

## *Evaluation of temporary cement to prevent bacterial contamination of the root canal after pulpectomy*

## *Avaliação da capacidade do cimento temporário prevenir a contaminação bacteriana do canal após pulpectomia*

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### ABSTRACT

To evaluate the ability of temporary cement (TC) and gutta-percha sticks (GP) to prevent bacterial contamination of the root canal through the coronal seal after pulpectomy. Eighty artificial primary maxillary central incisors were selected and randomly divided into 2 groups: TC (n = 40) and GP (n = 40). Endodontic access, rotary instrumentation, root canal filling, and coronal sealing were performed according to group allocation. The root canal opening was seeded with *S. mutans* and *E. faecalis*. Both groups were subdivided into 5 experimental time points (24, 48, 72, 96, and 120 hours), with 8 specimens per time-point group: 5 in which both root canal filling and coronal sealing were performed (with either TC or GP) and 3 controls (coronal sealing alone, without root canal filling). All specimens were incubated in an anaerobic jar at 37°C, and bacterial contamination was assessed in a spectrophotometer. ANOVA (t-test) was used to compare contamination and the Kruskal-Wallis test to compare filling scores between the experimental groups. A significant difference was observed in sealing in the first 24 hours between GP and controls (p = 0.046). There was no significant difference in the filling pattern between canals sealed with TC versus GP. Specimens sealed with GP showed less contamination than controls in the first 24 hours. At later time points, neither GP nor TC were effective at controlling bacterial contamination; both failed to provide adequate coronal sealing.

**Indexing terms:** Endodontic obturation. Pulpectomy. Tooth, deciduous.

### RESUMO

*Avaliar a capacidade do obturador provisório (OP) e da gutapercha em bastão (GP) de prevenir a contaminação bacteriana dos condutos radiculares. Foram selecionados 80 incisivos centrais superiores decíduos artificiais que foram divididos aleatoriamente em*

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2 grupos: OP (n = 40) e GP (n = 40). Foi realizado acesso endodôntico, instrumentação rotatória, preenchimento do canal radicular e selamento coronário conforme os grupos. Foi feita a sementeira de *S. mutans* e *E. faecalis* na entrada do canal radicular. Ambos os grupos foram subdivididos em 5 tempos experimentais (24, 48, 72, 96 e 120 horas), com 8 espécimes por tempo experimental: 5 submetidos a preenchimento do canal radicular e selamento coronário (com OP ou GP) e 3 controles (apenas selamento coronário, sem preenchimento do canal). Todos os espécimes foram incubados em jarras de anaerobiose a 37°C e a contaminação bacteriana foi avaliada em espectrofotômetro. Utilizou-se ANOVA (teste t) para a comparação da contaminação e o teste de Kruskal-Wallis para a comparação dos escores da obturação entre os grupos experimentais. Foi observada diferença significativa no selamento nas primeiras 24 horas entre GP e controles ( $p = 0,046$ ). Não houve diferença estatisticamente significativa no padrão de preenchimento entre os canais selados com OP versus GP. Os espécimes selados com GP apresentaram menor contaminação do que os controles nas primeiras 24 horas. Nos demais tempos experimentais, tanto GP quanto OP não foram eficientes no controle da contaminação bacteriana; ambos apresentaram falha no selamento coronário.

**Termos de indexação:** Obturação do canal radicular. Pulpectomia. Dente decíduo.

## INTRODUCTION

Endodontic treatment of primary teeth is indicated when the pulp canal is infected or necrotic due to caries or trauma [1,2]. The goal is to reduce the microbial burden in the root canal system, to restore the integrity of the dental tissue and its supporting structures, and to ensure that the primary tooth will remain in place until its physiological exfoliation [3,4].

Pathological mobility, a history of spontaneous toothache, presence of a fistula or abscess, excess bleeding, or pus issuing from the pulp chamber indicate the need to perform non-vital pulp therapy [1,5]. In addition, the crypt of the succedaneous tooth must be radiographically intact and the extent of root resorption cannot have exceeded one-third of the total root length; otherwise, pulpectomy cannot be performed [3,6].

Root canals can be prepared by manual or mechanical instrumentation prior to the endodontic treatment of primary teeth. Chemomechanical preparation of the root canal was first reported by Barr et al. [7] Since then, rotary instrumentation has been widely studied and used, and has proven effective for root canal cleaning and disinfection, providing a better taper and substantially shorter chair time than conventional techniques, which is an extremely relevant advantage in pediatric dentistry [8-12].

