

Comparison of root canals obturated with ProTaper gutta-percha master point using the active lateral condensation and the single-cone techniques: a bacterial leakage study

Anizabele Milet do Amaral Mercês¹, Carlos Menezes Aguiar², Neide Kazue Sakugawa Shinohara³,
Andréa Cruz Câmara⁴, Jose Antonio Poli de Figueiredo⁵

¹Private Practice, Recife, Brazil

²Associate Professor, Department of Prosthodontics and Oral and Maxillofacial Surgery, Dental School, Federal University of Pernambuco, Brazil

³Adjunct Professor, Keizo Asami Immunology Laboratory, Federal University of Pernambuco, Brazil

⁴Substitute Professor, Department of Prosthodontics and Oral and Maxillofacial Surgery, Dental School, Federal University of Pernambuco, Brazil

⁵Postgraduate program in Dentistry, Pontifical Catholic University of Rio Grande do Sul, PUCRS, Brazil

Abstract

Aim: The aim of the present study was to assess the recontamination time of root canals filled with ProTaper gutta-percha master point using the active lateral condensation and the single-cone techniques. **Methods:** Fifty premolars, after biomechanical preparation, were randomly divided into six groups according to the obturation technique and the diameter of the gutta-percha point. For leakage evaluation, the roots were mounted in a dual-chamber leakage model system using *Pseudomonas aeruginosa*, *Enterococcus faecalis*, *Escherichia coli*, and *Staphylococcus aureus*. The Kaplan-Meier and Long-Rank tests were employed at the .05 level of significance. **Results:** The recontamination was detected between the 13th and 80th days. The active lateral condensation produced less infiltration than the single cone technique, though without statistically significant differences between them. **Conclusions:** It may be concluded that there was no difference in bacterial penetration between the two root canal filling techniques. The mean root canal contamination time was 67 days.

Keywords: gutta-percha, microleakage, ProTaper gutta-percha master point, ProTaper Universal, single-cone technique.

Introduction

Successful root canal treatment requires proper cleaning and shaping of root canal, as well as hermetic sealing of root canal space with an inert, dimensionally stable, and biologically compatible material¹. When the coronal portion of the root canal is exposed to the oral environment, the root canal is a potential route for microorganisms to gain access to the periapical tissue. This situation may lead to endodontic failure². From a clinical standpoint, coronal exposure of the root canal obturation might be considered an indication for endodontic retreatment³.

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Correspondence to

Carlos Menezes Aguiar
Rua Aristides Muniz, 70/501, Boa Viagem,
Recife PE 51020-150 Brazil.
Phone: (+55) 81 3467 6821.
E-mail: cmaquiari.ufpe@yahoo.com.br

The influence of the quality of the coronal restoration is an important factor in achieving success in root canal treatment. The teeth with a satisfactory coronal restoration displayed significantly better periapical healing than those with an unsatisfactory restoration⁴⁻⁵.

Radiographically, the technical quality of endodontic treatment is significantly more important than the technical quality of the coronal restoration when the periapical status of the endodontically treated teeth is evaluated⁶. The biomechanical preparation with nickel-titanium (NiTi) rotary system was considerably quicker than the hand instrument series⁷. It would appear that rotary NiTi instruments have the potential to prepare root canals more efficiently, and to shape root canals with greater safety⁸, achieving a three-dimensional obturation of the root canal over its whole length without the time spent on lateral condensation⁹.

Cleaned and shaped root canals must be filled three-dimensionally, eliminating the empty space, which has the potential to be infected or reinfected¹⁰. The aims of the root canal sealer are to prevent penetration of bacteria and eliminate the risks of infection or reinfection of the root canal system³. Given the importance of the root canal filling and the control and prevention of endodontic infection, several studies have been published on the subject¹⁰⁻¹². Coronal leakage has been evaluated by different tests, among which is the analysis of bacterial penetration through the filling material. This method has been used because it has reached results similar to those obtained in daily practice¹³.

The purpose of this study was to investigate the time required for recontamination of root canals sealed with ProTaper gutta-percha cones after obturation with two different techniques and exposure to a simulated oral environment.

