

Mary Stefany Andrade Carvalho

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Dissertação apresentada à Faculdade de
Odontologia da Universidade Federal de
Uberlândia, para obtenção do título de Mestre
em Clínica Odontológica Integrada.

Uberlândia MG
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Orientadora: Prof^a Dra. Veridiana Resende Novais Simamoto

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UNIVERSIDADE FEDERAL DE UBERLÂNDIA
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ATA DE DEFESA - PÓS-GRADUAÇÃO

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Iniciando os trabalhos o(a) presidente da mesa, Dr(a). Veridiana Resende Novais Simamoto, apresentou a Comissão Examinadora e o candidato(a), agradeceu a presença do público, e concedeu ao Discente a palavra para a exposição do seu trabalho. A duração da apresentação do Discente e o tempo de arguição e resposta foram conforme as normas do Programa.

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RESUMO

Pacientes submetidos à radioterapia na região de cabeça e pescoço podem apresentar um dos efeitos adversos mais comuns após o tratamento, a cárie relacionada à radiação ionizante, e o manejo dessas lesões ainda é astante desafiador para o cirurgião-dentista. O objetivo deste trabalho foi avaliar o efeito do Diamino Fluoreto de Prata nas propriedades químicas e mecânicas da dentina humana irradiada in vivo. Para isso, foram coletados doze terceiros molares hígidos e doze pré-molares e molares de pacientes pós-radioterapia. Os dentes foram divididos em dois grupos (n=12) sendo um grupo controle, dentes hígidos, e o grupo com dentes irradiados. Em ambos os grupos, foi aplicado Diaminofluoreto de prata a 38% (SDF, SDI Limited, Victoria, Austrália). A análise da dentina ocorreu em duas etapas, antes da aplicação do SDF e após 1 mês. As hemiseções foram analisadas por Espectroscopia Infravermelha Transformada de Fourier (FTIR), teste de microdureza Knoop (KHN) e teste de rugosidade superficial. Os dados foram analisados por ANOVA two-way (irradiação e aplicação SDF), seguido do teste post hoc de Tukey. O FTIR revelou diferenças para o fator de irradiação ($p=0,047$) para a banda Carbonato, para o fator de aplicação SDF na banda Amida I ($p=0,006$), Carbonato ($p=0,008$) e todas as razões, C:F ($P=0,004$), M:M ($P=0,005$) e AmidaI:Amida III ($P=0,007$). Para KHN, houve diferença significativa no fator de irradiação ($p=0,047$) e aplicação de SDF ($p<0,001$) entre os grupos. A rugosidade superficial também apresentou diferença estatística para o fator de irradiação ($p<0,001$) e aplicação de SDF ($p<0,001$) entre os grupos. A utilização do SDF sobre a dentina irradiada causa alterações estruturais na porção orgânica, reorganização de minerais mais solúveis e formação/deposição de minerais. Assim, em última análise, aumentando as propriedades mecânicas da dentina irradiada e reduzindo sua rugosidade.

Palavras-chave: Cariostáticos; Dentina; Espectroscopia de Infravermelho Transformada de Fourier; Dureza; Neoplasias de Cabeça e Pescoço; Radioterapia.

ABSTRACT

Patients undergoing radiotherapy in the head and neck region may have one of the most common adverse effects after treatment, caries related to ionizing radiation, and the management of these lesions is still quite challenging for the dental surgeon. The aim of this work was to evaluate the effect of Silver Diamino Fluoride on the chemical and mechanical properties of human dentin irradiated in vivo. For this, twelve healthy third molars and twelve premolars and molars of post-radiotherapy patients were collected. The teeth were divided into two groups (n=12) being a control group, sound teeth, and the group with irradiated teeth. In both groups, 38% silver diaminofluoride (SDF, SDI Limited, Victoria, Australia) was applied. The dentin analysis took place in two steps, before SDF application and after 1 month. The hemisections were analyzed by Fourier Transformed Infrared Spectroscopy (FTIR), Knoop microhardness test (KHN) and surface roughness test. Data were analyzed by two-way ANOVA (irradiation and SDF application), followed by Tukey's post hoc test. The FTIR revealed differences for the irradiation factor ($p=0.047$) for the Carbonate band, for the SDF application factor in the Amide I band ($p=0.006$), Carbonate ($p=0.008$) and all ratios, C:F ($P=0.004$), M:M ($P=0.005$) and Amid I: Amide III ($P=0.007$). For KHN, there was a significant difference in the irradiation factor ($p=0.047$) and application of SDF ($p<0.001$) between groups. Surface roughness also showed statistical difference for the irradiation factor ($p<0.001$) and application of SDF ($p<0.001$) between groups. The use of SDF on irradiated dentin causes structural alterations in the organic portion, reorganization of more soluble minerals and formation/deposition of minerals. Thus, ultimately increasing the mechanical properties of irradiated dentin and reducing its roughness.