Zinc oxide/eugenol, iodoform, and calcium hydroxide are among the compounds used for root canal filling after pulpectomy [4,13]. Appropriate root canal fillers for primary teeth must have antimicrobial and anti-inflammatory effects and must be biocompatible with living tissue, radiopaque, and resorbable to allow physiological root resorption [14,15]. There is no consensus as to the optimal filling material; each has its advantages and disadvantages, and it is up to the dental practitioner to select the most suitable product [4,13,15,16].

After root canal filling, a small plug is commonly placed to isolate the filling material from the overlying restorative material, in an attempt to achieve a good marginal seal. This is usually done with temporary cement (TC) or gutta-percha sticks (GP). After endodontic treatment, root canal filling alone is not sufficient to prevent penetration of microorganisms into the periapical region, and coronal microleakage is the leading cause of recontamination of the periapical tissues [17]. The quality of the seal provided by the temporary restoration is just as important as the root canal filling itself [18,19].

In some cases, pediatric dentists are unable to complete endodontic treatment and final restoration in a single session; temporary restorative materials must then be used between visits. Also, the definitive restoration may come loose from the cavity, leaving the root canals protected only by the intermediate material. When this occurs, many dentists simply proceed with the definitive restoration without considering the potential for root canal contamination due to the defective remaining seal and the probable need for reinstrumentation and for a new round of disinfection of the canal.

Within this context, the objective of this in vitro study was to evaluate the ability of TC and GP to prevent bacterial contamination of the root canal through the coronal seal after pulpectomy. The null hypothesis was that both materials would prevent microbial contamination for at least 5 days.

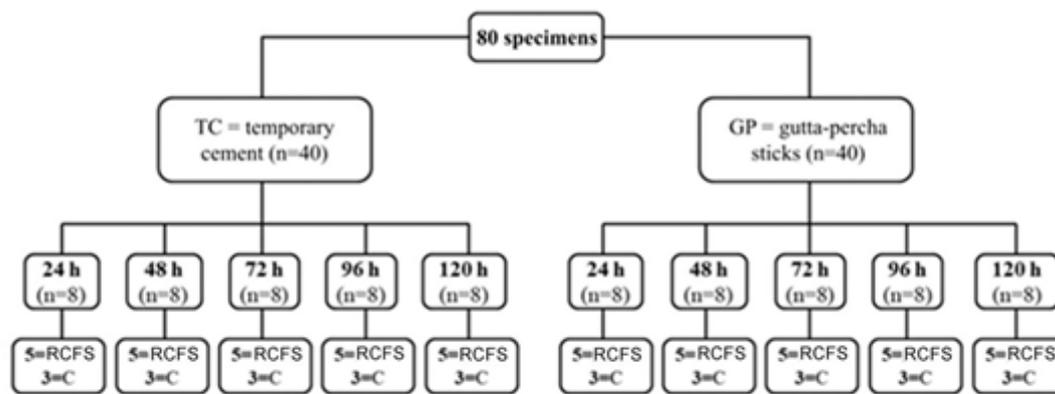
## METHODS

Eighty artificial primary maxillary central incisors (Denarte, São Paulo, Brazil) were selected and randomly divided into 2 experimental groups (n = 40 each) to receive either TC (Obturador Provisório®, Villevie, Joinville, Brazil) or GP (GuttaPercha Sticks®, Dentsply Sirona, São Paulo, Brazil) as a sealant. Endodontic access, rotary instrumentation, root canal filling, and coronal sealing were performed according to group allocation. The table 1 below shows the materials composition using in this study (Obturador Provisório® and GuttaPercha Sticks®) and Coltosol® (Coltene, Rio de Janeiro, Brazil) (the most known material similar to Obturador Provisório®).

