Coronal Microleakage in Root Canals Filled with Resilon or Gutta Percha Following Smear Layer Removal with EDTA or MTAD

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Abstract

Objective: By the advancements in root canal filling systems, a new sealer namely Epiphany self-etch sealer (Pentron Clinical Technologies, LLC, Wallingford, CT, USA) was recently introduced to the market that does not require self-etching primer application prior to the use of sealer. On the other hand, dentin surface conditioning with various irrigation solutions can cause chemical and structural dentinal changes. The present study sought to assess the effect of EDTA (final irrigation with distilled water or chlorhexidine) and MTAD on coronal microleakage in root canals filled with Resilon/Epiphany self-etch or AH Plus/Gutta Percha obturation materials.

Methods: In this ex-vivo study, a total of 140 extracted single rooted human teeth were selected and irrigated with 1.3% sodium hypochlorite during preparation. Samples were divided into 6 test groups and 2 positive and negative control groups. Smear layer was removed by EDTA and final irrigation with distilled water in groups 1 and 4, by EDTA and final irrigation with 2% chlorhexidine in groups 2 and 5 and by MTAD in groups 3 and 6. In groups 1, 2 and 3 canals were filled with AH Plus sealer and Gutta Percha while in groups 4, 5 and 6 canals were filled with Resilon and Epiphany self-etch sealer using lateral condensation method. After placing samples in a split-chamber and sterilizing them, the coronal part of teeth was contaminated with human saliva. Samples were controlled daily for 60 days. Chi square test, survival analysis and log-rank test were used for data analysis.

Results: Nine samples (45%) in group 1, 14 samples (70%) in group 2, 18 samples (90%) in group 3, 13 samples (65%) in group 4, 7 samples (35%) in group 5 and 13 samples (65%) in group 6 had microleakage after 60 days. Difference in resistance against microleakage between groups 1 and 3 and also 3 and 5 was statistically significant.

Conclusion: MTAD does not have a negative impact on sealing properties of Resilon/Epiphany self-etch sealer but EDTA can be an appropriate dentin conditioner before using AH Plus sealer and Gutta Percha. Also, Chlorhexidine can be a good final irrigant in root canals that are going to be filled with Epiphany self-etch sealer/Resilon.

Key words: Epiphany self-etch, Chlorhexidine, Resilon, Microleakage, Gutta Percha,MTAD, EDTA

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Introduction:

Bacteria are the causative agents of pulpal and peri-radicular diseases (1-3). Following an efficient root canal therapy, number of microorganisms in the root canals decreases significantly (4). Constant penetration of oral fluids can result in failure of root canal treatments (5). Therefore, root canal filling materials, despite having the characteristics of an ideal filling material, should act as a strong barrier against bacterial penetration and microleakage of oral fluid into the peri-radicular tissues. These materials should also bury any remnant bacteria in the root canal system (6, 7).

Gutta Percha is the most frequently used root canal filling material. Despite the great advancements in root canal filling systems, no technique has been able to provide a complete seal of the root canal system by using Gutta Percha (8, 9).

On the other hand, advancements in dentin bonding systems have resulted in production of a new soft resin endodontic obturation system. Resilon is a new synthetic root canal filling material that has been introduced to replace Gutta Percha and conventional sealers in endodontic treatments. Resilon/Epiphany endodontic obturation system consists of Resilon as a core material, epiphany sealer and self-etch primer (10, 11). On the other hand, a 2-component endodontic obturation system was also introduced to the market with the brand name of Resilon/Epiphany SE which is comprised of epiphany self-etch (SE) sealer and Resilonas the core material. Self-etch primer has been eliminated in this system.

The properties of Resilon are mostly similar to those of Gutta Percha and lateral condensation, vertical condensation, thermoplastic and injectable techniques can all be used for filling the root canals with Resilon. Resilon is a highly biocompatible, non-toxic and non-mutagenic material (12). The resin sealer bonds to the filling material from one side and to the dentinal root canal walls from the other. Therefore, the gaps that usually exist between the Gutta Percha and sealer or canal walls are not present in this system (monoblock)(13).

