

Microbial Leakage of Cavit, IRM, and Temp Bond in Post-prepared Root Canals Using Two Methods of Gutta-percha Removal: *An In Vitro Study*

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Abstract

The aim of this study was to evaluate the integrity of the coronal seal of Temp-Bond and compare it to Cavit and IRM after post space preparation using *S. faecalis* as a microbial tracer. In addition, the affect of two methods of gutta percha removal on the apical seal of root canal fillings was also evaluated. Forty extracted human single rooted teeth were prepared chemomechanically and obturated with gutta percha and AH26 sealer cement using the lateral cold condensation technique to a standardized working length of 15 mm. About 10 mm of the coronal gutta-percha was removed with either Peeso-reamer or a hot plugger. The roots were divided into three experimental groups of 10 roots and a control group. Each experimental group was subdivided equally into two groups of 15 each according to the method of post space preparation. Cavit, IRM, and Temp-Bond were used to seal the access opening. Each root was fixed in a cuvette containing Tryptic Soya Broth which, covered 2 mm of the root apex. Bacterial suspension was introduced through pipette. Fresh bacterial suspension was added every week, and the system was monitored daily for the growth of microorganisms for a period of one month. The results showed there was no significant difference in terms of coronal leakage between the three coronal materials used ($P=0.478$), but the methods of gutta-percha removal did have an impact on the apical leakage ($P=0.047$). The mean value showed the Peeso-reamer provided less leakage compared to using a hot plugger during the 30-day experimental time period. It was

concluded the temporary type of coronal seal of endodontically treated teeth will not prevent coronal leakage if left for a long period of time. In addition, permanent cementation of the post with the coronal restoration should be carried out as soon as possible to prevent recontamination of the root canal.

Keywords: *S. faecalis*, leakage of Cavit, IRM and Temp-Bond, gutta-percha removal

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Introduction

Proper coronal restoration is strongly recommended after root canal therapy.¹ This is usually done to prevent tooth fracture or microbial leakage, which will lead to therapy failure. Ray and Trop² had reported bacteria and bacterial products could penetrate the marginal gap of a leaky restoration and the interface between the root filling and the canal wall to reach the periapical region.

Endodontically treated teeth often lack sufficient support for a permanent restoration, where additional retention through the root canal is highly recommended. Traditionally, a post and core has been used for this purpose. In order to create space for a post part of the root canal filling material must be removed. The technique used to remove the filling should be safe, efficient, and not to disturb the apical seal. Three techniques are commonly used to remove gutta percha: (a) chemical, using a solvent such as chloroform, (b) thermal, using hot endodontic pluggers, and (c) mechanical, using a rotary instrument such as a Gates Glidden or a Peeso-reamer. However, reported studies have shown no significant difference in the amount of the apical leakage when the post space is prepared with, heated endodontic plungers, Peeso-reamers, or Gates Glidden burs.^{3,4}

It was reported the preparation of the root canal for post space might affect the rate and extent of canal contamination.^{5,6} The oral-micro flora present in saliva can inadvertently infiltrate during post space preparation and impression procedures. The empty canal space, coronal to the root canal filling material, may provide a suitable environment for promoting bacterial growth. A temporary coronal restoration such as

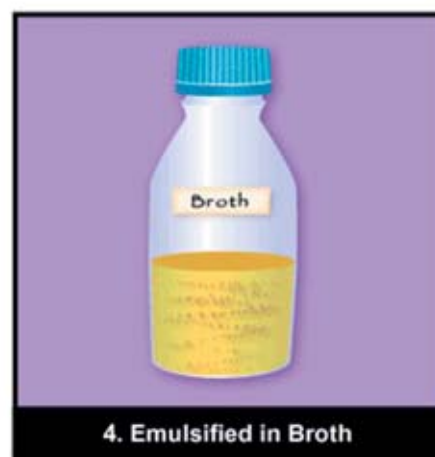
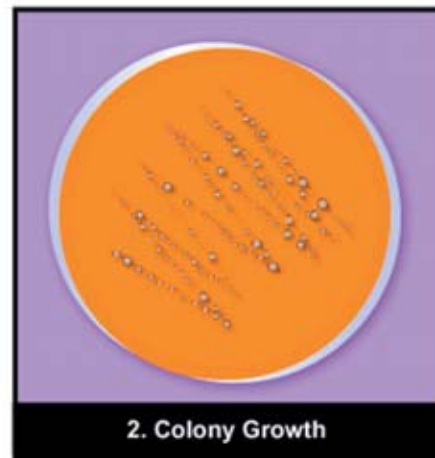
Cavit, IRM, or a temporary crown cemented with temporary filling material, such as Temp Bond, is usually used until the final cast post is ready for cementation. These temporary coronal materials are soluble and could leak irritants into the apical filling material. Thus, the integrity of the coronal seal is as important as that of the apical seal after post preparation.

