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Chemical analysis of irradiated root dentin and its interaction with resin cements

Dissertação apresentada à Faculdade de Odontologia da Universidade de Uberlândia, para obtenção do Título de Mestre em Odontologia na Área de Clínica Odontológica Integrada.

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Orientadora: Profa. Dra. Veridiana Resende Novais Simamoto

Banca Examinadora:
Profa. Dra. Profa. Veridiana Resende Novais Simamoto
Prof. Dr. Carlos José Soares
Prof. Dr. Daniel de Almeida Decurcio

Uberlândia, 2020

Alessandro Marcelo Pelloso

UNIVERSIDADE FEDERAL DE UBERLÂNDIA

Coordenação do Programa de Pós-Graduação em Odontologia
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DEDICATÓRIA

A Deus, por ter me concebido o dom da vida e de ser dentista, com todas as condições de desfrutá-las e também de poder ajudar os que necessitam.

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A todos os professores, que conseguem transmitir seus conhecimentos e fazer um mundo melhor.

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Desistir... eu já pensei seriamente nisso, mas nunca me levei realmente a sério; é que tem mais chão nos meus olhos do que o cansaço nas minhas pernas, mais esperança nos meus passos, do que tristeza nos meus ombros, mais estrada no meu coração do que medo na minha cabeça.

Cora Coralina

SUMÁRIO

RESUMO

O objetivo deste estudo foi investigar as alterações químicas da dentina radicular submetida à irradiação in vitro e como isso afeta a interação com cimentos resinosos. Para isso, 40 pré-molares humanos foram coletados e distribuídos aleatoriamente em dois grupos (n = 20): não irradiados e irradiados. Posteriormente, foram divididos em dois subgrupos de acordo com o tipo de cimento resinoso (n = 10): cimento convencional RelyX ARC (3M ESPE, São Paulo, MN, EUA) e autoadesivo RelyX U200 (3M ESPE, São Paulo , MN, EUA). Após tratamento endodôntico e cimentação do pino de fibra de vidro, as raízes foram seccionadas ao meio ao longo de seu eixo longitudinal, para a metodologia de espectroscopia no infravermelho transformada de Fourier (FTIR) e espectroscopia Raman. Para microscopia confocal de varredura a laser (CLSM), os espécimes foram cortados em duas fatias de 1,0 mm de espessura de cada região radicular, perpendiculares ao longo eixo do dente. O FTIR foi utilizado para verificar alterações na composição química da dentina radicular de acordo com terços (n = 08). Três amostras por grupo, que foram analisadas por FTIR, foram levadas à espectroscopia Raman para estimar a zona de difusão dos cimentos e sua interação química com a dentina. Para análise da interface de ligação, duas outras amostras por grupo foram preparadas com corantes e analisadas sob microscopia confocal. Os dados de FTIR foram analisados usando ANOVA one way, com medidas repetidas (terços) e teste de Tukey. Os dados de Raman foram analisados por ANOVA two way e teste de Tukey ($\alpha = 0,05$). Para imagens confocais, foi realizada uma análise descritiva da interação cimento-dentina. O FTIR revelou diferença significativa para a amida I apenas no terço ($p < 0,001$), e um valor mais alto para o terço apical em relação ao terço cervical no grupo não irradiado. Para fosfato e amida III, houve diferença significativa apenas para irradiação ($p = 0,015$ e $p = 0,038$, respectivamente). O teste de Tukey revelou que, no terço cervical, a dentina irradiada apresentava valores alterados desses componentes químicos. Para o carbonato, houve diferença significativa apenas na irradiação (p = 0,002), e o teste de Tukey mostrou que, nos terços cervical e apical, a dentina irradiada apresentava valores mais baixos de carbonato. Quanto às razões, houve diferença significativa para o terço (p <0,001) na relação mineral / matriz (M: M), enquanto a irradiação e a interação entre os fatores do estudo não apresentaram (p = 0,826 e p = 0,460, respectivamente). Para a razão carbonato / mineral (C: M), houve diferença significativa apenas na irradiação (p = 0,001). Em todos os terços, a dentina irradiada apresentou valores mais baixos da relação C:M quando comparada à não irradiada. Para a relação amida I / CH2, a ANOVA mostrou diferença significativa apenas para o terço (p = 0,007). Os dados da espectroscopia Raman foram analisados quanto a razões de 961/1458 cm-1 e revelaram que a irradiação (p = 0,818), cimento resinoso (p = 0,381) e a interação entre eles (p = 0,273) não influenciaram. A análise da interface adesiva pelo CLSM mostrou menor interação adesiva na dentina irradiada nos dois cimentos. Nas condições deste estudo in vitro, a radiação ionizante alterou as moléculas de dentina radicular, principalmente no terço cervical. Os cimentos resinosos apresentaram menor interação com a dentina radicular irradiada. O cimento resinoso convencional RelyX ARC apresentou maior interação com a dentina irradiada do que o cimento resinoso autoadesivo RelyX U200.

Palavras-chave: interface de união, análise química, dentina, radioterapia, cimento resinoso.

ABSTRACT

The aim of this study was to investigate the chemical changes of root dentin submitted to in vitro irradiation and how it affects the interaction with resin cements. For this, 40 human premolars were collected and randomly assigned into two groups (n=20): non-irradiated and irradiated. Subsequently, they were divided into two subgroups according to the type of resin cement (n=10): total-etch RelyX ARC and self-adhesive RelyX U200. After endodontic treatment and cementation of fiberglass post, the roots were sectioned. Fourier-transform infrared spectroscopy (FTIR) was used to verify chemical composition changes in root dentin according to thirds (n=08). Three samples per group were taken to Raman spectroscopy to estimate the diffusion zone of the cements and their chemical interaction with dentin. For bonding interface analysis, two other samples per group were prepared with colorants and analyzed under confocal laser scanning microscopy(CLSM). FTIR data were analyzed using One-way ANOVA with repeated measurement (thirds) and Tukey's test. Raman data were analyzed using Two-way ANOVA and Tukey's test (α=0.05). For confocal images, a descriptive analysis was done. FTIR revealed significant difference for amide I only for third (p<0.001), and a higher value for the apical in relation to the cervical third within the non-irradiated group. For phosphate and amide III, there was significant difference only for irradiation (p=0.015 and p=0.038, respectively). Tukey's test revealed that within the cervical third the irradiated dentin presented altered values of these chemical components. For carbonate, there was significant difference only for irradiation (p=0.002), and Tukey's test showed that within the cervical and apical thirds the irradiated dentin presented lower values of carbonate. Regarding the ratios, showed significant difference for third (p<0.001) in the mineral/matrix ratio (M:M), while irradiation and the interaction between the study factors did not (p=0.826 and p=0.460, respectively). For carbonate/mineral ratio (C:M), there was significant difference only for irradiation (p=0.001). In all thirds, the irradiated dentin presented lower values of C:M ratio when compared to the non-irradiated one. For amide I/CH_2 ratio, ANOVA showed significant difference only for third (p=0.007). Raman spectroscopy data were analyzed for 961/1458 cm^{-1} ratios and revealed that the irradiation (p=0.818), resin cement (p=0.381) and the interaction between them (p=0.273) had no influence. The analysis of the bonding interface by CLSM showed less adhesive interaction in the irradiated dentin in both cements. Under the conditions of this in vitro study, ionizing radiation altered the root dentin molecules, mainly in the cervical third. Resin cements had less interaction with irradiated root dentin. RelyX ARC conventional resin cement had greater interaction with irradiated dentin than RelyX U200 self-adhesive resin cement.

Keywords: bonding interface, chemical analysis, dentin, radiotherapy, resin cement.

1. INTRODUÇÃO E REFERENCIAL TEÓRICO

A radioterapia é uma modalidade terapêutica rotineiramente utilizada para o tratamento de neoplasias malignas na cavidade oral. No entanto, apesar de destruir as células tumorais, a radioterapia também pode causar danos a tecidos saudáveis, como ossos, mucosa, dentes e glândulas salivares localizadas no campo de radiação [1,2,3,4]. A função oral pode ser prejudicada e a visita do paciente ao dentista com mais frequência pode ser necessária [1,2,4].

Os efeitos colaterais da radioterapia, como hipossalivação e xerostomia, associados a alterações das propriedades químicas e mecânicas dos dentes, podem levar à rápida e severa destruição do esmalte e da dentina [2,5,6,7,8,9,10,11]. Nesses casos, a reabilitação pode ser desafiadora, uma vez que procedimentos cirúrgicos como extrações e inserção de implantes dentários geralmente não são recomendados devido ao risco de osteorradionecrose [4]. Portanto, o uso de pinos para restaurações diretas ou indiretas desses dentes com extensa destruição pode ser uma alternativa [5,6,7].

Pinos de fibra de vidro são comumente usados porque geram menores tensões dentro do canal radicular e menos deformação, reduzindo o risco de fraturas radiculares [15]. Núcleo metálico fundido, embora não exija interação química, produz mais estresse em uma dentina mais frágil [8]. A interação química de cimentos resinosos com dentina é importante para a retenção de pinos de fibra em canais radiculares [9]. Entretanto, procedimentos restauradores adesivos realizados em dentes irradiados são um desafio para a odontologia restauradora, provavelmente devido a uma frágil interface adesiva e a ocorrência frequente de defeitos coesivos na dentina [10,11,12]. Os cimentos convencionais demonstraram um ótimo padrão de adesão à dentina radicular e ao pino, mas são muito sensíveis à técnica devido às várias etapas, com maior risco de excesso de ataque ácido e/ou ressecamento [13,14]. Por outro lado, os cimentos resinosos autoadesivos têm sido amplamente utilizados na odontologia devido à sua facilidade de manuseio e ótimos resultados de adesão, na dentina saudável [15].