Mechanical preparation of root canals results in formation of a smear layer on dentinal walls (14-16). By removing this layer, sealer penetrates into the dentinal tubules and increases the bond strength between the resin-based sealer and dentinal walls (17). The most common method for smear layer removal is irrigation of root canals with EDTA 17% and NaOCl 5.25% after finishing canal preparation. EDTA dissolves the mineral content while NaOCl dissolves the organic matrix of the smear layer (18, 19).

Considering some hypotheses regarding the reduction of bond strength following application of NaOCl before filling the canals with Resilon, it has been suggested that after using EDTA, only normal saline, sterile water or chlorhexidine are to be used (12). However, Stratton et al, in 2006 demonstrated that after smear layer removal, application of CHX and NaOCl final irrigation solutions made no significant changes in microleakage of Resilon-filled root canals (20).

MTAD is another material developed by the combination of tetracycline isomer, citric acid and detergent for smear layer removal (21). The effect of MTAD on coronal microleakage of Resilon/Epiphany-filled root canals has not yet been evaluated. Thus, the present study aimed to assess the rate of coronal microleakage in canals filled with Resilon/Epiphany self-etch or AH Plus/Gutta Percha obturation systems after smear layer removal using various irrigation solutions (EDTA/distilled water, EDTA/chlorhexidine, and MTAD).

Methods:

In this ex-vivo study, a total of 140 single rooted extracted human teeth that were free of root fracture, resorption or curvature were selected and evaluated. After removal of tissue appendages from root surfaces, chloramine T 0.5% was used for teeth disinfection. Samples were stored in this solution until the examinations. Teeth crowns were then cut off leaving 13 mm of root length. After observing the tip of # 10 or # 15 file from the apical foramen, working length was determined by subtracting 1 mm from the used file length. Root canals were prepared using Mtworotary files (VDW, Munich, Germany). According to the
manufacturer’s instructions, 6 files from #10 to #35 were consecutively used till working length for canal preparation. During root canal preparation and with each change in file size, the canal was irrigated with 3 ml sodium hypochlorite 1.3%. Then, the teeth were randomly divided into 6 groups of 20 each as the test groups and 2 control groups of 10 each as positive and negative controls.

Group 1: After canal preparation in this group, 5 ml EDTA 17% was used for 1 minute for smear layer removal. Canals were irrigated with 5 ml distilled water and dried with paper points. The master cone (#35 Gutta Percha point with 2% taper, Gapadent Co. LTD, Korea) was soaked in AH Plus sealer (DENTSPLY, Maillefer) and placed inside the canal. Auxiliary Gutta Percha points (#20, with 2% taper) were also placed using lateral condensation technique until the whole root canal was filled. Excess Gutta Percha was removed with a hot instrument and a plugger was used for vertical compaction.

Group 2: For smear layer removal in this group 5 ml EDTA (17%) was used for 1 minute and root canals were irrigated with 5 ml chlorhexidine 2% (Consepsis, Ultradent Products Inc.). Canals were filled with Gutta Percha and AH Plus sealer similar to the first group.

Group 3: In this group, smear layer was removed using BioPure MTAD (DENTSPLY Tulsa Dental, Tulsa, OK) according to the manufacturer’s instructions. After root canal preparation, 1 mm MTAD was placed inside the canals for 5 minutes and the canals were then filled with Gutta Percha and AH Plus sealer similar to the first group.

Group 4: In this group, 5 ml EDTA 17% was used for 1 min for smear layer removal and root canals were irrigated with 5 ml distilled water. Canals were dried with paper points and filled using Resilon and Epiphany self-etch sealer (Pentron Clinical Technologies, LLC, Wallingford, CT). Epiphany self-etch sealer is packaged in a dual barrel syringe. The sealer was mixed and prepared. Resilon master cone (#35 with 2% taper) was soaked in Epiphany sealer and placed inside the canal. Resilonauxiliary points (#20 with 2% taper) were added using lateral condensation technique until the canal was filled completely. Excess Resilon was removed with a hot instrument. Vertical compaction was done using a plugger. According to the manufacturer’s instructions, the coronal part of the filling was light cured for 40 seconds in order to achieve coronal seal.

