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Thais Camponogara Bohrer

EFEITO DE CIMENTOS ENDODÔNTICOS NA RESISTÊNCIA ADESIVA DE PINOS DE FIBRA E NO GRAU DE CONVERSAO DE DOIS CIMENTOS RESINOSOS

Santa Maria, RS
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Dissertação apresentada ao Curso de Mestrado do Programa de Pós-Graduação em Ciências Odontológicas, Área de Concentração em Odontologia, ênfase em **Prótese Dentária**, da Universidade Federal de Santa Maria (UFSM, RS), como requisito parcial para obtenção do grau de **Mestre em Ciências Odontológicas**.

Orientador: Prof. Dr. Osvaldo Bazzan Kaizer

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Prof. Dr. Osvaldo Bazzan Kaizer (UFSM)
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RESUMO

EFEITO DE CIMENTOS ENDODÔNTICOS NA RESISTÊNCIA ADESIVA DE PINOS DE FIBRA E NO GRAU DE CONVERSAO DE DOIS CIMENTOS RESINOSOS

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O restabelecimento da função dentária após a perda da porção coronária depende, em grande parte, da resistência adesiva dos pinos intrarradiculares. Entretanto, vários fatores podem afetar a adesão entre a dentina radicular e o retentor intrarradicular utilizado, um deles é o cimento utilizado para obturação do canal radicular na endodontia. Em vista disso, o objetivo desse estudo é avaliar o efeito de dois cimentos endodônticos em diferentes tempos de armazenamento na resistência adesiva entre pinos de fibra e as paredes do canal radicular, e verificar a influencia de cimentos endodônticos no grau de conversão dos cimentos resinosos. Para a realização do estudo, 180 dentes bovinos foram tratados endodonticamente utilizando dois cimentos endodônticos, à base de eugenol (Endofill) ou à base de resina epóxica (AH Plus). A seguir, os espécimes foram armazenados em estufa a 37 °C nos tempos 24 h, 6 meses e 12 meses. Decorrido o tempo de armazenamento, os canais radiculares foram preparados para a cimentação do pino de fibra utilizando dois cimentos resinosos, RelyX U200 ou Multilink Automix. O teste de *push-out* e a análise das falhas foram realizados. Para a análise do grau de conversão foi utilizado o microscópio Raman. As amostras foram preparadas utilizando somente cimento resinoso ou cimento resinoso e cimento endodôntico. Finalmente, os dados do teste de *push-out* foram submetidos à análise estatística e as porcentagens do teste de grau de conversão foram calculadas. O cimento AH Plus obteve os maiores valores de *push-out* nos tempos 24 h e 6 meses. Não teve diferença significativa entre os cimentos AH Plus e Endofill após 12 meses ($p < 0,05$). Além disso, o grau de conversão foi afetado pelo cimento endodôntico à base de eugenol. Portanto, o uso de cimentos endodônticos à base de eugenol não é recomendado, pois afeta a resistência adesiva dos cimentos resinosos.

Palavras-chave: Cimentação. Cimentos endodônticos. Eugenol. Grau de conversão. Pinos de fibra. *Push-out*.

ABSTRACT

EFFECT OF ENDODONTIC SEALERS ON THE BOND STRENGTH OF FIBREGLASS POSTS AND THE DEGREE OF CONVERSION OF TWO RESIN CEMENTS

AUTHOR: Thais Camponogara Bohrer
ADVISOR: Osvaldo Bazzan Kaizer

Restoring dental function after partial loss of the coronary structure depends mainly of post retainer adhesive strength. However, several factors may affect the adhesion between the root dentin and the radicular post used including the endodontic sealers used for root canal obturation during endodontic treatment. Therefore, the objective of this study is to evaluate the effect of two endodontic sealers on push-out bond strength between fibreglass posts and root canal wall after different storage times and verify the influence of the endodontic sealers on the degree of conversion of resin cements. For this study, 180 bovine teeth were endodontically treated using two endodontic sealers, eugenol-based (Endofill) or epoxy resin-based (AH Plus). Subsequently, the specimens were stored in 100% relative humidity at 37 °C for 24 h, 6 or 12 months. After the respective storages times, the root canals were prepared for fibreglass post cementation using two resins cements, RelyX U200 or Multilink Automix. The push-out test and the analysis of failures were performed. For the degree of conversion analysis with a Raman microscope. The samples were prepared using only resin cement or resin cement and endodontic sealer. Finally, the push-out data were subjected to statistical analysis and the percentages of degree of conversion were calculated. The AH Plus sealer obtained the highest values of push out bond strength at 24 h and 6 months. There was no difference between the values of AH Plus and Endofill sealer at time 1 year ($p < 0.05$). Moreover, the degree of conversion was affected by the endodontic sealers used. Therefore, the use of eugenol-based sealer is not recommended because it affects the push-out strength of fibreglass posts.

Keywords: Degree of conversion. Endodontic sealer. Eugenol. Fibreglass post. Luting. Push-out bond strength test.

SUMÁRIO

1 INTRODUÇÃO

A cárie e as fraturas ocasionadas por traumas são as principais causas de perda extensa da porção coronária da estrutura dentária, o que pode impossibilitar a funcionalidade da mesma (DEMIRYÜREK et al., 2010; DIMITROULI et al., 2011). Para obter retenção mecânica e adequação biológica desta estrutura, geralmente é necessário o tratamento endodôntico e a posterior cimentação de um retentor intrarradicular possibilitando a reconstrução da estrutura coronária perdida (DEMIRYÜREK et al., 2010; ROSA et al., 2013; MANICARDI et al., 2011).

Há um crescimento da utilização de pinos de fibra de vidro na clínica odontológica, devido ao fato de possuírem um módulo de elasticidade similar ao da dentina e apresentarem melhores propriedades estéticas comparados com os pinos metálicos (GIACHETTI et al., 2009; NAUMANN et al., 2012; SARKIS-ONOFRE et al., 2014; SILVA et al., 2009). Além disso, a utilização desses retentores intrarradiculares associados a uma técnica de cimentação adesiva tem sido comumente aplicada, pois promove maior resistência de união comparado com outros tipos de cimentos, menor microinfiltração e maior resistência à fratura da restauração final (ALEISA et al., 2012; ALTINTAS, ELDENIZ, USUMEZ, 2008; DEMIRYUREK et al., 2010; ZHANG et al. 2008).

Para a simplificação da técnica de cimentação, foi introduzido no mercado o cimento resinoso chamado autoadesivo (FERRACANE, STANSBURY, BURKE, 2011; ÖZCAN et al., 2013; TOMAN et al., 2009). Ainda, uma recente revisão sistemática demonstrou que os cimentos resinosos autoadesivos apresentam uma técnica menos sensível comparada com os cimentos convencionais (SKUPIEN et al., 2015).

No entanto, vários fatores podem influenciar na retenção dos pinos intrarradiculares cimentados, entre eles a anatomia radicular, a técnica e os materiais utilizados para tratamento endodôntico e para a cimentação do pino intrarradicular (DIMITROULI et al., 2011). Há diversos estudos que comprovaram que os cimentos endodônticos utilizados para a obturação do canal radicular podem interferir na adesão dos pinos pré-fabricados (ALEISA et al., 2012; BARBIZAM et al.,2011; CECCHIN et al.,2012; LIMA et al., 2016; MOSHARRAF, ZARE, 2014; NGOH ET AL., 2001; ÖZCAN et al., 2013).