A investigação nesse campo é escassa e precisa ser feita para possibilitar a reabilitação oral em pacientes com câncer de cabeça e pescoço após radioterapia. Assim, o objetivo deste estudo é analisar as alterações químicas da dentina radicular submetida à radiação in vitro e como ela afeta a interação com os cimentos resinosos. As hipóteses nulas testadas foram: (1) a radiação in vitro não altera a composição química da dentina radicular, independentemente do terço radicular; (2) a radiação in vitro não altera a interação dos cimentos resinosos com a dentina radicular, independentemente do tipo de cimento resinoso.

2. CAPÍTULO 1

CHEMICAL ANALYSIS OF IRRADIATED ROOT DENTIN AND ITS INTERACTION WITH RESIN CEMENTS

CHEMICAL ANALYSIS OF IRRADIATED ROOT DENTIN AND ITS INTERACTION WITH RESIN CEMENTS

ABSTRACT

Objective: The aim of this study was to investigate the chemical changes of root dentin submitted to in vitro irradiation and how it affects the interaction with resin cements.

Methods: For this, 40 human premolars were collected and randomly assigned into two groups (n=20): non-irradiated and irradiated. Subsequently, they were divided into two subgroups according to the type of resin cement (n=10): convencional cement RelyX ARC (3M ESPE, St. Paul, MN, USA) and self-adhesive cement RelyX U200 (3M ESPE, St. Paul, MN, USA). After endodontic treatment and cementation of fiberglass post, the roots were sectioned in halves along their longitudinal axis, for the methodology of Fourier-transform infrared spectroscopy (FTIR) and Raman spectroscopy. To confocal laser scanning microscopy(CLSM), the specimens were cut in two 1.0-mm thick slices of each root region, perpendicular to the long axis of the tooth. FTIR was used to verify chemical composition changes in root dentin according to thirds (n=08). Three samples per group, which were analyzed by FTIR, were taken to Raman spectroscopy to estimate the diffusion zone of the cements and their chemical interaction with dentin. For bonding interface analysis, two other samples per group were prepared with colorants and analyzed under confocal laser scanning microscopy(CLSM). FTIR data were analyzed using One-way ANOVA with repeated measurement (thirds) and Tukey's test. Raman data were analyzed using Two-way ANOVA and Tukey's test (α=0.05). For confocal images, a descriptive analysis of the cement-dentin interaction was done.

Results: FTIR revealed significant difference for amide I only for third ($p<0.001$), and a higher value for the apical in relation to the cervical third within the non-irradiated group. For phosphate and amide III, there was significant difference only for irradiation ($p=0.015$ and $p=0.038$, respectively). Tukey's test revealed that within the cervical third the irradiated dentin presented altered values of these chemical components. For carbonate, there was significant difference only for irradiation ($p=0.002$), and Tukey's test showed that within the cervical and apical thirds the irradiated dentin presented lower values of carbonate. Regarding the ratios, showed significant difference for third ($p<0.001$) in the mineral/matrix ratio (M:M), while irradiation and the interaction between the study factors did not ($p=0.826$ and $p=0.460$, respectively). For carbonate/mineral ratio (C:M), there was significant difference only for irradiation ($p=0.001$). In all thirds, the irradiated dentin presented lower values of C:M ratio when compared to the non-irradiated one. For amide I/CH2 ratio, ANOVA showed significant difference only for third ($p=0.007$). Raman spectroscopy data were analyzed for 961/1458 cm^{-1} ratios and revealed that the irradiation ($p=0.818$), resin cement ($p=0.381$) and the interaction between them ($p=0.273$) had no influence. The analysis of the bonding interface by CLSM showed less adhesive interaction in the irradiated dentin in both cements.

Conclusion: Under the conditions of this in vitro study, ionizing radiation altered the root dentin molecules, mainly in the cervical third. Resin cements had less interaction with irradiated root dentin. Conventional cement had greater interaction with irradiated dentin than self-adhesive cement.

Clinical Significance: Radiotheraphy alters the chemical composition of irradiated dentin and understanding the interaction of resin cement with root dentin is critical to the successful rehabilitating use of the fiberglass post.

Keywords: bonding interface, chemical analysis, dentin, radiotherapy, resin cement.

INTRODUCTION

Radiotherapy is a therapeutic modality routinely used to treat malignant neoplasms in the oral cavity. However, despite destroying tumor cells, radiotherapy can also cause some damage to healthy tissues, such as bones, mucosa, teeth and salivary glands, located in the radiation field [1,2,3,4]. The oral function can be impaired, and the patient's visit to the dentist more often may be necessary [1,2,4].

The side effects from radiotherapy, like hyposalivation and xerostomia, associated with changes of chemical and mechanical properties of teeth can lead to rapid and severe destruction of enamel and dentin [2,5,6,7,8,9,10,11]. In these cases, the rehabilitation can be challenging, once surgical procedures like extractions and insertion of dental implants are generally not recommended because of the risk of osteoradionecrosis [4]. Therefore, the use of posts for direct or indirect restorations of these teeth with extensive destruction could be an alternative [12,13,14].

Fiberglass posts are commonly used because they generate lower stresses inside the root canal and less deformation, reducing the risk of root fractures [15]. The cast post, although not requiring chemical interaction, produces more stress on a more fragile dentin. [13]. The chemical interaction of resin cements with dentin is important for the retention of fiber posts in root canals [16]. However, adhesive restorative procedures performed on irradiated teeth is a challenge for restorative dentistry, probably due to a fragile adhesive interface and frequent occurrence of cohesive defects in dentine [17,9,18]. Conventional cements demonstrated an great adhesion pattern to the post and root dentin, but they are very sensitive to the technique due to higher steps, higher risk of overetching and/or overdrying [19,20]. By the other hand, self-adhesive resin cements have been largely used in dentistry due to their ease of handling and optimum adhesion results, in healthy dentin [21].

Investigation in these field is scarce and needed to be made to possibility the oral rehabilitation in the head and neck cancer patients after radiotherapy. Thus, the aim of this study is to analyze the chemical changes of root dentin submitted to in vitro radiation and how it affects the interaction with resin cements. The null hypothesis tested were: (1) *in vitro* radiation does not alter the chemical composition of root dentin, regardless of root third; (2) *in vitro* radiation does not alter the interaction of resin cements with root dentin, regardless of resin cement type.

MATERIALS AND METHODS

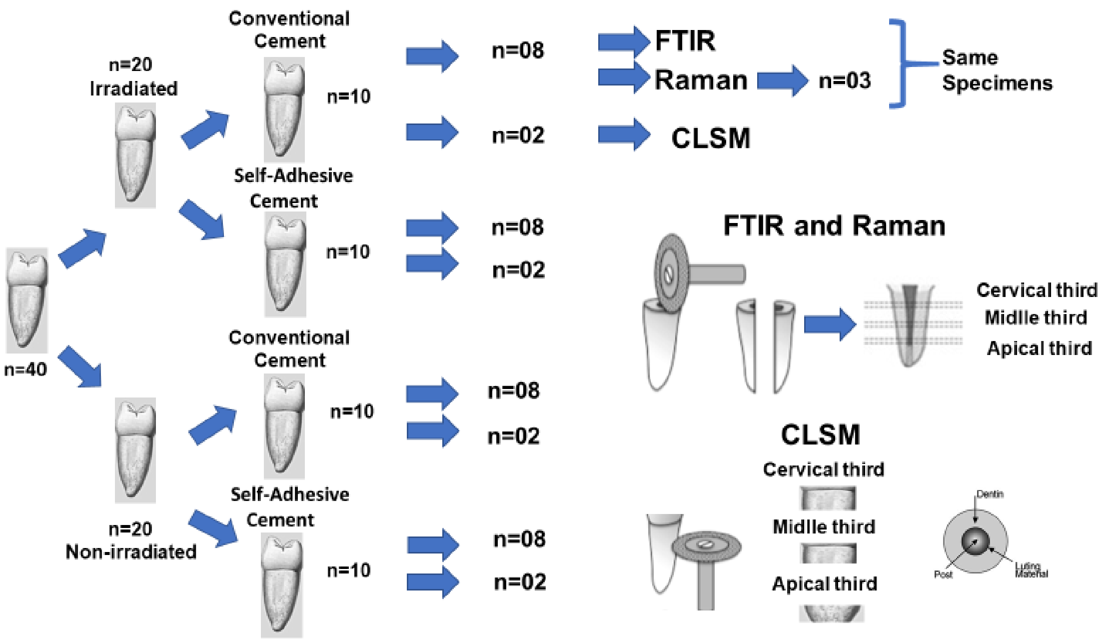

Fig. 1 Schematic illustration of the experimental design.

Teeth selection and irradiation

After approval by the Ethics Committee (protocol #11830919.0.0000.5152), 40 extracted single root human premolars with a ≥ 14-mm single root canal and complete apex formation were selected for this study. Exclusion criteria were the presence of resorption, root caries, fractures, or previous endodontic treatment. All teeth were cleaned with a curette and rubber bowl with pumice and water, coupled to hand piece and stored in deionized water at 4ºC for up to 3 months after extraction.