**Table 1** – Materials composition using in this study.

| Material   | Composition  |
|--|--|
| Coltosol® (Coltene, Rio de Janeiro, Brazil):             | Zinc oxide, Zinc sulfate, Calcium sulfate, Polyvinyl acetate, Menthol, Dibutylphthalate.                               |
| Obturador provisório® (Villevie, Joinville, Brazil)      | Zinc oxide, Calcium sulfate, Yellow iron oxide, Thickener, Zinc sulfate, Silicone oil, Flavoring, Orthodontic plaster. |
| Guttapercha Sticks® (Dentsply Sirona, São Paulo, Brazil) | Gutta-percha, Wax, Calcium Carbonate, Pharmaceutical Caolim, Zinc Oxide and Organic Pigments.                          |

Both the TC and GP groups (n = 40 each) were subdivided into 5 experimental time points (24, 48, 72, 96, and 120 hours), as shown in Figure 1. Sample size was calculated by analysis of variance (ANOVA). To achieve a minimum difference of 0.01 between treatment means, with a standard error of 0.0056, 3 treatments, a power of 80%, and an alpha of 0.05, a sample size of 8 specimens per time-point group was necessary: 5 specimens in which both root canal filling and coronal sealing were performed (with either TC or GP) and 3 specimens used as controls (coronal sealing alone, without root canal filling).



**Figure 1** – Distribution of specimens across the time points of analysis. RCFS: Specimens in which both root canal filling and coronal sealing were performed. C: Control specimens in which coronal sealing alone was performed.

## Specimen preparation

In all teeth, endodontic access was performed with #1012 diamond and #2 carbide burs (KG Sorensen, Cotia, São Paulo, Brazil) mounted on a high-speed micromotor (Kavo, Santa Catarina, Brazil) under constant water cooling. The convenience form was achieved with a #3082 tapered carbide bur (KG Sorensen, Cotia, São Paulo, Brazil), making the pulp chamber walls divergent to facilitate direct access to the root canal. Canal length was determined visually, and

patency was established with a #10 K-file (Dentsply Maillefer, Ballaigues, Switzerland). With the aid of a caliper (Mitutoyo, São Paulo, Brazil), the working length was set 1 mm short of the apex.

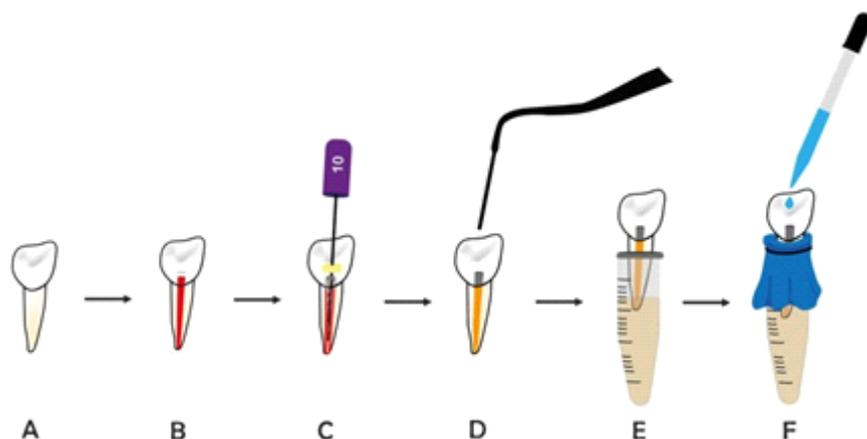
Rotary instrumentation was performed using the ProTaper Universal system (Dentsply Maillefer, Ballaigues, Switzerland), and all instruments were driven by an X-Smart™ endodontic motor (Dentsply Maillefer, Ballaigues, Switzerland) [20]. The S1 and S2 files (Dentsply Maillefer, Ballaigues, Switzerland) were used to prepare the cervical and middle thirds with a rotational speed of 300 rpm and 3 N/cm torque. The F1 and F2 files (Dentsply Maillefer, Ballaigues, Switzerland) were used at the full working length at the same rotational speed, with 2 N/cm torque. After instrumentation of each third, the root canal was irrigated with 5 mL of 1% sodium hypochlorite, for a total of approximately 15 mL per root canal. After completion of root canal preparation, all specimens were autoclaved at 121°C for 15 minutes in order to eliminate microorganisms that could be present and interfere with the results of the spectrophotometric analysis.

## Microbiological processing

All microbiological procedures were performed in a laminar flow hood. Root canals were filled with Feapex obturating cement (Fórmula & Ação, São Paulo, Brazil) using #15 K-files (Dentsply Maillefer, Ballaigues, Switzerland), interspersed with gentle pressure from sterile cotton balls (Cremer, São Paulo, Brazil) and compactors (Millenium Golgran, São Paulo, Brazil).