Um exemplo desses cimentos são os que contêm eugenol em sua composição, sendo ainda uma opção de grande parte dos endodontistas devido ao seu longo histórico de sucesso clínico e ao seu baixo custo comparado aos demais cimentos endodônticos disponíveis (DEMIRYÜREK et al., 2010). Entretanto, esses cimentos podem interferir no grau de polimerização dos cimentos resinosos, devido à presença de radicais livres no componente fenólico da molécula de eugenol que ao reagir com componentes do cimento resinoso poderá diminuir a conversão de ligações monoméricas (C=C) em ligações poliméricas (C-C), com isso poderão ser afetadas as propriedades físicas e mecânicas do polímero formado, no caso, o cimento resinoso (BEHR et al., 2004, DIAS et al., 2009).

Baldissara et al. (2006) sugeriram que resíduos de eugenol podem permanecer no canal radicular mesmo após a realização do preparo para o pino intrarradicular. Por outro lado, alguns autores sugerem que, durante a execução do preparo para o retentor, a maioria do eugenol remanescente no canal radicular é removido, não havendo a inibição da reação de polimerização dos cimentos resinosos (MOSHARRAF; ZARE, 2014; NGOH et al., 2001).

Outro aspecto que tem sido alvo de estudos é a influência do tempo decorrido entre a obturação do canal radicular e a cimentação do pino intrarradicular na resistência adesiva desse retentor (ALEISA et al., 2016; FARIA E SILVA et al., 2012; ROSA et al., 2013; VANO et al., 2006). Aleisa et al. (2016) demostraram que a cimentação do retentor intrarradicular nas primeiras 24 horas após a obturação obteve melhores valores de adesão comparado com 2 semanas.

Em contrapartida, Vano et al. (2006) verificaram que a cimentação do pino intrarradicular imediatamente após a obturação do canal radicular obteve os menores valores de resistência adesiva comparado com 24 horas e 1 semana, os quais não diferiram entre si. Já, Rosa et al. (2013) relataram que o tempo decorrido entre a obturação do canal e a cimentação do pino não influenciou na resistência adesiva do pino de fibra à dentina radicular.

De acordo com os estudos citados acima, podemos constatar que os resultados obtidos quanto à influência do tempo de espera pós-endodontia para a cimentação na retenção do pino intrarradicular são controversos (ALEISA et al., 2016). Ainda, Altmann et al. (2015) verificaram que há na literatura somente estudos com no máximo 15 dias de armazenamento, demostrando a necessidade de novos estudos.

De acordo com Goracci et al. (2007) para a determinação da resistência adesiva em superfícies dentinárias radiculares, podemos utilizar testes de cisalhamento, microtração e *push-out*. Porém, o teste de *push-out* é o mais utilizado pelos estudos na literatura e o que reproduz a melhor evidência disponível quando se testa materiais em dentina radicular, devido ao fato de promover tensões de cisalhamento na interface cimento resinoso e dentina e, também, cimento resinoso e pino intrarradicular (COLLARES et al., 2015; GORACCI et al., 2005).

Devemos lembrar, também, que uma das principais maneiras de se determinar as propriedades mecânicas de materiais à base de resina é por meio do teste do grau de conversão (KOMORI et al., 2010). Porém, até o presente momento, há na literatura uma deficiência de estudos que avaliem diretamente o grau de conversão dos cimentos resinosos utilizados atualmente em contato com cimentos endodônticos que possam vir a interferir no grau de polimerização desses materiais.

Desse modo, verificamos a importância da avaliação do grau de conversão dos cimentos resinosos, pois um insuficiente grau de conversão, além de influenciar as propriedades mecânicas dos materiais, diminui a estabilidade dimensional e a adesão entre cimento resinoso e a estrutura dentária (BAGIS; RUEGGEBERG, 2000; JANDA et al., 2007). Aumentando, também, a sorção e a solubilidade, os quais poderão influenciar diretamente na longevidade dos procedimentos restauradores (LOPES et al., 2015).

Porém, Faria e Silva et al. (2007) relataram que um baixo grau de conversão de polimerização do cimento resinoso não necessariamente reproduz uma menor resistência adesiva do pino intrarradicular. Em casos em que o fator-C é crítico, como nos canais radiculares, um baixo grau de conversão pode ser visto como uma vantagem, devido ao fato de reduzir a contração de polimerização nessa região, diminuindo, assim, a formação de estresse nessa região (BOUILLAGUET et al., 2003; CALIXTO et al., 2012; LE BELL et al., 2003).

Portanto, são necessários novos estudos buscando a relação entre a retenção adesiva dos pinos intrarradiculares e o grau de conversão dos cimentos resinosos utilizados na presença ou não de cimentos endodônticos contendo eugenol. Além do mais, verificar a influência do tempo de espera pós-endodontia para a cimentação desses retentores em um maior período de tempo do que os estudos existentes na literatura.

O presente trabalho será apresentado em forma de artigo, intulado como "**Effect of endodontic sealers on the bond strength of fibreglass posts and the degree of conversion of two resin cements**" tem como objetivo avaliar a resistência adesiva de pinos de fibra de vidro à dentina radicular em dentes bovinos obturados com cimento à base de óxido de zinco e eugenol ou com cimento à base de resina epóxica após diferentes tempos de armazenamento. Além disso, verificar a influência dos cimentos endodônticos testados no grau de conversão de dois cimentos resinosos.

2 ARTIGO - EFFECT OF ENDODONTIC SEALERS ON THE BOND STRENGTH OF FIBREGLASS POSTS AND THE DEGREE OF CONVERSION OF TWO RESIN CEMENTS

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EFFECT OF ENDODONTIC SEALERS ON THE BOND STRENGTH OF FIBREGLASS POSTS AND THE DEGREE OF CONVERSION OF TWO RESIN CEMENTS

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EFFECT OF SEALER IN ADHESION

Keywords: degree of conversion, endodontic sealer, eugenol, fibreglass post, luting. push-out bond strength test

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ABSTRACT

EFFECT OF ENDODONTIC SEALERS ON THE BOND STRENGTH OF FIBREGLASS POSTS AND THE DEGREE OF CONVERSION OF TWO RESIN CEMENTS

Aim To evaluate the effect of two endodontic sealers on push-out bond strength between fibreglass posts and root canal wall after different storage times and verify the influence of the endodontic sealers on the degree of conversion of resin cements.
Methodology A total of 180 bovine teeth were endodontically treated using two endodontic sealers, eugenol-based, Endofill, or epoxy resin-based, AH Plus. Subsequently, the specimens were stored in 100% relative humidity at 37 °C for 24 h, 6 months, or 12 months. After the respective storage times, the root canals were prepared for fibreglass post cementation using two resins cements, RelyX U200 or Multilink Automix. The push-out test and the analysis of failures were performed. For the degree of conversion analysis using a Raman microscope, samples were prepared using only resin cement or resin cement and endodontic sealer. Finally, the push-out data were subjected to statistical analysis and the percentages of degree of conversion were calculated.
Results The AH Plus sealer obtained the highest values of push-out bond strength at 24 h and 6 months. There was no difference between the values of AH Plus and Endofill sealer at 1 year ($p < 0.05$). Moreover, the degree of conversion was affected by the Endofill sealer.
Conclusion The use of a eugenol-based sealer is not recommended because it affects the push-out strength of fibreglass posts and degree of conversion of the resin cements.