The teeth were randomly divided into two groups (n=20): non-irradiated and irradiated. The teeth were maintained in deionized water at 4ºC changed weekly [8,22]. The irradiation protocol consisted of a total dose of 70 Gy, with 2 Gy daily applied 5 days per week, during 7 weeks with X-rays from a linear accelerator (Clinac 600C Varian® - Palo Alto, CA, USA, Beam 6 MV). During irradiation, the teeth were fixed in utility wax plates (Technew, Rio de Janeiro, RJ, Brazil) and immersed in artificial saliva (pH 7.0) a room temperature and completely covering the teeth. Every 15 days the artificial saliva was discarded and the specimens were maintained in new artificial saliva [8,22].

Specimen preparation

Teeth were sectioned with a double-faced diamond blade in a precision saw (Isomet 1000, Buehler, Lake Bluff, IL, USA) and the root length standardized at 14 mm. The root canal was located using a 15 K-file (Dentsply Sirona, Pirassununga, SP, Brazil) that was introduced into the root canal until it was visible at the apical foramen. The working length was set at 1 mm below of the apical foramen. Canal preparation was performed with a Reciproc R50 instruments, size 0.50mm and taper 0.05mm (VDW GmbH, Munich, Germany). The root canals were prepared following the manufacturer's recommendations, using a peck and pull movement of about 3 mm in amplitude with light apical pressure, and cleaned after three peck movements until reaching the working length. One reciproc instrument was used to prepare 5 specimens and then replaced. The root canals were irrigated with 2 mL of 2.5% sodium hypochlorite (Q-boa, Osasco, SP, Brazil) using a syringe between each preparation step. After the preparation, the final irrigation was performed with 2 mL of 17% ethylene diamine tetra-acetic acid (EDTA, Biodinâmica, Ibiporã, PR, Brazil) for 3 minutes and passive ultrasonic irrigation using the E1 - Irrisonic insert (Helse Ultrasonic, Santa Rosa de Viterbo, SP, Brazil) for 20 seconds, followed by irrigation with 5 mL of 2.5% NaOCl and 10 mL of deionized water. Root canals were dried by means of suction cannulas and tips of absorbent paper cones (Tanari Industrial Ltda., Manaus, AM, Brazil) [18]. The obturation was performed with standard gutta-percha cone number 50 (Dentsply Sirona) and AH Plus sealer (Dentsply Sirona) [22]. After the adaptation of the master cone and accessory cones by lateral condensation, the specimens were stored in 100% humidity at 4°C for 24 hours. The root canal was sealed with glass ionomer (Vidrion C, SS White, Rio de Janeiro, RJ, Brazil) after treatment. The same operator performed all procedures of preparation and obturation of the root canal for all the specimens.

After this period, the gutta-percha was removed to a depth of 10 mm with Gates-Glidden of sizes 2-4 (Dentsply Sirona Endodontics) and the 3 mm remaining filling material was confirmed by digital radiography. The roots were prepared with a post-specific drill (White Post DC #1; FGM, Joinville, SC, Brazil). The canal was irrigated with 2.5% NaOCl during drilling, and finally rinsed with deionized water and passive ultrasonic irrigation performed using the E1 - Irrisonic insert (Helse Ultrasonic) for 20 seconds. The canals were then dried with absorbent paper cones (Tanari Industrial Ltda.) and distributed into two subgroups according to the type of resin cement (n=10): Conventional cement and self-adhesive.

For cementation with conventional cement, the canals were conditioned with 37% phosphoric acid (Condac®, FGM, Joinville, Brazil) for 15 seconds and rinsing with 10 mL of deionized water followed by drying with absorbent paper cones (Tanari Industrial Ltda., Manaus, AM, Brazil). Adhesive Adper Single Bond 2 (3M ESPE, St. Paul, MN, USA) was then applied with microbrush (KG Sorensen, Cotia, SP, Brazil) to the entire surface of the root canal, followed by a jet of air and a second layer of adhesive on the same surface. After removing the excess, the photoactivation was made for 40 seconds at 1100 mW cm2 using a high-power LED (Valo, Ultradent, South Jordan, UT, USA). For cementation with self-adhesive cement, no previous etching was done.

Each fiberglass post (White Post DC #1; FGM, Joinville, SC, Brazil) was etched with 35% hydrogen peroxide (Whiteness HP, FGM, Joinville-SC, Brazil) for 1 minute [23], and rinsed with deionized water and then air dried for 30 seconds. A silane agent (3M ESPE, St. Paul, MN, USA) was subsequently applied to the post for 1 minute [24]. Prior to the cementation, the roots were wrapped with wax (Technew, Rio de Janeiro, RJ, Brazil) to prevent further cement polymerization. The cements were manipulated and inserted into the canal with the help of the Centrix syringe (Maquira, Maringá, PR, Brazil), in sufficient quantify to fill the entire root canal, thus avoiding the formation of void [25]. The fiberglass post was introduced into the canal and kept under digital pressure for 5 minutes for chemical activation [26]. After this time, polymerization was carried out at a power of 1100 mW/cm2 for 40 seconds on three sides of the specimen (occlusal, buccal and lingual), totaling 120 seconds. The specimens were stored in 100% humidity at 4˚C for 7 days. The resin cements used are described in Table 1.

Table 1 – Resin cements, batch number, manufacturer and composition.

Fourier transform infrared spectroscopy (FTIR)

Chemical composition was determined for non-irradiated and irradiated groups (n=10) using attenuated total reflectance/Fourier transform infrared spectroscopy (ATR/FTIR; Vertex 70, Bruker, Ettlingen, Germany). To conduct the test, the roots with fiberglass posts were sectioned in halves along their longitudinal axis. . The specimens were ultrasound with deionized water for five minutes to remove the smear layer formed after using the cutter's diamond disk. The dentin surfaces were positioned against the diamond crystal of the ATR/FTIR unit and pressed with a force gauge at a constant pressure to

Resin cement	Batch number	Manufacturer	Composition
Conventional cement	1833200207	3M Oral Care; St Paul, MN, USA	Tube A: Bis-GMA, TEGDMA, zirconia and silica, pigments, amine and photoinitiating system. Tube B: Bis-GMA, TEGDMA, zirconia e silica, benzoyl peroxide.
Self-adhesive cement	4380967	3M Oral Care; St Paul, MN, USA	Base Tube: silane treated glass powder, 2-propenoic acid, 2-methyl1,1 [1-(hydroxymethyl) -1,2-ethanodlyl] ester, TEGDMA, silane treated silica, fiberglass, sodium persulfate and t-butyl per-3,5,5-trimethylhexanoate. Catalyst Tube: silane-treated glass powder, substitute dimethacrylate, silane-treated silica, sodium p-tulenesulfonate, 1-benzyl-5-phenyl baric acid, calcium salts, 1,12-dodecane dimethacrylate, calcium hydroxide and titanium dioxide.

Abbreviations: Bis-GMA: bisphenol glycidyl methacrylate; TEGDMA: triethylene glycol dimethyl acrylate.

facilitate contact. The spectra were recorded in the range from 400 to 4000 cm^{-1} at a 4 cm^{-1} resolution. The specimens were scanned 32 times in each measurement, and the final spectrum acquired is the average of all these scans. Spectra were recorded and analyzed by OPUS 6.5 software (Bruker, Ettlingen, Germany). After baseline correction and normalization, the FTIR spectra were analyzed by calculating the following parameters: (1) mineral/matrix ratio M:M (the ratio of the integrated areas at 1035 and 1655 cm^{-1}, attributed to the v_1, v_3 vibration of phosphate ion and the C=O stretching of collagen amide I, respectively); (2) carbonate/mineral ratio C:M (the ratio of the integrated areas of carbonate v_2 at 872 cm^{-1} to the phosphate v_1, v_3 at 1035 cm^{-1}); (3) amide I/amide III ratio (the ratio of the integrated areas of amide I at 1655 cm^{-1} to the amide III at 1235 cm^{-1}); (4) amide I/CH_2 ratio (the ratio of the integrated areas of amide I at 1655 cm^{-1} to the CH_2 scissoring at 1450 cm^{-1}) [13,15].

Raman spectroscopy

Three samples per group were evaluated by a LabRam HR Evolution Raman spectrometer (Horiba LabRam, Villeneuve d'Ascq, France) to verify the diffusion zone of the resin cements and their chemical interaction with root dentin. The spectrometer operated at an excitation power of 20 mW with radiation emitted by a He-Ne laser (785 nm). Raman signal was acquired using a 600-line/mm grating centered between 480 and 1800 cm^{-1} with a 400-µm confocal hole. The roots with fiberglass posts were sectioned in halves along their longitudinal axis with a water-cooled diamond saw (Isomet 1000; Buehler). The root sections with exposed adhesive interfaces were rinsed with deionized water in an ultrasonic bath for 5 minutes to remove the debris smeared on the specimen surface during cutting. Since the post from one of the halves was consumed during cutting, only half of each root was available for the Raman analysis [16].