The seal provided by Cavit and IRM has been investigated using dye, radioisotope, and the bacteria leakage test.^{7,8,9} There is no study yet reported to investigate the sealing ability of Temp Bond after post space preparation. The aim of the present investigation was to evaluate the integrity of a coronal seal of Temp-Bond and compare it to Cavit and IRM materials after post space preparation using *S. faecalis* as a microbial tracer. In addition, the effect of two methods of gutta percha removal (hot plugger vs. Peeso reamer) on the apical seal was also evaluated.

Materials and Methods

Microorganism and Culture Media

The log phase culture of *S. faecalis* (ATCC 29212) was obtained from the Microbiology Laboratory of the King Faisal Specialist Hospital and Research Center, Riyadh, Saudi Arabia. Tryptic Soy Broth (Difco Laboratories, Detroit, MI, USA) was prepared according to the manufacturer's directions. Bacterial culture was streaked on Tryptic Soy agar (Difco Laboratories) to obtain single colony, which were re-streaked on fresh agar plates to obtain the fresh bacterial cultures in log or late-log phase of growth. The identity of this strain was verified on several occasions by doing a Gram staining. The growth of this organism was scrapped off the agar plate and emulsified in broth.



Preparation of the Teeth

Forty extracted mandibular premolars were used in this study. The teeth were extracted for orthodontic reasons. They were stored in 1% sodium hypochlorite solution (NaOCl) at room temperature until used. Teeth with cracks, root resorption, open foramen, calcified, and curved canals were excluded. The anatomic crowns of each tooth were resected at the level of CEJ with metal disk in a straight, slow speed hand piece under water irrigation. All roots were then shortened, to the standardized root length of 15 mm, with a high-speed hand piece using a straight diamond bur under water irrigation. The working length was designed as 1 mm shorter from a point at which #15k file passes the apical foramen. All the canals were instrumented to size 40k, irrigated with 1% NaOCl between each file size, and dried with paper points. Canals were obturated with gutta percha and AH26 “silver free”

as sealer cement (De Tray Dentsply, Milford, DE, USA) using the lateral cold condensation technique. Teeth were instrumented and obturated in the same manner by two investigators. The roots were radiographed from proximal view to confirm adequacy of obturation. The specimens were kept in a sealed jar that contained 2x2 inch sterile gauze pads soaked with physiological saline solution for 2 weeks to ensure 100% humidity and to allow complete setting of the sealer cement.

About 10 mm of coronal gutta-percha was removed either incrementally with a hot plugger or in one piece with a size 3 Peeso reamer, leaving about 5 mm of this material at apical third. Radiographs were taken to ensure the adequacy of the apical seal after post space preparation.

According to the coronal materials, roots were divided randomly into three experimental groups (10 roots each). Each experimental group was subdivided equally into two groups according to the method of post space preparation (5 prepared with a Peeso reamer and the other 5 with a heated plugger). Six roots were used as a positive control. Four roots were used as the negative control. A small sterile cotton pellet was placed 3 mm deep into the canal to act as a base for the coronal materials. In group 1 the orifice of the root access opening was closed with 3 mm of Cavit (ESPE America, INC., Norristown, PA, USA), group 2 with IRM (L.D., Caulk Division Milford, DE, USA), and group 3 with Temp Bond (Kerr, Italia, S.P.A). The coronal materials were mixed and/or placed according to the manufacturer's instructions. They were placed incrementally into the orifice of the root with a plastic instrument and the excess was removed. The orifice of the access opening in the positive control group did not receive any coronal material. On the other hand, the orifice of the root access opening of the negative control was closed with wax. The root surface, including the orifice and the apical foramen, were completely sealed with two layers of nail polish. The teeth in the three experimental groups and the positive controls received double layers of nail polish leaving the area of the canal's orifice and the apical foramen exposed.

Design and Construction of Model

Coulter cell counting cuvettes (Beckman-Coulter, Fullerton, CA, USA) were used as a base for this model. These cuvettes are made of polystyrene with a snap-on lid made of polypropylene. Three circular holes (3, 5, and 9 mm in diameter) were made, respectively, using a high-speed headpiece and a 171 tapered carbide bur. The 9 mm hole was made in the center of the lid, while the other two were made around it. A 5 mm piece of 5-ml plastic pipette was introduced in the 9 mm hole with 2-cm portion sticking outside of the lid. The inside end of this pipette piece was glued to the coronal part of the root of the tooth. The upper portion of this pipette was plugged with sterile gauze. The 5 mm hole was used to introduce a 7 cm long tubing from a 22-gauge butterfly extending to the bottom of the cuvette, while the leur-lock connection was left closed on the top of the lid. This connection was used for a weekly exchange of growth medium

using a sterile syringe. The tubes were glued very well to the lid with epoxy so there was no leakage. An epoxy resin was used as glue in this study due to its viscosity that allows easier visualization and, thereby, assuring complete coverage.¹⁰ The third hole (3 mm in size) was plugged with sterile cotton to serve as a ventilator and to keep uniform pressure inside and outside the cuvette.

Each model apparatus was sterilized in the Gamma Facility of King Faisal Specialist Hospital and Research Center by giving a dose of 25 Kilo Gray. To assure sterility, transfer of culture medium and bacterial inoculations were carried on in a Class II Biological Safety Cabinet. A volume of 7-8 ml of Tryptic Soya Broth media was added to the bottom of the cuvette so a minimum of 2 mm of the apical part of each root was immersed in it (Figure 1).

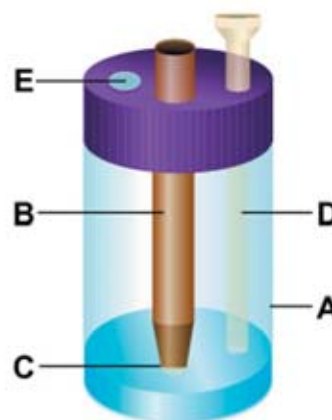


Figure 1. Schematic drawing of model apparatus, A= cuvette, B= pipette, C= root specimen, D= tubing, E= ventelature.

The bacterial suspension of the *S. faecalis* was introduced in the pipette piece placed in middle of the lid. After inoculation, the top part of this tube was closed with sterile gauze and incubated at 37°C with 50% relative humidity. Fresh bacterial suspension was added every week into this cavity. The system was monitored daily for the growth of microorganisms for a period of one month. The bacteriological medium at the bottom of the cuvette was checked visually on a daily basis for turbidity. If turbidity is noticed, then the identity of the microorganism was verified by microscopic observations and Gram staining.

Statistical Analysis

The data was entered into a PC using the FoxPro data base computer program. Statistical Package for Social Science (SPSS version 10) was utilized for statistical analysis. Parametric two-way analysis of variance (ANOVA) was utilized to test the main factors (the temporary filling material and methods of gutta-percha removal) and the interaction between them. The significance level was set at 0.05.

Results

Leakage was demonstrated in four specimens out of six (67%) of the positive controls. All but one negative control (75%) demonstrated no leakage for the entire 30-day experimental time period.

Cavit provided a better coronal seal compared to Temp Bond and IRM. Statistical analysis using two-way ANOVA showed no significant difference in terms of coronal leakage between the three coronal materials ($P=0.478$).

There was a significant difference in apical leakage between the two methods of gutta-percha removal ($P=0.047$). The mean value showed Pecho-reamer provided less leakage compared to hot plugger (Figure 2).

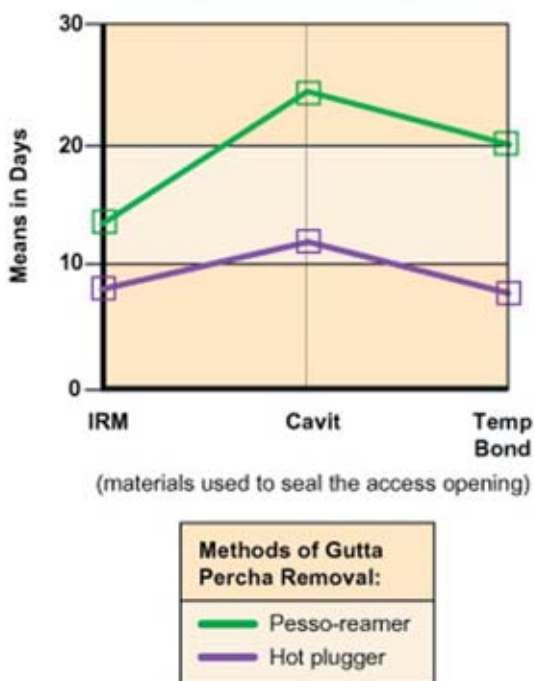
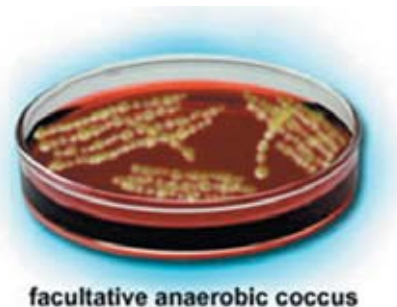


Figure 2. Line graph represents time periods at which the three materials start to leak using two methods of gutta-percha removal.