Material and methods

Specimen Selection and Preparation and Root Canal Filling

Fifty intact, caries-free, human mandibular premolars with single roots and mature apices (mean length of 21 mm) were used. The teeth were obtained from the Tooth Bank of the Department of Prosthodontics and Oral and Maxillofacial Surgery of the Federal University of Pernambuco, after approval of the Research Ethics Committee of the University's Center of Health Sciences. Buccolingual and mesiodistal radiographs were taken to confirm the presence of a single canal.

Once coronal access had been established, a K-File #10 (Dentsply/Maillefer, Ballaigues, Switzerland) was inserted into the root canal until the tip was just visible beyond the apex. The working length (WL) was determined at 1 mm short of this length. The root canals were prepared by the ProTaper Universal™ rotary system (Dentsply/Maillefer, Ballaigues, Switzerland). In groups 1 and 2, the root canals were instrumented up to instrument F4, and in the groups 3 and 4 the canals were prepared up to instrument F5. A single operator instrumented all root canals. The root canals were irrigated with a freshly prepared 1% sodium hypochlorite

solution (Roval, Recife, Brazil) in conjunction with ethylenediaminetetraacetic acid (Biodinâmica, Paraná, Brazil) as a chelating agent for irrigating the root canals, as well as the 3-mL FCF syringe system (FCF, Sao Paulo, Brazil) with a 30-gauge needle (Injecta, Diadema, Brazil). The irrigation was performed at the beginning of the instrumentation, between the changes of instrument, and at the end of the biomechanical preparation. The specimens were stored in glass test tubes and were individually sterilized in an autoclave at 121°C for 30 minutes. Ten samples were randomly chosen and immersed totally in bottles containing 10 mL autoclaved brain heart infusion (BHI; Acumedia, Lansing, MI). They were kept in an incubator at 37°C for 96 hours to check the sterilization's efficacy.

After preparation, the specimens were randomly divided into six groups as follows:

- Group 1: 10 specimens were filled using the single cone technique with the gutta-percha master point F4 (Dentsply/Maillefer, Ballaigues, Switzerland) and AH Plus sealer (Dentsply DeTrey, Konstanz, Germany).

- Group 2: 10 specimens were filled using the active lateral condensation technique with the gutta-percha master point F4 and 10 FF accessory cones (Dentsply/Maillefer, Ballaigues, Switzerland) and AH Plus sealer .

- Group 3: consisted of 10 specimens filled using the single cone technique with the gutta-percha master point F5 and AH Plus sealer.

- Group 4: 10 specimens were filled using the active lateral condensation technique with the gutta-percha master point F5 and 10 FF accessory cones and AH Plus sealer.

- Group 5 (positive control group): consisted of 5 specimens that did not have the canal filled.

- Group 6 (negative control group): consisted of 5 specimens that did not have the canal filled but was rendered impermeable by receiving an epoxy resin coating (Araldite, Brascola, São Bernardo do Campo, Brazil) along its entire length.

Three blinded endodontists evaluated the root fillings on radiographs in accordance with the criteria of Eckerbom and Magnusson¹⁴.

Leakage Apparatus Preparation

The apparatus used to evaluate saliva leakage was prepared as previously described by Gomes et al.¹¹, with modifications. Glass vials with rubber stoppers were adjusted for use in this experiment. Using a heated instrument, a hole was made through the center of every rubber stopper in which each tooth was inserted under pressure up to its cemento-enamel junction, so that its crown lay outside and its root inside the vial. The teeth were sealed with three layers: a layer of cyanoacrylate (Henkel Ltda, São Paulo, Brazil), a layer of epoxy resin (Pulvitec S.A. Indústria e Comércio, São Paulo, Brazil) and a final layer of cyanoacrylate were applied at the interface between the tooth and rubber. Thereafter the roots were sealed with two layers of cyanoacrylate, except for the apical 2 mm around the apical foramen. The cylinders prepared from 5-mL plastic syringes

(Plascap, São Paulo, Brazil) were adapted on the external surface of the stoppers to create a chamber around the crown of the tooth. The junction between the plastic cylinders and glass tube was sealed as described above. The apparatus was then sterilized with ethylene oxide gas (Galdi Produtos Hospitalares, Paulista – Brazil) for 12 h. The chamber of each apparatus was filled with BHI, so that approximately 2 mm of the root apex was immersed in the broth. To ensure sterilization, no growth was observed after the whole apparatus was incubated at $37^{\circ}\text{C} \pm 1$ for four days. Any test apparatus that showed signs of turbidity in the BHI broth was discarded.