Raman spectra were acquired at the cervical root area, mapping the interface starting from the resin cements and moving towards the dentin at 1 µm intervals by using the computer at 100X magnification and controlled *x–y–z* stage[16]. Spectra were collected along 1 line-scan with 20 μm long. Five 30-second accumulations were used for spectra readings. In order to avoid dehydration of the specimens, gauze soaked in deionized water was kept in contact with the dentin during data collection. OriginPro 2018 software (OriginLab Corporation, Northampton, MA, USA) was used to analyze the acquired Raman data. The diffusion zone of the cements into dentin was identified through the Raman band peak at 1113 cm^{-1} (*v*C—O—C), which is representative of the carbon chain in the resin monomer present in all materials. Chemical interactions at the interface were investigated through the analysis of characteristic peaks that represent the mineral component in dentin (961 cm^{-1} - v_1 phosphate symmetric stretch) and the CH_2 group of methacrylate monomers (1458 cm^{-1} - deformation δ of CH_2) (Figure 1). The ratio between these intensity peaks (961 cm^{-1}/1458 cm^{-1}) was calculated for every Raman mapping point on the interface [16].

Confocal laser scanning microscopy (CLSM)

Following the same adhesive protocols described above, two specimens per group were prepared for observation under confocal laser scanning microscopy (CLSM). The colorant rhodamine B (Sigma, St Louis, MO, USA) was incorporated into the resin cements and sodium-salt fluorescein (Sigma, St Louis, MO, USA) into the adhesive (1% of colorant was incorporated). After 7 days of cementation, the specimens were cut in two 1.0-mm thick slices of each root region and examined under a CLSM (LSM 510 Meta, Zeiss, Oberkochen, Germany). The rhodamine B was excited at 543 nm excitation line of helium ion laser, and the fluorescein was excited at 488 nm excitation line of argon ion laser. Each resin-dentin interface was completely investigated, and then representative optical images were captured (10X magnification).

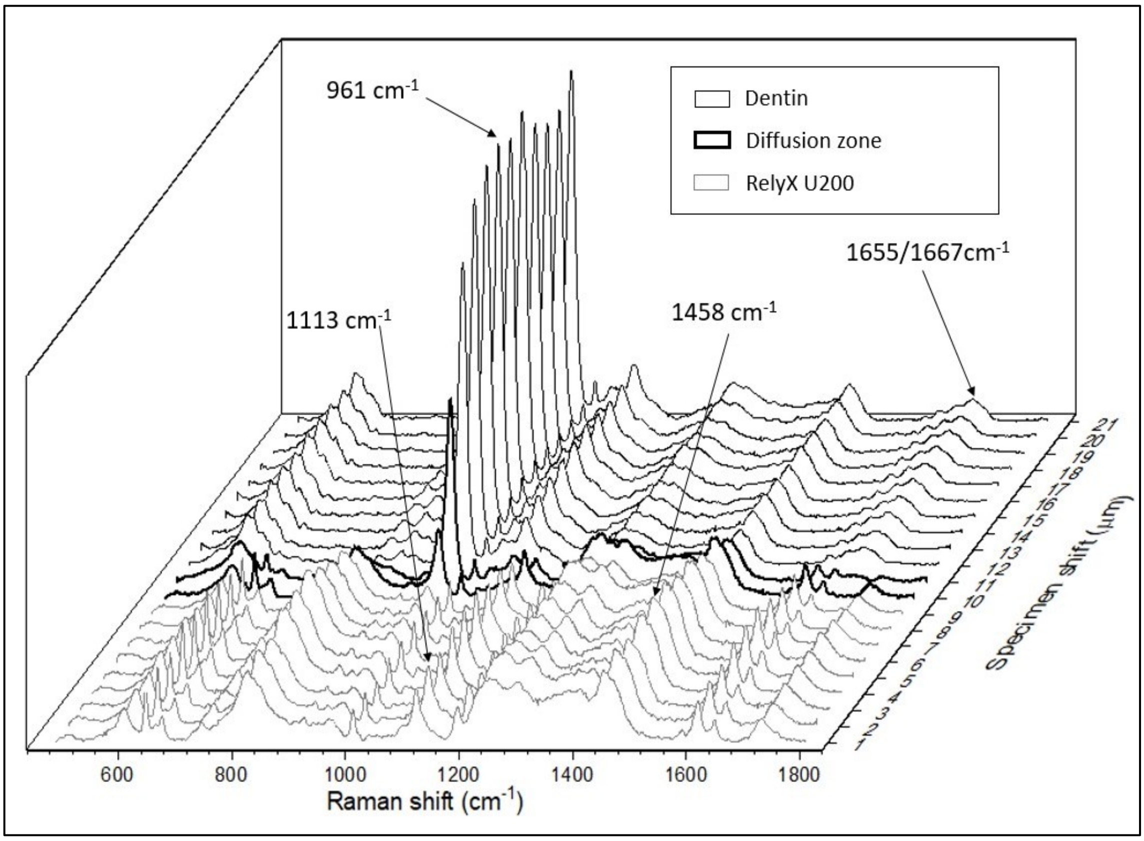

Figure 2 – Representative Raman spectra of the adhesive interface (in this case self-adhesive cement). Each measurement is represented by a numbered line on the z-axis (specimen shift). The gray lines represent cement, the black bold lines represent the diffusion zone, and the black slim lines the dentin.

Statistical analysis

Data were tested for normal distribution (Shapiro-Wilk, $p > 0.05$) and equality of variances (Levene's test, $p > 0.05$). FTIR data were analyzed using One-way ANOVA with repeated measurement (thirds), followed by Tukey's post hoc test. Raman data were analyzed using Two-way ANOVA and Tukey's test. The significance level was set at 5%. For confocal images, a descriptive analysis was done. All statistical analyses were performed using Sigma Plot statistical package (version 12.0, Systat Software, Inc., San Jose, CA, USA).

RESULTS

FTIR

The mean and standard deviation values for chemical parameters obtained by the ATR/FTIR are shown in Tables 2 and 3. For amide I, One-way ANOVA with repeated measurement showed significant difference only for third ($p<0.001$), and Tukey's test revealed a higher value for the apical in relation to the cervical third within the non-irradiated group. For phosphate and amide III, there was significant difference only for irradiation ($p=0.015$ and $p=0.038$, respectively). Tukey's test revealed that within the cervical third the irradiated dentin presented difference of values for the non-irradiated group of these chemical components. For CH_2, there was significant difference for third ($p=0.017$), and Tukey's test revealed a higher value for the apical in relation to the middle third within the non-irradiated group. For carbonate, there was significant difference only for irradiation ($p=0.002$), and Tukey's test showed that within the cervical and apical thirds the irradiated dentin presented lower values of carbonate.

Regarding the ratios, One-way ANOVA with repeated measurement showed significant difference in non-irradiated group for third ($p<0,001$) in the M:M ratio, but there was no difference between the study factors($p=0.826$). Within the non-irradiated group, Tukey's test revealed a higher ratio for the cervical in relation to the middle and apical thirds(Cervical/Middle $p=0.041$ e Cevical/Apical $p=0,001$). For C:M ratio, there was significant difference only for irradiation($p=0.001$), while between the root thirds there was no difference(Interaction between non-irradiated third $p=0.287$ and interaction between irradiated third $p=0.763$). In all thirds, the irradiated dentin presented lower values of C:M ratio when compared to the non-irradiated one. Amide I/amide III ratio showed no significant differences for any of the factors (irradiation $p=0.291$) and between the root thirds of the non-irradiated group $p = 0.051$ and irradiated $p = 0.068$). For amide I/CH_2 ratio, ANOVA showed significant difference only for third in group non-irradiated($p=0.007$). Among the thirds for the irradiated group ($p = 0.341$) and between the study factors ($p = 0.970$) there were no significant differences. Tukey's test revealed a lower ratio for the cervical in relation to the middle third within the non-irradiated group.

Table 2 – Mean and standard deviation of the integrated areas for each chemical component analyzed in FTIR.

	Amide I		Phosphate		Amide III		CH2		Carbonate	
	NI	IR	NI	IR	NI	IR	NI	IR	NI	IR
Cervical	1.53 (0.32) Ba	1.65 (0.26) Aa	15.64 (0.41) Aa	16.15 (0.43) Ab	0.23 (0.05) Aa	0.19 (0.03) Ab	0.21 (0.03) ABa	0.22 (0.06) ABa	0.28 (0.04) Aa	0.23 (0.05) Ab
Middle	1.77 (0.17) ABa	1.75 (0.13) Aa	15.71 (0.10) Aa	15.92 (0.43) Aa	0.22 (0.04) Aa	0.20 (0.03) Aa	0.19 (0.03) Ba	0.19 (0.02) Ba	0.28 (0.03) Aa	0.24 (0.02) Aa
Apical	1.99 (0.27) Aa	1.83 (0.19) Aa	15.64 (0.25) Aa	15.68 (0.13) Aa	0.22 (0.03) Aa	0.21 (0.02) Aa	0.22 (0.02) Aa	0.22 (0.03) Aa	0.29 (0.04) Aa	0.25 (0.02) Ab

	M:M ratio		C:M ratio		Amide I/ Amide III		Amide I/CH_2	
	NI	IR	NI	IR	NI	IR	NI	IR

***NI: non-irradiated; IR: irradiated.** Different uppercase letters indicate statistical difference between thirds (columms) and lowercase letters between irradiation (rows) ($p<0.05$).

Table 3 – Mean and standard deviation for the chemical ratios analyzed in FTIR.