Keywords: Cariostatic Agents; Dentin; Spectroscopy, Fourier Transform Infrared; Hardness; Head and Neck Neoplasms; Radiotherapy.

INTODUÇÃO E REFERENCIAL TEÓRICO

Os cânceres de cabeça e pescoço incluem um grupo heterogêneo de neoplasias com origem na cavidade oral, orofaringe, seios paranasais, nasofaringe e laringe (Schutte et al., 2020). A incidência do câncer na região de cabeça e pescoço tem aumentado em todo o mundo, sendo o sétimo câncer mais frequente, responsável por mais de 660.000 novos casos e 325.000 mortes anualmente (Sung et al., 2021; Johnson et al., 2020).

O tratamento para o câncer de cabeça e pescoço envolve a terapia radioterápica geralmente associada a cirurgia, quimioterapia e terapias sistêmicas (Kitamura et al., 2021). Infelizmente, a radioterapia pode causar danos aos tecidos sadios adjacentes à região tumoral, o que resulta em diversos efeitos colaterais na cavidade oral dos pacientes (Moore et al., 2020). Sinais e sintomas que incluem mucosite, hipossalivação, xerostomia, osteorradionecrose e cárie relacionada a radiação, aspectos que afetam diretamente a qualidade de vida desses pacientes (Villa e Akintoye, 2018; Ray-Chaudhuri et al., 2013).

A radioterapia também causa efeitos diretos nos tecidos dentais, a dentina como apresenta um maior teor de água em sua composição torna-se mais vulnerável aos efeitos da radiação, isso ocorre devido a formação de radicais livres reativos após a interação dos raios com as moléculas de água (Rodrigues et al., 2018; Fang et al., 2002). A cárie relacionada à radiação (RRC) é um dos efeitos adversos mais comuns após o tratamento radioterápico e está diretamente relacionada aos efeitos diretos da radiação na estrutura dentária e aos efeitos indiretos, como a hipossalivação associada à dieta cariogênica (Chopra et al., 2022).

A RRC geralmente ocorre após seis meses a conclusão da radioterapia na cabeça e pescoço, apresenta uma progressão rápida e destrutiva e diferem da lesão de cárie em pacientes não irradiados quanto a sua localização, afetam principalmente região cervical, incisais e pontas de cúspide, apesar da rápida progressão geralmente não apresentam dor aguda e manifestam com coloração amarronzada ou preta (Chopra et al., 2022; Moore et al., 2020).

O Fluoreto Diamino de Prata (SDF) é um produto odontológico de uso tópico que inibe a degradação do colágeno na dentina, propriedade anticariogênicas, remineralizantes e bactericidas (Brunnet et al., 2022; Zhao et al., 2018). O SDF apresenta quatro principais efeitos anti-cárie: o primeiro é a ação antibacteriana do íon prata; o

segundo é a precipitação de fosfato de prata e cloreto de prata, formando uma camada protetora; terceira ação é a formação da fluorohidroxiapatita pouco solúvel e resistente a ácidos e a quarta ação que é a capacidade de preservar o colágeno (Sayed et al., 2020; Mei et al., 2018; Zhao et al., 2018).

Desde 1969, o SDF tem sido utilizado para prevenir e controlar cáries em dentes decíduos de crianças, para prevenir cárie de fossas e fissuras de molares em irrupção e prevenção de cáries radiculares em idosos (Tan et al., 2010; Yamaga R e Yokomizo, 1969). O SDF é um produto que combina os efeitos antibacterianos da prata com os efeitos remineralizantes do flúor, sendo considerado um agente terapêutico muito útil no controle de lesões cariosas (Brunet et al., 2022).