INTRODUCTION

Fibreglass posts are frequently used to restore endodontically treated teeth with extensive loss of coronal structure mainly for their esthetic properties, elastic modulus similar to dentine, and favorable stress distribution that reduces the risk of root fracture (Stewardson 2001, Boschian Pest *et al*. 2002, Barjau-Escribano *et al*. 2006, Schmitter *et al*. 2007). For better retention, the adhesive cementation of the fibreglass posts has been indicated due to higher bond strength compared to other types of cements (Soares *et al*. 2012, Taira *et al*. 2007).

Different resin cements such as regular cements and self-adhesive cements have been used to cement fibreglass posts, the latter being a simplified approach (Mazzoni *et al*. 2009, Ferracane *et al*. 2011). Cementation with resin cements reduces the wedging effect and provides better retention, less microleakage, and more resistance to tooth fracture (Cheung 2005, Demiryurek *et al*. 2010, Aleisa *et al*. 2012).

Luting of posts is a critical procedure that can be affected by several factors, such as the sealer used for root canal endodontic treatment (Barbizam *et al*, 2011, Cecchin *et al*. 2012, Lima *et al*. 2016). Eugenol-based sealers are known to interfere with the polymerization of the resin cements and may affect the bond strength of the fibreglass posts (Schwartz 2006, Dias *et al*. 2009, Altmann *et al*. 2015). On the other hand, the resin-based sealers are described as the gold standard, since they do not affect the adhesion of the intraradicular posts (Baldissara *et al*. 2006, Brackett *et al*. 2006, Nunes *et al*. 2008).

The influence of the waiting time after root canal endodontic treatment on the bond strength of the cemented root posts remains unknown (Vano *et al*. 2006, Faria e Silva *et al*. 2012, Rosa *et al*. 2013, Aleisa *et al*. 2016). There is no consensus by the scientific community regarding the effect of waiting time after canal filling on post adhesion/retention .A systematic review by Altmaan *et al*. (2015) described that there are studies with a maximum of a 15 day waiting period for cementation.

With respect to the time between root canal filling and fibreglass post cementation, it is possible that prolonged contact of the endodontic sealer with the dentine walls may affect future adhesion to adhesive systems (Hagge *et al*. 2002, Izadi *et al*. 2013). Also, the retention of the fibreglass posts in the root canal is also derived from the coefficient of friction of the resin cements, which is directly affected by the mechanical properties of these cements (Goracci *et al*. 2005, Cury *et al*. 2006).

This highlights the importance of the degree of conversion for the longevity of the restorations once there is a direct relation between the degree of conversion and the increase of the mechanical properties (Janda *et al*. 2007, Nihei *et al*. 2008, Bahari *et al*. 2014). Eugenol-based sealers have free radicals, which in contact with the resin cements decrease the conversion of monomeric units to polymeric structures, directly influencing the degree of conversion (Altmann *et al*. 2015, Dias *et al*. 2009, Lima *et al*. 2016). Despite this, lack of studies evaluating the degree of conversion of resin cements in the presence of endodontic sealers.

The aims of this study were to evaluate the effect of two endodontic sealers on push-out bond strength between fibreglass posts and root canal walls after different storage times and verify the influence of the endodontic sealers on the degree of conversion of resin cements. The null hypotheses were: 1) there is no influence of the endodontic sealers and waiting time for fibreglass post cementation on the bond strength; and 2) the endodontic sealers do not influence the degree of conversion of the resin cements.

MATERIALS AND METHODS

Experimental design

To determine the number of teeth to be used in the present study, the sample size calculation was performed using the OpenEpi 3.01 program (Dean *et al*. 2013), with the parameters described in the study of Rosa *et al*. (2013) as references. The power of the study was considered to be 80% and the significance level was set at 0.05, resulting in 12 teeth per group. However, taking into account the variability of the root anatomy of bovine teeth, 15 teeth per group were used. Each procedure of this study was performed by a trained operator.

The cements used are listed in the Table 1.

The crown portions and cervical parts of the root of the 180 bovine incisors were sectioned to obtain roots with a length of 16 mm. The roots were selected according to the anatomical size of a size 80 K-file (Dentsply Maillefer, Ballaigues, Switzerland), in order to reduce the size variation between the root canals. After that, the roots were embedded in self-curing acrylic resin (VIPI, Pirassununga, São Paulo, Brazil). The samples were randomly assigned into groups based on three factors: endodontic sealer, post-endodontic waiting time and resin cement (Table 2).

Root canal endodontic treatment and storage condition

The working length was determined by inserting a size 15 K-file (Dentsply Maillefer, Ballaigues, Switzerland) into the root canal and reducing 1 mm of the real canal length. The preparation of the canals was performed using the stepback technique with second and third series endodontic files (Dentsply Maillefer, Ballaigues, Switzerland) and Gates-Glidden burs of size 3, 4 and 5 (Dentsply Maillefer, Ballaigues, Switzerland). Between each instrument change, the roots were irrigated with 2.5% sodium hypochlorite solution (NaOCl) (Asfer Indústria Química, São Caetano do Sul, São Paulo, Brazil). Next, the root canals were irrigated with 17% ethylenediaminetetraacetic acid (EDTA) (Maquira, Maringá, Paraná, Brazil) for 3 min and subsequently washed with sodium hypochlorite and dried with absorbent paper points (Dentsply Maillefer, Ballaigues, Switzerland).

The root canal was filled using one of two types of endodontic sealer (Endofill or AH Plus). The endodontic sealers were manipulated according to the manufacturer’s instructions and inserted in the root canal with the help of a lentulo spiral (Injecta, Diadema, São Paulo, Brazil), and using gutta-percha cones (Tanari, Manacapuru, Amazonas, Brazil). The compaction technique was cold lateral condensation standardized through a digital scale.

The root canals were filled on a digital scale and when the compaction force achieved 2000 g the force was immediately removed.

After the endodontic treatment, the entrance of the canal was closed with cotton and composite resin (Z250, 3M ESPE, Seefeld, Germany). The specimens were stored in deionized water in 100% relative humidity at 37°C for the storage times described in Table 2.