Bacterial Leakage Test

The bacterial stains used for analysis were *Pseudomonas aeruginosa* (American Type Culture Collection – ATCC 9027), *Enterococcus faecalis* (ATCC 19433), *Escherichia coli* (ATCC 8739) and *Staphylococcus aureus* (ATCC 6538). The microorganisms used in this study were selected because of their clinical importance and association with the endodontic infection^{10,15}.

The chamber of each apparatus was filled with 3 mL of sterile artificial human saliva (Farmácia Roval de Manipulações, Recife, Brazil) and mixed in BHI broth in a 3:1 (v/v) ratio. The saliva was replenished every 3 days for 3 months. The turbidity of the inocula was adjusted to approximately 10^8 UFC/mL, this concentration being obtained using the Neubauer Chamber technique¹⁶.

The apparatus was incubated at $35^{\circ}\text{C} \pm 1$ and checked daily for the appearance of turbidity in the BHI broth over a period of 90 days. The number of days it took for bacterial growth to appear was indicative of the total recontamination of the root canal by bacteria from the saliva. Following the appearance of broth turbidity, aliquots were removed and sowed in BHI, Agar blood, Agar Levine, Agar MacConkey and Agar Chapman, and the plates were incubated in a cabinet at $35^{\circ}\text{C} \pm 1$ for 24 hours. The bacterial identification was based on colony features, Gram-staining and cell morphology: catalase production test, Triple Sugar Iron Agar test (TSI), growth in 6.5% NaCl BHI broth, Esculin Bile Agar, and Hugh-Leifson's test, constituting the complementary identification tests¹⁷.

Statistical Analysis

The statistical analysis was performed using the Kaplan-Meier test for survival analysis, which includes calculation of the median time leakage and pairwise comparisons of groups. The Long-Rank test was used to compare the survival curves of the groups. A level of significance of .05 was adopted.

Results

All specimens in the positive control group showed broth turbidity within 13 days of incubation. By contrast, none of the negative control specimens leaked during the

experimental period (90 days), confirming the efficacy of rendering the whole root surface waterproof (Figure 1).

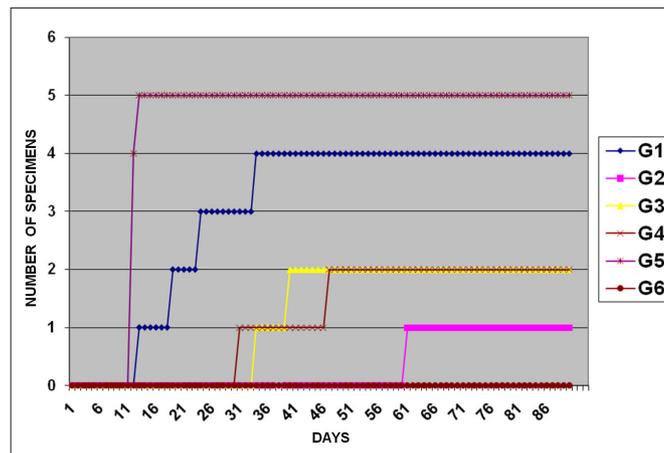


Fig. 1- Number of teeth contaminated at 90 days.

Four specimens in group 1 showed contamination at 36 days. The first specimen that showed microbial leakage was observed at 13 days. In group 2, 1 sample showed contamination at 61 days. Two specimens in group 3 showed contamination at 41 days. The first specimen that showed microbial leakage was observed at 34 days. In group 4, 2 samples showed contamination at 46 days. The first specimen that showed microbial leakage was observed at 34 days. There was no significant statistical difference among the tested techniques. The results of the Kaplan-Meier test are shown in Table 1.