Cervical	10.60 (2.24) Aa	10.09 (2.04) Aa	0.018 (0.002) Aa	0.014 (0.003) Ab	6.96 (2.23) Aa	8.75 (1.96) Aa	7.30 (1.53) Ba	8.15 (2.68) Aa
Middle	8.95 (0.96) Ba	9.16 (0.81) Aa	0.018 (0.002) Aa	0.015 (0.002) Ab	8.28 (1.40) Aa	8.96 (0.91) Aa	9.58 (1.20) Aa	9.16 (1.25) Aa
Apical	8.01 (1.07) Ba	8.64 (0.89) Aa	0.018 (0.003) Aa	0.016 (0.001) Ab	9.32 (1.46) Aa	8.80 (1.36) Aa	8.96 (1.26) ABa	8.60 (1.63) Aa

	Conventional Cement	**Self-adhesive Cement**
Non-irradiated	2.60 (0.18) Aa	2.69 (0.21) Aa
Irradiated	3.12 (1.11) Aa	2.34 (0.60) Aa

NI: non-irradiated; IR: irradiated.

Different uppercase letters indicate statistical difference between thirds (columms) and lowercase letters between irradiation (rows) ($p<0.05$).

Raman spectroscopy

The mean and standard deviation values for 961/1458 cm^{-1} are shown in Table 3. Two-way ANOVA revealed that the irradiation ($p=0.818$), resin cement ($p=0.381$) and the interaction between them ($p=0.273$) had no influence on 961/1458 cm^{-1} ratio. Figure 2 show the mean and minimum and maximum values of the diffusion zone in each sample for the ratios analyzed. Analyzing the amplitudes of each sample, we observed a heterogeneous behavior between maximum and minimum values.

Table 4 – Mean and standard deviation values for 961/1458 cm^{-1} obtained at the diffusion zone by Raman spectroscopy.

Different uppercase letters (analysis in columns) and lowercase letters (analysis in rows) represent significant differences ($p<0.05$).

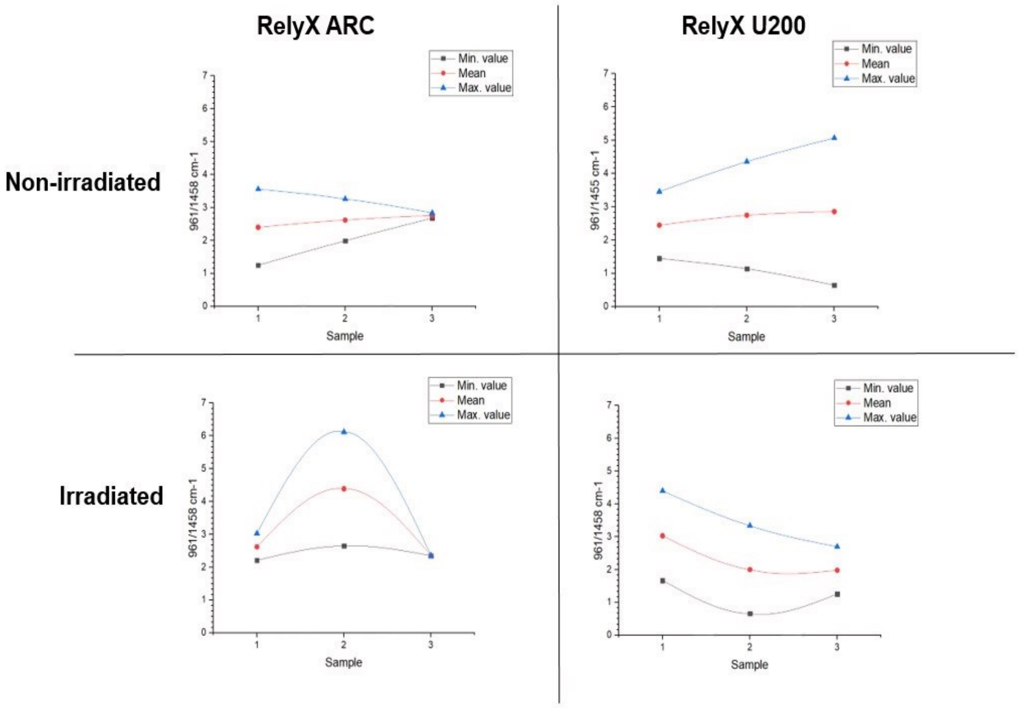

Figure 3 – Mean and minimum and maximum values of the diffusion zone in each sample for 961/1458 cm^{-1} ratio.

CLSM

Figure 4 show the interaction pattern of resin cement with dentin in each group, in the cervical third of the root, through magnifications of 10X. It is possible to observe the different interaction pattern for both cements in the irradiated dentin.

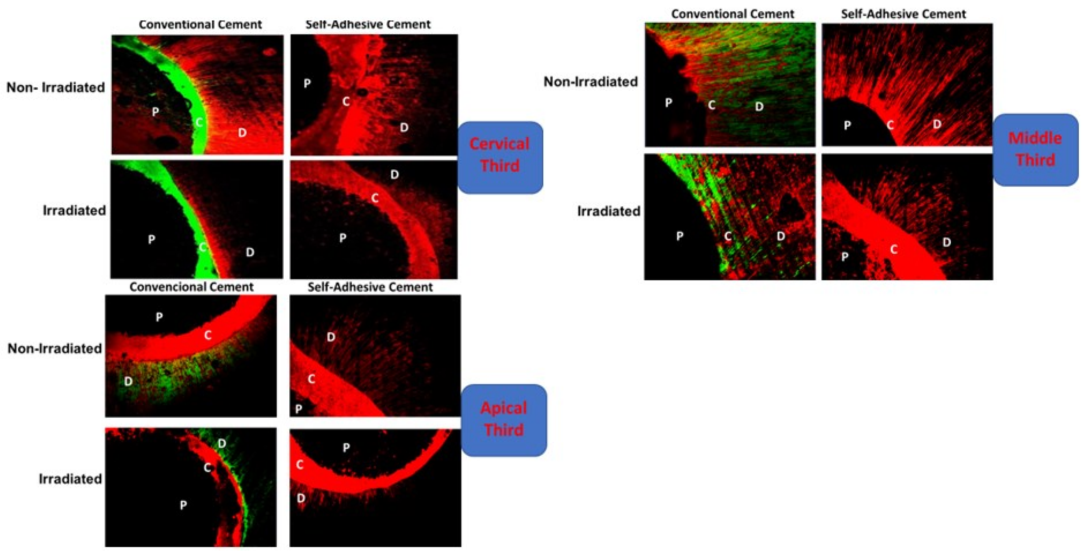

Figure 4 – Confocal images representing the interaction pattern of resin cement with dentin in each group (magnifications of 10X). Colorant rhodamine B= Red (resin cements); Colorant sodium-salt fluorescein = Green (adhesive); D= dentine; C= cement; P= post

DISCUSSION

The two null hypothesis were rejected, since in vitro radiation altered the chemical composition of root dentin (especially in the cervical third) and changed the interaction pattern of resin cements with root dentin, as can be seen in maximum and minimum amplitude values in each sample in Raman spectroscopy and CLSM images. Therefore, dentists should be aware of the difficulties in treating these patients undergoing radiotherapy and their implications for oral rehabilitation in order to obtain a good dental treatment outcome [27].

The first null hypothesis, was rejected probably due to interaction of ionizing radiation with dental tissues by exciting the molecules, releasing free radicals, for example oxygen ions(O^{-2}), hydroxyl (OH^{-}), hydrogen ($+^{1}$) and these bind to other molecules and reorganize with a change in dentin's chemical components, changing the conformation of the molecules[28]. Therefore, the ionization energy released by radiotherapy can cause a molecular reshuffling in dental hard tissues [29}. Understanding the effects of radiotherapy on the organic and inorganic components of dentin is critical to establishing in-depth analysis for the dental organ[29]. Our results showed higher values of phosphate ions in the cervical third of irradiated group when compared to the non-irradiated. Meanwhile, carbonate ions showed lowest values in the cervical and apical thirds of irradiated root dentin, which caused changes in the C:M ratio of these same irradiated thirds. The cervical third of root is probably the region most affected by ionizing radiation, because it is a region more exposed to the oral environment and less protected by alveolar bone and gingival tissues [5]. This change caused by ionizing radiation could cause an imbalance to phosphate and carbonate ions, especially in the cervical third of the root, facilitating ion exchange with artificial saliva[8]. Decreasing carbonate ion makes the tissue more susceptible to acid, causing imbalance, degradation and accelerating the dissolution process of the tooth structure [30,31,11]. These changes in substrate composition can potentially interfere with the pattern of interaction with resin cements, especially during the fiberglass bonding process [18].

Amide I is the main absorption band in protein and is very important in the organization of protein structure. This is determined by the conformation of the skeleton and the hydrogen bonding pattern [32]. The reason for the higher values of amide I found in the apical third compared to the cervical third is because in the cervical third of the root there is a higher density of dentinal tubules with a greater presence of peritubular dentin than in the apical third. Peritubular dentin surrounds the dentinal tubules is much more mineralized than intertubular dentin. Intertubular dentin has a higher amount of organic content than tubular dentin [33,32]. For the phosphate / amide I ratio (M: M), there was a difference between the cervical third and the middle and apical thirds of the non-irradiated group, justified by the higher mineral quantity in relation to the organic components of the cervical third root.