Fibreglass post cementation

After storage, the composite resin was removed from the entrance of the root canal with a #3216 high-speed diamond bur (KG Sorensen, Cotia, Brazil). First, 12 mm of the gutta-percha was removed from inside of root canal using a hot instrument and the root canals were prepared to a 12 mm length with the #3 drill of the fibreglass post system (Exacto, Ângelus, Londrina, Brazil). The root canals were irrigated with deionized water for 15 s and dried with absorbent paper cones (Tanari, Manacapuru, Amazonas, Brazil) and each group received root canal and fibreglass post treatment according to the manufacturer's instructions, as described below:

- *Cementation with RelyX U200 resin cement*: Fibreglass posts were cleaned with 70% alcohol and kept untouched for 1 min for alcohol evaporation. Then, a silane coupling agent (RelyX Ceramic Primer S, 3M-ESPE, Seefeld, Germany) was applied on the post surface, waiting 5 min for evaporation as recommended by the manufacturer.
- *Cementation with Multilink resin cement*: The fibreglass posts were cleaned with 70% alcohol and Monobond Plus (Ivoclar Vivadent, São Paulo, Brazil) was applied. The Primer A and B were mixed as recommended and the primer mixture was applied inside of the canal using a micro-brush.

Resin cements RelyX U200 (3M-ESPE, Seefeld, Germany) and Multilink Automix (Ivoclar Vivadent, São Paulo, Brazil) were handled according to the manufacturer's instructions and inserted into the canal, followed by insertion of the fibreglass post. The cement excesses were removed and photo-activation (Radiical, SDI, Victoria, Australia) was performed for 40 s, with 10 s of photo-activation on each face (buccal, mesial, distal and lingual). Subsequently, the specimens were stored in 100% relative humidity at 37 °C for 24 h until the push-out test was performed.

Push-out test

Specimens were fixed on a metal base in the cutting machine (Isomet 1000 Precision Saw, Buehler, Warwick, UK) and then sectioned perpendicular to the long axis of the root. Four slices (1.5 ± 0.5 mm of thickness) per specimen were obtained, and the first slice was discarded.

Subsequently, the slices were subjected to the push-out test (1 mm/min) in a universal testing machine (DL 2000, Emic, São José dos Pinhais, Brazil). For this each slice was positioned with the coronal portion downward, in a metal device with an opening of approximately 4 mm in diameter, where the root canal orifice of each slice was centralized. Afterwards, a cylindrical metallic tip (diameter 0.8 mm) applied a load in the center of the fibreglass post until fibreglass post failure.

The bond strength (σ) in MPa (Mega Pascal) was obtained using the formula $\sigma = F/A$, where *F* is force for the rupture of the specimen (N) and *A* is the bonded area (mm^2). To determine the *A*, the formula used to calculate the lateral area of a circular straight cone with parallel based was applied. The formula is defined as $A = \pi \cdot g \cdot (R1 + R2)$, where π = 3.14, g is the slant height, *R*1 and *R*2 account for the smaller the larger radius respectively. To determine the slant height the following calculation was used:

$$\boldsymbol{g^2} = \boldsymbol{h^2} + [\boldsymbol{R2} - \boldsymbol{R1}]^2$$

where *h* is the sectioned height and *R*1 and *R*2 were obtained by measuring the internal diameters of the smaller and larger bases, respectively, corresponding to the root canal diameter. The diameters and the height (*h*) of each slice were measured with a digital caliper (Starrett 727; Starrett, Itu, São Paulo, Brazil).

After testing, all specimens were analysed under a stereomicroscope (Stereomicroscope Discovery V20; Carl Zeiss, Gottingen, Germany) at 10x magnification to determine the failure type: adhesive failure between cement and dentine, adhesive failure between cement and post, cohesive failure of cement, cohesive failure of the post, cohesive failure of dentine or mixed failure. Importantly, only adhesive failures were used for the statistical analysis.

After normality and homogeneity analysis, the data were submitted to parametric tests. First, a general analysis of the factors was performed through the 3-way ANOVA. Then, each factor was analysed separately using an appropriate test (2-way ANOVA or Test T).

Sample preparation for degree of conversion analysis

Sample preparation was performed according to the group: resin cement only, using RelyX U200 (3M-ESPE, Seefeld, Germany) or Multilink (Ivoclar Vivadent, São Paulo, Brazil), resin cement in contact with Endofill or resin cement in contact with AH Plus (n=3). For standardization, rectangular matrices made in addition silicone (Express XT, 3M-ESPE, Seefeld, Germany) with standardized dimensions of height of 3 mm, width of 8 mm and thickness of 10 mm were used.

For the preparation of samples with resin cement only, a polyester Mylar strip was placed on a glass plate in order to obtain a smooth surface. Then, the fabricated silicone matrix was positioned and filled with RelyX U200 (3M-ESPE, Seefeld, Germany) resin cement or Multilink Automix resin cement (Ivoclar Vivadent, São Paulo, Brazil) handled according to manufacturers' recommendations.

On this, a polyester Mylar strip and a glass plate were again placed, and then a pressure of 500 *g* was applied. After removal of excesses, the material was light-cured (Radiical, SDI, Victoria, Australia) for 40 s in the upper and lower portion of the sample and, after the glass plate was removed, the sample was photo-polymerized for a further 20 s on each side.

For the samples in which an endodontic sealer was used together with resin cement, the endodontic sealer was manipulated according to the instructions of each manufacturer. The sealer was inserted and accommodated only in the middle of silicon matrix, thus was positioned on a glass plate and polyester Mylar strip, as described above. Subsequently, the sets were stored in 100% relative humidity at 37 °C for 24 h the endodontic sealer to achieve complete setting of the sealer. After the storage time, the resin cements used in each group were manipulated and photo-polymerized as previously described in the space reserved for this material.

Prior to undertaking the degree of conversion test, all the samples were stored in an oven at 37 °C for 24 h.

Degree of conversion test

The degree of conversion was estimated by comparison of relative heights of the C=C and C-C stretching peaks in the vibrational spectra. Vibrational spectra for all samples in the region between 1500 and 2630 cm^{-1} were collected with a Bruker Senterra Raman microscope (Ettlingen, Germany) using a 785 nm laser-line at 25 mW of power with an exposure time of 10 s as the radiation source. The best spectra were collected with a 20x objective.

The groups containing polymerized resin cements, the samples placed on a mirror to perform the focus and analysed. For standardization of the area of analysis in all the samples, orientation points were marked in 4 mm along the length of the sample on one of its side faces. For the test, the laser was directed at the demarcated region, on the center of the upper face of the sample.

In the samples containing two components, the marking of the points of analysis was performed in the region of the interface of the cements and at 8 mm (opposite point) along the length of the sample. Then, the test was performed on the mirror in the region of the markings.

The interface region points were selected at different distances in relation to the endodontic sealer. These points were called D1, D2 and D3, with D1 being closest to the endodontic sealer (Figure 1). In addition, the non-polymerized cements were analysed and used as references for the calculations. The analysis was performed in the dark to avoid any interference of external light in the polymerization of the pastes.

After the collection of the vibrational spectra, the peaks of the bands at 1608 cm^{-1} and 1638 cm^{-1} related to the C-C and C=C stretching modes were analysed. The calculation of the degree of conversion (DC) was performed using the following formula:

$$DC(\%) = 100 \cdot \left[1 - \left(\frac{R_{polimerized}}{R_{unpolimerized}}\right)\right]$$

where *R* represents the ratio of peaks at 1638 cm^{-1} to 1608 cm^{-1}, polymerized cements and unpolymerized cements.

RESULTS

Push-out results

Three-way ANOVA depicted that, independently of all other factors, the endodontic sealer and post-endodontic waiting time had statistically significant effects on the push-out bond strength results, while the resin cement factor had no effect.

