

Journal section: *Clinical and Experimental Dentistry*
Publication Types: *Research*

doi:10.4317/medoral.16.e556
<http://dx.doi.org/doi:10.4317/medoral.16.e556>

An in vitro evaluation of two dentine adhesive systems to seal the pulp chamber using a glucose penetration model

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González-Castillo S, Bailón-Sánchez ME, González-Rodríguez MP, Poyatos-Martínez R, Ferrer-Luque CM. An in vitro evaluation of two dentine adhesive systems to seal the pulp chamber using a glucose penetration model. *Med Oral Patol Oral Cir Bucal*. 2011 Jul 1;16 (4):e556-60.
<http://www.medicinaoral.com/medoralfree01/v16i4/medoralv16i4p556.pdf>

Received: 13/03/2010
Accepted: 25/06/2010

Article Number: 16980 <http://www.medicinaoral.com/>
© Medicina Oral S. L. C.I.F. B 96689336 - pISSN 1698-4447 - eISSN: 1698-6946
eMail: medicina@medicinaoral.com

Indexed in:

Science Citation Index Expanded
Journal Citation Reports
Index Medicus, MEDLINE, PubMed
Scopus, Embase and Emcare
Indice Médico Español

Abstract

Objectives: To evaluate the sealing capability of Cavit™ G with or without Clearfil™ S3 Bond and Prime & Bond NT placed in the pulp chamber.

Study Design: Forty single rooted premolars, extracted for orthodontic and periodontal reasons, with intact coronal surface and mature apices, were standardized to a length of 15 mm. The teeth were instrumented, filled with a gutta-percha master cone and divided into three groups to obturate the pulp chambers: Cavit™ G; Clearfil™ S3 Bond plus Cavit™ G and Prime & Bond® NT plus Cavit™ G. A glucose leakage model was used for evaluating the coronal microleakage. The Mann-Whitney test was used to evaluate the differences in the means of the glucose leakage.

Results: An increase in glucose penetration was observed during the first week in groups Cavit™ G and Cavit™ G+PBNT. The glucose penetration values of all groups were similar at 30 and 45 days, and there were no significant differences among them in both time periods ($p=0.736$ and $p=0.581$, respectively).

Conclusions: The adhesive systems did not improve the capability of Cavit™ G to seal the pulp chamber over time.

Key words: Cavit™ G, Clearfil™ S3 Bond, Prime & Bond® NT, coronal leakage, glucose penetration model, pulp chamber.

Introduction

Different studies have shown that coronal microleakage is a significant factor in the prognosis of root canal treatment (1,2). The amount of coronal leakage that occurs within a relatively short time (3 days) should be considered as a potential etiological factor in root canal treatment failure (3). Therefore, the sealing capability of temporary restoratives plays a key role in shielding the pulp chamber and root canal from bacterial infection (4). This is especially true for root canal treatments involving multiple visits, since infection during treatment can delay both healing and the conclusion of root canal therapy. Magura et al. (5) suggested retreatment of filled root canals exposed to the oral cavity for three or more months.

Various materials have been tested for providing a coronal barrier to prevent microleakage in root canal therapy (4, 6-10), though most studies have focused on the sealing capability of IRM[®], Cavit[™] or similar materials (7,8). None of them was able to avoid coronal leakage completely, but Cavit[™] demonstrated a better sealing capacity of the coronal access (9,11).

The use of dentin adhesives in sealing pulp chamber walls (12) has been evaluated and compared with temporary restoration materials as a secondary barrier to prevent microfiltration within the pulp chamber (7,13). Previous studies do not evaluate the capacity of adhesive agents used together with temporary restoration materials for sealing the pulp chamber.

The purpose of this study was to evaluate capability of Cavit[™] G to seal the pulp chamber, used alone, with Clearfil[™] S3 Bond or Primer & Bond[®] NT adhesive systems, at 24 hours, 7, 30 or 45 days.

The null hypothesis was that the sealing capability of Cavit[™] G used as the only filling material would be no

different than Cavit[™] G used with Clearfil[™] S3 Bond or Primer & Bond[®] NT, at 24 hours, 7, 30 or 45 days.

Materials and Methods

Selection and preparation of teeth

Forty recently extracted human maxillary premolars, extracted for orthodontic and periodontal reasons, with intact coronal surface and mature apices, were used in this study. Soft tissues and calculus were removed mechanically from the teeth and were stored in 2% thymol solution at room temperature before testing.

To ensure uniformity in tooth length, the samples were standardized to a length of 15 mm. First, the crowns of the teeth were cut to leave 4 millimetres in coronal length from the enamel-cementum junction using an Accutom-50 diamond cutter (Accutom Hard Tissue Microtome, Struers, Denmark) under running water (Fig. 1-2°). Coronal access was achieved using high-speed fissure diamond burs under water cooling. The cavity access size was standardized to 2.5 mm in width, 3.5 mm in length and a depth of 4 mm from the cavo-surface margin (Fig. 1-3°).

