

Er:YAG laser effect on bovine enamel microhardness after erosive challenge

Efeito do laser Er:YAG na microdureza do esmalte bovino após desafio erosivo

Abstract

Purpose: The aim of this study was to evaluate longitudinal enamel microhardness after treatment with laser and fluoride followed by exposure to an erosive challenge.

Methods: Twenty-four specimens were obtained from bovine enamel (2x3 mm), polished and randomly divided in to four groups (n=6) according to the preventive treatments: G1 – Er:YAG laser; G2 – laser + fluoride; G3 – fluoride; and G4 – fluoride + laser. Half of the enamel surface was covered with nail varnish (control area) and the other half exposed to the preventive treatments and erosive challenge. The laser was irradiated for 10s at 80 mJ/2Hz, and the fluoride gel was applied for 4 min. Each specimen was individually exposed to the soft drink Coca-Cola 4 times/day for 1 minute for a total of 5 days. The specimens were embedded in resin, sectioned in half, flattened and polished. After microhardness analysis, the results were averaged using the Kruskal-Wallis test.

Results: Groups 2 and 4 had decreased enamel demineralization compared to the other groups but did not differ from each other: G1 = -30.95; G2 = 19.62; G3 = -20.5; and G4 = 29.67.

Conclusion: It can be concluded that laser irradiation associated with fluoride application can effectively reduce enamel demineralization.

Key words: Tooth erosion; laser; fluoride

Resumo

Objetivo: O objetivo do estudo foi analisar a microdureza do esmalte bovino, previamente tratado com laser e fluoreto e exposto a desafio erosivo.

Metodologia: Vinte e quatro espécimes de esmalte utilizados. Uma área de 2x3 mm foi delimitada na superfície. Os espécimes foram distribuídos em 4 grupos (n=6): Laser Er:YAG (G1); Laser+Flúor(G2); Flúor (G3) e Flúor+Laser (G4). Metade da superfície do espécime foi isolada com esmalte de unha (área controle) e metade submetida aos tratamentos erosão. O laser foi irradiado por 10s, 80mJ/2Hz; o flúor fosfato acidulado foi aplicado por 4min. Cada espécime foi individualmente exposto a Coca-Cola 4 vezes/dia, durante 1 min, por 5 dias. As amostras foram incluídas em resina, seccionadas ao meio, planificadas e polidas. Posteriormente as amostras foram submetidas ao teste de microdureza Knoop na subsuperfície e os dados foram analisados pelo teste Kruskal-Wallis.

Resultados: A diferença de dureza entre área experimental e controle mostrou os seguintes resultados: G1 = -30,95; G2 = 19,62; G3 = -20,5; G4 = 29,67.

Conclusão: Os espécimes tratados com laser Er:YAG em associação com flúor (G2 e G4) promoveram maior resistência a erosão do que os tratados isoladamente (G1 e G3), os quais não foram eficazes em inibir o processo erosivo.

Palavras-chave: Erosão dentária; laser; fluoretos

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Introduction

Dental erosion is a multifactorial disease and is defined as the irreversible loss of tooth surface due to the interaction of chemical, biological and behavioral factors and the involvement of microorganisms (1).

The chemical process is due to acids with pH values less than 4.5 that cause the dissolution of hydroxyapatite and fluorapatite in the dental enamel (2). Biological factors, such as saliva flow, buffer capacity, saliva composition and dental anatomy, are related to the pathogenesis of dental erosion (3). In addition, behavioral factors, such as anorexia nervosa, bulimia, gastro-esophageal reflux, and eating habits that include acidic foods and drinks (soft drinks, fruit juices, teas and sports drinks), can also trigger the process of dental erosion (1).

The acid erosive effect of diet on the dental tissue can be influenced by a number of factors, such as the pH, pK, acidity, temperature, concentration and potential chelation. In addition to the frequency, time of ingestion, and length of time in the mouth, changes in tooth structure, especially in relation to fluoride content, are considered important to understand dental erosion caused by acids in the diet (4).

When the erosion is diagnosed, methods to control dental erosion and/or restorative procedures to restore erosion lesions that compromise the function, appearance esthetic and/or integrity of the dental structure are important to develop are necessary. The first control method is attempting to remove the acid source causing the erosion process and reduce the consumption of acidic foods and beverages. These patients should be referred for medical care (5).

The main method used for injury prevention of enamel erosion is the topical application of fluoride. The possible role of fluorine in the prevention of dental erosion may be to strengthen the tooth surface and increase resistance to acid (6). Although many authors (7,8) report that fluoride acts to protect the teeth against erosion, other authors (2,9) say that it is not effective. So, we have sought new ways to control the activity of erosive acids, such as using a laser (10,11).

It has been demonstrated that lasers can significantly enhance the acid resistance of enamel by changing the crystallinity, solubility, and permeability of the enamel (12). One hypothesis regarding the benefits to prevention is that laser irradiation causes the loss of water and carbonate and decomposes organic matter, which leads to the formation of micro-spaces that are filled by the product of this decomposition. Therefore, sealing reduces the degree of demineralization (13). Thus, different lasers have been used to control dental erosion, and its association with fluorine has also shown good results (10,14-16). Of the different wavelengths, the Er: YAG laser has been particularly studied (14).

Thus, the objective of this study was to evaluate the subsurface microhardness of enamel that has been previously treated with Er: YAG laser and fluoride and exposed to an erosive challenge.

Methodology

The experimental units consisted of 24 enamel slabs cut from the buccal surface of bovine incisors. The specimens were coated with an acid-resistant nail varnish (Colorama-Maybelline, Procosa Produtos de Beleza Ltda., São Paulo, SP, Brazil. and wax, except for their outer surface (2x3 mm), which was divided in half. One part of this surface was the control area (no treatment) and the other half was subjected to the proposed treatment and subsequent erosive challenge.

