Polymicrobial Leakage of Four Root Canal Sealers at Two Different Thicknesses

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Abstract

The present study investigated the sealing ability of four root canal sealers at two different thicknesses. There were 82 maxillary incisors roots prepared with Gates Glidden drills up to size 6 and divided into four groups: Pulp Canal Sealer, EndoREZ, Sealapex, and AH Plus. Each group was divided in two subgroups and gutta-percha cylinders 1.5 or 1 mm wide were used, respectively. The roots were mounted in a bacterial leakage model and the system was checked daily during the following 12 wk. Data were analyzed by Log-Rank test and Student *t*-test. In the thin layer samples, the sealers demonstrated similar results while, in the thick layer samples, AH Plus revealed the best performance. Generally, greater sealer thickness influenced negatively the sealing ability of the root canal filling, except in AH Plus samples. *(J Endod 2006;32: 998–1001)*

Key Words

Bacterial leakage, root canal sealer, sealer thickness

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Canal filling in root canal treatment is used to prevent any communication between the oral cavity and the periapical tissues. The ingress of oral or tissue fluids via such communication may maintain the viability of residual bacteria that survive the treatment (1). Although sealers enhance sealing ability, the optimal obturation aims at maximizing the volume of the core material while minimizing the amount of sealer between the inert core and the canal wall (2–5). In contrast to gutta-percha, which is chemically and dimensionally stable, the areas filled by sealer are more vulnerable because it can dissolve over time (4, 6). However, laboratory studies have shown that gutta-percha seals significantly better when used in combination with a sealer (7). Consequently, the sealing quality of a root canal filling depends strongly on the sealing ability of the sealer used (8).

The relationship between sealer thickness and the quality of apical sealing has been previously studied (6, 8). It is worth mentioning that, to the best of the authors' knowledge, no experimental procedure using a bacterial tracer has been reported in the literature. Hence, the purpose of the present study was to investigate the sealing ability of four root canal sealers with two different thicknesses using a polymicrobial leakage model.

Materials and Methods

Instrumentation

A sample of 82 straight maxillary central incisors was selected. Crowns were removed at the cemento-enamel junction with a #4138 diamond bur (KG Sorensen, Zenith Dental ApS, Agerskov, Denmark). The root tips were removed in the same way, leaving roots 8 mm in length. The root canals were then prepared with Gates Glidden drills (Maillefer Instruments SA–CH 1338, Baillaigues, Switzerland) up to size 6 (ISO size 150). At this point, all prepared root canals were straight, round in cross-section and measured 1.5 mm in diameter. A final irrigation with 5.25% NaOCl was used.

Gutta-Percha Cylinders

Thirty-six gutta-percha cylinders 8 mm high × 1.5 mm wide were made by injection of thermoplasticized gutta-percha (Obtura II System, Obtura Corp., Fenton, MO) into the lumen of a metal mold with the same diameter (6). After cooling, the formed gutta-percha cylinder was pushed out of the mold. In the same manner, another set of 36 gutta-percha cylinders 8 mm high × 1 mm wide was prepared using a 1-mm diameter mold.

Obturation and Sample Grouping

There were 72 teeth randomly divided into four groups (n = 18 teeth *per* group):

A. Pulp Canal Sealer EWT (Kerr Corp., Orange, USA);
B. EndoREZ (Ultradent Products, Inc., South Jordan, UT);
C. Sealapex (Kerr Corp., Orange, USA);
D. AH Plus (Dentsply DeTrey Gmbh, Germany).

Each group was divided into two subgroups (n = 9 teeth *per* subgroup). A predetermined amount of sealer (1.5 cm^3) was put into the root canals with a #80 endodontic file (Maillefer Instruments SA). After every two roots filled, a fresh mix of sealer was used. All sealers tested were prepared following proportions and recommendations of the manufacturers. The filled roots were stored at 37°C and 100%

humidity for 14 days to guarantee setting of the sealer. The specimens in subgroups 1 (A1, B1, C1, and D1) were filled with gutta-percha cylinders 1.5 mm in diameter. Because this cylinder just fitted the size of the canal, a slight pressure was applied to accommodate the gutta-percha, leaving a very thin layer of sealer. The specimens in subgroups two (A2, B2, C2, and D2) were filled with gutta-percha cylinders 1 mm in diameter, that were carefully inserted in the center of the canals.

Five teeth with standard access and patent apical foramens served as a positive control group, and five healthy teeth served as a negative control group. Two coats of nail varnish were applied on the external surface of all teeth, except on the apices and coronal ends. In the negative control group, teeth were completely covered with nail varnish.