Differences were obtained for amide III values when compared the non-irradiated and irradiated group. It can be explained by the great molecular instability of amide III submitted to ionizing radiation. Amide III is a protein that participates in the formation of collagen fibers and acts in the conformation between the organic and mineral components. The disorganization of amida III may result in increased solubility and decreased root resistance [31]. The amide I / amide III ratio represents the organization of collagen in dentin [34]. This ratio was similar to the non-irradiated and irradiated group, probably because the teeth were immersed in wax at the cementum-enamel junction and covered with artificial saliva, making it difficult to expose the roots directly to ionizing radiation. This model was used to simulate what happens clinically in radiotherapy patients, where teeth are protected by periodontal tissues and in constant contact with saliva [31]. The quality of collagen can be represented by the amide I / CH2 ratio [34,35]. There was no difference between the non-irradiated and irradiated group. This shows that ionizing energy did not change the quality of organic material [34,35,31]. Within the non-irradiated group, this proportion differed between the cervical and middle thirds. The variation in tubular density and dentin permeability could explain this fact. Regarding CH2 values, we found similarity between the non-irradiated and irradiated groups and the difference between the middle and apical thirds, suggesting a higher collagen concentration due to the higher amount of intertubular dentin in the apical third of the root [33,32]. CH2 represents the intensity of side chain vibrations of the molecular conformation of the polypeptide chain. Therefore, the irradiation did not influence the organization and quality of collagen, interfering more in the mineral than in the organic matrix of root dentin.

The interface between cement and root dentin was evaluated by Raman spectroscopy, comparing etch-and-rinse and self-adhesive mode of interaction of resin cement in non-irradiated and irradiated teeth. Knowledge about the interaction of resin cements with irradiated dentin is of paramount importance for the dentist's decision on dental rehabilitation [18]. The 961/1458 cm^{-1} ratio by Raman spectroscopy evaluate the relation of phosphate group of dentin to CH2 monomer of resin cement. This ratio analyzes the interaction quality of cement with root dentin [16]. This study showed no difference of the mean values between the non-irradiated and irradiated group and neither to the resin cements tested. However, the authors also evaluated the amplitude to show the heterogeneity of minimum and maximum values of the samples, and it was possible to seem the heterogeneity for conventional resin cement on irradiated dentine. It can be probably explained because of technical sensibility of conventional cement, that needs acid etching to expose collagen fibers and moisture-lowering solvents associated with adhesives that imbue collagen fibers and inside dentinal tubules to form the dentin hybrid layer [21,19,20], associated with an altered dentine by irradiation.

CLSM images showed for middle and apical thirds a similar pattern between non-irradiated and irradiated groups. Cervical third showed that the interaction of resin cement with the root dentine was markedly lower in the irradiated groups. The behavioral pattern of self-adhesive cement in the irradiated group had less interaction than the self-adhesive cement in the non-irradiated group. The hybrid layer formed by the conventional resin cement from the irradiated group was smaller compared to the non-irradiated dentin. These results were also shows in the FTIR spectroscopy analysis, which showed a change in phosphate, carbonate and amide III molecules, and C: M ratio in the cervical root third. This disorganization can justify a lower interaction of cements in the cervical third of the irradiated teeth. The amplitude oscillation between the maximum and minimum values related to Raman spectroscopy was observed mainly in the cervical third of the irradiated group of conventional cement. This lesser interaction of the self-adhesive cement with the irradiated dentin was probably due to the alterations observed in the mineral components of the irradiated dentin, essential to an adequate adhesion for this class of materials, as these depend mainly on the interaction with the mineral components of the dentin. For the conventional cement of the irradiated group, the lesser interaction of the cement was due to the alteration of the mineral components and changes in the organization of the organic components of the irradiated dentin.

Regarding the methodologies employed in this work, spectroscopy is very effective in determining the chemical components that make up the structures under analysis. FTIR spectroscopy measures the infrared light excitation, wavelength and intensity of light absorption of a given structure [9]. Using software (Opus 6.5), a graph with a given spectrum is obtained and the frequencies of the various modes of vibration of the organic and inorganic molecules are read. ATR (attenuated total reflectance) is a form of internal reflection spectroscopy, in which a sample is placed in contact with a high refractive index, for example, a diamond crystal [10]. As a complementary methodology to the FTIR spectroscopy, Raman spectroscopy utilizes vibrational energy levels, governed by light-scattering processes that can achieve a greater reading depth, since it depends on the phenomenon of dispersion and reflection of light. Thus we can estimate the diffusion zone of the cements and their chemical interaction with dentin [11]. In these methodologies, both Raman spectroscopy and ATR / FTIR samples maintain original conditions and can be used for further analysis [11]. For the CLMS methodology, the colorant impregnated with resin cement shows, through magnification with the use of CLMS, the cement-dentin interaction seen by the images.

Changes on root irradiated dentine indicate that further studies evaluating adhesion should be performed to better understand the adequate oral rehabilitation treatment of head and neck cancer patients. Studies evaluating the bond strength of cements to root dentin are important. Other cements that behave differently than those used in the research should be tested. Clinical studies evaluating cementation behavior under the influence of oral function and environment are necessary for the definition of fiber post cementation protocols.

CONCLUSION

Ionizing radiation altered the root dentin molecules, mainly in the cervical third. Resin cements had less interaction with irradiated root dentin. The etch-and-rinse and self-adhesive resin cements did not present difference influence on chemical interaction between root dentine and resin cement.

REFERENCES

1. Vissink A, Jansma J, Spijkervet FK, Burlage FR, Coppes RP. Oral sequelae of head and neck radiotherapy. Crit Rev Oral Biol Med. 2003; 14(3):199-212. https://doi.org/10.1177/154411130301400305
2. Tolentino ES, Centurion BS, Ferreira LHC, Souza AP, Damante JH, Rubira-Bullen IRF. Oral adverse effects of head and neck radiotherapy: literature review and suggestion of a clinical oral care guideline for irradiated patients. J Appl Oral Sci. 2011; 19(5):448-54. https://doi.org/10.1590/S1678-77572011000500003
3. Thiagarajan A, Iyer NG. Radiation-induced sarcomas of the head and neck. World J Clin Oncol. 2014 Dec 10;5(5):973-81. https://doi.org/10.5306/wjco.v5.i5.973

4. Jawad H, Hodson NA, Nixon PJ. A review of dental treatment of head and neck cancer patients, before, during and after radiotherapy: part 1. Br Dent J. 2015; 218(2):65-8. https://doi.org/10.1038/sj.bdj.2015.28
5. Gupta N, Pal M, Rawat S, Grewal MS, Garg H, Chauhan D, Ahlawat P, Tandon S, Khurana R, Pahuja AK, Mayank M, Devnani B. Radiation-induced dental caries, prevention and treatment - A systematic review. National Journal of Maxillofacial Surgery. 2015; 6, 160-6. https://doi.org/10.4103/0975-5950.183870
6. Soares CJ, Roscoe MG, Castro CG, Santana FR, Raposo LH, Quagliatto PS, Novais VR Effect of gamma irradiation and restorative material on the biomechanical behaviour of root filled premolars. Int Endod J. 2011 Nov;44(11):1047-54. https://doi.org/10.1111/j.1365-2591.2011.01920.x
7. Naves LZ, Novais VR, Armstrong SR, Correr-Sobrinho L, Soares CJ. Effect of gamma radiation on bonding to human enamel and dentin. Support Care Cancer. 2012 Nov;20(11):2873-8. https://doi.org/10.1007/s00520-012-1414-y
8. Gonçalves, L. M. N., Palma-Dibb, R. G., Paula-Silva, F. W. G., de Oliveira, H. F., Nelson-Filho, P., da Silva, L. A. B., & de Queiroz, A. M. Radiation therapy alters microhardness and microstructure of enamel and dentin of permanent human teeth. Journal of Dentistry. 2014; 42(8), 986–992. https://doi.org/10.1016/j.jdent.2014.05.011
9. Rodrigues RB, Soares CJ, Junior PCS, Lara VC, Arana-Chavez VE, Novais VR. Influence of radiotherapy on the dentin properties and bond strength. Clin Oral Investig. 2018 Mar;22(2):875-883. https://doi.org/10.1007/s00784-017-2165-4
10. Lopes, C. de C. A., Limirio, P. H. J. O., Novais, V. R., & Dechichi, P. Fourier transform infrared spectroscopy (FTIR) application chemical characterization of enamel, dentin and bone.2018; Applied Spectroscopy Reviews, 2018; 53(9), 747–769. https://doi.org/10.1080/05704928.2018.1431923
11. de Miranda RR, Silva ACA, Dantas NO, Soares CJ, Novais VR. Chemical analysis of in vivo-irradiated dentine of head and neck cancer patients by ATR-FTIR and Raman spectroscopy. Clin Oral Investig.2019 Aug; 23(8):3351-3358. https://doi.org/10.1007/s00784-018-2758-6