Group 5: In this group, smear layer removal was done similar to group 2 and root canals were filled with Resilon and Epiphany SE sealer just like group 4.

Group 6: Smear layer was removed using BioPure MTAD similar to group 3 and canals were filled with Resilon and Epiphany SE sealer just like group 4.

Positive control group: In this group, #35 Gutta Percha points were placed in 5 root canals and in the remaining 5, #35 Resilon points were inserted without sealer.

Negative control group: In this group, 5 root canals were filled with Gutta Percha and AH Plus sealer using lateral condensation technique while the remaining 5 were filled with Resilon/Epiphany SE sealer.

All canal preparations and fillings were done by one individual. All samples were radiographed mesiodistally and buccolingually for assessing the quality of fillings. Samples were stored at 37°C and 100% moisture for 7 days in an incubator. All external surfaces of test and control samples were covered with 2 coats of nail polish (Arcancil, France) except for the last 3 mm of tooth apex. In negative control group, all root surfaces especially the coronal surface were completely covered with 2 layers of nail polish.

In order to assess the rate of bacterial microleakage a split-chamber model was used. Two millimeters of the end part of a plastic Eppendorf tube was cut and each root was placed in one tube in a way that their apices exited the cut-end part of the Eppendorf tubes. The gap between the plastic tube and root was sealed by adhesive wax from inside and by cyanoacrylate glue from outside. For sterilization, samples underwent 25K Gray of gamma radiation exposure. The Eppendorf tubes containing samples were placed in a glass tube containing 20 ml BHI (Merck, Germany) medium in sterile conditions (under a hood) in a way that at least 2 mm of the apices were in the BHI medium. The gap between the Eppendorf tube and glass tube was perfectly sealed with
adhesive wax. To ensure sterilization, samples were kept in an incubator at 37°C for 3 days. In case of observing turbidity in the BHI broth, the sample would be sterilized again. After ensuring that the samples were sterile, 2 ml human saliva was poured in the upper chamber of the microleakage assessment apparatus. The saliva would be refreshed every 3 days. Human saliva was provided by a volunteer who did not brush his teeth for a minimum of 12 hours before saliva collection (22).

Samples were kept in an incubator at 37°C for 60 days and were checked every day for presence of turbidity in the BHI medium in the lower chamber. Paired comparison of mean coronal microleakage between groups was done using chi square test. Survival analysis and Log-rank test were employed for comparison of the mean survival of various groups.

Results:

All positive control samples showed signs of microleakage within 4 days after initiation of study. None of the negative controls had microleakage within the 60 days. Number and percentage of samples that had microleakage are presented in Table 1. Chi square test demonstrated a significant difference in terms of number of samples with microleakage after the 60-day period between the understudy groups (P<0.006). Paired comparison of groups also showed a significant difference in number of samples with positive microleakage between MTAD/ Gutta Percha and EDTA and distilled water/Gutta Percha groups (P<0.032). Also, a significant difference was observed between MTAD/Gutta Percha and EDTA and chlorhexidine/Resilon groups in terms of samples with positive microleakage (P<0.004).

Survival analysis and log-rank test showed that after the 60-day period survival rate between groups 3 and 1 and between groups 3 and 5 was significantly different (P<0.05). Resistance in group 3 (MTAD/Gutta Percha) against salivary microleakage during the 60-day period was significantly lower than in groups 1 and 5 while the difference in this regard between other groups was not statistically significant (P>0.05). The mean period of time required for recontamination of coronally sealed canals (survival time) in different groups is demonstrated in Table 2.