The specimens were randomly assigned to 4 groups (n=6) according to the following surface treatments: Er:YAG laser (G1); Er:YAG followed by fluoride – 1.23% acidulated fluoride gel (DFL S.A., Rio de Janeiro, RJ, Brazil) (G2); Fluoride (G3); and Fluoride followed by Er:YAG (G4). The equipment used for the surface treatment with the Er:YAG laser was the Kavo Key Laser II (Kavo Co, Biberach, Germany), emitted at a wavelength of 2.940 μm . The specimens were irradiated with 80 mJ of energy in the noncontact mode at 2 Hz for 10s, and the tip was positioned 4.0 mm from the enamel surface (unfocused mode) (17). The 1.23% acidulated fluoride gel was applied for 4 min on the exposed enamel area and then removed with gauze.

After the treatments, the erosive challenge was performed 4 times a day for 5 days. During the cycles, the specimens were individually kept in Coca-Cola® at 4°C (pH 2.5) and stirred for 2 min. After each cycle, the specimens were rinsed with deionized water and then soaked in artificial saliva at $37 \pm 0.5^\circ\text{C}$ until the next erosive challenge, which is similar to the protocol described by Amaechi et al. (1999) (18). The artificial saliva was changed daily.

Throughout the erosive challenge, the specimens were embedded in polyester resin (Milflex Indústria Química Ltda., São Bernardo do Campo, SP, Brazil) and cut in half so that both areas were in the section (control area/treated area). In the sequence, the sections were flattened while water-cooling on a polishing machine with Al_2O_3 papers, and then polished with alumina suspension on a felt disc. After finishing and polishing, Knoop microhardness measurements were performed using an indentation tester (Microhardness Tester HMV-2, Shimadzu Corporation, Kyoto, Japan) with a 25 g load applied for 10 seconds. Three indentations were made in both areas of the section (control and treated) at different depths (20, 40 and 60 μm from the enamel margin).

The statistical evaluation of the data was based on the overall average of each specimen (control and experimental area), and the differences between the treated and control areas were analyzed. Because the data did not show a normal distribution, a nonparametric Kruskal Wallis test was used for the analyses.

Results

The analysis of the data found significant differences among the treatments. Groups 1 and 3 showed a decrease in the microhardness, which was significantly different from groups 2 and 4. Groups 2 and 4 showed increased

microhardness in the treated area compared to the control area. Therefore, both groups demonstrated a surface that was less demineralized than the control area (Table 1).

Table 1. Difference of microhardness - Median (KHN) of studied groups.

Group	Difference of microhardness – Median (KHN)*
G1	-30.95 a
G2	19.62 b
G3	-20.50 a
G4	29.67 a

* Different letters indicate statistically significant difference of microhardness change.

Discussion

The effects of fluoride in the prevention of caries and as a vehicle for controlling the progression of dental erosion are known (19). Its action is mainly attributed to precipitation of CaF₂ on the tooth surface, which reduces the action of acid on the surface and consequently acts to partially reduce enamel erosion (20).

Lasers have also been used to prevent the enamel demineralization caused by dental caries and have shown good results (17,21). The Er:YAG laser has been shown to reduce or prevent the demineralization of tooth enamel (14). Furthermore, when associated with fluoride, it leads to a reduction in mineral surface loss (22).

Other wavelengths (488, 514.5, 532, 633, 670, 830 and 1064 nm) associated with fluoride have shown an increased resistance to dental enamel erosive challenge compared to the control group, which was not irradiated or exposed to fluoride (11).

In this study, surfaces treated with the combination of Er:YAG laser and a topical fluoride application showed a higher hardness value than the control surface, and significant protection was provided to the enamel compared to the individual treatments. Therefore, there was a protective effect associated with laser fluoride therapy, which confirms the work of Steiner-Oliveira et al. (10), who observed a minor loss of enamel and dentin after CO₂ laser treatment

and topical application of fluoride in teeth subjected to erosive challenge. However, according to these authors, this effect was not synergistic.

The present study showed a tendency for higher, but not statistically significant, hardness values in the group treated with fluoride prior to irradiation, which is similar to the findings from a previous study (23). This comparison, however, should not be considered in its entirety because different experimental models were used in the previous study. However, this work did not show the protective effect of laser or fluoride individually in the prevention of dental erosion, which can be explained by the aggressive nature of the erosion cycle used in these experiments. As a result, the benefits provided by laser treatment or fluoride would be lower with the proposed erosive challenge.

The CaF₂ deposited on the tooth surface when fluoride is applied topically is responsible for the beneficial effect towards the prevention of erosive lesions (20). It is believed that the temperature variations induced by laser irradiation are able to promote chemical modifications and the formation of organic compounds in morphological and/or inorganic substrates, which make enamel less likely to demineralize (24). Despite the favorable result, little is known about the synergy of the laser and fluoride. The laser may provide structural alterations that favor the absorption of fluoride from the tissue, or may be conducive to the formation of compounds that are more stable despite a high risk of erosion. Despite the favorable results with the combination of the Er:YAG laser and fluoride, more studies are necessary to understand the action of this association on the tissue and determine the best parameters for clinical applications to avoid causing major changes in the substrate.

Conclusions

In this *in vitro* study, Er:YAG laser irradiation and acidulated fluoride gel promoted inhibition of the erosive process, while the isolated treatments (Laser or Fluoride) did not effectively inhibit the process. Therefore, the concomitant use of the laser and fluoride represents a new and effective procedure to reduce the solubility of enamel in cases of dental erosion.

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