12. Soares CJ, Monise Rodrigues MP, Faria-e-Silva AL, Santos-Filho PCF, Veríssimo C, Kim H-C, Versluis A. How biomechanics can affect the endodontic treated teeth and their restorative procedures? Braz. Oral Res. 2018;32(suppl):e76 169-183; https://doi.org/10.1590/1807-3107bor-2018.vol32.0076
13. Santos-Filho PC, Veríssimo C, Soares PV, Saltarelo RC, Soares CJ, Martins LRM. Influence of ferrule, post system, and length on biomechanical behavior of endodontically treated anterior teeth. Journal of Endodontics. 2014; 40, 119-23. https://doi.org/10.1016/j.joen.2013.09.034
14. Novais VR, Rodrigues RB, Simamoto Júnior PC, Lourenço CS, Soares CJ. Correlation between the Mechanical Properties and Structural Characteristics of Different Fiber Posts Systems. Braz Dent J. 2016 Jan-Feb;27(1):46-51. https://doi.org/10.1590/0103-6440201600377
15. Wang X, Shu X, Zhang Y, Yang B, Jian Y, Zhao K. Evaluation of fiber posts vs metal posts for restoring severely damaged endodontically treated teeth: a systematic review and meta-analysis. Quintessence Int. 2019;50(1):8-20.
16. Ubaldini ALM, Benetti AR, Sato F, Pascotto RC, Medina Neto A, Baesso ML, Peutzfeldt A. Challenges in luting fibre posts: Adhesion to the post and to the dentine. Dent Mater. 2018 Jul; 34(7):1054-1062. https://doi.org/10.1016/j.dental.2018.04.001
17. Aggarwal V. An in vitro evaluation of effect of ionizing radiotherapy on push-out strength of fiber posts under cyclic loading. J Endod. 2009 May;35(5):695-8. https://doi.org/10.1016/j.joen.2009.01.010
18. Yamin PA, Pereira RD, Lopes FC, Queiroz AM, , Saquy PC, Sousa-Neto MD. Longevity of bond strength of resin cements to root dentine after radiation therapy. Int Endod J. 2018 Nov; 51(11):1301-1312. https://doi.org/10.1111/iej.12945
19. Farina AP, Chiela H, Carlini-Junior B, Mesquita MF, Miyagaki DC, Randi Ferraz CC, Vidal CM, Cecchin D. Influence of Cement Type and Relining Procedure on Push-Out Bond Strength of Fiber Posts after Cyclic Loading. J Prosthodont. 2016 Jan; 25(1):54-60. https://doi.org/10.1111/jopr.12271

20. Rodrigues RV, Sampaio CS, Pacheco RR, Pascon FM, Puppin-Rontani RM, Giannini M. Influence of adhesive cementation systems on the bond strength of relined fiber posts to root dentin. J Prosthet Dent. 2017 Oct;118(4):493-499. https://doi.org/10.1016/j.prosdent.2017.01.006

21. Amaral M, Rippe MP, Bergoli CD, Monaco C, Valandro LF. Multi-step adhesive cementation versus one-step adhesive cementation: push-out bond strength between fiber post and root dentin before and after mechanical cycling. Gen Dent. 2011 Sep-Oct; 59(5): e185-91.

22. Martins, C. V., Leoni, G. B., Oliveira, H. F., Arid, J., Queiroz, A. M., Silva, L. A. B., & Sousa-Neto, M. D. Influence of therapeutic cancer radiation on the bond strength of an epoxy- or an MTA-based sealer to root dentine. International Endodontic Journal. 2016; 49(11), 1065–1072. https://doi.org/10.1111/iej.12556

23. Menezes MS, Faria-e-Silva AL, Silva FP, Reis GR, Soares CJ, Stape TH, Martins LR. Etching a fiber post surface with high-concentration bleaching agents. Oper Dent. 2014 Jan-Feb;39(1):E16-21. https://doi.org/10.2341/12-270-L

24. Valdivia AD, Novais VR, Menezes Mde S, Roscoe MG, Estrela C, Soares CJ. Effect of surface treatment of fiberglass posts on bond strength to root dentin. Braz Dent J. 2014;25(4):314-20. https://doi.org/10.1590/0103-6440201300143

25. Silva NR, Rodrigues MP, Bicalho AA, Soares PBF, Price RB, Soares,CJ. Effect of resin cement mixing and insertion method into the root canal on cement porosity and fiberglass post bond strength. J Adhes Dent. 2019; 21:37-46.

26. Pereira RD, Valdívia AD, Bicalho AA, Franco SD, Tantbirojn D, Versluis A, Soares CJ. Effect of Photoactivation Timing on the Mechanical Properties of Resin Cements and Bond Strength of Fiberglass Post to Root Dentin. Oper Dent. 2015; 40, 206-21. https://doi.org/10.2341/14-115-L

27. Fromm L, Gotfredsen K, Wessel I, Øzhayat EB. Oral health-related quality of life, oral aesthetics and oral function in head and neck cancer patients after oral rehabilitation. J Oral Rehabil. 2019 Aug;46(8):738-746. https://doi.org/10.1111/joor.12806

28. Seredin P, Goloshchapov D, Prutskij T, Ippolitov Y. Phase transformations in a human tooth tissue at the initial stage of caries. PLoS One. 2015; 10(4):e0124008. https://doi.org/10.1371/journal.pone.0124008
29. Velo MMAC, Faha ALH, Santos PSS, Shiota A, Sansavino SZ, Souza AT, Honório HM, Wang L. Radiotherapy alters the composition, structural and mechanical properties of root dentin in vitro. Clin Oral Investig.2018 Nov; 22(8):2871-2878. https://doi.org/10.1007/s00784-018-2373-6
30. Tartari T, Bachmann I, Maliza AGA, Andrade FBA, Duarte MAH, Bramante CM. Tissue dissolution and modifications in dentin composition by different sodium hypochlorite concentrations. J Appl Oral Sci. 2016; 24(3):291-8. https://doi.org/10.1590/1678-775720150524
31. Campi LB, Lopes FC, Soares LES, de Queiroz AM, de Oliveira HF, Saquy PC, Sousa-Neto MD. Effect of radiotherapy on the chemical composition of root dentin. Head Neck. 2019 Jan;41(1):162-169. https://doi.org/10.1002/hed.25493
32. Xu C, Wang Y Chemical composition and structure of peritubular and intertubular human dentine revisited. Arch Oral Biol. 2012 Apr;57(4):383-91. https://doi.org/10.1016/j.archoralbio.2011.09.008
33. Schellenberg, U., Krey, G., Bosshardt, D., & Ramachandran Nair, P. N. Numerical density of dentinal tubules at the pulpal wall of human permanent premolars and third molars. Journal of Endodontics, 1992; 18(3), 104–109. https://doi.org/10.1016/S0099-2399(06)81308-7
34. Salehi, H, Terrer E, Panayotov I, Levallois B, Jacquot B, Tassery H, Cuisinier F. Functional mapping of human sound and carious enamel and dentin with Raman spectroscopy. J. Biophotonics. 2013; 6, 765-774. https://doi.org/10.1002/jbio.201200095
35. Toledano M, Aguilera FS, Osorio E, Cabello I, Toledano-Osorio M, Osorio R. Functional and molecular structural analysis of dentine interfaces promoted by a Zn-doped self-etching adhesive and an in vitro load cycling model. J Mech Behav Biomed Mater.2015; 50:131–149. https://doi.org/10.1016/j.jmbbm.2015.05.026

3. REFERÊNCIAS

1. Vissink A, Jansma J, Spijkervet FK, Burlage FR, Coppes RP. Oral sequelae of head and neck radiotherapy. Crit Rev Oral Biol Med. 2003; 14(3):199-212.
2. Tolentino ES, Centurion BS, Ferreira LHC, Souza AP, Damante JH, Rubira-Bullen IRF. Oral adverse effects of head and neck radiotherapy: literature review and suggestion of a clinical oral care guideline for irradiated patients. J Appl Oral Sci. 2011; 19(5):448-54.
3. Thiagarajan A, Iyer NG. Radiation-induced sarcomas of the head and neck. World J Clin Oncol. 2014 Dec 10;5(5):973-81.
4. Jawad H, Hodson NA, Nixon PJ. A review of dental treatment of head and neck cancer patients, before, during and after radiotherapy: part 1. Br Dent J. 2015; 218(2):65-8.
5. Soares CJ, Monise Rodrigues MP, Faria-e-Silva AL, Santos-Filho PCF, Veríssimo C, Kim H-C, Versluis A. How biomechanics can affect the endodontic treated teeth and their restorative procedures? Braz. Oral Res. 2018;32(suppl):e76 169-183;
6. Santos-Filho PC, Veríssimo C, Soares PV, Saltarelo RC, Soares CJ, Martins LRM. Influence of ferrule, post system, and length on biomechanical behavior of endodontically treated anterior teeth. Journal of Endodontics. 2014; 40, 119-23.
7. Novais VR, Rodrigues RB, Simamoto Júnior PC, Lourenço CS, Soares CJ. Correlation between the Mechanical Properties and Structural Characteristics of Different Fiber Posts Systems. Braz Dent J. 2016 Jan-Feb;27(1):46-51.
8. Santos-Filho PC, Veríssimo C, Soares PV, Saltarelo RC, Soares CJ, Martins LRM. Influence of ferrule, post system, and length on biomechanical behavior of endodontically treated anterior teeth. Journal of Endodontics. 2014; 40, 119-23.
9. Ubaldini ALM, Benetti AR, Sato F, Pascotto RC, Medina Neto A, Baesso ML, Peutzfeldt A. Challenges in luting fibre posts: Adhesion to the post and to the dentine. Dent Mater. 2018 Jul; 34(7):1054-1062.