Comparing the post-endodontic waiting times, it can be observed that: a) for the RelyX U200 cement, the fibreglass post cementation after 24 h promoted the highest push-out values, independently of the endodontic sealer, and b) for the Multilink cement, the time factor appears to be influenced by the endodontic sealer (Table 3).

Comparing the endodontic sealer, it was noted that the AH Plus sealer obtained the highest push-out values after 24 h and 6 months for the two resin cements when compared with the eugenol-based sealer. In contrast, no statistical difference between the two endodontic sealers was seen after 12 months (Table 3).

With regard to resin cements, it was noted that the RelyX U200 obtained higher push-out values than Multilink, after a period of 24 h for both endodontic sealers and after a period of 6 months for the Endofill sealer. Multilink promoted higher values after a period of 6 months and 12 months for the AH Plus sealer. No statistically significant difference between RelyX U200 and Multilink were noted for the 12 months group using the Endofill sealer.

Failure mode distribution is illustrated in Table 4.

Degree of conversion results

When testing the effects of the eugenol-based sealer, it was observed that this sealer reduces the degree of conversion of the resin cements. Also, it was noted that the degree of conversion depends on the distance from the point of contact between resin cement and sealer – a lower conversion was observed when the cement was closer to the endodontic sealer, with the conversion increasing as the distance became larger (Table 5).

On the other hand, no decrease in the degree of conversion of resin cements was observed when the epoxy resin-based sealer was tested (Table 5).

DISCUSSION

The present study verified that the endodontic sealer and post-endodontic waiting time affect the push-out bond strength of the fibreglass posts. In addition, the degree of conversion of the cements was reduced by the eugenol-based sealer, while the resin-based sealer did not affect it. Therefore, the null hypotheses were rejected.

Eugenol-based sealers are still widely used for endodontic filling (Demiryurek *et al*. 2010, Zhou *et al*. 2013, Sarkis-Onofre *et al*. 2014). Endofill groups at 24-h and 6-months times presented lower values compared to AH Plus groups. These results are in accordance with the literature (Vano *et al*. 2008, Aleisa *et al*. 2012, Izadi *et al*. 2013, Ozcan *et al*. 2013, Altmann *et al*. 2015, Aleisa *et al*. 2016), demonstrating that eugenol-based sealers can interfere in the bond strength of fibreglass posts; this is in contrast with other studies that found no significant influence of eugenol on the bond strength of the fibreglass post (Demiryurek *et al*. 2010, Manicard *et al*. 2011).

Degree of conversion analysis confirms that the eugenol-based sealers can interfere in the polymerization of the resin cements, possibly because eugenol is a phenolic compound that releases free radicals that interact with the monomers of resinous compounds, inhibiting the initiation of the polymerization or accelerating the finishing of the same (Cohen *et al*.

2002, Altman *et al*. 2015). Therefore, it can be suggested that the results found for the push-out test may have been caused by a decrease in resin cement polymerization.

Also, most of the failures that occurred for the groups filled with eugenol-based sealer were adhesive between cement and dentine. This indicates a poor connection between resin cement and dentine (Ozcan *et al*. 2013).

The AH Plus sealer presented better results in push-out strength, which can be explained by its similarity in chemical composition to the resin cement due to presence of epoxy resin and absence of substances affecting the polymerization, improving the bond strength of the posts (Asmussen & Peutzfeldt 2005, Cechin *et al*. 2011, Sonmez *et al*. 2013). In contrast, the results in the 12-months groups demonstrated that no significant differences between the endodontic sealers. This could suggest that eugenol does have a smaller effect on the degree of polymerization of the resin cement after 12 months of endodontic treatment.

Analysing the resin cements, it was noted that they behave differently depending on the endodontic sealers used. The different values of push-out tests can be explained by the composition of the cements (Table 1), principally by amounts of TEGDMA in the RelyX U200, and BISGMA in the Multilink, which affect the viscosity and consequently the degree of conversion. The filler weights of the two cements may have also affected the flexural properties and shrinkage behavior (Di Francescantonio *et al*. 2013, Pulido *et al*. 2016). Another explanation is that the treatment procedures for the fibreglass post and root canals were performed differently for the two resin cements used. In the RelyX U200 group, was used a silane coupling agent in the fibreglass post, and the root canal did not receive any additional treatment. In the Multilink cement group, Monobond was used in the fibreglass post and primers inside the root canal. This difference could influence the bond strength of the fibreglass posts.

It should be noted that a higher degree of conversion in close environments, such as the root canal cavity (high C-factor), is a disadvantage because it increases the contraction stress of polymerization (Bouillaguet *et al*. 2003, Calheiros *et. al*. 2006, Feng & Suh 2006, Sarkis-Onofre *et al*. 2014), damaging the fibreglass post–root dentine adhesion. This can be explained because the analysis of the degree of conversion in this study showed that samples with Multilink obtained the highest values. However, in the push-out test, the Multilink only obtained higher values for the 6-month a 12-month storage groups for AH Plus sealer.

Comparing the times after endodontic filling with Endofill sealer and luting with RelyX U200, the results show that the 24-h storage group had a higher bond strength compared with the 6- and 12-month storage groups. The literature explains that a long interval between the obturation and the cementation of the posts favors the eugenol molecules, allowing them to penetrate the dentinal tubules (Hagge *et al*. 2002, Izadi *et al*. 2013) and causing greater damage to the fibreglass post bonding. According to Maseki *et al*. (1991), the greatest amount of eugenol is released from the sealer during the first two weeks, favoring the cementation of the posts after 24 h of obturation.

Confirming these results, when filling with Endofill sealer and luting with Multilink, the lowest push-out values were found for the 6-month storage group. Surprisingly, Multilink resin cement showed an increase in bond strength after 12 months. This may be due to the eugenol molecules gradually losing their effect on polymerization. Thus, at the 12-month storage timeframe, there was no difference between the endodontic sealers.

When cementation was performed with AH Plus and RelyX, it could be observed that the 24-h storage period group had higher push-out strength than the other two groups. This can be explained because, in the 24-h storage group, the sealer was still setting and there was insufficient time for significant penetration into the dentinal tubules, making its removal from the root canal easier (Vano *et al*. 2006, Hagge *et al*. 2002, Izadi *et al*. 2013).

For the groups cemented using Multilink with AH Plus, the 6-month storage period had a higher bond strength suggesting that the presence of the resin-based sealer increased the bond strength, due to the fact that this sealer presents a characteristic of low solubility and great adhesion in the walls of the root canals (Asmussem & Peutzfeldt 2005, Sonmez *et al*.

2013), making its removal difficult after complete cure of this sealer. At 12 months, it is possible that this sealer would have no further effect on the resistance of the posts.

Analyses of the failures in the study demonstrated the predominant modes of failure were adhesive between resin cement and fibreglass post in the Multilink groups filled with AH Plus sealer, our findings were similar to those reported by Perdigão *et al*. (2006). In addition, in the group 24-h filled with AH Plus and luting with RelyX U200 there is predominant modes of failures cohesive of dentine demonstrated that the actual adhesive force generated at the bonding interface was higher to the obtained bond strength values (Taira *et al*. 2007).

