Effect of Er,Cr:YSGG laser irradiation on the apical sealing ability of AH Plus/gutta-percha and Hybrid Root Seal/Resilon Combinations

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Objective. The objective of this study was to evaluate the sealing ability of 2 different resin-based endodontic filling systems after smear layer removal with 2 different techniques.

Study design. Extracted human single-rooted teeth (n = 74) were instrumented using HERO Shaper rotary instruments and irrigated with 1 mL of 2.5% NaOCl between each instrument. Additionally, the canals received either an extra 3-minute rinse with 2 mL of 17% EDTA or a 40-second Er,Cr:YSGG laser treatment. The root canals were filled with either Hybrid Root Seal/Resilon combination or AH Plus/gutta-percha combination using lateral condensation technique (n = 11). Apical leakage quantity was measured with the computerized fluid filtration meter at 1 and 4 weeks. One root from each group, which was not submitted to the fluid filtration test, was selected for scanning electron microscopy (SEM) analysis. Statistical analysis was performed using 3-way ANOVA and the post hoc Tukey HSD test (α = 0.05).

Results. A significant decrease was observed in the microleakage values of all the experimental groups tested with time (P < .0001). EDTA + AH Plus/gutta-percha combination exhibited the least microleakage, whereas laser irradiation + Hybrid Root Seal/Resilon combination showed the greatest microleakage at each of the 2 time periods. Each experimental combination exhibited architecture in SEM that seemed to correlate with its sealing performance.

Tulsa, OK) are used clinically. Among these chemical agents, EDTA has been largely used as a resource to remove smear layer because of its calcium chelating capacity. Because EDTA acts by dissolving the inorganic components of the smear layer, several authors have recommended its use in combination with NaOCl (0.50%-5.25%) to remove organic remnants, thereby increasing the cleaning effect.

In addition to the routinely used chemical substances, other technologies have been investigated for the treatment of root canal dentin, such as laser irradiation. Recently, erbium lasers have been studied for applicability in smear layer removal. Treatment of the radicular dentinal walls with erbium lasers have been shown to promote cleaner surface, which might result in better adaptation of the filling material to the root canal walls. On the contrary, Mello et al. demonstrated that smear layer removal with the erbium: yttrium aluminum garnet (Er:YAG) laser showed no apparent advantage when compared with EDTA-T (EDTA + detergent) in terms of apical leakage.

Improvements in adhesive technology have prompted attempts to incorporate adhesive dentistry into endodontics. The recent introduction of Resilon (Resilon Research LLC, Madison, CT) as an alternative root filling material offers the promise of adhesion to root dentin. This thermoplastic polymer is presented as a root canal filling material having bonding ability to methacrylate-based resin sealers through the inclusion of dimethacrylate monomers. This core material can couple to a variety of dentin adhesives and resin-type sealers, including Hybrid Root Seal (Sun Medical, Moriyama, Shiga, Japan).

A 4-methacryloxyethyl trimellitate anhydride (4-META) containing polymethyl methacrylate-based endodontic sealer Hybrid Root Seal has recently been introduced to the market. Hybrid Root Seal is a self-etching sealer and has a dual-curable characteristic. Although it is believed that this dual-curable character gives a further advantage in sealing, previous studies demonstrated that Hybrid Root Seal showed similar leakage values as epoxy-resin–based AH Plus sealer (Dentsply De Trey, Konstanz, Germany).

AH Plus sealer has shown to have excellent sealing properties and is considered as the gold standard against which all new sealers and bondable root canal filling materials must be compared.

Thus, the objective of this study was to evaluate in vitro apical sealing abilities of root canals filled with Hybrid Root Seal/Resilon and AH Plus/gutta-percha combinations at 1 and 4 weeks after treated with either 17% EDTA solution or Er,Cr:YSGG laser irradiation. The null hypotheses tested are that the Er,Cr:YSGG laser treatment does not influence the sealing ability of the filling materials compared with EDTA application and the sealing ability of the Hybrid Root Seal/Resilon combination is not superior to that of the AH Plus/gutta-percha combination.