O Diamino Fluoreto de Prata é um agente terapêutico promissor para o tratamento de lesões de cárie, diante disso, estudos que investiguem o comportamento do SDF sobre a estrutura dental irradiada são urgentemente importantes e necessários. Nesse contexto, o objetivo deste trabalho foi avaliar o efeito do SDF nas propriedades químicas e mecânicas da dentina humana irradiada in vivo.

CAPÍTULO ÚNICO

Este trabalho será submetido à periódico Qualis/ CAPES vigente estrato A4 ou superior.

Efeito do Diamino Fluoreto de Prata nas propriedades da dentina irradiada *in vivo*

ABSTRACT

Patients undergoing radiotherapy in the head and neck region may have one of the most common adverse effects after treatment, caries related to ionizing radiation, and the management of these lesions is still quite challenging for the dentist. The objective of this work was to evaluate the effect of Silver Diamino Fluoride on the chemical and mechanical properties of human dentin irradiated *in vivo*. For this, twelve healthy third molars and twelve premolars and molars of post-radiotherapy patients were collected. The teeth were divided into two groups (n=12) being a control group, sound teeth, and the group with irradiated teeth. In both groups, 38% Silver Diamino Fluoride (SDF, SDI Limited, Victoria, Australia) was applied. The dentin analysis took place in two stages, before the application of the SDF and after 1 month. The hemisections were analyzed by Fourier transform infrared spectroscopy (FTIR), Knoop microhardness test (KHN) and Surface roughness test. Data were analyzed using two-way ANOVA (irradiation and SDF application), followed by Tukey's post hoc test. The FTIR revealed differences for the irradiation factor for Carbonate ($p=$), for the SDF application factor to Amide I band ($p=$), Carbonate ($p=$) and all ratios, C:F ($P=0.004$), M:M ($P=0.005$) and AmidI:Amide III($P=0.007$). For KHN, there was a significant difference in the irradiation factor ($p=0.047$) and SDF application ($p<0.001$) between the groups. Surface roughness also showed statistical difference for the irradiation factor ($p<0.001$) and application of SDF ($p<0.001$) between groups. The use of SDF on irradiated dentin causes structural changes in the organic portion, reorganization of more soluble minerals and formation/deposition of minerals. Thus, ultimately increasing the mechanical properties of irradiated dentin and reducing its roughness.

Keywords: Dentin; Radiotherapy; silver diamine fluoride; Hardness; Fourier transform infrared spectroscopy.

Introduction

The incidence of cancer in the head and neck region has increased worldwide, being the seventh most common cancer, responsible for more than 660,000 (Sung et al., 2021) and generally the adopted treatment protocols are known to be multimodal including surgery, radiotherapy in the head and neck region and chemotherapy, used alone or in combination (Schutte et al., 2020). Radiotherapy is a treatment that, through ionizing radiation, eliminates tumor cells, but causes undesirable changes in the tissues surrounding the irradiation area. Adverse events in the head and neck region include oral mucositis, hyposalivation, radiation-related caries, osteoradionecrosis and trismus, effects that directly impact the quality of life of patients (Bohn et al., 2021; Kawashita et al., 2020).

Patients with head and neck cancer, after radiotherapy treatment, have a higher risk of radiation-related caries, the incidence is 30% among cancer patients and may increase when no preventive measures are taken before and during radiotherapy (Bohn et al., 2021). The development of radiation-related caries is directly linked to hypofunction of the salivary glands after radiation, the reduction in salivary flow impairs the buffering capacity of saliva and, as a result of this imbalance, a drop in pH occurs, favoring a more cariogenic oral microbiota (Moore et al., 2020; Palmier et al., 2020; Bohn et al., 2021). There are also direct radiotherapy damages to the tooth structure, changes in the mechanical properties and chemical composition of the tissues, especially at the dentinal-enamel junction, factors that contribute to biomechanical failure and enamel delamination after radiotherapy (Gupta et al., 2015).

Silver diamine fluoride (SDF) is a therapeutic agent for the treatment of carious lesions, the product combines the antibacterial effects of silver and the remineralizing effects of fluoride that impede the progress of carious lesions and prevent the development of future caries (Crystal et al., 2019; Burgess et al., 2018). SDF has been an option in the management of conventional caries, as it combines remineralizing and antimicrobial effects in deciduous and permanent teeth (Oliveira et al., 2019; Varughese et al., 2022).