Table 1 - Contamination time of each group.

Group	Mean contamination time (days)	Standard error	Confidence interval
1	67	10	(47; 88)
2	87	3	(82; 92)
3	79	7	(66; 93)
4	80	7	(67; 93)
5	11	0	(11; 12)
6	90	0	-

The Long-Rank test revealed no statistically significant differences between the root canal filled techniques and the diameters of the gutta-percha master points employed.

Discussion

Methods to measure microleakage have included the use of dyes¹⁸, human saliva¹⁹, microorganisms²⁰ and bacterial toxins²¹. The inadequacies of each of these methods have been highlighted and their clinical significance questioned. Although the use of dyes, radioisotopes, fluid filtration, bacteria, and endotoxins penetration techniques to evaluate sealing capacity of endodontic materials, the bacteria leakage model has proved to be the most clinically important one^{12,22}.

Facultative anaerobic and aerobic microorganisms have

been used, although they are found in only 10 to 20% of root canals with a chronic periapical reaction. Such microorganisms are found interacting with strict anaerobes, causing changes in nutritional relationships, shifts in redox potential and oxygen tension, thereby establishing the microbial survival relationship²³. Determining the sealing capacity of endodontic materials against these microorganisms is essential.

The endodontic sealers showed degradation with oral fluids. AH Plus showed the smallest weight loss in water and artificial saliva with different pH values²⁴. This is because AH Plus is an epoxy resin-based sealer, and provides a tight seal of the root canal system¹. These properties justified its choice and use in this study, corroborating the findings of Tronstad et al.⁶.

Complete obturation of the root canal system is an important factor in achieving success in root canal therapy. Ideally, root canal filling should be a complete, homogenous mass that fills the prepared root canal completely. The conventional root-filling technique taught in most dental schools²⁵ and used by dentists²⁶ has been the cold lateral compaction technique using the .02 tapered gutta-percha point as the master cone, supplemented with accessory cones. However, several practitioners regard it as time-consuming and difficult to master. The alternative to root canal filling is the single-cone technique⁹. This tendency to optimize endodontic treatment led to the evaluation of the sealing capacity of these techniques, using the ProTaper gutta-percha master points.

The teeth obturated using the vertical compaction technique exhibited a dense fill throughout the canal space with the aim of preventing the leakage of microorganisms²⁰. The results of the present study showed no statistically significant differences between the methods employed.

Although the cold lateral compaction of gutta-percha has been regarded as the gold-standard obturation technique, the results of the research were in agreement with those of Britto et al.²⁷, which showed microleakage of microorganisms.

Yücel et al.³ demonstrated that all samples obturated with cold lateral compaction showed bacterial penetration after 60 days of observation. Their results are at odds with those observed in the present study, in which 15% of the specimens showed bacterial leakage after 60 days. The reason for the discrepancies between the two studies was determined by enlarging the apices.

At 90 days, bacterial penetration was observed in 22.5% of all the specimens, in contrast with the results of Ricucci and Bergenholtz²⁸, who demonstrated that well-prepared filled root canals resist bacterial penetration even after long-standing oral exposure to caries, fracture or loss of restoration.

Although West²⁹ stated that apical enlargement with #40 or #50 files ensures maximum cleaning of the foraminal constriction and increases apical sealing, the results of the present study showed no significant differences between apical enlargement and apical seal.

The present study demonstrated that there is no filling material or root canal filling technique able to completely prevent the entry of microorganisms into the root

canal space when the teeth are in an oral environment. These results are in agreement with the findings of Timpawat et al.¹, Britto et al.²⁷, Yücel et al.³, and Monticelli et al.²².

This study showed that an increased apical diameter did not hinder bacterial penetration, neither did it minimize root canal recontamination time. The cold lateral compaction and single-cone technique appears to be effective in prevent the leakage of microorganisms.

It may be concluded that there was no difference in bacterial penetration between the root canal filling techniques. The mean root canal contamination time was 67 days.

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