Checking Sealer Thickness

Three samples randomly selected from each subgroup were sectioned horizontally at 3 mm from the apical foramen using a low-speed saw. The metallographic preparation was performed as previously described (9). Samples were examined under a light optical microscope (Carl Zeiss Vision, Hallbergmoos, Germany) and the images submitted to computer-assisted evaluation. The Axiovision 4.0 program (Carl Zeiss Vision) was used for image analysis and processing; sealer thickness was measured at four different sites. The measurements obtained were repeated twice to ensure reproducibility.

Polymicrobial Leakage

The apparatus used to evaluate bacterial leakage was modified from that described previously (10). Briefly, 10-ml glass assay tubes (BD Vacutainer, Juiz de Fora, Minas Gerais, Brazil) with rubber stoppers were adjusted for use in this experiment. By using a heated instrument, a hole was made through the center of every rubber stopper in which a cylinder prepared from insulin syringes (BD Vacutainer) was inserted. The tooth crown was tightly fitted into a rubber tube that was fixed using cyanoacrylate. Syringe cylinders were then adapted on the other side of the rubber tube to create a reservoir for saliva. Cyanoacrylate was applied in all junctions of the system.

The testing apparatus was sterilized overnight in ethylene oxide gas (BIOXXI Esterilization Services Ltd., Rio de Janeiro, Brazil). The set up was made in a laminar airflow hood (Bioprotector Plus 09, Veco, Campinas, São Paulo, Brazil), where the glass assay tubes were filled with 3 ml sterile Brain Heart Infusion (BHI–Oxoid Ltd., Basingstoke, UK), so that ≈2 mm of the resected root was immersed in the broth. The whole apparatus was incubated at 37°C for 4 days to ensure sterilization.

Afterwards, the reservoirs were filled with human saliva (20 ml) mixed in BHI broth in a 1:1 (v/v) ratio and replenished every 3 days (11). Human saliva was collected from one individual and the volunteer did not brush for at least 12 h before collection (12). The system was incubated at 37°C and checked daily for the appearance of turbidity in the BHI broth during the following 9 wk.

Statistical Analysis

The Log-rank test was used to analyze the leakage data (13). The Student *t*-test was used to compare results for different thicknesses for each sealer. The level of significance in all tests was set at $p < 0.05$. SPSS (Statistics 4.0, SPDD, Gorinchem, The Netherlands) was also used as analytical tool.

Results

Sealer Thickness

In samples with a thin layer, sealer thickness varied from 6.35 to 91.35 μm. In samples with a thick layer, sealer thickness varied from 91.36 to 385.87 μm. An image with the sealer thickness measured is showed in Fig. 1. The wide dispersion of the thickness value is mainly because of the difficulty in centering the gutta-percha cylinder, as seen in Fig. 1*B*. No significant differences (*t*-test, $p > 0.05$) among the groups presenting similar sealer thickness were found.

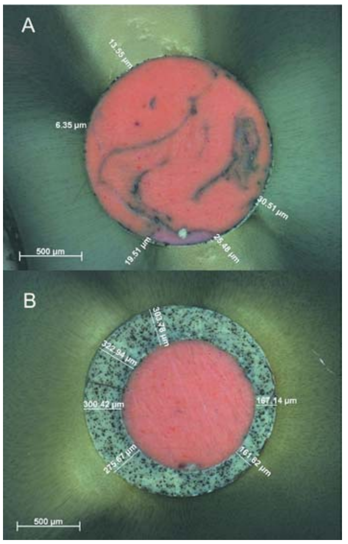

Figure 1. (*A*) Thin layer of Pulp Canal Sealer sample with sealer thickness varying from 6.35 μm to 30.51 μm. (*B*) Thick layer of Pulp Canal Sealer sample with sealer thickness varying from 161.82 μm to 322.94 μm.

Leakage Analysis

No growth was observed when checking the sterilization of the whole apparatus. No contamination was found in any sample of the negative control group. All specimens of the positive control group showed broth turbidity until the third day of incubation. The data were analyzed searching for differences between different sealers and different sealer thicknesses. The Log-rank test was used to analyze the leakage data for all groups after 3, 6, and 9 wk. The leakage pattern of each group is shown in Table 1.

The following results were found for the thin layer subgroups: After 9 wk, bacterial leakage was observed in five samples (55.6%) of Pulp Canal Sealer, four samples of EndoREZ (44.4%), five samples of Sealapex (55.6%), and six samples of AH Plus (66.6%). Statistical analysis showed significant differences among Sealapex, Pulp Canal Sealer, and EndoREZ ($p < 0.05$), but only after 9 wk, and not after 3 or 6 wk.