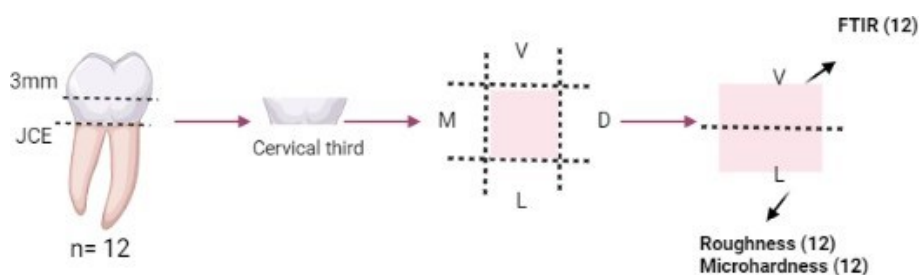
Radiation-related caries represents a major challenge for professionals regarding prevention and treatment measures, and SDF can be a promising option for its

management. Therefore, studies that investigate the interaction of SDF on the irradiated tooth structure are urgently important and necessary. Thus, the objective of this work was to evaluate the effect of SDF on the chemical and mechanical properties of human dentin irradiated *in vivo*. The null hypothesis tested is that SDF does not interact with irradiated dentin, altering its chemical and mechanical properties.

Methods

Specimen Preparation

This study was approved by the Ethical Committee in Research of the Federal University of Uberlândia (Protocol 59076222.6.0000.5152). Twelve premolars and molars from post-radiotherapy patients and twelve healthy third molars were collected, cleaned and stored in distilled water at a temperature of 4 °C for up to 3 months after extraction (Miranda, Silva, Dantas, Soares, & Novais, 2019). All teeth had a clinical indication for extraction and patients had previously signed an informed consent form. The teeth were divided into two groups (n=12), being a control group consisting of sound teeth, and the other group with irradiated teeth. In both groups, 38% SDF (Riva Star, SDI Limited, Victoria, Australia) was applied. The dentin analysis took place the SDF application: before SDF application and after 1 month of SDF application.



Teeth were cut at the cemento-enamel junction using a diamond disk (Isomet, diamond 15HC; Buehler Ltd., Lake Bluff, IL, USA) mounted on a precision saw (Isomet 1000; Buehler Ltd., Lake Bluff, IL, USA) with refrigeration. Subsequently, the crown was sectioned mesiodistally, resulting in two halves: buccal and lingual. The enamel present in the sample was also removed, only the dentin was analyzed. The mesiobuccal hemisections were analyzed by Fourier transform infrared spectroscopy (FTIR). The

distobuccal hemisections were submitted to the Knoop microhardness (KHN) test and surface roughness. The samples were stored in distilled water at 4°C during the pre-application test period of 38% SDF, after application of the product they were incubated at 37°C.

Experimental treatments

After the first analyses, the protocol for applying Riva Star (38%) was applied following the manufacturer's recommendations on the buccal and lingual side in the cervical region. The sample was properly dried with a light jet of air, a drop of Riva Star Step 1 (38% SDF solution) was applied using micro brush in an active manner for 60 seconds, then 2 drops were applied of Riva Star Step 2 (Potassium iodine solution) rubbed for 60 seconds, and to finish, absorbent paper was used in the region to remove excess product and rinsed with distilled water.

ATR/FTIR

The chemical composition of the samples was determined using the Fourier Transform Infrared Spectrometer (Vertex 70, Bruker, Ettlingen, Germany) using the Attenuated Total Reflection (ATR) technique. The tested surface was placed in contact with the diamond crystal of the ATR unit and constantly pressed by a dynamometer to increase the contact of the sample with the crystal of the equipment. Thus, the spectra will be recorded in the range of 400 to 4000 cm^{-1} , with a resolution of 4 cm^{-1} . Each specimen was scanned 32 times, and the final spectrum acquired was the average of these scans. Spectra were recorded and analyzed by OPUS 6.5 software (Bruker, Ettlingen, DEU).

The spectra acquired through FTIR were analyzed by calculating the following parameters: organic matrix /mineral ratio (M/M), given by the ratio between the bands at 1035 cm^{-1} and 1655 cm^{-1} , attributed to the vibration of the phosphate ion and to the C=elongation amide I in collagen, respectively; carbonate/mineral ratio (C/M), given by the ratio of integrated areas of carbonate at 872 cm^{-1} to phosphate at 1035 cm^{-1} and amide I/amide III ratio, indicated by the ratio of integrated areas of amide I at 1655 cm^{-1} for amide III at 1235 cm^{-1} (Lopes, Limirio, Novais, & Dechichi, 2018).