Discussion

Many *in vitro* methods have been used to evaluate the sealing quality of endodontic filling materials. The most frequent methods used are dye penetration or radioisotopes techniques. These molecules are smaller than bacteria and may not simulate *in vivo* conditions.^{11, 12} *S. faecalis* (facultative anaerobic Gram +ve coccus) was chosen for use in this study because it is often involved in persistent endodontic infections¹³ and is one of the most resistant species found in the oral cavity, having the ability to survive under unusual environmental stresses.¹⁴



Lateral cold condensation of gutta percha (GP) with sealer was used in this study since this has been the most popular root canal filling technique.¹⁵ In addition other studies^{4, 16} have showed alternate obturation techniques yielded no statistical difference in apical leakage provided 4 to 5 mm of GP remained at the apex. In this study and during post space preparation 5 mm of gutta-percha was left at the apical third of the canal to comply with the recommendation of several investigators.^{16, 17, 18}

A cotton pellet was placed about 3 mm deep into the canal to provide adequate space for the coronal materials and to comply with the recommendation of Webber et al.¹⁹, who found a 3.5-mm thickness of Cavit was the minimum thickness necessary to prevent total leakage of the dye molecule.

The model used in this study was almost similar to the one used by Kayat et al.²⁰ with a slight modification. This model system and others could not address many *in vivo* conditions. However, all used models were consistent in their findings as to the potential for substance penetration of filled canals. Indeed, some *in vivo* variables such

as the influence of periapical tissues and fluids, thermal oral changes, intraoral pressure, diversity of oral flora, and nutrients available could not be addressed in the used model system. However, the model gave partial stimulation of the complex and dynamic nature of the oral cavity. This was achieved by adding fresh bacterial suspension and growth medium every week through the pipette.

The result of the present study indicated the method of gutta-percha removal has a greater impact on the apical leakage than the type of the coronal restoration. Removal of gutta-percha with Peeso-reamer resulted in less disturbance of the apical seal than the removal with hot plugger. Similar results have been reported in a previous study.²¹ The frictional heat produced by rotary instruments plasticized the gutta-percha and removed it without excessive pull. In addition, slight apical pressure during the removal of gutta-percha may act as vertical condensation, which might improve the apical seal.

An increase in the apical leakage, when hot plugger was used to remove the gutta-percha, could be due to the shrinkage of the material. This result contradicted Haddix et al.²², who indicated removal of gutta-percha with a warm instrument results in less disturbance of the apical seal than removal with the mechanical methods.

Most of the samples in the positive control group showed leakage in 2 to 6 days, except for two samples which did not show evidence of leakage until the end of the experimental period. There are two possible technical errors which could have caused the latter, either these samples may have been switched by mistake with one of the negative controls or the canal orifice and/or the apical foramen of these two samples were sealed completely with nail polish. Although there was no significant difference in terms of coronal leakage between the three coronal materials used, the mean values showed that Cavit provided a higher coronal seal compared to IRM. The favorable sealing characteristic of Cavit in endodontic access preparations has been previously reported.^{7, 8, 23} This property of

Cavit may be attributed to its relatively higher linear expansion resulting from water absorption during setting.²⁴ This expansion enhances the contact between the material and cavity, which will improve the seal. The other reason might be that the material is premixed and it reduces the inconsistencies related to chair side mixing. IRM showed the lowest coronal seal, and the inability of IRM to prevent microleakage has been reported.^{8, 9, 23} The poor sealing ability of IRM may be linked to the fact powder and liquid have to be mixed together to produce the paste to be inserted, this mixing is the cause of reduced homogeneity.

Temp Bond consists of 10 parts zinc oxide and one part eugenol and is usually used as a temporary crown and bridge cement. The relatively better sealing ability of this material compared to IRM could be due to the form of the material. Temp Bond is available as a "paste-paste" system and the mixing of the two paste ingredients produces a more homogenous mix. If a temporary crown is cemented with Temp Bond, then the coronal sealing would be much better. However, in this study Temp Bond was used as coronal material without a temporary crown to mimic some of the clinical cases in which the temporary crown fell off and only remnants of such cement were left in the coronal part of the root canal.

Conclusion

Within the parameters of this *in vitro* study, the following conclusions can be drawn:

1. The temporary type of coronal seal will not prevent the coronal leakage if left for long periods of time.
2. The apical seal of the root canal filling was not affected adversely by the type of the coronal restoration, but rather with the methods of post space preparation.
3. Peeso-reamer provided a significantly better apical seal compared to hot plugger when used for post space preparation.
4. The need for an immediate and proper coronal restoration after post space preparation is reinforced.

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