Instrumentation and obturation of root canals

The working length was established by placing #15 K-Flexofile file (Dentsply Maillefer, Ballaigues, Switzerland) into the canal until the tip was visible at the apical foramen, then subtracting 1 mm (Fig. 1-4°). The canals were prepared sequentially with #15-40 K-Flexofiles (Dentsply Maillefer) and coronal flaring was accomplished with Gates Glidden burs, sizes 2 and 3 (Dentsply Maillefer), to create a uniform canal size and to overcome the variation in natural morphology. The root canals were flushed with 2 mL of 2.5% NaOCl solution between files. After root canal preparation, each specimen was rinsed with 5 mL of 25% citric acid for 2 min

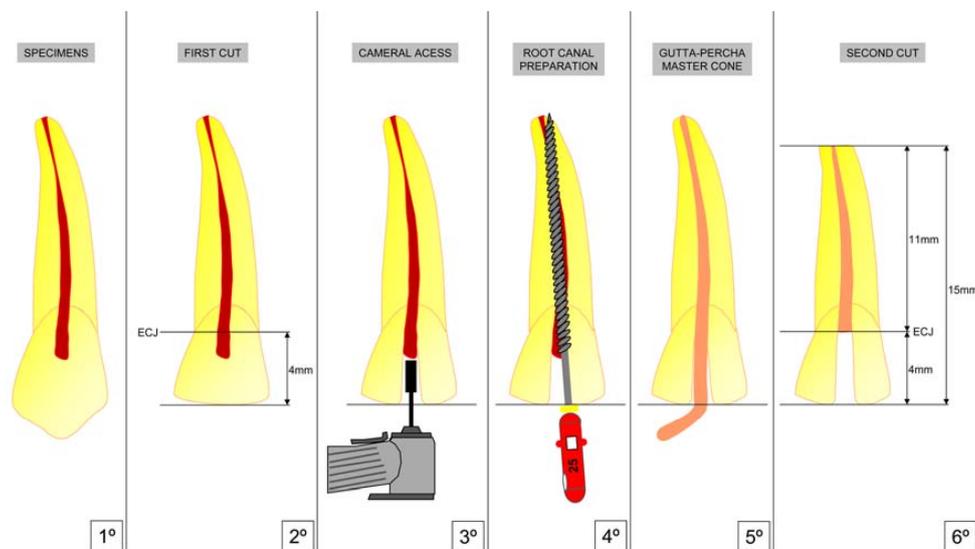


Fig. 1. Diagram showing tooth preparation.

Table 1. Glucose leakage results in mmol L⁻¹ at different time periods (mean and standard deviations).

Materials	Glucose leakage			
	1 day	7 days	30 days	45 days
Cavit™ G	0.14±0.09	4.61±4.17 ^a	7.08±3.45	9.76±0.01
Cavit™ G + Clearfil™ S3 Bond	0.11±0.00	1.21±3.03 ^{a,b}	8.56±1.27	9.46±0.65
Cavit™ G + Primer & Bond® NT	0.16±0.10	4.33±4.72 ^b	8.09±2.31	9.76±0.01

Read vertically, the same letter indicates presence of significant differences.

to remove the smear layer, and then a final flush with 5 mL of deionized water.

The canals were dried with paper points (Dentsply Maillefer) and filled with a gutta-percha master cone (Dentsply Maillefer) without sealer (Fig. 1-5°). Excess gutta-percha master cone was cut with a heated instrument and vertical pressure was applied with standard endodontic pluggers. A periodontal probe was used to measure the depth of the opening to ensure it could accommodate at least 4 mm of the temporary filling material. Approximately 11 mm of the root length apical to the cementum-enamel junction was left intact, and the apical part was sectioned and removed (Fig. 1-6°).

Access cavity filling

The teeth were randomly divided into three experimental groups (n= 10) and two control groups (n= 5).

Group 1: the cavities were obturated with Cavit™ G (3M ESPE Seefeld, Germany). The material was placed incrementally in the access cavity with a plastic instrument, condensed with a plugger, and the excess material was removed with a sterile cotton pellet lightly dampened with sterile saline.

Group 2: the pulp chambers were treated with Clearfil™ S3 Bond (CS3B) (Kuraray Medical Inc., Okayama, Japan) for 20s, gently air-dried for 3 to 5s, and then cured with a visible light activator (Bluephase Ivoclar/Vivadent Schaan, Liechtenstein) for 10 seconds. Afterwards, Cavit™ G was placed in the coronal access in the same manner as in the first group.