Microhardness test

To evaluate the Knoop microhardness (KHN) of the dentin, the samples were embedded in polystyrene resin (Resin AM 190; Aerojet, São Paulo, SP, Brazil) and the surfaces sanded with silicon carbide sandpaper (n° 600, 800, 1200 and 2000; Norton S/A, Campinas, SP, Brazil) for 60 seconds, polished with felt discs and metallographic diamond pastes (6, 3, 1 and $\frac{1}{4}$ μm granulation; Arotec, São Paulo, SP, Brazil) (Novais et al., 2016 ; Rodrigues et al., 2018). The entire process on the polishing machine was timed and performed by a single operator. Knoop indentation values were determined with a micro-durometer (FM 700; FutureTech Corp., Kawasaki, Japan) applying a load of 100 g for 15 s. Three indentations with a distance of 100 μm between them were made on each sample, and these measurements were subsequently calculated to determine the KHN of the samples.

Surface roughness

Each sample was subjected to a Surf Test SJ-301 Rugosimeter computerized roughness tester (Mitutoyo Corporation, Japan). Measurements were performed at a constant speed of 0.25 mm/s, measurement force of 4 mN and the cut-off value was set to 0.25 mm. The numerical parameters obtained were the mean surface roughness (R_a).

Statistical analysis

Data were tested for normal distribution (Shapiro-Wilk, $p > 0.05$) and equality of variance (Levene test, $p > 0.05$). Data were analyzed using two-way ANOVA (irradiation and SDF application), followed by Tukey's post hoc test. The level of statistical significance was 0.05, and all statistical analyses were performed using a statistical package (SigmaPlot® System, version 12.0, Systat Institute Inc.; San Jose, CA, USA).

Results

ATR/FTIR

The means and standard deviations of the integrated area of each chemical component analyzed are presented in Table 1. Considering the chemical components, two-way ANOVA showed statistical significance in relation to the SDF application factor

for amide I ($p = 0.006$) and carbonate ($p = 0.008$), which also showed statistical significance for the irradiation factor ($p = 0.047$).

Table 1. Means and the standard deviations of the integrated area of each chemical component by ATR/FTIR spectroscopy.

	Amide I		Phosphate		Amide III		Amide II		Carbonate	
SDF applica tion	non-irradiate d	irradiate d	non-irradiate d	irradiate d	non-irradiate d	irradiate d	non-irradiated	Irradiated	non-irradiate d	irradiate d
Before	2,988 +-	3,027 +-	21,933 +-	20,837 +-	0,408 +-	0,409 +-	0,588 +-0,100 Aa	0,611 +-0,080 Aa	2,572 +-	2,067 +-
	0,487 Aa	0,654 Aa	0,779 Aa	1,552 Aa	0,069 Aa	0,109 Aa			0,216 Aa	0,380 Ab
After 1 month	2,360 +-	2,291 +-	21,283 +-	21,312 +-	0,374 +-	0,405 +-	0,567 +-0,091 Aa	0,605 +-0,141 Aa	2,182 +-	1,899 +-
	0,406 Ba	0,685 Ba	0,485 Aa	0,861 Aa	0,056 Aa	0,075 Aa			0,270 Ba	0,520 Bb

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

For the ratios, two-way ANOVA showed statistical significance for the SDF application factor for all ratios, C/F ($p = 0.004$), M/M ($p = 0.005$) and Amide I / Amide III ($p = 0.007$), presented in Table 2.

Table 2. Means and the standard deviations for M/M, C/M and Amide I / Amide III by ATR/FTIR spectroscopy, comparing irradiated and non-irradiated and SDF application.

	C/F		M/M		Amide I / Amide III	
SDF application	non-irradiated	irradiated	non-irradiated	irradiated	non-irradiated	irradiated
Before	0,119 +- 0,011 Aa	0,103 +- 0,015 Aa	0,135 +- 0,019 Aa	0,146 +- 0,033 Aa	7,114 +- 0,550 Aa	7,067 +- 1,211 Aa
After 1 month	0,101 +- 0,009 Ba	0,090 +- 0,021 Ba	0,115 +- 0,012 Ba	0,109 +- 0,027 Ba	6,355 +- 0,577 Ba	6,454 +- 0,401 Ba

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

Means and standard deviations of KHN values are shown in Table 3. Two-way ANOVA showed a significant difference between groups for the irradiation factor ($p=0,047$) and SDF application factor ($p<0,001$).