A limitation of this study is that no primers were used on the Multilink resin cement for the degree of conversion tests, thus the cement became self-curing with additional light-curing. This may explain the higher values for the degree of conversion for this cement. In addition, the drill of the fibreglass post system may have removed the dentine affected by the sealer used modifying the bond strength (Izadi *et al*. 2013).

Thereby, there appears to be a relationship between the degree of conversion and bond strength. New studies are necessary to confirm this theory, and further studies to evaluate the degree of conversion at different times and to evaluate the penetration of eugenol in the dentinal tubules over time are needed.

CONCLUSIONS

Within the limitations of this study, the following conclusions were drawn:

1. AH Plus sealer obtained the highest puh-out values in 24 hours and 6 months and did not interfere in the degree of conversion of the resin cements. In contrast, the eugenol-based sealer decreases the bond strength between fibreglass posts and root dentine and reduces the degree of conversion of the resin cements.

2. The resin cement is time-sealer dependent, obtaining different values of bond strength according to the variation of the time and the type of endodontic sealer.

3. The post-endodontic waiting time of the 24 hours obtained the highest of push-out values, except for the Multilink when root canals were filled with AH Plus sealer.

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FIGURE LEGENDS

Figure 1 Distribution of the analysis points in the sample interface region.

TABLES

Table 1 Materials used and theirs composition.

Material	Composition
Endofill (Dentsply, Maillefer, Ballaigues, Switzerland)	Zinc oxide, hydrogenated resin, barium sulfate, eugenol.
AH Plus (Dentsply, Maillefer, Ballaigues, Switzerland)	Epoxy-amine resin-based root canal sealer, opacifier –zirconium oxide.
Multilink Automix (Ivoclar-Vivadent, São Paulo, Brazil)	Bis-GMA, HEMA, 2-dimethylaminoethyl methacrylate, ethyoxylated bisphenol A dimethacrylate, UDMA, barium glass, ytterbium trifluoride, spheroid mixed oxide (filler weight: 65% to 70%) **Primer A:** Water, initiators, sulfonate amine **Primer B**: HEMA, phosphonic acid acrylate, mehacrylate modified polyacrylic acid, stabilizers
RelyX U200 (3M ESPE, Seefeld, Germany)	Glass powder treated with silane, 2-propenoic acid, 2-methyl 1, 1'-(1- [hydroxymetil]-1,2- ethanodlyl) ester dimethacrylate, TEGDMA, silica-treated silane, glass fibre, sodium persulfate and per-3,5,5-trimethyl hexanoate *t*-butyl, substitute dimethacrylate, sodium *p*-toluenesulfonate, 1- benzyl-5-phenyl-acid barium, calcium, 1,12- dodecane dimethacrylate, calcium hydroxide, and titanium dioxide (filler weight: 45%)

Bis-GMA: bisphenyl A glycidyl methacrylate; HEMA: 2-hydroxyethyl methacrylate; TEGDMA: triethyleneglycol dimethacrylate; UDMA: urethame dimethacrylate.

Table 2 Design of the study for push-out test, considering 3 factors: endodontic sealer, post-endodontic waiting time and resin cement.

Endodontic sealer	Post-endodontic waiting time	Resin cement
Endofill	24 hours	RelyX U200
		Multilink
	6 months	RelyX U200
		Multilink
	12 months	RelyX U200
		Multilink
AH Plus	24 hours	RelyX U200
		Multilink
	6 months	RelyX U200
		Multilink
	12 months	RelyX U200
		Multilink

Table 3 Means and standard deviation of the push-out data (MPa), considering the studied factors.

Endodontic sealer	Resin cement	Post-endodontic waiting time		
		24 hours	6 months	12 months
Endofill	**RelyX U200**	6.9 ± 2 aA*	4.6 ± 2.1aB*	4.1 ± 2.7aB*
	Multilink	4.9 ± 2.2bA*	2.8 ± 1.4bB*	6.1 ± 2.8aA*
AH Plus	**RelyX U200**	11.2 ± 3.2aA**	6.0 ± 2.3bB**	3.9 ± 2.6bB*
	Multilink	6.9 ± 2.1bB**	9.5 ± 2.3aA**	6.1 ± 1.9aB*

Uppers case letters compare the ‘waiting time’ factor (lines), keeping unaltered the endodontic sealer and resin cement.
Lower case letters compare the two resin cements (column), keeping unaltered the endodontic sealer and time.
The symbols *, ** compare the two endodontic sealers, within the same time and resin cement, same symbols indicate similarity and different symbols indicate differences.

Table 4. Failure mode distribution after the push-out test.

Endodontic sealer	**Post-endodontic waiting time**	**Resin cement**	**Types of failures**					
			Adhes c/d	Adhes c/p	Cohes c	Cohes p	Cohes d	M
Endofill	24 hours	RelyX U200	52	-	-	-	8	-
		Multilink	33	25	-	-	2	-
	6 months	RelyX U200	57	1	-	-	2	-
		Multilink	48	9	-	-	1	-
	12 months	RelyX U200	53	2	-	-	3	-
		Multilink	30	30	-	-	-	-
AH Plus	24 hours	RelyX U200	18	8	-	2	31	-
		Multilink	28	31	-	-	1	-
	6 months	RelyX U200	54	5	-	-	1	-
		Multilink	10	29	-	-	20	-
	12 months	RelyX U200	57	-	-	1	-	-
		Multilink	7	50	-	-	2	-
	Total		447 (62,87%)	190 (26,72%)	-	3 (0,42%)	71 (9,99%)	-

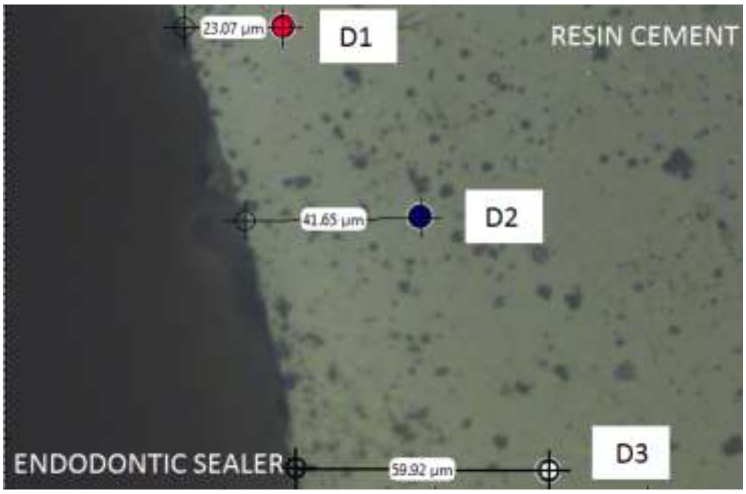
Adhes c/d = adhesive failure between cement and dentine; Adhes c/p = adhesive failure between cement and post; Cohes c = cohesive failure of cement; Cohes p = cohesive failure of post; Cohes d = cohesive failure of dentine; M = mixed failure.

Table 5 Degree of conversion (%) of the resin cement.