10. Aggarwal V. An in vitro evaluation of effect of ionizing radiotherapy on push-out strength of fiber posts under cyclic loading. J Endod. 2009 May;35(5):695-8.
11. Rodrigues RB, Soares CJ, Junior PCS, Lara VC, Arana-Chavez VE, Novais VR. Influence of radiotherapy on the dentin properties and bond strength. Clin Oral Investig. 2018 Mar;22(2):875-883.
12. Yamin PA, Pereira RD, Lopes FC, Queiroz AM, , Saquy PC, Sousa-Neto MD. Longevity of bond strength of resin cements to root dentine after radiation therapy. Int Endod J. 2018 Nov; 51(11):1301-1312.
13. Farina AP, Chiela H, Carlini-Junior B, Mesquita MF, Miyagaki DC, Randi Ferraz CC, Vidal CM, Cecchin D. Influence of Cement Type and Relining Procedure on Push-Out Bond Strength of Fiber Posts after Cyclic Loading. J Prosthodont. 2016 Jan; 25(1):54-60.
14. Rodrigues RV, Sampaio CS, Pacheco RR, Pascon FM, Puppin-Rontani RM, Giannini M. Influence of adhesive cementation systems on the bond strength of relined fiber posts to root dentin. J Prosthet Dent. 2017 Oct;118(4):493-499.
15. Amaral M, Rippe MP, Bergoli CD, Monaco C, Valandro LF. Multi-step adhesive cementation versus one-step adhesive cementation: push-out bond strength between fiber post and root dentin before and after mechanical cycling. Gen Dent. 2011 Sep-Oct; 59(5): e185-91.

4. ANEXOS

UFTM - HOSPITAL DAS CLÍNICAS DA UNIVERSIDADE FEDERAL DO TRIÂNGULO

PARECER CONSUBSTANCIADO DO CEP

Elaborado pela Instituição Coparticipante

DADOS DO PROJETO DE PESQUISA

Título da Pesquisa: Análise in vitro da resistência de união de cimentos resinosos à dentina radicular de dentes irradiados

Pesquisador: Veridiana Resende Novais

Área Temática:

Versão: 1

CAAE: 11830919.0.3001.8667

Instituição Proponente: Hospital de Clínicas da Universidade Federal do Triângulo Mineiro

Patrocinador Principal: Financiamento Próprio

DADOS DO PARECER

Número do Parecer: 3.441.997

Apresentação do Projeto:

O presente projeto pretende analisar a resistência de união de cimentos resinosos à dentina radicular de dentes irradiados. Para isso, 60 dentes serão coletados e divididos em dois grupos (n=30): irradiado e não irradiado. Posteriormente serão subdivididos em dois subgrupos de acordo com o tipo de cimento resinoso (n=15): cimento resinoso convencional e cimento resinoso autoadesivo. Após tratamento endodôntico e cimentação de pinos de fibra de vidro, as raízes serão seccionadas em seis fatias (duas fatias para o terço cervical, duas para o terço médio e duas para o terço apical), sendo que uma fatia de cada terço será analisada imediatamente e outra seis meses após a cimentação. A resistência de união será testada por meio de ensaio mecânico de push-out (n=10). Em seguida, as amostras serão levadas em microscópio eletrônico de varredura para avaliação do padrão de falha. Para avaliar a zona de difusão do cimento e interação química deste com a dentina, três amostras por grupo serão submetidas à espectroscopia Raman.

Para análise da interface adesiva, duas amostras de cada grupo serão levadas ao microscópio confocal. Após a realização das metodologias, caso os dados das análises quantitativas apresentem distribuição normal será empregada 3-way Anova e, caso ocorra diferença entre os grupos, será empregado o teste de Tukey. Caso não apresentem distribuição normal, os dados serão analisados com teste não-paramétrico de Kruskal-Wallis. Nas microscopias, uma análise descritiva das imagens será feita.

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A hipótese desse estudo é que a irradiação e o momento da avaliação influenciam na resistência e interface de união de cimentos resinosos à dentina radicular. Os dentes deverão ter indicação clínica de exodontia e prévio consentimento dos pacientes que estarão cientes do uso nesta pesquisa e assinarão o termo de consentimento livre e esclarecido. As metodologias serão realizadas no Centro de Pesquisa Odontológico Biomecânico, Biomateriais e Biologia Celular (CPBio), da Faculdade de Odontologia da UFU. A irradiação das amostras será realizada no Hospital de Clínicas da Universidade Federal do Triângulo Mineiro (UFTM).

Objetivo da Pesquisa:

Objetivo Primário:

Avaliar a resistência e a interface de união de cimentos resinosos na dentina radicular de dentes irradiados e não irradiados, variando o tipo de cimento e o momento da avaliação.

Objetivos Secundários:

Avaliar a resistência de união e o padrão de falha de cimento resinoso convencional e autoadesivo à dentina radicular irradiada ou não, imediatamente e 6 meses após a cimentação;

Avaliar a zona de difusão dos cimentos resinosos na dentina radicular (interface adesiva);

Avaliar a interação química dos cimentos resinosos com a dentina radicular.

Avaliação dos Riscos e Benefícios:

Segundo os pesquisadores:

Riscos: A coleta dos dentes será realizada pelo mestrando Alessandro Marcelo Pelloso, após o atendimento clínico dos pacientes nas clínicas de cirurgia da Faculdade de Odontologia da Universidade Federal de Uberlândia. O único risco é a identificação dos participantes, o que contraria a Resolução 466/12, porém a equipe executora se compromete em não revelar em nenhum momento a identidade dos pacientes e assim, serão utilizados códigos para minimizar este risco. Os dentes utilizados serão dentes com indicação clínica de exodontia e os pacientes que concordarem farão a doação destes dentes por escrito ao assinarem o termo de consentimento livre e esclarecido.

Benefícios: Por meio desse trabalho, os pesquisadores buscam comparar a resistência de união de pinos de fibra de vidro nos condutos radiculares de prémolares extraídos e submetidos à radiação ionizante. Portanto, busca-se verificar o desempenho de dois cimentos resinosos (convencional e autoadesivo), avaliados imediatamente após a cimentação e 06 meses depois. Isso tudo se deve ao fato de que pinos de fibra de vidro são comumente utilizados na reconstrução de coroas dentárias severamente destruídas, bastante comuns em pacientes que se submetem ao tratamento radioterápico e são acometidos por cáries relacionadas à radiação. Os resultados obtidos possibilitarão aos cirurgiões-dentistas compreenderem de maneira mais aprofundada as alterações que ocorrem nos tecidos dentais desses pacientes, bem como sua interação com os materiais resinosos utilizados na prática odontológica.

Comentários e Considerações sobre a Pesquisa:

Pesquisa acadêmica visando avaliar a resistência e a interface de cimentos resinosos na dentina de dentes irradiados e não irradiados, conforme o tipo de cimento utilizado e o momento da avaliação. Pesquisa realizada na Universidade Federal de Uberlândia sendo a parte de radiação ionizante realizada no serviço de radioterapia do HC-UFTM.

Considerações sobre os Termos de apresentação obrigatória:

Apresentados adequadamente, tanto os originais apresentados no CEP-UFU quanto os necessários para tramitação no CEP-HC-UFTM.

Recomendações:

Não há recomendações

Conclusões ou Pendências e Lista de Inadequações:

De acordo com as atribuições definidas na Resolução CNS 466/12 e norma operacional 001/2013, o colegiado do CEP-HC/UFTM manifesta-se pela aprovação do protocolo de pesquisa proposto, situação definida em reunião do dia 04/07/2019.

O CEP-HC/UFTM não se responsabiliza pela qualidade metodológica dos projetos analisados, mas apenas pelos pontos que influenciam ou interferem no bem-estar dos participantes da pesquisa conforme preconiza as normas da Comissão Nacional de Ética em Pesquisa – CONEP.

Considerações Finais a critério do CEP:

Conforme prevê Norma Operacional Nº01/2013 CNS, e resolução 466/2012 CNS, o acompanhamento dos projetos na Plataforma Brasil é de inteira responsabilidade dos pesquisadores, não podendo ser alegado desconhecimento de pendências como justificativa para não cumprimento de prazos.

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Obs:

• A secretaria do CEP-HC/UFTM está à disposição para quaisquer esclarecimentos sobre trâmites e funcionalidades da Plataforma Brasil, durante os dias de segunda a sexta-feira, das 07 às 16 h. Telefone: 34 3318-5319. e-mail: cep.hctm@ebserh.gov.br.

Este parecer foi elaborado baseado nos documentos abaixo relacionados:

Tipo Documento	Arquivo	Postagem	Autor	Situação
Informações Básicas do Projeto	PB_INFORMAÇÕES_BÁSICAS_DO_PROJETO_1363220.pdf	06/06/2019 13:30:00		Aceito
TCLE / Termos de Assentimento / Justificativa de Ausência	TCLE.pdf	12/04/2019 17:57:09	Veridiana Resende Novais	Aceito
Projeto Detalhado / Brochura Investigador	Projeto_Detalhado.pdf	02/04/2019 16:23:21	Veridiana Resende Novais	Aceito
Outros	Links_para_Curriculos_Lattes_dos_pesquisadores.pdf	05/02/2019 15:32:27	Veridiana Resende Novais	Aceito
Outros	Calculo_Amostral.pdf	05/02/2019 15:31:38	Veridiana Resende Novais	Aceito

Situação do Parecer:
Aprovado

Necessita Apreciação da CONEP:
Não

UBERABA, 05 de Julho de 2019

Assinado por:
GILBERTO DE ARAUJO PEREIRA
(Coordenador(a))