Table 3. Means and the standard deviations for dentin KHN values before and after SDF application.

SDF application	non-irradiated	irradiated
Before	58,9 +- 6,24 Ba	49,6 +- 8,66 Bb
After 1 month	61,0 +- 6,55 Aa	60,1 +- 4,51 Ab

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

Surface roughness

Table 4 resents the means and standard deviations of the Ra values of the dentin surface performed by profilometry. Two-way ANOVA showed a significant difference between groups ($p < 0,001$) for the irradiation factor and the SDF application factor ($p < 0,001$).

Table 4. Mean and standard deviation (SD) of roughness values for irradiated and non-irradiated dentin surfaces before and after SDF application.

SDF application	non-irradiated	irradiated
Before	0,44 +- 0,056 Aa	0,39 +- 0,047 Ab
After 1 month	0,33 +- 0,030 Ba	0,27 +- 0,042 Bb

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

Discussion

The null hypothesis was rejected since the results indicated that after the application of SDF there is an interaction between the product and the irradiated dentin with significant values. The present study evaluated the effect of SDF on chemical and mechanical properties in healthy human dentin and in vivo irradiated dentin. This probably occurred because the irradiated dentin presented a change in its molecular conformation due to the effects of ionizing radiation (Fränzel, Gerlach, Hein, and Schaller, 2006).

In dentin, after the application of SDF, there is a reaction of the product with the mineral and organic phase of the structure. The silver reacts with the mineral phase and results in the formation of silver phosphate (Suzuk et al., 1974). The fluoride present in the compound reacts by being incorporated by hydroxyapatite, through ionic exchange between fluorine and hydroxyl ions resulting in the formation of fluorine-hydroxyapatite crystals (Mei et al., 2018), in addition to the formation of calcium fluoride. (Lou et al., 2011).

In the literature, the mechanism of action of SDF on deciduous and permanent teeth is clearly described regarding its remineralizing and antibacterial effects, however, regarding irradiated dentin, there are few studies that analyze the effect of SDF on the structure treated by radiation. De Souza et al. (2021) concluded that the SDF solution was able to reduce the development of radiation-induced dentin caries in vitro after microbiological analysis. In the present study, microhardness, chemistry and surface roughness were analyzed.

The methodologies used in this study provided information on the chemical and mechanical composition of dentin through non-destructive methods, allowing the use of the same method for different evaluations. The most intense absorption band in proteins is amide I, detected by the vibrations of the C=O (70-85%) and C-N (10-20) groups and is related to the conformation and pattern of hydrogen bonding (Xu and Wang, 2012). Amide I showed a significant decrease for the irradiated and non-irradiated groups. The alterations observed in the Amida groups suggest that the SDF favors a change in the organization of the collagen structure and also contributes to a dehydration of the collagen fibers (Miles et al., 2005; Belbachir et al., 2009). In view of this, treatment with SDF

generates a crosslinking effect on the collagen structure that contributes to mineral treatment movements (Cifuentes et al., 2023), a scenario that contributes to a lower reading in the Amida I band.

The carbonate band showed a decrease in value after SDF application for the non-irradiated and irradiated groups. Carbonate groups are more soluble than phosphate groups (Otsuka et al., 2012). In the present study the treatment with SDF altered the mineral phase of dentin, increasing electively in compounds of higher crystallinity and a possible greater dissolution of carbonates of lower crystallinity occurred (Driscoll et al., 2002). This may facilitate an ideal scenario for the precipitation of low solubility fluorohydroxyapatite (Hu et al., 2018). The radiation factor also showed a significant difference between the non-irradiated and irradiated groups, with a decrease in the carbonate value for the irradiated group. This probably occurred because the radiation interacted with chemical components of the tooth structure and somehow altered the chemical bonds, irradiated patients have an acidic environment in the oral cavity, which favors the dissolution of carbonate in the structure (Rodrigues et al., 2018 ; Lopes, Soares et al., 2018)

The ratios that illustrate the relationships between organic and inorganic components was calculated, in order to provide more information about the structure of collagen and hydroxyapatite. The FTIR showed that the C/F ratio (Carbonate/Phosphate) showed a significant decrease in value after the application of SDF in the non-irradiated and irradiated groups. The C/F ratio is important because it represents the mineral carbonate content in the hydroxyapatite (Toledano et al., 2015) and the decrease in the carbonate value directly impacts the ratio. It is inferred that these compositional variations influence the microstructural characteristics of hydroxyapatite crystals, contributing to mechanical changes in the structure.