Group 3: the specimens were etched with 37% phosphoric acid for 15s, washed for 10s and gently air-dried for 5s. A thin layer of Primer & Bond® NT (PBNT) (DeTrey/Dentsply) was applied for 20s with a brush and light-cured for 10s. Cavit™ G was placed in the coronal access as in the previous groups.

After, all teeth were stored in relative humidity for 48h to allow the materials to set. In the positive control

group (n=5), the teeth were instrumented and filled with gutta-percha without sealer, and the coronal accesses were not obturated with temporary restorative material or any adhesive system. In the negative control group (n=5), the intact teeth were not treated and they were covered with two layers of nail varnish.

Glucose penetration model

A glucose penetration model (14) was used for the quantitative evaluation of leakage. A total of 100µL of solution was drawn from the glass bottle using a micropipette at 1, 7, 30 and 45 days. After drawing the sample, 100 µL of sterile water was added to the glass bottle reservoir to maintain a constant volume of 2 mL. The samples were then analyzed with Glucosa Kit (Glucosquant Glucose/HK, Roche/Hitachi 917/ACN 549. Roche Diagnostics, Basel, Switzerland) in a Roche/Modular P: ACN 668 autoanalyzer (Roche Diagnostics, Basel, Switzerland) at a wavelength of 340 nm. The results of leakage were calculated as mmol L⁻¹ at each time interval following filling.

Statistical analysis

Mean and standard deviations of glucose penetration were determined for each group. The Shapiro-Wilk test was used to assess the distribution of the data. Given that the results for each group did not follow a normal distribution, the Mann-Whitney U test was used for pairwise comparisons. The level of statistical significance was set at P<0.05. All statistical analyses were performed by means of SPSS 15.0 software (SPSS Inc, Chicago, IL.).

Results

The results are given in table 1 and fig. 2. All positive controls showed the highest levels of glucose leakage at 24 hours. The negative control group showed no glucose leakage at any of the evaluated times. The lowest glucose level for which the current procedure proved ef-

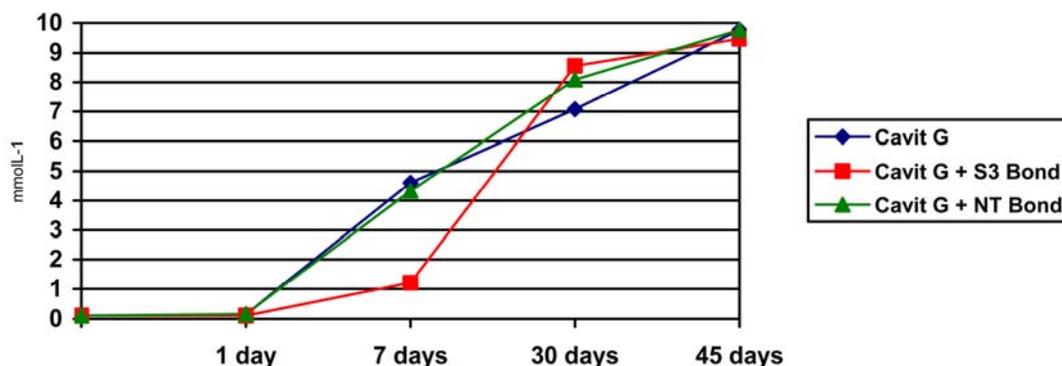


Fig. 2. Mean glucose leakage in mmol L⁻¹ after 45 days.

fective was 2mg/dL (0.11 mmol L⁻¹), for which reason readings lower than this were rejected. The amount of glucose penetration of three experimental groups increased significantly over time.

A significant increase in glucose penetration was observed during the first week in groups CavitTM G (4.61 mmol L⁻¹) and CavitTM G+PBNT (4.33 mmol L⁻¹) ($p=0.008$ and $p=0.005$, respectively). The group CavitTM G+CS3B showed significantly less glucose penetration (1.21 mmol L⁻¹) than the other two groups ($p=0.023$ and $p=0.005$, respectively). The glucose penetration values of all three study groups were roughly comparable at thirty days [CavitTM G (7.08 mmol L⁻¹), CavitTM G+CS3B (8.56 mmol L⁻¹) and CavitTM G+PBNT (8.09 mmol L⁻¹)] ($p=0.736$).

At 45 days, CavitTM G and CavitTM G+PBNT showed both the same value of glucose penetration, 9.76 mmol L⁻¹, and CavitTM G+CS3B gave 9.46 mmol L⁻¹; and no significant differences were found among them ($p=0.581$).

Discussion

Most studies used to evaluate the sealing quality of temporary restoration materials involve bacteria or dyes (7,9). The use of a glucose penetration model, for the quantitative, nondestructive measurement of leakage following a longitudinal study protocol can enhance reliability, reproducibility and comparability of results (15).