Root canals were sealed at the opening of the canal, 3 mm short of the pulp chamber, as measured with compactors (Millenium Golgran, São Paulo, Brazil), rubber stops (Jon, São Paulo, Brazil), and a millimeter ruler (Dentsply Maillefer, Ballaigues, Switzerland). In the TC group, the root canal opening was sealed with the aid of a spatula (Millenium Golgran, São Paulo, Brazil) and pressure with cotton balls. In the GP group, the sticks were heated over the flame of a bunsen burner (Prolab, São Paulo, Brazil) and compacted with the aid of a spatula (Millenium Golgran, São Paulo, Brazil) and amalgam condensers (Millenium Golgran, São Paulo, Brazil).

After completion of the filling and sealing steps, a device was fabricated using an Eppendorf microcentrifuge tube (Eppendorf do Brasil, São Paulo, Brazil), cyanoacrylate adhesive (Three Bond, São Paulo, Brazil), and a rubber dam (Madeitex, São Paulo, Brazil) to keep all specimens in a vertical position with the root apex immersed in brain heart infusion (BHI) broth (Acumedia, Indaiatuba, Brazil). A schematic diagram illustrates the experiment (figure 2).



**Figure 2** – Schematic diagram of the experimente. A – Primary maxillary central incisor. B – Endodontic access. C – Canal length determination and establishment of patency with a #10 K-file. D – Root canal filling and sealing at the opening of the canal, 3 mm short of the pulp chamber. E – Tooth secured vertically in the positioner device, with the root apex immersed in BHI broth. F – 10-µL aliquot of *S. mutans* and *E. faecalis* is seeded at the opening of the pulp chamber.

All specimens were seeded with a 10- $\mu$ L aliquot of *Streptococcus mutans* and *Enterococcus faecalis* using a micropipette (Uniscience do Brasil, São Paulo, Brazil) at the opening of the root canal, followed by agitation for 10 seconds in a tube shaker (Phoenix, Araraquara, São Paulo, Brazil) to ensure that the drop would spread throughout the pulp chamber. The specimens were evaluated in a spectrophotometer at 5 different time points (24, 48, 72, 96, and 120 hours) after inoculation, according to subgroup allocation. All specimens were incubated under anaerobic conditions in a microbiological incubator at 37°C using the candle jar method. Spectrophotometry was performed to assess bacterial contamination at each time point of analysis.

## Analysis of root canal filling patterns

After completion of the microbiological stage, specimens from both the TC and GP groups were sectioned along the buccolingual axis with #4137 and #4138 diamond burs (KG Sorensen, Cotia, São Paulo) mounted on a high-speed handpiece to expose the filled root canal. The sectioned specimens were photographed for evaluation of the filling pattern. Three independent examiners, all specialist endodontists, received the same images acquired from the samples sectioned in half and analyzes and classified in the scores, them on a scale of 1 to 5 as follows: 1) canal not filled; 2) canal less than 50% filled; 3) canal 50% filled; 4) canal more than 50% (but still incompletely) filled; and 5) completely filled canal (Figure 3). To assess the calibration between examiners, the Kappa test was used. The scores given by each examiner were averaged and used for analysis. The purpose of this assessment was to establish a correlation between the pattern of root canal filling obtained in each group and the degree of contamination observed.



**Figure 3** – Score classification. 1 – Canal not filled. 2 – Canal less than 50% filled. 3 – Canal 50% filled. 4 – Canal more than 50% filled. 5 – Completely filled canal.

## Statistical analysis

BioEstat 4.0 was used for data analysis. The normality of data distribution was assessed by the Shapiro-Wilk test. The results of microbiological analysis were normally distributed and analyzed by parametric tests (ANOVA, t-test) to compare bacterial contamination between the experimental groups. Because the results for filling scores were not normally distributed, the nonparametric Kruskal-Wallis test was used to compare filling scores between the experimental groups, as an indicator of filling patterns. The level of significance was set at 5% for all analyses.

## RESULTS

In the first 24 hours, a significant difference was observed in sealing between the GP group and the control group ( $p = 0.046$ ). At all other time points, there was no statistically significant difference between the GP group and controls (table 2).