The M/M ratio (Matrix/Mineral) evaluates the amount of organic matrix in relation to the inorganic matrix (Lopes, Soares et al., 2018). The M/M ratio presented a significant decrease after the application of SDF in the non-irradiated and irradiated groups. The lower ratio is explained due to a lower detection of amide I, that occurs due to the change in the collagen structure and the greater precipitation of minerals. After

treatment with SDF, there is a reaction with the hydroxyapatite that composes mineral dentin and calcium fluoride and insoluble silver phosphate salts are formed, minimizing the Amide I reading (Cifuentes et al., 2023). The Amide I/Amide III ratio evaluates the organization of collagen within the sample (Toledano et al., 2015) and the value of this ratio showed a significant decrease after the application of SDF in the non-irradiated and irradiated groups, suggesting a reorganization in the structure of the collagen.

There was an increase in the microhardness value after the application of SDF. The current result is similar to the study by Mante et al. (2022) and of Cifuentes et al. (2023) who showed an increase in the KHN value after treatment with SDF. The SDF reaction in dentin leads to the formation of a layer with a high silver content and deposits containing silver on the surface and in the dentinal tubules (Hiraishi et al., 2010; Willershausen et al., 2015). The incorporation of silver in hydroxyapatite, compounds containing silver in the tubules and the high formation rate of fluorohydroxyapatite may contribute to the increase in surface hardness (Mei et al., 2018; Mei et al., 2013). The higher concentration of SDF in the structure generates an inhibitory effect on metalloproteinases, a scenario that corroborates to prevent collagen degradation, reinforcing the mechanical properties (Cifuentes et al., 2023; Mei et al., 2012).

The irradiation factor also showed a significant difference, the non-irradiated group had a lower KHN value than the non-irradiated group. Exposure to radiation alters the composition and structure of dentin, radiation causes oxidation of water molecules producing hydrogen free radicals and highly reactive oxygen species, collaborating for the denaturation of organic components (Velo et al., 2018; Fränzel et al., 2006).

Surface roughness showed a significant decrease in values after application of SDF. We can infer that the fluorine and silver components of the SDF, when reacting with the dentin structure, protect the dentin surface with the formation of fluoroapatite, calcium fluoride and insoluble silver products such as silver phosphate, silver chloride or metallic silver, thus, there is a decrease in roughness (Ainoosah et al., 2020). When there is the analysis of the radiation factor, a significant decrease in roughness in the irradiated group is observed when compared to the non-irradiated group, this can be explained due to the action of radiation generating microcracks in the hydroxyapatite that occurs due to the decrease in the interaction of apatite crystals and organic matrix and increased carbon

dioxide production, resulting in a rough dentin surface (Liang et al., 2016).

Clinical studies are needed to confirm the results of this in vitro study and later collaborate for the creation of a protocol with the objective of minimizing the effects of radiation to dental tissues.

Conclusion

The use of silver diamine fluoride with high concentration on the irradiated dentin surface causes structural alterations in the organic portion, removal of more soluble minerals and formation/deposition of minerals more resistant to demineralization. Thus, ultimately increasing the mechanical properties of irradiated dentin and reducing its roughness.

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CONSIDERAÇÕES FINAIS

A cárie relacionada à radiação é um dos efeitos colaterais da radioterapia na região de cabeça e pescoço mais complexo e desafiador. O tratamento e prevenção dessas lesões apresentam um grande desafio aos profissionais, dessa forma a busca por alternativas que auxiliem o manejo e tratamento desse efeito adverso são de extrema importância. Este trabalho demonstrou que o uso do SDF é promissor para as lesões de cárie relacionada à radiação, o produto provoca alterações na porção orgânica e inorgânica, aumentando as propriedades mecânicas da dentina irradiada e reduzindo sua rugosidade.

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