MATERIALS AND METHODS

A total of 74 extracted human mandibular premolars, macroscopically similar in size and with straight and single root canals (as verified radiographically), were used. Each tooth was placed in 5.25% sodium hypochlorite (NaOCl) for 2 hours for surface disinfection, and then stored in distilled water until testing was carried out.

Sample preparation (n = 64)

The crown of each tooth was sectioned at the cementoenamel junction using a water-cooled diamond disk. The working length was determined visually by subtracting 1 mm from the length of a size 10 K-file (Maillefer, Ballaigues, Switzerland) at the apical foramen. The middle and coronal thirds were prepared using ISO size 1, 2, 3, and 4 Gates Glidden drills (Produits Dentaires, Vevey, Switzerland) with a low-speed handpiece. All teeth were instrumented with a crown-down technique using HERO Shaper rotary instruments (Micro-Mega, Besançon, France) as follows: a #30 file with a .06 taper was initially introduced in two thirds of the working length. Shaping was completed with a #30 file with .04 taper at the working length. One milliliter of 2.5% NaOCl was used to irrigate between each instrument. A lubricant (Glyde File Prep., Dentsply, Montigny-le-Bretonneux, France) was used throughout the cleaning and shaping of the root canal. In groups 1 and 3, the canals received an extra 3-minute rinse with 2 mL of 17% EDTA (pH 7.4).

In groups 2 and 4, the canals received a 40-second Er,Cr:YSGG laser treatment (Biolase Technology, San Clement, CA), with a Z3 endodontic tip (0.32 mm in diameter, Biolase Technology) and the following parameters: 1.50 W, 20 pulses per second, 30% water, and 50% air. Groups 5 and 6 received no further treatment and served as positive control groups. Finally, the root canals were flushed with 2 mL of distilled water and then dried with paper points before filling.

Root canal filling

Group 1 (EDTA + AH Plus/gutta-percha, n = 11). The roots were filled with AH Plus sealer and .04 Taper gutta-percha points (Diadent, Chongju, Korea) using the cold lateral condensation technique. AH Plus sealer was introduced into the root canal using a paper point. An ISO size 30 master gutta-percha cone was lightly coated with AH Plus sealer and placed into the canal to working length. A size 25 finger spreader (Dentsply Maillefer,
Ballaiques, Switzerland) was then inserted into the canal to a level approximately 1 mm short of working length. Lateral condensation with fine accessory gutta-percha cones was performed until the entire root canal was filled. The excess gutta-percha was removed with a heated instrument and then compacted vertically using a size 11 plugger (Dentsply Maillefer, Ballaiques, Switzerland).

Group 2 (laser irradiation + AH Plus/gutta-percha, n = 11). The roots were filled with AH Plus sealer and .04-taper gutta-percha points in the same manner as in group 1.

Group 3 (EDTA + Hybrid Root Seal/Resilon, n = 11). The roots were filled with Hybrid Root Seal and .04 Taper Resilon points (Pentron, Wallingford, CT) using the cold lateral condensation technique. As specified by the manufacturer, 3 drops of liquid and 1 scoop of powder were dispensed onto a pad and mixed with a spatula. The homogeneous mixture was introduced to the root canal using a paper point. An ISO size 30 master Resilon cone was lightly coated with Hybrid Root Seal and placed into the canal to working length. A size 25 finger spreader was then inserted into the canal to a level approximately 1 mm short of working length. Lateral condensation with fine accessory Resilon points was performed until the entire root canal was filled. The excess Resilon was removed with a heated instrument and then compacted vertically using a size 11 plugger. The coronal surface of the root filling was light-cured for 20 seconds to create an immediate coronal seal, as per the manufacturer’s recommendations.

Group 4 (laser irradiation + Hybrid Root Seal/Resilon, n = 11). The roots were filled with Hybrid Root Seal and .04 Resilon points in the same manner as in group 3. The canal orifice was light-cured for 20 seconds.

Group 5 (no treatment + gutta-percha, n = 10). The roots were filled with .04 Taper gutta-percha points using the cold lateral condensation technique and served as a positive control group.

Group 6 (no treatment + Resilon, n = 10). The roots were filled with .04 Taper Resilon points using the cold lateral condensation technique and served as a positive control group.