Cements		No sealer	Endofill Distance from Interface - DC%	AH Plus Distance from Interface - DC%
RelyX U200	Sample 1	28.5%	D1: 23 µm – 8.8% D2: 41µm – 12% D3: 59 µm – 21% OP: 8 mm – 29.4%	D1: 31 µm – 41% D2: 32 µm – 33.8% D3: 34 µm – 39.4% OP: 8 mm – 27.8%
	Sample 2	29.4%	D1: 20 µm – 3.3% D2: 104 µm – 22.2% D3: 122 µm – 25.3% OP: 8 mm – 35.3%	D1: 62 µm – 55% D2: 63 µm – 48% D3: 64 µm – 52% OP: 8 mm – 21%
	Sample 3	20%	D1: 18 µm – 2.3% D2: 83 µm – 11.5% D3: 149 µm – 8.1% OP: 8 mm – 33.3%	D1: 28 µm – 46.8% D2: 67 µm – 56.2% D3: 126 µm – 51.9% OP: 8 mm – 35.2%
Multilink	Sample 1	69.2%	D1: 40 µm – 25.5% D2: 53 µm – 27.3% D3: 101 µm – 44.9 OP: 8 mm – 69.3%	D1: 53 µm – 79.5% D2: 54 µm – 77.1% D3: 55 µm – 80.3% OP: 8 mm – 73.7%
	Sample 2	69.3%	D1: 17 µm – 15.1% D2: 43 µm – 32.5% D3: 45 µm – 30.1% OP: 8 mm – 68.6%	D1: 53 µm – 72.3% D2: 54 µm – 73.2% D3: 56 µm – 70.1% OP: 8 mm – 72%
	Sample 3	69.2%	D1: 34 µm – 31.6% D2: 53 µm – 34.7% D3: 90 µm – 46.6% OP: 8 mm – 66.9%	D1: 28 µm – 68.8% D2: 36 µm – 67.6% D3: 49 µm – 71.1% OP: 8 mm – 72.6%

DC= degree of conversion; D = distance; OP= opposite point.

FIGURES



Source: Personal archive

3 CONCLUSÃO

A retenção e a resistência ao deslocamento do pino intrarradicular são importantes fatores para o sucesso da restauração final (FARID et al., 2013) . No entanto, há relatos na literatura que existem vários fatores que influenciam na resistência adesiva de pinos intrarradiculares, sendo um deles os cimentos endodônticos (ALEISA et al., 2012; ALTMANN et al., 2015; BARBIZAM et al., 2011; DIMITROULI et al., 2011; MOSHARRAF, ZARE, 2014)

Cimentos endodônticos à base de eugenol são amplamente utilizados na endodontia (DEMIRYÜREK et al., 2010). Porém, a molécula de eugenol contém radicais livres que interferem na polimerização dos cimentos resinosos, influenciando, assim, na resistência adesiva dos pinos de fibra (BEHR et al., 2004, DIAS et al.,2009).

Contudo na literatura há uma deficiência de estudos que avaliem diretamente o grau de polimerização dos cimentos resinosos em contato com o cimento endodônticos e sua influência na resistência adesiva dos retentores intrarradiculares. Neste estudo, foi avaliado o grau de conversão desses materiais em contato, e foi verificada a diminuição do grau de conversão à medida que o ponto de análise se aproximava do cimento endodôntico à base de eugenol, confirmando a influência desse cimento na polimerização dos cimentos resinosos.

Além disso, através do teste de *push-out* foi verificado, também, a diminuição na resistência adesiva de pinos de fibra de vidro quando as raízes foram obturadas com cimento à base de eugenol em comparação com raízes obturadas com cimento à base de resina epóxica. Sendo, então, confirmada a influência do eugenol sobre a resistência adesiva dos pinos intrarradiculares.

Por outro lado, o cimento à base de resina epóxica AH Plus, é considerado padrão ouro e, devido a sua composição, não interfere na polimerização dos cimentos resinosos (COHEN et al., 2002; CECCHIN et al., 2011; SONMEZ, SONMEZ, ALMAZ, 2013). O que é confirmado pelo presente estudo, pois não houve diminuição do grau de conversão dos cimentos resinosos em contato com o AH Plus.

Outro aspecto a ser estudado e que ainda não há consenso na literatura é a influência do tempo de espera entre a obturação e a cimentação do pino na resistência adesiva do retentor intrarradicular. O presente estudo avaliou a resistência ao *push-out* em três diferentes tempos: 24 horas, 6 meses e 12 meses. Foram utilizados para a cimentação do pino dois cimentos resinosos que, conforme o tempo de espera pós-endodontia e cimento endodôntico utilizado, se comportaram de maneira diferente. Para o cimento RelyX U200 os maiores valores de resistência foram no tempo de 24 horas após obturação, independente do cimento endodôntico utilizado.

Já para o cimento Multilink Automix, quando as raízes foram obturadas com o cimento Endofill, os tempos 24 horas e 12 meses após a obturação obtiveram os melhores valores de resistência adesiva, não havendo diferença significativa entre os dois grupos. Quando o cimento AH Plus foi utilizado, o melhor tempo para a cimentação do pino foi de 6 meses após a obturação.

Portanto, os fatores cimento endodôntico e tempo de espera pós-endodontia tiveram influência significativa na resistência adesiva dos pinos de fibra. Além disso, em uma análise mais detalhada foi verificado que os cimentos resinosos foram influenciados de maneira diferente pelos fatores estudados.

Contudo, é verificada a necessidade de novos estudos para analisar o comportamento de outros cimentos resinosos utilizados na prática clínica, bem como de estudos que avaliem diretamente o grau de conversão em diferentes tempos, para que possa ser confirmada a influência desses fatores no grau de conversão dos cimentos resinosos ao longo do tempo.

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ANEXO A – NORMAS PARA A PUBLICAÇÃO NO PERIÓDICO *INTERNATIONAL ENDODONTIC JOURNAL*

MANUSCRIPT FORMAT AND STRUCTURE

Format

Language: The language of publication is English. It is preferred that manuscript is professionally edited. A list of independent suppliers of editing services can be found at http://authorservices.wiley.com/bauthor/english_language.asp. All services are paid for and arranged by the author, and use of one of these services does not guarantee acceptance or preference for publication

Presentation: Authors should pay special attention to the presentation of their research findings or clinical reports so that they may be communicated clearly. Technical jargon should be avoided as much as possible and clearly explained where its use is unavoidable. Abbreviations should also be kept to a minimum, particularly those that are not standard. The background and hypotheses underlying the study, as well as its main conclusions, should be clearly explained. Titles and abstracts especially should be written in language that will be readily intelligible to any scientist.

Abbreviations: International Endodontic Journal adheres to the conventions outlined in Units, Symbols and Abbreviations: A Guide for Medical and Scientific Editors and Authors. When non-standard terms appearing 3 or more times in the manuscript are to be abbreviated, they should be written out completely in the text when first used with the abbreviation in parenthesis.

Structure

All manuscripts submitted to *International Endodontic Journal* should include Title Page, Abstract, Main Text, References and Acknowledgements, Tables, Figures and Figure Legends as appropriate

Title Page: The title page should bear: (i) Title, which should be concise as well as descriptive; (ii) Initial(s) and last (family) name of each author; (iii) Name and address of department, hospital or institution to which work should be attributed; (iv) Running title (no more than 30 letters and spaces); (v) No more than six keywords (in alphabetical order); (vi) Name, full postal address, telephone, fax number and e-mail address of author responsible for correspondence.

