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Effect of cavity preparation with Er:YAG laser and fluoride releasing materials on the prevention of caries lesions

Ana Carolina Tedesco JORGE^a, Mario Alberto Marcondes PERITO^b, Patricia Moreira de FREITAS^c, Alessandra CASSONI^d, José Augusto RODRIGUES^d

^aDDS, MS student, Dental Research and Graduate Studies, Division Department of Restorative Dentistry, UnG – Guarulhos University, 07023-070 Guarulhos - SP, Brazil
^bDDS, MS, ScD student, Dental Research and Graduate Studies, Division Department of Restorative Dentistry, UnG – Guarulhos University 07023-070 Guarulhos - SP, Brazil
^cDDS, MS, PhD, Special Laboratory of Lasers in Dentistry, Department of Restorative Dentistry, School of Dentistry, USP – University of São Paulo 05508-000, São Paulo - SP, Brazil
^dDDS, MS, ScD, Dental Research and Graduate Studies, Division Department of Restorative Dentistry, UnG – Guarulhos University, 07023-070 Guarulhos - SP, Brazil

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Resumo

O objetivo desse estudo foi avaliar a influência da técnica de preparo cavitário e de materiais restauradores contendo flúor na prevenção da secundária. Dentes humanos foram seccionados em 72 blocos e distribuídos em dois grupos. Cavidades com 1,6 mm de diâmetro foram preparadas com pontas diamantadas ou laser de Er:YAG laser (6 Hz, 300 mJ, 47 J.cm⁻²). Cada grupo foi dividido em três subgrupos e restaurados com cimento de ionômero de vidro, ionômero de vidro modificado por resina ou uma resina composta. Os espécimes foram termociclados e submetidos a ciclagem de pH. As lesões de cárie artificial foram ranqueadas utilizando uma escala ordinal por inspeção visual. Os testes de Kruskal-Wallis e Dunn ($\alpha = 0,05$) não demonstraram diferenças no desenvolvimento de lesões entre as cavidades resturadas com o mesmo material e preparadas com pontas diamantadas ou laser de Er:YAG laser. O laser de Er:YAG utilizado para o preparo cavitário com 6 Hz, 300 mJ, 47 J.cm⁻² não demonstrou a habilidade de garantir maior ácido resistência aos preparos.

Palavras-chave: Laser de érbio; cárie dentária; resinas compostas; cimento de ionômero de vidro; flúor; cárie secundária.

Abstract

The objective of this study was to evaluate the influence of the cavity preparation technique and fluoride-containing restorative materials on the prevention of the secondary caries. Human teeth were sectioned into 72 blocks and distributed into 2 groups. Cavities measuring 1.6 mm were performed with diamond burs or Er:YAG laser (6 Hz, 300 mJ, 47 J.cm⁻²). Each group was divided into 3 sub-groups, and restored with a glass-ionomer cement, resinmodified glass-ionomer, or composite resin. The specimens were thermal cycled and submitted to pH cycling. Artificial caries were scored using an ordinal scale based on visual inspection. Kruskal-Wallis and Dunn test ($\alpha = 0.05$) showed no differences in the caries lesion development between the cavities restored with the same material and prepared with diamond burs or Er:YAG laser. The Er:YAG laser used for cavity preparation used with 6 Hz, 300 mJ, 47 J.cm⁻² did not show the ability to guarantee significantly more acid-resistance against acid challenge.

Keywords: Erbium laser; dental caries; composite resins; glass ionomer cement; fluoride; secondary caries.

INTRODUCTION

The metabolic processes in bacterial biofilm are physiological phenomena that may lead to enamel mineral loss and subsequent cavity formation because of the imbalance in the dynamic equilibrium between tooth mineral and plaque fluid, determining caries lesion development, which may be visually diagnosed by the presence of a white, opaque lesion¹. To avoid caries development an individual preventive treatment based on the patient's caries risk should be implemented¹. Secondary caries is the lesion at the margin of an existing restoration, similar to the primary lesion, but it may also show lines of demineralized tissue running along the cavity wall². The presence of fluoride in the oral cavity may inhibit the demineralization phenomena due to acid produced by bacteria in the biofilm. Therefore, the use of topical fluorides and fluoride-releasing restorative materials, such as glass ionomerbased materials are a useful way to prevent secondary caries, as well as caries in enamel located at a considerable distance from the margin³⁻⁶.