**Table 2** – Arithmetic means and standard deviations comparing gutta-percha sticks (GP) versus control (C) at different time points.

| Groups | 24 hours                | 48 hours                | 72 hours                | 96 hours                | 120 hours               | p*   |
|--------|-------------------------|-------------------------|-------------------------|-------------------------|-------------------------|------|
| GP     | 0.13±0.09 <sup>Aa</sup> | 0.01±0.16 <sup>Aa</sup> | 0.28±0.12 <sup>Aa</sup> | 0.29±0.24 <sup>Aa</sup> | 0.20±0.12 <sup>Aa</sup> | 0.07 |
| C      | 0.40±0.05 <sup>Ba</sup> | 0.36±0.25 <sup>Aa</sup> | 0.42±0.13 <sup>Aa</sup> | 0.48±0.08 <sup>Aa</sup> | 0.35±0.12 <sup>Aa</sup> |      |
| p*     | 0.046                   | 0.0540                  | 0.1845                  | 0.2367                  | 0.1516                  |      |

Different uppercase letters in the same column indicate statistically significant differences. Same lowercase letters in the same row indicate no significant difference. \*t-tests.

There was no significant difference between the TC group and the control group at any of the time points of analysis ( $p > 0.05$ ) (table 3).

There was no significant difference between the TC and GP groups in the quality of filling pattern (table 4).

**Table 3.** Arithmetic means and standard deviations comparing temporary cement (TC) versus control (C) at different time points.

| Groups | 24 hours                | 48 hours                | 72 hours                | 96 hours                 | 120 hours               | p*   |
|--------|-------------------------|-------------------------|-------------------------|--------------------------|-------------------------|------|
| TC     | 0.36±0.16 <sup>Aa</sup> | 0.42±0.19 <sup>Aa</sup> | 0.07±0.32 <sup>Aa</sup> | 0.22±0.17 <sup>Aa</sup>  | 0.21±0.09 <sup>Aa</sup> | 0.10 |
| C      | 0.41±0.13 <sup>Aa</sup> | 0.45±0.06 <sup>Aa</sup> | 0.14±0.16 <sup>Aa</sup> | 0.16±0.004 <sup>Aa</sup> | 0.44±0.01 <sup>Aa</sup> |      |
| p*     | 0.66                    | 0.83                    | 0.36                    | 0.45                     | 0.29                    |      |

Same lowercase letters in the same column or row indicate no significant difference. \*t-tests.

**Table 4** – Medians and interquartile deviations comparing gutta-percha sticks (GP) versus temporary cement (TC) at different time points

| Groups | 24 hours             | 48 hours             | 72 hours             | 96 hours             | 120 hours            | p*     |
|--------|----------------------|----------------------|----------------------|----------------------|----------------------|--------|
| GP     | 4.0±1.0 <sup>A</sup> | 4.0±1.0 <sup>A</sup> | 4.0±0.0 <sup>A</sup> | 5.0±1.0 <sup>A</sup> | 4.0±1.0 <sup>A</sup> | 0.8088 |
| TC     | 4.0±3.5 <sup>A</sup> | 4.0±4.5 <sup>A</sup> | 4.0±4.0 <sup>A</sup> | 4.0±4.5 <sup>A</sup> | 5.0±4.5 <sup>A</sup> |        |
| p*     | 0.2506               | 1.000                | 0.9168               | 0.6015               | 0.2563               |        |

Same uppercase letters in the same column or row indicate no significant difference.

\*Kruskal-Wallis test.

## DISCUSSION

Studies focusing on the use of sealants after pulpectomy are scarce in pediatric dentistry, and this study adds to the current literature by providing relevant information for the endodontic treatment of primary teeth. The present study aimed to compare the ability of 2 temporary sealing materials (TC and GP) used after endodontic treatment of primary teeth to prevent microbial contamination of the root canal. The results showed that both sealants failed to provide adequate coronal sealing and to control bacterial contamination across the 24- to 120-hour assessment periods. Therefore, the null hypothesis was rejected.

Endodontic treatment cannot always be completed in a single session, making the newly filled root canal more vulnerable to damage due to exposure. Therefore, several temporary restorative materials are used to seal the root canal between sessions. However, to what extent these materials actually protect the obturated root canal from bacterial contamination and infiltration remains unknown. In addition, the restorative material may detach from the cavity, leaving only a thin layer to protect the root canal. When this occurs, many pediatric dentists choose to simply place another coronal seal, without any intervention in the root canal or decontamination of the pulp chamber that was exposed to oral fluids. For this reason, the present study sought to evaluate whether there is a time window of safe exposure to oral fluids before contamination occurs.

Plasticized GP sticks have long been used as the sealant of choice in the endodontic treatment of primary teeth [8,14]. However, they are now being replaced by TC, which is easier to manipulate and to place into the pulp chamber [22]. Both materials must be placed into the pulp chamber as a thin layer, separating the canal filler from the definitive restoration.