**Table 1- Number and percentage of samples with positive microleakage at the end of the 60-day study period**

<table>
<thead>
<tr>
<th>Method of smear layer removal/ canal filling material</th>
<th>Number (percentage) of samples with positive microleakage</th>
</tr>
</thead>
<tbody>
<tr>
<td>EDTA and distilled water/Gutta-Percha and AH Plus sealer</td>
<td>9 (45%)</td>
</tr>
<tr>
<td>EDTA and chlorhexidine/Gutta Percha and AH Plus sealer</td>
<td>14 (70%)</td>
</tr>
<tr>
<td>MTAD/ Gutta Percha and AH Plus sealer</td>
<td>18 (90%)</td>
</tr>
<tr>
<td>EDTA and distilled water/Resilon and Epiphany sealer</td>
<td>13 (65%)</td>
</tr>
<tr>
<td>EDTA and chlorhexidine/Resilon and Epiphany sealer</td>
<td>7 (35%)</td>
</tr>
<tr>
<td>MTAD/Resilon and Epiphany sealer</td>
<td>13 (65%)</td>
</tr>
</tbody>
</table>

**Table 2- The mean period of time required for recontamination of coronally sealed canals in different study groups**

<table>
<thead>
<tr>
<th>Method of smear layer removal/canal filling material</th>
<th>Mean±SD</th>
</tr>
</thead>
<tbody>
<tr>
<td>EDTA and distilled water/Gutta Percha and AH Plus sealer</td>
<td>35.85±6.0</td>
</tr>
<tr>
<td>EDTA and chlorhexidine/Gutta Percha and AH Plus sealer</td>
<td>30.5±5.16</td>
</tr>
<tr>
<td>MTAD/Gutta Percha and AH Plus sealer</td>
<td>22.5±4.54</td>
</tr>
<tr>
<td>EDTA and distilled water/Resilon and Epiphany sealer</td>
<td>31.55±5.42</td>
</tr>
<tr>
<td>EDTA and chlorhexidine/Resilon and Epiphany sealer</td>
<td>40.9±4.87</td>
</tr>
<tr>
<td>MTAD/Resilon and Epiphany sealer</td>
<td>31.6±4.92</td>
</tr>
</tbody>
</table>

Discussion:

Based on the present study results, no significant difference was detected between Resilon (after smear layer removal with EDTA/distilled water
or MTAD) and Gutta Percha/AH Plus (after smear layer removal with MTAD or EDTA/water) endodontic obturation systems. It has been reported that in thermoplastic root canal filling technique, there is no difference between Gutta Percha/AH Plus and Resilon/Epiphany in terms of resistance against Enterococcus faecalis microleakage (23-25). However, some studies have reported better results for adhesive resin root canal filling materials compared to Gutta Percha after smear layer removal by EDTA and final irrigation with water or normal saline (26,23,27). Controversy in this respect can be due to difference in length of root canal filling (28), duration of application, the amount of EDTA used, study methodology (11) and test duration (29, 30). Geometric configuration of the root canal system does not provide a suitable environment for adhesive resin root canal filling materials (31). Some of the factors that affect the seal of the root canal system filled with adhesive resin materials include high configuration factor (C factor) that accumulates the stress caused by the polymerization shrinkage of adhesive resin materials (31), incomplete removal of smear layer (26), and incomplete penetration of resin into the demineralized dentin (32). Therefore, achieving a monoblock without any gap that may result from the bonding of Epiphany sealer to Resilon would be impossible (23). Polycaprolactone is the primary component of Resilon. Thus, the invading microorganisms will disintegrate (33). Lipase released by microorganisms can cleave the ester bonds of polycaprolactone (34,35). Also, polycaprolactone is susceptible to alkaline and enzymatic hydrolysis (36,37). However, in the present study Resilon samples did not show worse results compared to Gutta Percha although they were exposed to saliva for 2 months. Thus, further investigations are required to evaluate the outcome of Resilon hydrolysis by salivary enzymes and microorganisms.