Abstract for Original Scientific Articles should be no more than 250 words giving details of what was done using the following structure:

-Aim: Give a clear statement of the main aim of the study and the main hypothesis tested,if any.

-**Methodology**: Describe the methods adopted including, as appropriate, the design of the study, the setting, entry requirements for subjects, use of materials, outcome measures and statistical tests.

-Results: Give the main results of the study, including the outcome of any statistical analysis.

- **Conclusions**: State the primary conclusions of the study and their implications. Suggest areas for further research, if appropriate.

Main Text of Original Scientific Article should include Introduction, Materials and Methods, Results, Discussion and Conclusion

Introduction: should be focused, outlining the historical or logical origins of the study and gaps in knowledge. Exhaustive literature reviews are not appropriate. It should close with the explicit statement of the specific aims of the investigation, or hypothesis to be tested.

Material and Methods: must contain sufficient detail such that, in combination with the references cited, all clinical trials and experiments reported can be fully reproduced.

When experimental animals are used the methods section must clearly indicate that adequate measures were taken to minimize pain or discomfort. Experiments should be carried out in accordance with the Guidelines laid down by the National Institute of Health (NIH) in the USA regarding the care and use of animals for experimental procedures or with the European Communities Council Directive of 24 November 1986 (86/609/EEC) and in accordance with local laws and regulations.

All studies using human or animal subjects should include an explicit statement in the Material and Methods section identifying the review and ethics committee approval for each study, if applicable. Editors reserve the right to reject papers if there is doubt as to whether appropriate procedures have been used.

(iii) Suppliers: Suppliers of materials should be named and their location (Company, town/city, state, country) included.

Results: should present the observations with minimal reference to earlier literature or to possible interpretations. Data should not be duplicated in Tables and Figures.

Discussion: may usefully start with a brief summary of the major findings, but repetition of parts of the abstract or of the results section should be avoided. The Discussion section should progress with a review of the methodology before discussing the results in light of previous work in the field. The Discussion should end with a brief conclusion and a comment on the potential clinical relevance of the findings. Statements and interpretation of the data should be appropriately supported by original references.

Conclusion: should contain a summary of the findings.

Acknowledgements: *International Endodontic Journal* requires that all sources of institutional, private and corporate financial support for the work within the manuscript must be fully acknowledged, and any potential conflicts of interest noted. Grant or contribution numbers may be acknowledged, and principal grant holders should be listed. Acknowledgments should be brief and should not include thanks to anonymous referees and editors. See also above under Ethical Guidelines.

References

It is the policy of the Journal to encourage reference to the original papers rather than to literature reviews. Authors should therefore keep citations of reviews to the absolute minimum.and recommend the use of a tool such as EndNote or Reference Manager for reference management and formatting. The EndNote reference style can be obtained upon request to the editorial office (iejeditor@cardiff.ac.uk). Reference Manager reference styles can be searched for here: www.refman.com/support/rmstyles.asp

In the text: single or double authors should be acknowledged together with the year of publication, e.g. (Pitt Ford & Roberts 1990). If more than two authors the first author followed by *et al.* is sufficient, e.g. (Tobias *et al.* 1991). If more than 1 paper is cited the references should be in year order and separated by "," e.g. (Pitt Ford & Roberts 1990, Tobias *et al*. 1991).

Reference list: All references should be brought together at the end of the paper in alphabetical order and should be in the following form.

(i) Names and initials of up to six authors. When there are seven or more, list the first three and add *et al.*

(ii)Year of publication in parentheses

(iii) Full title of paper followed by a full stop (.)

(iv) Title of journal in full (in italics)

(v) Volume number (bold) followed by a comma (,)

(vi) First and last pages

Examples of correct forms of reference follow:

Tables, Figures and Figure LegendsTables: Tables should be double-spaced with no vertical rulings, with a single bold ruling beneath the column titles. Units of measurements must be included in the column title.

Figures: All figures should be planned to fit within either 1 column width (8.0 cm), 1.5 column widths (13.0 cm) or 2 column widths (17.0 cm), and must be suitable for photocopy reproduction from the printed version of the manuscript. Lettering on figures should be in a clear, sans serif typeface (e.g. Helvetica); if possible, the same typeface should be used for all figures in a paper. After reduction for publication, upper-case text and numbers should be at least 1.5-2.0 mm high (10 point Helvetica). After reduction, symbols should be at least 2.0-3.0 mm high (10 point). All half-tone photographs should be submitted at final reproduction size. In general, multi-part figures should be arranged as they would appear in the final version. Reduction to the scale that will be used on the page is not necessary, but any special requirements (such as the separation distance of stereo pairs) should be clearly specified.

Unnecessary figures and parts (panels) of figures should be avoided: data presented in small tables or histograms, for instance, can generally be stated briefly in the text instead. Figures should not contain more than one panel unless the parts are logically connected; each panel of a multipart figure should be sized so that the whole figure can be reduced by the same amount and reproduced on the printed page at the smallest size at which essential details are visible.

Figures should be on a white background, and should avoid excessive boxing, unnecessary colour, shading and/or decorative effects (e.g. 3-dimensional skyscraper histograms) and highly pixelated computer drawings. The vertical axis of histograms should not be truncated to exaggerate small differences. The line spacing should be wide enough to remain clear on reduction to the minimum acceptable printed size.

Figures divided into parts should be labelled with a lower-case, boldface, roman letter, a, b, and so on, in the same typesize as used elsewhere in the figure.

Lettering in figures should be in lower-case type, with the first letter capitalized. Units should have a single space between the number and the unit, and follow SI nomenclature or the nomenclature common to a particular field. Thousands should be separated by a thin space (1 000). Unusual units or abbreviations should be spelled out in full or defined in the legend. Scale bars should be used rather than magnification factors, with the length of the bar defined in the legend rather than on the bar itself. In general, visual cues (on the figures themselves) are preferred to verbal explanations in the legend (e.g. broken line, open red triangles etc.)

Figure legends: Figure legends should begin with a brief title for the whole figure and continue with a short description of each panel and the symbols used; they should not contain any details of methods.

Permissions: If all or part of previously published illustrations are to be used, permission must be obtained from the copyright holder concerned. This is the responsibilty of the authors before submission.

Preparation of Electronic Figures for Publication: Although low quality images are adequate for review purposes, print publication requires high quality images to prevent the final product being blurred or fuzzy. Submit EPS (lineart) or TIFF (halftone/photographs) files only. MS PowerPoint and Word Graphics are unsuitable for printed pictures. Do not use pixel-oriented programmes. Scans (TIFF only) should have a resolution of 300 dpi (halftone) or 600 to 1200 dpi (line drawings) in relation to the reproduction size (see below). EPS files should be saved with fonts embedded (and with a TIFF preview if possible). For scanned images, the scanning resolution (at final image size) should be as follows to ensure good reproduction: lineart: >600 dpi; half-tones (including gel photographs): >300 dpi; figures containing both halftone and line images: >600 dpi.