Temporary filling should seal the endodontic access cavity to avoid reinfection of the root canal system during endodontic treatment. In several studies (8,11) CavitTM was seen to exhibit good sealing properties, when compared with different temporary restoration materials, due to water absorption and expansion during setting. In addition, this material is premixed, which reduces inconsistencies related to chairside mixing.

In endodontics, dentine adhesive systems have been evaluated in the prevention of leakage in filled root ca-

nals, for perforation repairs, as root-end barriers and in sealing pulp chambers (12,13). However, their use as a secondary coronal barrier to prevent microleakage in temporary restoration after the treatment of root canals is uncommon.

The results of this study indicate an adequate sealing capacity at 24 hours when a dentine adhesive system is used (CS3B or PBNT), although no significant differences were found with the CavitTM G group. At 7 days, the group CavitTM G+CS3B showed a significantly lower microleakage in comparison with the other two groups. These results according to Belli et al. (13) could be attributed to a better curing of the self-etching adhesive on the gutta-percha with respect to a total-etch adhesive system. The adhesive systems that are placed in the pulp chamber should be able to adapt and polymerize on the gutta-percha, especially in single root teeth that have no pulpal floor, so that more than half the area of adhesion is on the gutta-percha.

The Prime & Bond[®] NT used in our study contains acetone, a solvent which may leach some components from the gutta-percha that inhibit polymerization (13). Meanwhile, ClearfilTM S3 Bond has water as the solvent; hence its polymer is hydrophilic and more capable of absorbing water in the dentine-adhesive interface (16). This fact, together with the capability of CavitTM to absorb water during hardening, may have contributed to the reduction of microleakage in this group. Moreover, the use of strong phosphoric acid with Prime & Bond[®] NT could cause 'over etching' and the subsequent collapse of the collagen network, reducing the penetration of adhesive resin and therefore resulting in a weak hybrid layer (17). Kijssamanmith et al. (18) found that when the superficial dentine was less demineralised and peritubular dentine matrix was removed with a self-etching adhesive (ClearfilTM SE Bond), the bond was stronger than the 'one-bottle' system (Prime & Bond[®] NT).

The better results of the group CavitTM G+CS3B at 7 days of study could also be due to the influence of irrig-

ant solutions during root canal treatment and their effect on the adhesive system/pulp chamber bond (19,20). Chemical irrigants such as sodium hypochlorite (NaOCl), commonly used in endodontic treatment, can adversely affect the bond strengths of adhesive systems in contact with dentine pulp chamber walls after endodontic treatment (19) when NaOCl was applied previous to the use of a self-etching priming system (Clearfil™ SE Bond, Prompt™ L-Pop™), better results were obtained than when it was used with a total-etching adhesive (Prime & Bond® NT).

Belli et al.(13) quantitatively assessed the capability of four different filling materials in sealing the pulp chamber after root canal therapy: a self-etching primer adhesive system (Clearfil™ SE Bond), a wet bonding system (One-Step®), a self-curing adhesive system (C&B Metabond®) or zinc oxide-eugenol (IRM®); and the quantitative evaluation of leakage was measured with a fluid filtration method. Clearfil™ SE Bond and C&B Metabond® polymerized well on top of gutta-percha but One-Step® did not, suggesting that adhesive resins provide excellent seals for as long as 1 month. In our study, CS3B plus Cavit™ G showed the best results at 7 days of contact with the system of glucose penetration; however these values equaled results of the other groups at 30 and 45 days, since the main barrier was a temporary material, not a definitive restoration material as in the study of Belli et al. (13).

Ozturk et al. (12) compared, in vitro, the sealing properties of five dentine adhesive systems (Prime & Bond® NT; Prompt™ L-Pop™; Clearfil™ SE Bond; Scotchbond™ Multi Purpose Plus and EBS™-Multi Adhesive System) plus resin composites, applied inside the pulp chamber, using a fluid filtration method for quantitative evaluation of leakage. Although none of the materials created a perfect seal of the pulp chamber, Prime & Bond® NT and Prompt™ L-Pop™ were more successful than the other systems in the short term, but no long-term differences were observed. Likewise, in our study, the differences between dentin adhesive systems were reduced over time (30 or 45 days). Hence, our working hypothesis must therefore be accepted at 30 and 45 days: the sealing ability of Cavit™ G when used as the only filling material was similar to Cavit™ G used with Clearfil™ S3 Bond or Prime & Bond® NT adhesive systems.

Under the conditions of this study, the use of a self-etching (Clearfil™ S3 Bond) or a total-etch (Prime & Bond® NT) adhesive system applied in the pulp chamber does not improve the sealing capacity of Cavit™ G over time. Further investigation is needed to corroborate these findings in single- and multi-rooted teeth.

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