This study showed that MTAD/Gutta Percha samples had a greater bacterial microleakage compared to Gutta Percha groups after smear layer removal with EDTA/distilled water and Resilon groups after smear layer removal with CHX/EDTA. On the other hand, MTAD did not have an adverse effect on resistance of Resilon/Epiphany against microleakage. Method of dentin surface conditioning by MTAD can be related to these findings. Tay and colleagues (2001) demonstrated that MTAD forms 10-12 micrometer thick collagen matrices while EDTA 17% creates 4-6 micrometer thick collagen matrices (38). Collagen matrices created by MTAD do not provide a suitable environment for dentin hybrid layer formation after the application of hydrophilic resin-based sealers like Epiphany (39). Also, Tween 80 as a detergent present in MTAD increases the surface free energy and wettability of dentin. This can have a negative impact on the adhesion of AH Plus sealer that has a hydrophobic property (40). Therefore, increased salivary penetration in MTAD/Gutta Percha samples can be due to the incomplete penetration of AH Plus hydrophobic sealer into the demineralized dentin.

Hashem et al, in 2009 reported that MTAD as the final irrigating solution before filling the root canal with Gutta Percha/AH Plus sealer significantly decreased the bond strength compared to EDTA (40). In the present study, MTAD/Resilon group had a lower microleakage (but not significant) compared to MTAD/Gutta Percha samples. This can be due to the increased wettability of dentin by Tween 80 detergent present in MTAD and penetration of Epiphany SE hydrophilic sealer into the rich collagen matrices created by MTAD (namely hybrid layer), or the good chemical bond between Epiphany SE sealer and Resilon. This subject needs to be further evaluated.

Dentin characteristics following application of EDTA can be a good justification for higher resistance in EDTA/Gutta Percha group against bacterial microleakage compared to MTAD/Gutta Percha group. It has been reported that EDTA decreases the wettability of dentin (41) and creates a suitable dentinal surface for adhesion to a hydrophobic sealer like AH Plus sealer (40). However, similar results obtained in EDTA/Gutta Percha and EDTA/Resilon groups cannot be justified by these reasons since despite decreasing the dentinal wettability, EDTA had a good efficacy in Resilon/Epiphany hydrophilic sealer group. The present study results were in contrast with those of Ghoddusi et al, in 2007; because they failed to demonstrate a significant difference between EDTA/Gutta Percha and AH Plus sealer and MTAD/Gutta
Percha and AH Plus sealer groups. This controversy can be due to the application of NaOCl 5.25% following the use of EDTA or difference in the methods employed for microleakage assessment (42). The lowest rate of microleakage in the present study (the highest survival rate) after 60 days of study period belonged to the EDTA and chlorhexidine/Resilon group. Small rate of microleakage in this group can be explained by the fact that the surfactant present in the formulation of chlorhexidine increases the surface free energy of dentin and results in subsequent improvement of its wettability (43, 44). As a result, a suitable environment would be created for the bonding of hydrophilic adhesive resin filling materials (like Epiphany hydrophilic sealer). Also, chlorhexidine bonds to the dentin hydroxyapatite and fights the microorganisms even after a period of time (45). Therefore, the highest resistance against salivary microleakage in the EDTA and chlorhexidine/Resilon group may be justified by the mentioned facts. It seems that final irrigation of canal with chlorhexidine compared to distilled water confers a higher resistance to the EDTA and chlorhexidine/Gutta Percha group; whereas, this study could not find a significant difference between groups that used chlorhexidine or distilled water as the final irrigation solution after application of EDTA and were filled with Resilon or Gutta Percha.

Conclusion:

This study demonstrated that Resilon along with the new Epiphany SE sealer can act similar to Gutta Percha and AH Plus sealer in prevention of salivary microleakage after smear layer removal by EDTA and final irrigation with distilled water. Also, smear layer removal by MTAD increased the salivary microleakage in canals filled with Gutta Percha and AH Plus sealer compared to EDTA. Final canal irrigation with chlorhexidine after smear layer removal by EDTA decreases the salivary microleakage in canals filled with Resilon/Epiphany SE. Thus, we recommend using EDTA along with final irrigation with distilled water or chlorhexidine for smear layer removal in canals that are going to be filled with Gutta Percha and AH Plus sealer; whereas, we can use MTAD or MTA along with final irrigation with distilled water or chlorhexidine for smear layer removal when canals are going to be filled with Resilon and Epiphany SE sealer.

References: