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ANÁLISE DA PRESENÇA DE *GAP* ATRAVÉS DO MARCADOR NITRATO DE PRATA EM DENTES BOVINOS OBTURADOS POR DIFERENTES CIMENTOS ENDODONTICOS E APÓS O SELAMENTO DO CANAL PROTÉTICO COM RESINA *FLOW*

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PORTO ALEGRE

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ORIENTADOR: Prof. Dr. José Antônio Poli de Figueiredo

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RESUMO

RESUMO

A longevidade do tratamento endodôntico está na dependência de inúmeras variáveis, entre elas o comportamento do material obturador ao longo do tempo e o selamento do canal radicular, especialmente quando da exposição aos fluidos bucais. Em face ao acima exposto, o objetivo do presente estudo foi avaliar in vitro a presença de *gap* através do marcador nitrato de prata, em canais radiculares de dentes bovinos obturados por diferentes cimentos endodônticos e armazenados por 24 meses, e após o selamento do canal protético com resina *flow*. Sessenta incisivos bovinos tiveram as coroas dentais seccionadas e os canais radiculares preparados através da técnica seriada. Na sequência os espécimes foram randomicamente divididos em 6 grupos experimentais: (AH) 10 raízes obturadas com o cimento AH Plus; (CP) 10 raízes obturadas com o cimento Endo CPM-Sealer; (EN) 10 raízes obturadas com o cimento Endofill; (RE) 10 raízes obturadas com o cimento RealSeal SE; (SS) 10 raízes obturadas com o cimento AH Plus e preparo do canal protético; (CS) 10 raízes obturadas com o cimento AH Plus, preparo do canal protético e selamento com resina flow. Os canais radiculares foram obturados pela técnica da condensação lateral utilizando cones de guta-percha e um dos cimentos endodônticos descritos. Nos grupos SS e CS a desobturação parcial do canal radicular foi realizada imediatamente após a conclusão da obturação. Após a realização do preparo do canal protético, 10 espécimes escolhidos aleatoriamente tiveram a porção superficial do remanescente obturador selado com resina do tipo *flow*. A seguir, a porção coronária de todos os espécimes foi selada com cimento restaurador provisório e a saída foraminal obliterada com cera utilidade. Na sequência, as raízes dos grupos AH, CP, EN e RE, e SS e CS foram armazenadas em umidade 100% por 24 meses e 24 horas, respectivamente. Após esse período os espécimes foram imersos em solução de nitrato de prata e analisados no microscópio eletrônico de varredura (MEV). As áreas de união dentina-material obturador foram observadas para avaliar a presença de *gap* marcado por nitrato de

prata. Os resultados mostraram que houve diferença estatisticamente significativa entre os grupos experimentais AH, CP, EN e RE, quanto à presença de *gaps* marcados pelo de nitrato de prata ($p<0,05$). Os cimentos AH Plus, Real Seal SE e Endo CPM Sealer apresentaram menor quantidade de *gaps* marcados pelo nitrato de prata em comparação ao Endofill ($p<0,05$). Já entre os grupos SS e CS, os resultados mostraram que não houve diferença estatisticamente significativa em relação a presença de *gaps* marcados ($p> 0,05$).

Ao analisar os resultados pode-se concluir que todos os canais radiculares apresentaram *gap* marcado por meio do nitrato de prata; os cimentos endodônticos AH Plus, Endo CPM Sealer e RealSeal SE obtiveram menores escores de *gap* marcados que o cimento Endofill e o selamento do canal protético com resina *flow* não apresentou menor quantidade de *gap* marcado quando comparado ao grupo sem selamento.

DESCRITORES[1]

Nitrato de prata, Endodontia, Microscopia eletrônica de varredura; Preparo de canal radicular; Obturação do canal radicular.

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[1]DeCS- Descritores em Ciências da Saúde, disponível em HTTP://descs.bvs.br

ABSTRACT

ABSTRACT

The longevity of endodontic treatments depends on many variables, including the aging of the filling material and the sealing of the root canal, especially when there is exposure to oral fluids. The aim of this study was to evaluate in vitro the silver nitrate gap penetration in root canals of bovine teeth filled with different endodontic sealers and sealing of the post space preparation with flow resin. The dental crowns of sixty bovine incisors were sectioned and root canals were prepared using a serial technique. The specimens were randomly divided into 6 groups (n= 10 specimens each) according to the root canal sealer used: (AH) AH Plus; (CP) Endo CPM sealer; (EN) Endofill; (RE) RealSeal SE; (SS) AH Plus and post space preparation; (CS) AH Plus and post space preparation and sealing with resin flow. The root canals were filled by lateral condensation using gutta-percha and one of the sealers described above. In groups SS and CS the post space was performed immediately after filling. Ten specimens were randomly selected and the remaining filling was sealed with flow resin. The roots of groups AH, CP, EN and RE, and SS and CS were stored at 100% humidity for 24 months and 24 hours, respectively. After this period, the specimens were immersed in a solution of silver nitrate and prepared for electron microscope (SEM). The areas of filling material-dentin were observed to assess the degree of penetration by silver nitrate. The results show that there were statistically significant differences between the experimental groups AH, CP, EN and RE ($p<0,05$). The sealers AH Plus, Real Seal SE and Endo CPM Sealer showed less infiltration of silver nitrate compared to Endofill ($p<0,05$). Among groups SS and CS, the results showed no statistically significant difference in the penetration of silver nitrate ($p > 0.05$).

By analyzing the results we can conclude that all root canals showed some degree of gap penetration through the marker silver nitrate. AH Plus, Endo CPM Sealer and RealSeal SE obtained lower gap penetration scores than Endofill. The sealing of the post space

preparation with flow resin did not prevent gap formation when compared to the group without sealing.

DESCRIPTORS[2]

Silver nitrate, Endodontic, Scanning electron microscopy, Root canal preparation, Root canal obturation.

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[2]MsSH – Medical Subject Headings, available at: www.nlm.nih.gov/mesh

LISTA DE ANEXOS

SUMÁRIO

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INTRODUÇÃO GERAL

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O prognóstico favorável da terapia endodôntica está intimamente relacionado com a execução precisa de todas as etapas do mesmo, bem como um correto diagnóstico. É fundamental a obtenção da sanificação do sistema de canais radiculares através do preparo químico-mecânico e do uso da medicação intra-canal, assim como a realização de uma adequada obturação do canal radicular.

A completa sanificação dos canais radiculares ainda é um grande desafio para a Endodontia. Apesar do emprego de materiais e técnicas que propiciam a remoção e ou inativação de microrganismos e seus produtos, é inevitável a permanência de algumas bactérias em determinadas áreas da anatomia endodôntica [1-2].

Além do binômio sanificação/modelagem almejado com o preparo químico-mecânico do canal radicular, a terapia endodôntica objetiva, através do material obturador, impedir que ocorram infiltrações tanto na porção apical como coronal. A infiltração nos condutos e sua consequente contaminação[3] podem interferir de forma negativa no prognóstico do tratamento endodôntico.

Quando a restauração do elemento dentário exige a confecção de um retentor intra-radicular, a quantidade de material obturador responsável pelo selamento do canal reduz consideravelmente, aumentando os riscos de infiltração [4]. O preparo do canal protético é uma etapa importante que deve ser realizada por profissionais que conheçam as particularidades anatômicas dos canais radiculares e disponham de suficiente conhecimento técnico para manter as condições assépticas do tratamento endodôntico [5-6].

Para a realização de obturações satisfatórias são necessárias técnicas aprimoradas e, principalmente, materiais obturadores que proporcionem um bom selamento e respeitem os tecidos apicais e periapicais [7]. Na constante busca do material ideal têm-se utilizado para obturar os canais radiculares, cones de guta-percha ou resina associados a um cimento endodôntico.

A importância do cimento na realização das obturações endodônticas se reflete no grande número de estudos que avaliam a capacidade de selamento destes materiais [8-9-10-11-12-13-14-15-16-17-18-19-20-21-22-23-24-25-26]. Dentre os cimentos resinosos, o AH Plus é um dos mais estudados, demonstrando uma adequada capacidade de selamento nos estudos de infiltração [10-11-12-17-21]. Outros cimentos, como o RealSeal (a base de resina metacrilato) e o Endo CPM sealer (a base de MTA), também tem sido avaliados em estudos que verificam as propriedades do material obturador [21-23-27-28].

A longevidade do tratamento endodôntico [29] está na dependência de inúmeras variáveis, assim o comportamento do material obturador ao longo do tempo[30] também pode ter interferência na qualidade do mesmo, especialmente quando da exposição aos fluidos bucais.

A escolha do cimento e uma adequada técnica de obturação não são garantia de que uma infiltração não ocorra. Nos casos em que seja necessário o preparo para retentor intra-radicular o volume de material obturador diminui consideravelmente comprometendo o selamento apical. Quando a obturação fica restrita no terço apical, favorece a infiltração, o que permite a invasão e proliferação bacteriana [31].

A possibilidade de infiltrações coronárias em dentes tratados endodonticamente sucita a pesquisa de métodos para impedir ou minimizar este risco. Desta forma inúmeros materiais e técnicas de selamento da embocadura dos canais radiculares, ou mesmo do interior destes no caso de preparo protético, são evidenciados na literatura [32-33-34-35-36-37-38].

Estudos demonstram que a infiltração coronária é uma via de contaminação [39] ou recontaminação do sistema de canais radiculares, ocasionando o fracasso da terapia endodôntica. A infiltração em dentes humanos extraídos tem sido pesquisada por meio de diferentes marcadores, como corantes [8-40-10-11-12-15-17], bactérias [41-42-43-44-12], radioisótopos [45], métodos como a filtragem de fluidos [13-46-47] e, mais recentemente, o nitrato de prata [19-27].

Apesar do grande número de estudos realizados com o objetivo de avaliar a capacidade de selamento de diferentes materiais obturadores, as metodologias aplicadas são tão variadas que a comparação entre os resultados obtidos nos estudos torna-se limitada [48].

O emprego do nitrato de prata como marcador de infiltração tem por objetivo verificar a presença de espaços no canal radicular obturado, mesmo que de dimensões até nanométricas. Utilizado em diversos experimentos, o nitrato de prata permite a marcação de possíveis falhas no selamento quando analisado através da microscopia eletrônica de varredura ou de transmissão [49-50-51-19-51-27-53-54].

Em face ao acima exposto, o objetivo do presente estudo foi avaliar in vitro a presença de *gap* através da marcação pelo nitrato de prata, em canais radiculares de dentes bovinos obturados por diferentes cimentos endodônticos e canais protéticos selados com resina do tipo *Flow*.

OBJETIVOS

Objetivo geral

O objetivo do estudo foi avaliar in vitro a presença de *gap* por meio do marcador nitrato de prata em canais radiculares de dentes bovinos obturados com diferentes cimentos endodônticos, armazenados por 24 meses e em canais protéticos selados com resina do tipo *Flow*, através da microscopia eletrônica de varredura.