However, some patients at high caries risk need additional care and preventive treatments to avoid the development of primary or secondary caries^{1,5}. Some studies have shown the potential of laser irradiation to produce morphological and chemical change in dental enamel by organic matrix decomposition and reduction in carbonate content, resulting in a less acid-permeable enamel with improved resistance to bacterial acid⁶⁻⁸. The most used lasers for preventive procedures are CO₂ and Erbium lasers⁶⁻⁸. Although, they are classified as high intensity lasers, the energy densities needed for caries preventive treatment are low and enamel ablation is avoided⁷⁻⁹.

Ablation is a phenomenon that occurs when the laser energy is absorbed by water molecules and hydrous organic components of biological tissues, and the water vapor produced induces an increase in the internal pressure within the tooth tissue, resulting in microexplosions, which cause dental tissue removal¹⁰. Thus, ablative parameters are used to remove carious tissue and perform cavity preparations, which have the advantage of significantly reduced need for local anesthesia, no vibratory or auditory irritation, perceived by patients as being more comfortable,^{11,12} when compared with conventional bur preparations.

Although the energy densities used for cavity preparation are higher than those used for caries prevention, heat is produced during ablation and transmitted through the cavity margins and this non-ablated surface may be fused or melted with enamel recrystallization resulting in a substrate that is less permeable to bacterial acid diffusion^{13,14}. Nevertheless, is not known whether sufficient heat accumulates to thermal modify the chemical composition of enamel and improve its acid resistance, as occurs by direct laser irradiation with subablative energy densities.

Thus, if such increase in the acid resistance of enamel cavity margins were possible, it could act synergistically with restorative materials that release fluorides, to prevent caries lesion development. Therefore, the aim of the present study was to conduct an in vitro investigation, by means of visual evaluation of the effect of cavity preparation with Er:YAG laser, on the inhibition of secondary caries around cavities filled with fluoridereleasing restorative materials.

MATERIAL AND METHODS

The Ethics Research Committee approved the research protocol that included a factorial design to test the effects of the 3 restorative materials and 2 cavity preparation techniques - with diamond burs or Er:YAG laser, used in human teeth, which resulted in 6 experimental groups (Table 1).

A total of 72 dental blocks (n = 12/group) were restored in 12 stage. At each stage, 2 restorations of each restorative system were made in a cavity prepared with a diamond bur and in a cavity prepared with Er:YAG laser, according to a randomized complete block design with 1 replication per block. The qualitative variable response "development of artificial caries-like lesion" was evaluated blindly and independently by 3 calibrated examiners using an ordinal scale based on visual examination.

Table 1. Restorative systems and cavity preparation

Groups	Cavity preparation	Restorative systems	
G 1	Diamond burs	Conventional glass ionomer cement (GI)	
61	(#2094, KG Sorensen, Barueri, SP, Brazil)	(Ketac-Fil,3M/ ESPE, Seefeld, Germany)	
G 2	Diamond burs	Resin-modified glass ionomer (RM)	
	Diamond burs	(Vitremer, 3M/ESPE, St. Paul, MN, USA)	
G 3	Diamond burs	Composite resin (CR)	
	Diamond burs	(Z250, 3M/ESPE, St. Paul, MN, USA)	
5 .	Laser Er:YAG	Conventional glass ionomer cement	
G 4	(Kavo Key II; Kavo, Biberach, Germany)		
G 5	Laser Er:YAG	Resin-modified glass ionomer	
G 6	Laser Er:YAG	Composite resin	

To prepare the blocks, unerupted third molars were selected and stored in a 0.1% thymol solution. The teeth were soft-tissue debrided and cleaned with water/pumice slurry and rubber cups in a low-speed handpiece (Kavo do Brasil, Joinville, SC, Brazil). The crowns were sectioned to obtain 72 dental enamel/dentin blocks ($4 \times 4 \times 3 \text{ mm}^3$) from the middle of the crowns, using double-faced diamond discs #7020 (KG Sorensen, Barueri, SP, Brazil, 06454-920). After this, the blocks were stored in 100% humidity until the cavity preparations were performed.

The blocks were distributed into two halves; one half had Cylindrical class V cavities of approximately 1.6 mm in diameter and 1.6 mm deep prepared with diamond burs #2292 at high speed, using constant water spray coolant.

The other half had the cavities prepared with Er:YAG laser working at 294 0 nm in the Special Laboratory of Lasers in Dentistry (LELO - University of São Paulo - USP, São Paulo, Brazil). The output power and pulse rate ranged from 60–500 mJ and 1–15 Hz, respectively. Working at a distance of 12 mm from the lased surface, a handpiece (#2056) with a 0.63 spot size, and energy of 300 mJ with a repetition rate of 6 Hz, and an approximate energy density of 47 J.cm⁻² was used in a focused mode to prepare the cavities under a continuous water spray (5 mL/min).

The prepared blocks were randomly assigned among the 3 restorative material subgroups (Table 1). Restorations were made in 12 blocks, in which one block per subgroup was filled. The restoration sequence was randomly determined, the materials were inserted in accordance with the manufacturers' instructions and light activated with an Optilux 501 device (Demetrom/Kerr, USA) with a mean of 700 mW.cm⁻².

In cavities filled with Ketac-Fil, the Ketac conditioner was applied for 10 seconds, rinsed and dried for 10 seconds. Ketac-Fil was prepared within 20-25 seconds, inserted into the cavity with a centrix injector, protected with a lead strip for 5 minutos, coated with Vitremer Finish Gloss and light activated for 20 seconds to maintain the ionomer water stability. For the Vitremer restoration, the Primer was applied for 30 seconds, dried for 5 seconds and light activated for 20 seconds. Vitremer was prepared within 45 seconds, inserted into the cavity with a centrix injector, light activated for 40 seconds, coated with Vitremer Finish Gloss and light activated for 20 seconds. In cavities filled with composite resin, the 3M Scotch Bond etchant was applied for 15 seconds, rinsed for 10 seconds and air-dried. Two coats of 3M Adper Single Bond 2 were applied, air-dried for 5 seconds and light activated for 10 seconds. The Z250 composite resin was inserted and light activated for 20 seconds.

All restored blocks were stored in 100% humidity for 24 hours and then polished using the Sof-lex (3M ESPE) disk system for 15 seconds with each disk under water-cooling at low speed.

The blocks were individually immersed in 1 mL of deionized distilled water to avoid ionic changes and thermal cycled together for 1000 cycles in water at a temperature ranging between $5 \pm 2 \,^{\circ}$ C and $55 \pm 2 \,^{\circ}$ C, with a dwell time of 2 minutes for each bath and a 15 seconds transfer time between baths⁴.

A uniform area of exposed enamel surrounding the restorations was obtained by covering the remainder of the

dental block with red wax. To simulate in vivo high caries risk conditions, the restored blocks were submitted to a dynamic demineralization/remineralization model, as proposed by Featherstone et al.^{4,5,15}.

This model simultaneously measures the net result of the inhibition of demineralization and the enhancement of remineralization. The demineralization stage uses an acid buffer containing 2 mmol.L⁻¹ Ca, 2 mmol.L⁻¹ PO₄, 0.075 mol.L⁻¹ acetate at pH 4.3. The remineralization solution contains calcium and phosphate at a known degree of saturation, to mimic the remineralizing properties of saliva, and 50 mmol.L⁻¹ KCl, 1.5 mmol.L⁻¹ Ca 0.9 mmol.L⁻¹ PO₄, 20 mmol.L⁻¹ tri-hydroxymethyl-aminomathan buffer at pH 7.0⁵⁻¹⁵.

The blocks were immersed separately in 15 mL of demineralization solution for 6 hours, washed with deionized distilled water, immersed in 15 mL of remineralization solution for 18 hours, washed and immersed in demineralization solution, thereby initiating a new cycle. The pH cycles were conducted for 14 days with 10 daily cycles. On the 6th, 7th, 13th, and 14th days of the cycle, the blocks were kept in the remineralization solution only^{4,5,15}.

After 14 days the wax was removed, the blocks were airdried for 15 seconds and standardized images were obtained from each block using a Nikon D70 digital camera with lens #105. Three calibrated examiners independently and blindly evaluated the images of all the blocks projected in a dark room, at approximately $100 \times$ magnification. The examiners evaluated these specimens scoring the presence and severity of caries-like lesions according to an ordinal scale ranked from 0 to 3, based on visual examination, as described in Figure 1⁴.

A median was obtained from scores given by the 3 examiners for each block. Differences among the medians were analyzed by Kruskal-Wallis non-parametric test at a 95% confidence level and the Dunn test. Examiner calibration was verified by the Kappa test.

RESULTS

The intra and inter-examiner kappa values are showed in Table 2, and may be considered to have good or excellent agreement.

The exploratory values to estimate the effect (medium) and variation (amplitude) and the results of Dunn test are showed in Table 3. The most extensive development of artificial caries lesions was in G3, which was prepared with DB and restored with CR, and showed statistical differences from G1, G2, G4, and G5. The G6 did not differ from G3 or from the other groups. The lowest incidence of artificial caries was observed in G4.

DISCUSSION

By visual examination, the examiners evaluated the presence and severity of caries lesion development around cavities prepared with burs or Er:YAG laser irradiation. Visual inspection is widely used to quantify opacities, fluorosis and white spot lesions resulting from enamel demineralization in laboratory and clinical studies^{4,5,15-18}. Although this method may be considered subjective when compared with other methods, such as microradiography, polarized light microscopy or microhardness testing, visual inspection is simple, facilitates laboratory investigation and allows the total net area to be inspected, providing a general evaluation. It also makes it easy to conduct studies in less time and at lower costs, and presents correlation with other sophisticated methods^{4,16}. Furthermore, the examiners made the diagnoses in a way similar to that of clinical diagnosis, by evaluating the absence or presence of white spot lesions, and quantifying their activity and severity, considering that the opacity of the lesion increases as the mineral content decreases. For this purpose, a four-point ordinal scale was used with the advantage of magnification and standardized room conditions^{4,5,16}.

In the present study the Er:YAG laser used for cavity preparation was not shown to be capable of changing the enamel surface and guaranteeing significantly more acid-resistance to acid challenge, than the bur preparation. The pH cycling model used to create the acid challenge and promote artificial carieslike lesions was similar to the acid challenge found in a patient at high caries risk and showed a correlation with the onset and



Figure 1. Scores used to quantify artificial caries-like lesion development around restorative materials.

Table 2. Kappa	intra	and	inter-exam	iners v	values
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Examiners	1	2	3
1	0.797	-	-
2	0.831	0.733	-
3	0.832	0.812	0.929

progression of caries lesions^{15,19}. This method simulates the demineralization and remineralization phenomena occurring in the oral environment and it has often been recommended for investigating the effects of different substances on dental caries prevention, with the goal of correctly predicting clinical outcomes¹⁵⁻¹⁹.

There is agreement about fluoride released from restorative materials being able to inhibit secondary caries development^{1,5,20-22}. Among the groups in which cavities were prepared with burs, Group G1 restored with the glass ionomer cement showed the least artificial caries development. This result is in agreement with some previous studies that have described the potential of glass ionomer cements to prevent secondary caries, which is well established^{4,5,22}.

Furthermore, some studies have demonstrated that the resinmodified glass ionomer materials, which are hybrid materials, exhibit intermediate properties between their precursors glass ionomer cements and light polymerizable composite resin^{4,5,23}. This result was observed in the present study, as G2 and G1 showed a similar anticariogenic effect, and this effect was also observed among the lased preparations.

Neither the composite resin nor the adhesive system used in the present study contained fluorides in their formulations, so it was observed that all blocks prepared with burs and restored with the composite resin showed artificial caries development, with scores ranging from 2 to 3. This result is in agreement with other studies that demonstrated that Z-250 did not present any cariostatic effect because has no fluoride in its composition^{4,5,16,20,25}.

Chimello et al.²⁵ (2008) revealed that after in situ caries development, the Er:YAG laser did not differ from conventional cavity preparation with regard to enamel microhardness when restored with a composite resin. Moreover, a Polarized Light Microscopy analysis showed no differences, irrespective of the Er:YAG laser parameters, when compared with the conventional bur cavity preparation¹⁶. However, after visual inspection of the specimens by image presentation in a dark room, Chimello et al.¹⁶ (2008) observed that inhibition zone scores showed significant difference among groups, which was ascribed to the control group in which cavities were prepared with diamond burs, and suggested a lower degree of demineralization at the restoration margin of the irradiated samples¹⁶. Although no statistically

 Table 3. Exploratory results of medium scores, median post, range from minimum to maximum scores (min-max), and Dunn test results per group (different letters indicate statistical significant differences at 5% level)

Restorative material	GI		RM		CR	
Cavity preparation	DB	LA	DB	LA	DB	LA
Group	G1	G4	G2	G5	G3	G6
Median	1	1	1	1	3	3
Median post	27.6	24.5	29.3	32.5	58.8	46.0
Min - max	0-3	0-2	0-3	0-3	2-3	0-3
Dunn test	А	А	А	А	В	AB

Glass-ionomer cement (GI), resin-modified glass-ionomer (RM), composite resin (CR), diamond bur (DB), Er:YAG laser (LA).

significant differences were found between the groups restored with composite resin (G3 and G6), all blocks in Group G3 presented caries development (scores 2-3) and the blocks prepared with Er:YAG laser (G6) ranged from 0 to 3. The presence of blocks without caries development in this group suggests some acid-resistance was gained by enamel due to laser preparation, which prevented the artificial caries development. This theory may be strongly reinforced by the absence of differences between the group prepared by Er:YAG laser and restored with composite resin (G6) and the group prepared with burs and restored with glass ionomer cement (G1). Furthermore, from the comparison of score ranges of groups G1 and G4 restored with glass ionomer cement, it can be observed that G1 presented scores from 0 to 3 and G4 showed no advanced active caries-like lesions (score 3), which could also suggest that some acid-resistance may have been promoted by laser preparation.

Additionally, some studies have shown that erbium lasers used with low energy densities may improve enamel acid-resistance,^{7,24} and a clinical trial showed that after six months, cavities prepared with Er,Cr:YSGG presented no secondary caries at the margins of the preparation sites¹².

In a previous study Perito et al.²⁶ (2009) found less caries lesion development around Er:YAG laser-prepared cavities than

around the cavities prepared with diamond burs. However, no synergistic cariostatic effect was observed between the Er:YAG laser and glass-ionomer cement²⁶.

Although some evidence of gain in acid-resistance was suggested, under the experimental conditions, no synergic effect of glass ionomers materials, or a simple improvement in the enamel acid-resistance after Er:YAG cavity preparation were statistically confirmed.

CONCLUSION

In the present study, the Er:YAG laser used for cavity preparation did not show the ability to change the enamel surface and guarantee significantly more acid-resistance against acid challenge than the bur preparation.

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CORRESPONDING AUTHOR

José Augusto Rodrigues

UnG – Universidade Guarulhos, Praça Tereza Cristina, 229, Centro, 07023-070 Guarulhos - SP, Brazil e-mail: jrodrigues@prof.ung.br

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