime; sealer used; or evaluated segments and there was no statistical significant difference.

TABLE (6): Mean difference and significance level using One sample t test

<table>
<thead>
<tr>
<th>Segment</th>
<th>Mean</th>
<th>SD</th>
<th>P-Value (P&lt;0.05)*</th>
</tr>
</thead>
<tbody>
<tr>
<td>Coronal</td>
<td>32.77</td>
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</tr>
<tr>
<td>Apical</td>
<td>23.93</td>
<td>2.75</td>
<td></td>
</tr>
</tbody>
</table>

Assessment of marginal adaptation of different segments across all group: results showed that only group I (Adseal + NaOCl\EDTA), showed presence of significant marginal gaps (P < 0.05), the highest mean was recorded in coronal segments (32.77) followed by middle segments (32.60) and apical (23.93), the significance difference was (0.00000*) for coronal; middle; and apical respectively. Group II (Adseal + NaOCl\HEDP); group III (Endoseal + NaOCl\EDTA); and group IV (Endoseal + NaOCl\HEDP) showed uniform adaptation without any marginal gaps regardless of the irrigation regime; sealer used; or evaluated segments and there was no statistical significant difference.
DISCUSSION

Many authors found higher concentration of NaOCl provided higher antimicrobial activity and better disruption of bacterial biofilm disruption. However, higher concentration increases the chances of toxicity to surrounding periodontal and periapical tissue. In our study we used a 3% NaOCl following the manufacturer instruction, several studies showed that HEDP was readily reactive with the active chlorine content of NaOCl solution that had concentration above 5% which adversely affected the dissolving and antimicrobial activity of NaOCl.

Alteration between NaOCl & EDTA effectively removed the smear layer, nevertheless it also affected dentin flexural strength, resulting in a higher incidence of vertical root fracture. NaOCl removes the organic part from dentin in a concentration and time-dependent manner, while EDTA then removes the mineral part resulting in dentin erosion. In the present study 1 mL of 17% EDTA was used only once as a final flush after finalizing the canal preparation and enlargement procedure.

The continuous chelation concept was introduced in 2005 using a mixture of NaOCl and a weak chelating agent such as Etidronic acid. In this study, Dual Rinse HEDP salt was mixed with 3% NaOCl just before clinical use, then 1 mL of the mixture was used to irrigate the canals after every preparation cycle, hence removing the smear layer as it forms without affecting the NaOCl organic-dissolving or antimicrobial activities. Moreover, Authors found that Dual Rinse HEDP had no effect on cytotoxic and genotoxic effect of NaOCl. Several authors showed that adding Dual rinse HEDP rendered the mixture hypertonic with a high surface tension leading to bacterial cell death and reduction in the cohesion of biofilms matrices.

Fig. (1) Scanning electron microscope (SEM) cross-section (CS) images showing marginal gaps at sealer-dentin interface; (A1) Adseal+NaOCl\EDTA cervical CS (A2) Adseal+NaOCl\EDTA middle CS (A3) Adseal+NaOCl\EDTA Apical CS (B1) Adseal+NaOCl\HEDP cervical CS (B2) Adseal+NaOCl\HEDP middle CS (B3) Adseal+NaOCl\HEDP Apical CS (C1) Endoseal+NaOCl\EDTA cervical CS (C2) Endoseal+NaOCl\EDTA middle CS (C3) Endoseal+NaOCl\EDTA Apical CS (D1) Endoseal+NaOCl\HEDP cervical CS (D2) Endoseal+NaOCl\HEDP middle CS (D3) Endoseal+NaOCl\HEDP Apical CS
Presence of marginal gaps at the sealer-canal wall interface is responsible for bond strength deterioration; it is agreed upon that removal of the smear layer allowed better sealer adaptation and adhesion. However, the process itself remains debatable; some authors believe that penetration into the dentinal tubules adds to the bond strength of the sealer-canal wall interface and increases sealing ability. Meanwhile, others authors found that the depth of sealer penetration into the dentinal tubules was more related to the characteristics of the sealers used and their ability to flow. In disagreement, several authors concluded that there was no significant correlation between sealability and sealer penetration into dentinal tubules, thus rejecting the hypothesis relating sealability with sealer amount and depth within the dentinal tubules. In similarity, more authors found that the dentinal tubules had a minor role in the dentin adhesion process with adhesive sealers, they concluded that the main retention is provided by micromechanical interactions of the sealer with the collagen matrix and the underlying demineralized zone in the intertubular dentin. Furthermore, recent studies showed that powerful chelating agents such as EDTA produced a demineralized dentin zone too deep to allow effective sealer penetration and adhesion in comparison to a weaker chelating agent like Dual Rinse HEDP. Accordingly, failure of the sealer to infiltrate the entire depth of the demineralized dentinal tubule leaves behind a collapsed collagen matrix that allows fluid movement between the hybrid layer and unfilled demineralized dentin leading to bond deterioration, which could explain the significant difference and marginal gaps that our results showed for Group 1 that had Adseal sealer and was irrigated with NaOCl\EDTA in comparison to group 2 that had Adseal sealer and was irrigated with NaOCl\HEDP mixture that showed absence of marginal gaps.

Groups 3 & 4 canals that had EndoSeal MTA sealer did show any marginal gaps and had uniform adaptation regardless of the irrigation regime used which could be related to their self-adhesiveness ability; the tendency to form a chemical bond with dentin through the formation of hydroxyapatite layer; high flow ability regardless of the canal moisture condition or presence of smear layer; high push-out bond strength; and deeper penetration into dentinal tubules values when compared to epoxy resin sealers.

Our results also partially contradicted those studies that compared epoxy resin sealers versus bioceramic sealers and concluded that both had similar and acceptable results, such was only true when we used NaOCl\HEDP mixture irrigation (Groups 2 & 4) which could be related to our study design because we were only concerned with the evaluation of sealer adaption through assessment of marginal gaps presences against root canal walls, sealer penetration into the dentinal tubules was not a parameter in this study in comparison to those studies that found epoxy resin and bioceramic sealers provided similar outcome.

CONCLUSION

Within the limitation of this study; it can be concluded that Endoseal MTA Sealer provided better sealer adaptation to root canal walls regardless of the irrigation regime. Also Adseal sealer showed better adaptation when NaOCl & Dual Rinse mixture was used for irrigation instead of NaOCl & EDTA final flush.

ACKNOWLEDGMENT

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Ethical statement

This study has the ethical clearance from the research ethics committee Faculty of Medicine Fayoum University (R-174 13/06/2021)
Declaration of Competing Interest

The authors have no conflict of interest to declare, this research did not receive any specific grant from funding agencies in the public, commercial, or not-for-profit sectors.

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