Group 7 (negative control group, n = 10). The nonprepared and unfilled roots were totally coated with 3 layers of nail varnish covering the root canal orifices as well as the apical foramina.

All 74 roots were then stored in gauze dampened with sterile saline and enclosed in sealed tubes for 7 days to allow the sealer to set. The coronal portion of each root was sectioned perpendicular to its long axis and discarded to create roots of uniform (10.00 ± 0.05 mm) length (Fig. 1) using a water-cooled diamond blade on an Isomet machine (Buehler, Lake Bluff, IL). The coronal parts of all the roots were surface sealed using the 2-step self-etch adhesive system (Clearfil SE Bond, Kuraray Medical, Okayama, Japan) with a nano-composite resin (Clearfil Majesty Esthetic, Kuraray Medical).

**Computerized fluid filtration technique**

Microleakage was measured with the computerized fluid filtration meter (Fig. 1) that was originally described by Ciucchi et al.23 and later used in different studies.24–27 Root sections were inserted into the plastic tube from the apical side and connected to an 18-gauge stainless steel tube. The cyanoacrylate adhesive (Zapit, Dental Venture of America, Anaheim Hills, CA) was applied circumferentially between the root and plastic tube. O₂ from a pressure tank of 120 kPa (1.2 atm) was applied at the apical side. Using a microsyringe, water was retracted approximately 2 mm. In this way, an air bubble was created and then adjusted to a suitable position in the micropipette. Through one side of the micropipette inside the device, an infrared light was passed. Two light-sensitive photodiodes were used to detect any movement of an air bubble inside the micropipette. All operations were controlled with PC-compatible software (Fluid Filtration 03, Konya, Turkey). Measurements of fluid movement were automatically made at 2 minutes during 8 minutes for each sample by using PC-compatible software. All specimens were stored at 37°C in water. The quality of seal of each specimen was measured at 1 and 4 weeks. Leakage quantity was expressed as μL/cmH₂O/min⁻¹ and means determined.

**Scanning electron microscope evaluation**

One root from each experimental group, which was not submitted to the fluid filtration test, was selected for analysis by scanning electron microscope (SEM) after 1 week. One root was cross-sectioned into two 3-mm-thick segments to obtain 1 specimen from each apical and middle third of the root. Specimen preparation for SEM observation followed routine procedure (specimens were fixed in 10% buffered formaldehyde overnight, carried through ascending alcohol concentrations to 100% alcohol and finally critical-point dried [BioRad E-3000, BioRad Microscience Ltd., Hertfordshire, UK]). Samples were mounted in stubs, sputter coated with gold, and then observed under SEM (Zeiss Evo 50; Carl Zeiss, Oberkochen, Germany) to assess the contact and adaptation between the root canal walls and filling materials.

Statistical analysis was performed using 3-way analysis of variance (ANOVA) and the post hoc Tukey HSD test. The level of statistical significance was set at α = 0.05.

**RESULTS**

The means and standard deviations of the microleakage values of the tested materials are shown in Table I.
The 3-way ANOVA revealed that microleakage values varied statistically according to the smear layer removal methods \( (P < .0001) \), the filling material combinations \( (P < .0001) \), and the time of testing \( (P < .05) \). The statistical analysis also demonstrated significant interactions between the smear layer removal method and the filling material combination \( (P < .0001) \), the smear layer removal method and the time of testing \( (P < .0001) \), and the filling material combination and the time of testing \( (P < .01) \). The interaction among these 3 factors was not significant \( (P > .05) \).

Both positive control groups indicated significantly more fluid filtration than the experimental groups throughout the experimental period \( (P < .0001) \). The varnish-coated negative controls had no measurable bubble movement at both time periods.

All the experimental groups allowed fluid to flow along the root dentin-filling material interface at the both time periods (1 and 4 weeks). There were statistically significant differences between the periods of the study, which presented a significant decrease in the microleakage values of all the experimental groups tested with time \( (P < .0001) \).

The computerized fluid filtration meter.