Coronal marginal microleakage is defined as the passage of microorganisms and toxins through the interface between the restorative material and the cavity walls [21-23]. Subsequent bacterial infection, which is the most common cause of pulp disease, is closely related to how long the temporary restoration stays in place and the quality of the coronal seal. Root canal recontamination can lead to endodontic treatment failure. In view of the foregoing, we compared specimens sealed with GP and TC with controls (without root canal filling, only coronal sealing) as part of the rationale for identifying whether coronal sealing alone was able to prevent contamination or whether contamination could be prevented only by the combination of root canal filling and coronal sealing.

The contamination of root canals observed in this study can be explained by the difficulty in ensuring that the sealant adheres to the walls of the pulp chamber in order to avoid the presence of fluids that would contaminate the root canal system. The analysis of root canal filling patterns showed a predominance of scores 4 and 5, i.e., root canals that were more than 50% filled or completely filled. It is therefore reasonable to assume that the microleakage observed here occurred due to coronal sealing failure.

Coltosol®, a temporary sealing material similar to the Obturador Provisório® cement used in our study, is a hygroscopic cement composed of zinc oxide and calcium sulfate, with the ability to expand on contact with moisture [23,24]. In the present study, there was no statistically significant difference between specimens sealed with TC and controls at any of the assessment time points (table 2,3), which indicates that root canals sealed with this material are susceptible to microleakage and recontamination as early as 24 hours after endodontic treatment. The sealing ability of TC increases with its hygroscopic expansion, which allows the material to better fit against the chamber walls. Application of a water-soaked cotton ball after placement of the TC layer is essential for this expansion to occur. In this study, we believe that TC did expand successfully on contact with the filling material, which is a wet paste. Nevertheless, the TC failed to prevent subsequent microbial contamination.

GP is a composite material made of zinc oxide, beeswax, Japan wax, calcium carbonate, kaolin, and organic dyes [25]. The results of the present study showed that coronal sealing with GP sticks was ineffective after 24 hours (figure 1); however, before 24 hours, this material provided significantly superior sealing compared with controls. Although GP is characterized by excellent adaptation to the pulp chamber walls, thermoplastic nature, and physicochemical stability [26], it lacks adhesiveness, which may explain the occurrence of microleakage.

The present study used standard strains of *E. faecalis* and *S. mutans* to evaluate the quality of the coronal seal. *Enterococcus* is a genus of facultatively anaerobic, catalase-negative bacteria, several species of which inhabit the human gastrointestinal tract and oral cavity [27]. *E. faecalis* is commonly found in endodontic infections and is even more prevalent in persistent infections, which denote failure of endodontic treatment [28]. *E. faecalis* is able to form biofilms even under limited conditions, nutritional stress, a wide range of alkaline pH values, and at temperatures from 10°C to 45°C [28].

Several methods have been described in the literature to assess the coronal sealing ability of restorative materials. Dye infiltration, with methylene blue or India ink used as a visual marker of coronal leakage, has been widely used in qualitative studies [22,23,29]. In the present study, microbial counts in the BHI broth were evaluated quantitatively by spectrophotometric analysis of turbidity. Previous studies that have used this technique for the same purpose as ours include those of Jafari & Jafari [19].

In contrast to our study, Naseri et al. [22] observed Coltosol® used for coronal sealing preventing contamination for up to 1 week, while we found the TC was unable to do so for 24 hours. Therefore, based on the results of the present study and previous literature, more studies are needed to find a capable material that can prevent coronal infiltration altogether, because that 2 temporary restorative materials used in this study are unable to provide an adequate seal against the passage of bacteria.

## CONCLUSION

Coronal sealing with GP sticks was only able to prevent bacterial contamination of the root canal for 24 hours. At later time points, neither GP nor TC were effective at preventing bacterial contamination; both failed as coronal sealing materials. When root canal filling patterns were compared between TC and GP, no statistically significant differences were observed, making it impossible to draw any correlation between failure in root canal filling and the subsequent degree of contamination.

## Collaborators

LM Pereira, SMM Naves, JM Costa and IO Moraes, methodology, investigation, writing – original draft, review & editing, final approval of the article. SREP Silva and CE Fontana, conceptualization, supervision, project administration, writing – original draft, review & editing, final approval of the article. SL Pinheiro: conceptualization, supervision, project administration, writing – original draft, review & editing, formal analysis, final approval of the article. All authors have read and approved the final version of the article and contributed equally.

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## ERRATUM

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