Objetivos específicos

a) Avaliar a presença de *gap* por meio do marcador nitrato de prata em canais radiculares de dentes bovinos obturados com os cimentos endodônticos AH Plus, Endo CPM Sealer, Endofill e RealSeal SE, armazenados por 24 meses.

b) Observar a presença de *gap* por meio do marcador nitrato de prata em canais radiculares de dentes bovinos obturados e com o canal preparado para pino, com o remanescente do material obturador selado com resina do tipo *Flow*.

CAPÍTULO I

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ARTIGO 1

Silver nitrate gap penetration in bovine teeth filled with endodontic sealers under long term storage.

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Title: Silver nitrate gap penetration in bovine teeth filled with endodontic sealers under long term storage.

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Abstract

Objectives: Longevity of endodontic treatment depends on many variables and the behavior of the filling material over time may lead to decreased longevity, especially when there is exposure to oral fluids. The aim of this study was to evaluate in vitro the silver nitrate gap penetration in root canals of bovine teeth filled with different endodontic sealers. **Methods:** The dental crowns of forty bovine incisors were removed and the root canal was instrumented up to K file #80. The specimens were randomly divided into four experimental groups according to root sealer used (n=10 for each group): (AH) AH Plus; (CP) Endo CPM sealer; (EN) Endofill; (RE) RealSeal SE. The roots were stored at 100% humidity for 24 months. The bonding areas of dentin-filling material were evaluated through silver nitrate gap penetration under SEM. **Results:** The results showed that there were statistically significant differences among the four experimental groups in terms of silver nitrate penetration ($p > 0.05$). The sealers AH Plus, Real Seal SE and Endo CPM Sealer presented lower silver nitrate gap penetration compared to Endofill. Pairwise comparisons denoted an evident discrepancy in the sealing ability of the Endofill sealer in comparison with the others. In all comparisons there was a statistically significant difference in the presence of silver nitrate gap penetration ($P > 0.05$). When the sealers AH Plus, RealSeal SE and Endo CPM Sealer were compared, a statistical difference was only observed between RealSeal SE and Endo CPM Sealer. **Conclusions:** All root canals showed some degree of silver nitrate gap penetration and the sealers AH Plus, Endo CPM Sealer and RealSeal SE obtained significantly lower scores for gaps marked by silver nitrate than Endofill sealer.

Keywords: Silver nitrate, Endodontic, Scanning electron microscopy, Root canal preparation.

Introduction

The favorable prognosis of endodontic therapy is closely related to a correct diagnosis and the precise execution of all phases. It is essential to adequately clean the root canal system by chemical-mechanical preparation and intracanal medication, especially in cases of pulp necrosis. Classic studies of endodontic literature have shown over the years, the microbial colonization of the root canal and its relationship with the establishment and maintenance of periapical inflammatory disease (1-2).

The complete cleaning of root canals continues to be a significant challenge in Endodontics. Despite the use of materials and techniques that allow the removal and or inactivation of microorganisms and their products, the permanence of some bacteria in the complex root canal system is inevitable (3). The sealing of the root canal system after cleaning and shaping aims to avoid apical or coronal leakage. The leakage and contamination of the root canals (4) can adversely affect the prognosis of endodontic treatment.

The longevity of the endodontic treatment (5) depends on a number of variables, including the behavior of the filling material over time (6) which may lead to decrease longevity, especially when there is exposure to oral fluids. The importance of sealer for the performance of endodontic fillings is reflected in the large number of studies that evaluate the sealing ability of these materials (7-11). Among the resin sealers, AH Plus is one of the most studied, demonstrating an excellent sealing ability in studies of leakage (8). Other resin and MTA-based sealers, such as the RealSeal SE (resin) and Endo CPM sealer (based on MTA) have also been studied (12-23).

Leakage has been investigated by different markers, dyes (7-8), bacteria (8), radioisotopes (24), fluid transport (25) and more recently, silver nitrate (9-12). Even with a large number of studies aiming to evaluate the sealing ability of different filling materials, the methods applied are so varied, that the comparison between the results obtained in studies

becomes limited (26). The use of silver nitrate as a marker of infiltration allows the verification of spaces in the root canal fillings, even to nanometer dimensions. Used in several experiments, silver nitrate allows marking of possible failures in sealing, when analyzed by scanning or transmission electron microscopy (9, 12, 27-30).

Identifying and understanding the variables that affect longevity and success of endodontic treatment contribute to the development of materials and techniques that can provide more favorable prognoses for a number of clinical conditions. Thus, the aim of this study was to evaluate in vitro the silver nitrate gap penetration in root canals of bovine teeth filling with different endodontic sealers.

Material and Methods

Specimen Preparation

The methodology was approved by the Scientific and Ethics Committee (CEEFO) of Pontifícia Universidade Católica do Rio Grande do Sul (RS, Brazil). Forty bovine incisors, obtained from animals aged approximately one year and killed for commercial reasons, were removed immediately after death and stored in sodium hypochlorite solution 1% (Novaderme Manipulation Pharmacy, Santa Maria, Brazil) during 24 hours, and then replaced with distilled water renewed every 48 hours.

The dental crowns were removed with the aid of a double-sided blade (KG Sorensen, Barueri, Brazil), so that the length of roots was standardized to 16.0 mm. The dental pulp was removed with a barbed broach and irrigation probe with a 1% sodium hypochlorite solution. Subsequently, the specimens were fixed in a vise (Mundial 60mm F-315) and the root canals were prepared with the standard technique.

Root canal filling

The specimens were randomly divided into four experimental groups: AH: 10 roots filled with AH Plus, CP: 10 roots filled with cement Endo-CPM Sealer, EN: 10 roots filled with cement Endofill, RE: 10 roots filled with RealSeal SE.

Prior to filling, the root canals were irrigated with 2.0 mL solution of 17% trisodium EDTA (Novaderme Manipulation Pharmacy, Santa Maria, Brazil) for 5 minutes. Then there was a final irrigation with 2.0 mL of distilled water (Novaderme - Dispensing pharmacy, Santa Maria - RS). The canals were dried with absorbent paper points (EndoPoints Industria e Comercio Ltda., Baron Creek, Paraíba do Sul, Rio de Janeiro). Root canals were filled by lateral condensation along with one of the following sealers: AH Plus (Dentsply De Trey GmbH, Konstanz, Germany), Endo CPM-Sealer (EGEO Laboratory

SRL, Buenos Aires, Argentina), Endofill (Dentsply Industria e Comercio Ltda., Petrópolis, RJ, Brazil) or RealSeal Self Etch sealer (SybronEndo Corporation, CA, USA) prepared according to the manufacturer's instructions.

After lateral condensation, a radiograph was performed in order to check the quality of the filling. Next, the cones were sectioned with a heated condenser (DUFLEX - SS White Dental Articles Ltda., Rio de Janeiro, RJ).

The coronal portion of the specimens was sealed with temporary cement (Cimpat - Septodont Brazil Ltda, Barueri, SP) and the apical foramen was obliterated with wax (Polidental, Cotia, SP). Subsequently, the roots were immersed in a glass containing distilled water and stored at 37 ° C for 24 hours. Then the specimens were removed from the water and stored in 100% humidity for 24 months.

Marking with silver nitrate and SEM evaluation

The temporary cement was removed e the specimens were covered with two layers of nail varnish (L'Oreal Brazil, Rio de Janeiro, RJ) and submerged in a solution of silver nitrate 50% / water 50% for 24 hours in the dark. Then, the roots were rinsed thoroughly in running water (1 hour) and immersed in a developing solution (Kodak, Rochester, NY, USA) for 8 hours. The roots were sectioned with a double face diamond wheel. The apical and middle third were discarded and the remaining root was prepared, providing the specimens (SPs). After this protocol, the polishing of SPs was performed to expose the union dentin / filling material. For this, water sandpapers 800, 1200, 1500, 2000 and 2500 (3M, Minnesota, USA) were mounted on a polishing machine (Buehler Ltd., Lake Bluff, IL, USA) and final polishing was carried out with a 1/4μm granulation paste (Arotec, Sao Paulo , Brazil).

SPs were submitted to slow dehydration with silica gel, mounted on stubs designed for JEOL 5800 (Jeol, Tokyo, Japan) and coated with gold / palladium in coater (MED 010,

Balzers Union, Balzers, Liechtenstein, Germany). The bonding areas of the dentin-filling material were analyzed to evaluate the degree of silver nitrate gap penetration, using an increase of approximately 700x.

A review of the silver nitrate gap penetration was performed by identifying and rating scores, using the following criteria:

1. Absence of gap.
2. Presence up to 20% of gap.
3. Presence up to 50% of gap.
4. Presence of more than 50% of gap.

Statistical Analysis

The nominal values of gap penetration scores were tabulated into spreadsheets and analyzed using descriptive statistics in SPSS (Statistical Package for Social Sciences, version 18.0) program. The 4 groups composed of different cements were compared by Kruskal-Wallis test followed by paired comparisons using the Mann-Whitney test at a significance level of 5%.

Results

The results presented in Table 1 show that there were statistically significant differences among the four experimental groups in terms of silver nitrate gap penetration (p> 0.05). The sealers AH Plus, Real Seal SE and Endo CPM Sealer presented lower gaps compared to Endofill (Table 2). Pairwise comparisons displayed an evident discrepancy in the sealing ability of the Endofill (Figure 1) when compared to the others. In all comparisons there was statistically significant difference in the presence of silver nitrate (P> 0.05). When sealers AH Plus, RealSeal SE and Endo CPM Sealer were compared, the only observed statistical difference was between RealSeal SE and Endo CPM Sealer, as shown in Table 3.

Discussion

Adequate sealing of the root canal system and maintenance of its integrity has been of high interest to researchers as has been the search for materials and techniques that are capable of impeding infiltration of liquids, microorganisms and their products. Results of the present study with regards to aging of the treatment (8,9), through storage of the specimens during 24 months, were not in conflict with previous studies from the literature for leakage into the filling material. Despite being a short period relatively to the period which endodontic treatments are designed to last, two years is frequently employed as a parameter in the evaluation of procedural outcomes. The presence of gaps in the dentin/filling material interface in the different cements tested demonstrated the same behavior found in studies verifying this condition immediately after completion of the filling.

Although the validity of methods utilized to evaluate infiltration in the experiments has been the target of critical scientific discussion, it is undeniable that silver nitrate, as a marker that has been validated in other studies, allows the tracking of gaps in the filling material, up to nanometric dimensions. The sealers evaluated also demonstrated results that are in line with the available literature, independent of the methodology utilized. Endofill presented the greatest presence of gaps, which is consistent with its composition as a zinc oxide-eugenol-based sealer (31).

It is important to note that the infiltration scores for Endo CPM Sealer, a mineral trioxide aggregate sealer, were similar to those found for the resin sealers AH Plus and RealSeal SE, even though the statistical analysis demonstrated the superiority of ReaSeal SE. As it is a powder/liquid cement, without a resin matrix, i.e., with lower viscosity and fluidity, it was hypothesized that it would present inferior results (30). Future experiments should be carried out with an aim to establish a reliable methodological standard in order to refine

available methods of evaluation allowing the comparison of results and their application in clinical settings.

Disclosure of conflicts of interest

We declare the absence of any conflict of interest regarding this paper.

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Table Legends

Table 1 – Statistically significant differences among the four experimental groups for silver nitrate gap penetration (p> 0.05)

	SCORES
Chi-square	25,565
df	3
Asymp. Sig.	,000

Table 2 – Mean Ranks of endodontic sealers AH Plus, Endo CPM Sealer, Endofill and RealSeal SE.

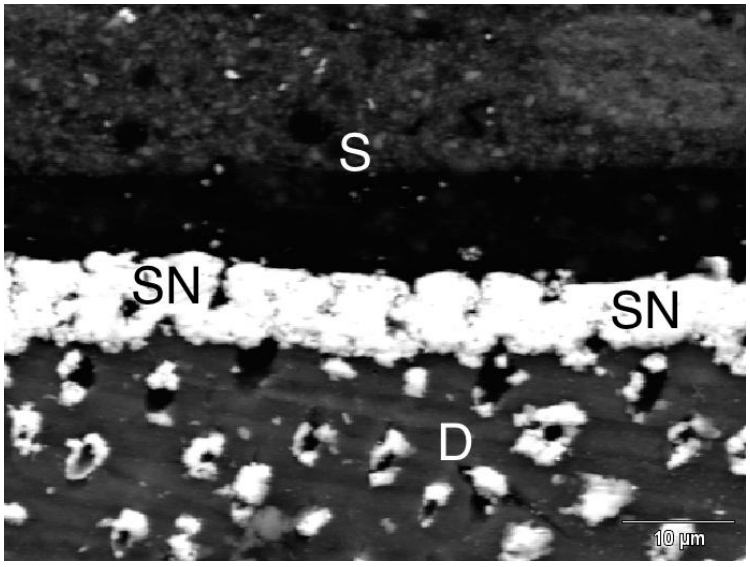
	GROUP	N	Mean Rank
SCORES	AH PLUS	10	14.25
	Endo CPM Sealer	10	20.45
	Endofill	10	34.60
	ReaSeal SE	10	12.70
	Total	40	

Table 3 – Statistically significant differences among the groups Endo CPM Sealer and Real Seal SE for silver nitrate gap penetration (p> 0.05)

	GROUP	N	Mean Rank	Sum of Ranks
SCORES	Endo CPM Sealer	10	13.00	130.00
	RealSeal SE	10	8.00	80.00
	Total	20		

Figure legends

Figure 1 – Silver nitrate gap penetration in Endofill group: sealer (S), dentin (D) and silver nitrate (SN).



CAPÍTULO II

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ARTIGO 2

Silver nitrate gap penetration in bovine teeth following post space preparation using hybrid technique and flow resin

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Title:

Silver nitrate gap penetration in bovine teeth following post space preparation using hybrid technique and flow resin

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Abstract

Objectives: Longevity of endodontic treatments depends on many variables and the behavior of the filling material over time may lead to decreased longevity, especially when there is exposure to oral fluids. The aim of this study was to evaluate in vitro the silver nitrate gap penetration in root canals of bovine teeth following post space preparation followed by sealing with hybrid technique and flow resin **Methods:** The dental crowns of twenty bovine incisors were cut and the root canal was instrumented with K file #80. The root canals were filled by lateral condensation, using gutta percha points and AH Plus sealer. Immediately after filling, post space preparation was performed. After of the prosthetic space preparation, 10 specimens were randomly assigned to receive sealing with flow composite resin. The specimens were submerged in a solution of silver nitrate and the bonding areas of dentin-filling material were evaluated for the degree of silver nitrate gap penetration. **Results:** The results showed that there were no statistically significant differences among the two experimental groups for silver nitrate gap penetration. **Conclusions:** All root canals showed some degree of gap penetration through the marker silver nitrate and the sealing of the post space preparation with flow resin did not present statistical differences in the markup of infiltration by silver nitrate, when compared to the group without sealing.

Keywords: Silver nitrate, Endodontic, Scanning electron microscopy, Root canal preparation, Aging.

Introduction

The sealing of the root canal system after cleaning and shaping aims to avoid apical or coronal leakage. The leakage and contamination of the root canals (4) can adversely affect the prognosis of endodontic treatment.

When the tooth restoration requires the post space preparation, the amount of filling material responsible for sealing the coronal third of the root canal reduces considerably, increasing the risks of leakage (5). The preparation of the prosthetic canal is an important step that should be performed by professionals (endodontists and prosthodontists) who know the anatomical particularities of root canals and have sufficient technical knowledge to maintain aseptic conditions in the endodontic treatment (6). Concern about the possibility of coronary infeccions in endodontically treated teeth has led to studies of methods to prevent or minimize the risk of infiltration. In this way, numerous materials and techniques for coronary sealing, or even the interior of the canals, in the case of prosthetic preparation, are found in the literature (7-13).

Leakage has been investigated by different markers, dyes (14-18), bacteria (14,19), radioisotopes (20), fluid transport (16,21) and more recently, silver nitrate (22,23). Even with a large number of studies aiming to evaluate the sealing ability of different filling materials, the methods applied are so varied, that the comparison between the results obtained in studies becomes limited (24) The use of silver nitrate as a marker of infiltration allows the verification of spaces in the root canal fillings, even to nanometer dimensions. Used in several experiments, silver nitrate allows marking of possible failures in sealing, when analyzed by scanning electron microscopy or transmission (22, 23, 25-30). Thus, the aim of the study was to evaluate the influence of sealing the post space preparation with flow resin on silver nitrate gap penetration in bovine teeth with filled roots by scanning electron microscopy (SEM).

Material and Methods

Specimen Preparation

The methodology was approved by the Scientific and Ethics Committee (CEEFO) of Pontifícia Universidade Católica do Rio Grande do Sul (RS, Brazil). Twenty bovine incisors, obtained from animals ageing approximately one year and killed for commercial reasons, were removed immediately after death and stored in sodium hypochlorite solution 1% (Novaderme Manipulation Pharmacy, Santa Maria, Brazil) during 24 hours, and then replaced with distilled water renewed every 48 hours.

The dental crowns were cut with the aid of a double-sided blade (KG Sorensen, Barueri, Brazil), so that the length of roots was standardized to 16.0 mm. The dental pulp was removed with a barbed broach and irrigation with a solution of 1% sodium hypochlorite. Subsequently the specimens were fixed in a vise (World 60mm F-315) and the root canals were prepared by starndar technique.

Root canal filling

The specimens were randomly divided into two experimental groups: Group I: 10 root canals without sealing of the post space preparation; Group II: 10 root canals with sealing of the post space preparation with flow resin.

Prior to filling, the root canals were irrigated with 2.0 mL solution of 17% trisodium EDTA (Novaderme Manipulation Pharmacy, Santa Maria, Brazil) for 5 minutes. Then there was a final irrigation with 2.0 mL of distilled water (Novaderme - Dispensing pharmacy, Santa Maria - RS). The canals were dried with absorbent paper points (EndoPoints Industria e Comercio Ltda., Baron Creek, Paraíba do Sul, Rio de Janeiro).

The root canals were filled by the lateral condensation, using gutta percha and AH Plus sealer (Dentsply De Trey GmbH, Konstanz, Germany).

After lateral condensation, a radiograph was performed in order to check the quality of the filling. Next, the cones were sectioned with a heated condenser (DUFLEX - SS White Dental Articles Ltda., Rio de Janeiro, RJ).

Post Space Preparation and sealing with Resin Flow

The procedures described below were carried out with the use of a operative microscope (Alliance, São Paulo, Brasil). The preparation of the prosthetic space was performed immediately after completion of the filling. The removal procedure was considered satisfactory based on observation in the control radiograph measuring 4.0mm of remaining filling material.

After the prosthetic space preparation, 10 specimens were randomly assigned to receive sealing with flow composite resin (Tetric Flow , Ivoclar Vivadent, Schaan, Liecgtenstein). The root canal was conditioned with 37% phosphoric acid (37 Condac, FGM Dental Products Ltd. , Joinville , SC / Brazil) for 15 seconds. Vigorous washing was then performed for 20 seconds, using a triple syringe. The canal was dried with absorbent paper points and adhesive (Scotchbond, 3M ESPE, Sumaré, São Paulo, SP) applied with a microbrush (Brush KG, KG Sorensen, Medical Industry and Trade Burs and Surgical Burs Tips Ltda., São Paulo, SP) on the filling material and adjacent dentin.

Next, the flow composite resin was applied on the remaining material using the syringe itself, to form a layer of approximately 1.0mm. The trapped material was activated by use of curing light for 20 seconds.

The coronal portion of the specimens was sealed with temporary cement (Cimpat - Septodont Brazil Ltda, Barueri, SP) and the apical foramen was obliterated with Wilson wax (Polidental, Cotia, SP). Next, the roots were immersed in a glass containing distilled water and stored at 37 ° C for 24 hours.

Marking with silver nitrate and SEM evaluation

The temporary cement was removed e the specimens were covered with two layers of nail varnish (L'Oreal Brazil, Rio de Janeiro, RJ) and submerged in a solution of silver nitrate 50% / water 50% for 24 hours in the dark. Then, the roots were rinsed thoroughly in running water (1 hour) and immersed in a developing solution (Kodak, Rochester, NY, USA) for 8 hours. The roots were sectioned with a double face diamond wheel. The apical and middle third were discarded and the remaining root was prepared, providing the specimens (SPs). After this protocol, the polishing of SPs was performed to expose the union dentin / filling material. For this, water sandpapers 800, 1200, 1500, 2000 and 2500 (3M, Minnesota, USA) were mounted on a polishing machine (Buehler Ltd., Lake Bluff, IL, USA) and final polishing was carried out with a 1/4μm granulation paste (Arotec, Sao Paulo , Brazil).

SPs were submitted to slow dehydration with silica gel, mounted on stubs designed for JEOL 5800 (Jeol, Tokyo, Japan) and coated with gold / palladium in coater (MED 010, Balzers Union, Balzers, Liechtenstein, Germany). The bonding areas of the dentin-filling material were analyzed to evaluate the degree of silver nitrate gap penetration, using an increase of approximately 700x.