**Table 1.** Mean microleakage \( (\mu L/cmH_2O/min^{-1}) \) value and SD for experimental groups

<table>
<thead>
<tr>
<th>Groups</th>
<th>1-week Mean microleakage ± SD</th>
<th>4-week Mean microleakage ± SD</th>
<th>( P )</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>( (\mu L/cmH_2O/min^{-1}) )</td>
<td>( (\mu L/cmH_2O/min^{-1}) )</td>
<td></td>
</tr>
<tr>
<td>Group 1 (EDTA + AH Plus/gutta-percha)</td>
<td>0.000638 ± 0.000308\textsuperscript{a,1}</td>
<td>0.000312 ± 0.000168\textsuperscript{c,2}</td>
<td>&lt; .0001</td>
</tr>
<tr>
<td>Group 2 (laser irradiation + AH Plus/gutta-percha)</td>
<td>0.000865 ± 0.000272\textsuperscript{b,3}</td>
<td>0.000464 ± 0.000292\textsuperscript{d,4}</td>
<td>&lt; .0001</td>
</tr>
<tr>
<td>Group 3 (EDTA + Hybrid Root Seal/Resilon)</td>
<td>0.000971 ± 0.000321\textsuperscript{b,5}</td>
<td>0.000375 ± 0.000197\textsuperscript{d,6}</td>
<td>&lt; .0001</td>
</tr>
<tr>
<td>Group 4 (laser irradiation + Hybrid Root Seal/Resilon)</td>
<td>0.000996 ± 0.000252\textsuperscript{b,7}</td>
<td>0.000495 ± 0.000186\textsuperscript{d,8}</td>
<td>&lt; .0001</td>
</tr>
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Groups with same superscript letters do not significantly differ (reading vertically) \( (P > .05) \). Groups evaluated in different time periods with same superscript number do not significantly differ (reading horizontally) \( (P > .05) \).
Group 1 (EDTA/VAH Plus/gutta-percha) exhibited the least microleakage, whereas group 4 (laser irradiation/Hybrid Root Seal/Resilon) showed the greatest microleakage at each of the 2 time periods. The mean microleakage values for group 1 and group 4 were significantly different at the both 1-week (P < .0001) and 4-week (P < .01) periods of the study. The mean levels of microleakage in group 2 (laser irradiation/AH Plus/gutta-percha) and group 3 (EDTA/VAH Root Seal/Resilon) were significantly higher than group 1 (P < .0001) at the 1-week period (Table I).

The SEM observations were in accordance with the computerized fluid filtration analysis, and showed that the root canal walls treated with 17% EDTA and filled with AH Plus/gutta-percha combination (group 1) had the best adaptation to the apical (Fig. 2, A) and middle (Fig. 2, B) portions of the root canal walls. The lack of continuity between the filling material and the root canal walls increased and could clearly be seen in group 2 (laser irradiation/AH Plus/gutta-percha) (Fig. 2, C and D), group 3 (EDTA/VAH Root Seal/Resilon) (Fig. 3, A and B), and group 4 (laser irradiation/Hybrid Root Seal/Resilon) in direct proportion to higher microleakage values (Fig. 3, C and D).

**DISCUSSION**

Several methods have been used to evaluate the sealing ability of root canal filling materials. In this study, we used the fluid filtration technique, which has been described as one of the best methods for quantitating microleakage of filling materials or measuring apical seal.28 One advantage of this fluid transport model for studying apical leakage is that root specimens are not destroyed and can be re-measured. The use of positive pressure removed entrapped air or fluid, which may skew the outcomes in dye penetration stud-
Additionally, the computerized fluid filtration method has the advantage that the movement of the air bubble can be observed by laser diodes that are computer controlled rather than visual following.

In light of current findings, we must accept the null hypotheses: Er,Cr:YSGG laser treatment does not influence the sealing ability of the sealers compared with EDTA application. The root canal adaptation and sealing ability of the Hybrid Root Seal/Resilon combination is not superior to that of the AH Plus/gutta-percha combination. The sealing ability of all filling material combinations used in the present study changed significantly after 4 weeks of storage. The decrease in leakage with time suggests that some voids are “dead ends” and that flow decreases as these types of voids are progressively filled with water. Additionally, the reported expansion of AH Plus sealer and gutta-percha by water sorption over time may have facilitated their better long-term sealing ability.