The following results were found for the thick layer subgroups: After 9 wk, bacterial leakage was observed in nine samples (100%) of Pulp Canal Sealer, eight samples (88.9%) of EndoREZ, nine samples (100%) of Sealapex, and four samples (44.4%) of AH Plus. Significant differences were found between AH Plus and Pulp Canal Sealer as well as AH Plus and Sealapex ($p < 0.05$), but only after 9 wk, and not after 3 or 6 wk.

For all groups, except AH Plus, thick layers showed more bacterial leakage than thin layers after 3, 6, and 9 wk ($p < 0.05$).

Discussion

The Pulp Canal Sealer group demonstrated a high incidence of infiltration. Samples were prepared similarly to a study by Georgopoulou et al. (8) that used a fluid transport method and found similar results for this sealer in particular. However, other studies have found that Pulp Canal Sealer gave less infiltration (14, 15). This difference might be attributed to the obturation technique. In the present work, a

TABLE 1. Summary tables of statistical results

3 WEEKS

	GA1	GA2	GB1	GB2	GC1	GC2	GD1	GD2
GA1	55.6%	X						
GA2	X	100%						
GB1			22.2%	X				
GB2			X	88.9%				
GC1					33.3%	X		
GC2					X	100%		
GD1							55.6%	
GD2								44.4%

6 WEEKS

	GA1	GA2	GB1	GB2	GC1	GC2	GD1	GD2
GA1	55.6%	X						
GA2	X	100%						
GB1			44.4%	X				
GB2			X	88.9%				
GC1					33.3%	X		
GC2					X	100%		
GD1							55.6%	
GD2								44.4%

9 WEEKS

	GA1	GA2	GB1	GB2	GC1	GC2	GD1	GD2
GA1	55.6%	X	X					
GA2	X	100%						X
GB1	X		44.4%	X	X			
GB2			X	88.9%				
GC1			X		55.6%	X		
GC2					X	100%		X
GD1							66.6%	
GD2		X				X		44.4%

Percent values: fraction of samples in the respective subgroup that showed leakage. *X marks:* significant difference ($P < 0.05$) between respective subgroup pairs. *Grey cells:* no significant difference ($P \geq 0.05$) between respective subgroup pairs. *White cells:* untested subgroup pairs.

cylinder of gutta-percha was only inserted into the root canal, without any compaction or condensation.

The Sealapex group also presented high levels of infiltration even though some studies have related good results with this sealer (16, 17). Some studies demonstrated that Sealapex presented good sealing ability at first, but very poor sealing after being stored in water for a long time (6, 8). Initial satisfactory results may be related to the volumetric expansion after setting, whereas loss of sealing ability may be related to sealer dissolution over time (5, 18, 19). Sonat (20) has described similar results. Siqueira et al. (10) observed that liberation of hydroxyl ions is rapid but limited and seems to be related to sealer solubility and disintegration in an aqueous environment. Hence, it seems that the high solubility of Sealapex is a determinant factor in microleakage control.

Resin-based sealers have shown good results both in the present study and in other investigations, demonstrating high-quality properties (18, 21, 22). Even after long periods of immersion in water, resin-based sealers have shown good sealing ability (8, 19, 23). It is believed that such good performance is because of low solubility of these materials (10, 24). In the present work, AH Plus presented the best results, in agreement with Timpawat et al. (25). These authors observed that although sealing ability of AH Plus decreased after 14 days, it still presented the best results. Several investigations using AH Plus have demonstrated good properties such as lower solubility (23, 26, 27).

EndoREZ has been recently introduced in the market with very few reports about its properties. As in the present study, Zmener (28) also related good performance when comparing EndoREZ with other resin-based sealers. Sevimay and Kalayci (29) compared AH Plus and EndoREZ, the former showing better sealing ability and adaptation to dentine walls.

The present study, as well as some another papers (6, 8), has demonstrated that a thicker layer of sealer negatively influences sealing ability. The only exception was for AH Plus, which did not present significant differences when comparing different thicknesses of sealer. This may be explained by characteristics inherent to the sealer such as volumetric expansion and high antimicrobial activity (7, 21, 25).

The better sealing ability of AH Plus in the thick layer was also observed in similar studies using resin cements. It was related to the shrinkage stress of the resin cement could be reduced by flow of the material when in a thick layer because the transition from paste to solid took 34 h. When the layer of the material was very thin, however, such relief by flow may be limited (6, 8).

In conclusion, in the thin layer groups, the sealers demonstrated similar results and in thick layer groups, AH Plus revealed the best performance. Overall, greater sealer thickness negatively influenced sealing ability of the root canal filling, except in AH Plus samples.

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