A review of the silver nitrate gap penetration was performed by identifying and rating scores, using the following criteria:

1. Absence of gap.
2. Presence up to 20% of gap.
3. Presence up to 50% of gap.
4. Presence of more than 50% of gap.

Statistical Analysis

The nominal values of infiltration scores were tabulated into spreadsheets and analyzed using descriptive statistics in the SPSS (Statistical Package for Social Sciences, version 18.0) program. The two different groups representing the presence or absence of sealing were compared by Mann-Whitney test, significance level of 5%

Results

The results presented in Tables 1 and 2 showed that there were no statistically significant differences among the two experimental groups for silver nitrate gap penetration (Figure 1).

Discussion

Adequate sealing of the root canal system and maintenance of its integrity has been an important problem in routine clinical practice. Many authors propose the use of protection of the filling material in order to avoid penetration of fluids and consequent contamination of the root canal system. Reports from literature show a number of materials that have been used to seal the prosthetic canal. Flow resin was chosen for the present study due to its easy application, good viscosity and compatibility with superior endodontic sealers.

After SEM analysis, the presence of gaps marked by silver nitrate on the interface of the filling material was observed in both groups. Although the area marked by silver nitrate was slightly greater on the group without sealant, this difference was not statistically significant.

In the present study, both the preparation of the prosthetic canal and the sealing with flow resin were carried out with the aid of a clinical microscope in order to provide greater technical. Silver nitrate was chosen as a marker of gaps in the dentin/filling interface or even in other sealed areas of of the prosthetic canal, due to its capacity to permeate spaces of nanometric dimensions, as shown in previous studies (25, 29, 30).

Based on the results, it is probable that the presence of gaps in both groups was due to a failure in the three-dimensional filling of the root canal. In some specimens in the group that received hybridization with flow resin, the silver nitrate marking was greater than in the group that did not receive sealant. This may be explained by two factors: inadequate photopolymerization and acid etching directly on the remaining gutta-percha. Inadequate photopolymerization is due to the difficulty of light to reach the depths of the prosthetic canal, leading to the lack of sealing of the flow resin. In terms of acid etching, it was observed that in the group with sealant, the remaining filling material was less compact, with the presence

of empty spaces in the portion below the protective plug, which also contributed to greater silver nitrate marking.

Flow resin has not been used as sealing material in other studies that utilize a plug, and in most of these studies materials such as silver amalgam, cyanoacrylate and varnishes were used. Although protective plugs are commonly employed, the conditions that they provide should be analyzed, such as easiness of application, good sealing capacity and facile removal.

Due to the lack of studies in the literature related to the utilization of flow resin with protective plugs and considering that it is the only material that requires acid conditioning and photopolymerization of the adhesive, more studies are required to understand the results regarding the employment of this treatment alternative as well as to contribute to unveil the interactions that occur among the materials utilized in this study.

Disclosure of conflicts of interest

We declare the absence of any conflict of interest regarding this paper.

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Table Legends

Table 1 – Mean Rank of group with and without resin flow sealing.

GROUP	N	Mean Rank	Sum of Ranks
With Sealing	10	8.25	82.50
Without Sealing	10	12.75	127.50
Total	20		

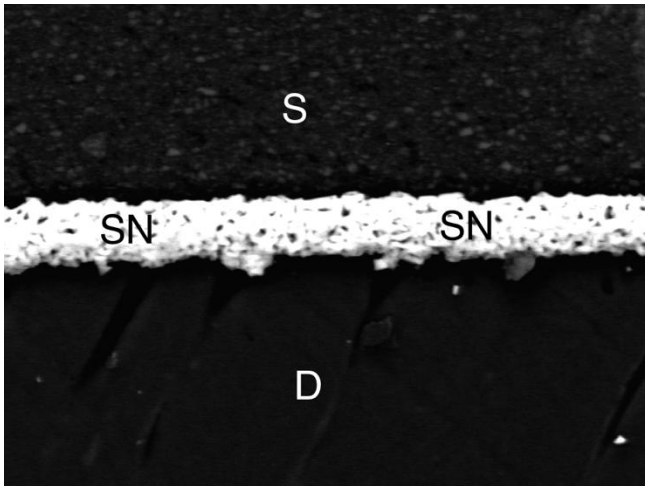
Table 2 – Silver nitrate gap penetration statistics (p> 0.05)

Test Statisticsb

	SCORE
Mann-Whitney U	27,500
Wilcoxon W	82,500
Z	-1,813
Asymp. Sig. (2-tailed)	,070
Exact Sig. [2*(1-tailed Sig.)]	,089

Figure legends

Figure 1 – Silver nitrate gap penetration in the group without flow resin sealing: sealer (S), dentin (D) and silver nitrate (SN).



DISCUSSÃO GERAL

DISCUSSÃO GERAL

O selamento adequado do sistema de canais radiculares e a manutenção de sua integridade tem sido de grande interesse para pesquisadores, assim como a busca de materiais e técnicas capazes de impedir a infiltração de fluidos, microrganismos e seus produtos.

Embora a validade dos métodos empregados para avaliar a infiltração nos experimentos tem sido alvo de discussão científica, é inegável que o nitrato de prata, validado como marcador em outros estudos, permite o rastreamento de *gaps* no material obturador até mesmo de dimensões nanométricas. A microscopia eletrônica de varredura permite a aproximação da imagem em áreas de contato entre dentina e material selador proporcionando maior precisão dos achados.

A utilização de dentes bovinos contribui para diminuir falhas relacionadas ao uso de dentes humanos. A idade dos espécimes pode ser padronizada e o sistema de túbulos dentinários é de mais fácil manuseio e controle. É de se especular que, considerando a facilidade do procedimento clínico, se ocorre formação de *gap* em dentes bovinos preparados e obturados em condições ideais e em laboratório, mais facilmente isso ocorrerá na clínica odontológica.

Os cimentos avaliados demonstraram resultados de acordo com a literatura disponível, independente da metodologia utilizada. O cimento Endofill apresentou a maior presença de *gaps*, o que é coerente com a composição de um cimento à base de óxido de zinco e eugenol (31).

É importante observar que os escores de infiltração para o Endo CPM Sealer, um cimento de agregado trióxido mineral, foram semelhantes aos encontrados para os cimentos resinosos AH Plus e RealSeal SE, embora a análise estatística tenha demonstrado a superioridade do cimento ReaSeal SE. Como o Endo CPM Sealer é um cimento pó / líquido,

sem uma matriz de resina, isto é, com menor viscosidade e fluidez, foi levantada a hipótese de que ele iria apresentar resultados inferiores (30).

Alguns autores propõem a utilização de proteção do material obturador a fim de evitar a penetração de fluidos e a consequente contaminação do sistema de canais radiculares. Os relatos da literatura mostram um grande número de materiais que têm sido utilizados para selar o canal protético. A resina *flow* foi escolhida para o presente estudo devido à sua aplicação fácil, boa viscosidade e compatibilidade com os melhores cimentos endodônticos .

Após a análise no MEV, a presença de *gaps* marcados por nitrato de prata na interface do material obturador foi observada em ambos os grupos. Embora a área marcada por nitrato de prata foi ligeiramente maior no grupo sem o selamento com resina *flow*, esta diferença não foi estatisticamente significativa.

No presente estudo, tanto a preparação do espaço protético quanto o selamento com resina flow foram realizados com o auxílio de um microscópio clínico, a fim de proporcionar maior rigor na técnica. O nitrato de prata foi escolhido como um marcador dos *gaps* na interface dentina / material obturador ou mesmo em outras áreas seladas do canal protético, devido à sua capacidade de penetração em espaços de dimensões nanométricas, como demonstrado em estudos anteriores (25, 29, 30) .

Com base nos resultados, é provável que a presença de *gaps*, em ambos os grupos ocorreu devido a uma falha no preenchimento tridimensional do canal radicular. Alguns espécimes do grupo que recebeu a hibridação com resina flow, apresentaram a marcação de nitrato de prata maior do que no grupo que não recebeu o selamento. Isto pode ser explicado por dois fatores: fotopolimerização inadequada ou o condicionamento ácido diretamente sobre a guta-percha remanescente. A fotopolimerização inadequada pode ser resultado da dificuldade de penetração da luz na profundidade do canal protético, o que culminou na falha do selamento. Quanto ao condicionamento ácido, observou-se que no grupo com o selamento,

o material obturador remanescente apresentou espaços vazios na porção abaixo da resina flow, o que também pode ter contribuído para uma maior marcação do nitrato de prata.

A resina flow não foi empregada como material de vedação em outros estudos que utilizam este protocolo, sendo que na maior parte destes foram utilizados o amálgama de prata, o cianoacrilato entre outros.

Devido à falta de estudos na literatura relacionada com a utilização de resina flow com *plug* de proteção e sendo que é o único material que requer condicionamento ácido e fotopolimerização do adesivo, mais estudos são necessários para entender os resultados referentes ao emprego deste alternativa de tratamento, bem como para contribuir para uma maior compreensão das interações que ocorrem entre os materiais utilizados neste estudo.

Futuros experimentos devem ser realizados com o objetivo de estabelecer um padrão metodológico confiável, a fim de aperfeiçoar os métodos disponíveis de avaliação, permitindo a comparação dos resultados e sua aplicação na prática clínica. E de se especular que, utilizando complementarmente a microscopia eletrônica de transmissão, o efeito do material obturador e do selamento protético sobre a estrutura do colágeno dentinário possam ser avaliados, constituindo em campo promissor de investigação.

CONCLUSÕES

CONCLUSÕES

A partir dos resultados do presente estudo pode-se concluir:

1. Todos os canais radiculares apresentaram algum grau de presença de *gap* através do marcador nitrato de prata;
2. Os cimentos endodônticos AH Plus, Endo CPM Sealer e RealSeal SE obtiveram menores escores de presença de *gap* marcado pelo nitrato de prata que o cimento Endofill, sendo esta diferença estatisticamente significante;
3. O selamento do canal protético com resina *flow* não apresentou diferenças estatísticas quanto a presença de *gap* marcado através do nitrato de prata, quando comparado ao grupo sem selamento.

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ANEXOS

ANEXOS A

Aprovação pela Comissão Científica e de Ética da Faculdade de Odontologia/PUCRS

Comissão Científica e de Ética
Faculdade da Odontologia da PUCRS

Porto Alegre 02 de Abril de 2014

O Projeto de: Tese

Protocolado sob nº:	0004/14
Intitulado:	Avaliação in vitro da capacidade de selamento do canal protético hibridizado com resina flow.
Pesquisador Responsável:	Prof. Dr. José Antonio Poli de Figueiredo
Pesquisadores Associados:	Charles da Cunha Pereira
Nível:	Tese / Doutorado

Foi ***aprovado*** pela Comissão Científica e de Ética da Faculdade de Odontologia da **PUCRS** em *Dois de Abril de Dois Mil e Quatorze*

Profa. Dra. Luciane Macedo de Menezes
Coordenadora da Comissão Científica e de Ética da
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