Controversy exists as to whether or not smear layer removal is necessary for root canal filling materials to penetrate dentinal tubules. Many studies have concluded that it is mandatory for its removal to attain proper sealer penetration into dentinal tubules. Er,Cr:YSGG laser was selected for the present study because it has been shown to promote cleaner surfaces when compared with a combination of NaOCl and EDTA, which resulted in filling of a greater number of root canal ramifications by gutta-percha and/or sealer. However, in spite of the absence of smear layer, no significant difference in respect to leakage of root canal fillings between laser-treated and conventionally treated root canals has also been demonstrated previously. In accordance with these studies, Er,Cr:YSGG laser treatment did not influence the sealing ability of the sealers compared with EDTA application in this study. When the results for groups with the same filling material were compared, group 1 (EDTA + AH Plus/gutta-percha) had
higher sealing ability than group 2 (laser irradiation + AH Plus/gutta-percha) at the both periods of the study. In line with this, group 3 (EDTA + Hybrid Root Seal/Resilon) had higher sealing ability than group 4 (laser irradiation + Hybrid Root Seal/Resilon) at both periods of the study. Although the differences were mostly not significant ($P > .05$), it is reasonable to speculate that EDTA performed better than Er,Cr:YSGG laser treatment in terms of apical sealing ability (Table I).

According to Kimura et al., the removal of smear layer and debris by lasers is possible; however, it is hard to clean all root canal walls, because the laser is emitted straight ahead, and in interrupted pulses making it almost impossible to irradiate the lateral canal walls. Additionally, despite the better cleanliness effect, Altundasar et al. demonstrated areas of thermal injury, including carbonization and partial melting when treating canals of permanent teeth with the Er,Cr:YSGG laser.

The most recent studies that have compared Hybrid Root Seal/Resilon root canal fillings to more conventional gutta-percha techniques have shown similar or inferior results, and these authors have concluded that Hybrid Root Seal/Resilon offers no apparent advantage over gutta-percha combined with various sealers. When the present results for groups with the same smear layer removal method were compared, group 1 (EDTA + AH Plus/gutta-percha) had higher sealing ability than group 3 (EDTA + Hybrid Root Seal/Resilon) at both periods of the study. In line with this, group 2 (laser irradiation + AH Plus/gutta-percha) had higher sealing ability than group 4 (laser irradiation + Hybrid Root Seal/Resilon) at both periods of the study (Table I). Consistent with the previous studies, our results based on the fluid filtration method indicate that Hybrid Root Seal/Resilon combination did not provide a better seal than AH Plus/gutta-percha combination.

There are a number of reasons why methacrylate-based sealers may not provide perfect seals: factors such as incomplete resin infiltration into the demineralized dentin and polymerization shrinkage stresses may affect the adaptation and the penetration of the materials and thereby may play an important role in sealing capability.

In the present study, each experimental combination exhibited architecture in SEM that seemed to correlate with its sealing performance. However, these observations should be interpreted cautiously, because gaps observed between the filling material and dentin and cracks within root might be artifacts that caused by vacuum desiccation during SEM examination. Within the limitations of the present study, cracks within the roots detected by SEM analysis could be partially attributable to shrinking artefacts (Fig. 3, B). The separation between the Hybrid Root Seal/Resilon combination and dentin was greater than observed with AH Plus/gutta-percha combination (Figs. 2 and 3). In accordance with this result, a study by Lawson et al. using the push-out test design and scanning electron microscopy analysis showed patent dentinal tubules and the absence of resin-rich zones in samples filled with MetaSEAL (marketed as Hybrid Root Seal in Japan; Parkell, Farmingdale, NY) and gutta-percha combination. Recent studies have also shown that MetaSEAL was not aggressive enough to partially demineralize dentin, even when the smear layer was completely absent.

Sealing capability is one of the main aspects of the quality of root canal sealer. Further investigation of long-term sealing abilities and other features of these products are required.

REFERENCES


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