Original Article

Effects of Er:YAG Laser Irradiation and Topical Fluoride Application on Inhibition of Enamel Demineralization

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ABSTRACT

Objective: The aim of this study was to evaluate the effect of the Er:YAG laser and acidulated phosphate fluoride application on enamel solubility around orthodontic brackets via atomic absorption spectrometry.

Materials and Methods: Twenty freshly extracted upper premolar teeth were divided into 2 halves. Each half was covered with a nail varnish, excluding 1 mm (width) of space around each side of the bracket base. Orthodontic brackets were bonded in the center of the isolated area. Four groups were generated. The first group was the control group, and no treatment was performed. In group II (F), only fluoride was applied. Groups III and IV were laser and fluoride combination groups. In Group III (LF), fluoride was applied to the laser-irradiated surface, whereas laser irradiation was performed on the fluoridated enamel surface in Group IV (FL group). Afterwards, samples were demineralized in an acidic solution for 96 hours, and calcium assessment was performed using atomic absorption spectrometry. Comparisons of the calcium ion release values of the groups were performed with Kruskal-Wallis and Tukey honestly significant difference post hoc tests. The statistical significance level was set at *p* < 0.05. **Results:** The least amount of calcium release was observed in the LF group (median, 112.7 ppm), while the parameter of the control group was the highest (median, 217.9 ppm). The difference was recorded between the control and FL groups.

Conclusion: Laser treatment followed by topical application of acidulated phosphate fluoride gel resulted in the lowest calcium dissolution from the enamel surface. (*Turkish J Orthod* 2013;26:30–35)

KEY WORDS: Demineralization, Fluoride, Laser

INTRODUCTION

White spot lesion (WSL) is one of the most challenging complications of orthodontic treatment and results from inadequate oral hygiene and prolonged plaque accumulation around the brackets. In this regard, several agents or methods have been used to prevent or reduce WSL formation.

Fluoride is stated to be the most important agent to prevent decalcification and inhibit lesion progression.¹ Also, fluoride ions in dental plaque induce remineralization of enamel with the formation of fluorapatite. The integration of fluoride to the crystalline lattice of dental enamel results in a structure that is more resistant to dissolution.² Fluoride can be used topically (fluoridated tooth paste, mouth rinse, gel, and varnish) or incorporated into cements, elastomeric modules, or chains.³

Sognnaes and Stern⁴ were the first to advocate the potential of lasers to decrease enamel solubility. Since that study, several studies have been conducted with argon, Nd:YAG, Er:YAG, Er,Cr:YSSG, and carbon dioxide lasers.⁵ The most accepted hypothesis about the ideal way to enhance enamel

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resistance to acid is heating the enamel surface to the range of 40–1000°C, reducing the amount of bound carbonate and, thus, decreasing enamel solubility.⁶ In recent years, numerous studies have suggested that the acid resistance of dental enamel could be increased with laser irradiation.^{7–14} However, there is scarce information about the effect of Er:YAG laser on decreasing enamel solubility. Cecchini *et al.*⁵ used an Er:YAG laser with different parameters of irradiation and reported that lower energies (subablative dose) decreased enamel solubility. The authors suggested that the Er:YAG laser application is a very promising treatment for acid resistance of enamel.

Synergetic effects of fluoride application in addition to laser treatment have been also investigated. It has been demonstrated that a combination of acidulated phosphate fluoride and Er,Cr:YSGG laser treatment resulted in lower enamel demineralization compared to either laser or fluoride treatments alone.¹⁵ Fox *et al.*¹⁶ used Er,Cr:YSGG and 3 different chemical agents, one of which was fluoride. They found that the enamel dissolution was almost totally inhibited with the use of these combinations. Bevilácqua *et al.*¹⁷ evaluated the effects of topical fluoride and laser irradiation on enamel demineralization. They reported that topical flouride application after laser treatment is a promising treatment for increasing enamel acid resistance.

The purpose of this study was to quantitatively evaluate the effectiveness of the Er:YAG laser combined with the use of fluoride gel on prevention of enamel solubility around the brackets via the atomic absorption spectrometry method.

MATERIALS AND METHOD

For this study, 20 freshly extracted upper premolar teeth were cleaned with pumice and stored in 0.1% thymol solution for no more than 1 week. Teeth with caries, cracks, or enamel irregularity were excluded. The crowns were separated from the roots, and the separated crown was cut along the whole length of the crown by a low-speed diamond saw. In this way, 2 sections were obtained from 1 tooth in order to use both buccal and lingual surfaces. For each patient, 4 sections were obtained by dividing 2 premolars (right and left upper premolars) into 2 parts.

In the center of the buccal/palatal side of each section, a surface area was isolated using an acidresistant nail varnish. The sections were covered with a nail varnish, excluding the enamel 1 mm (width) around each side of the bracket base. Samples were kept in a humid environment until the time of the experiment.

The buccal/lingual surfaces of each tooth were acid etched with 37% orthophosphoric acid (3M Dental Products, St Paul MN, USA) for 15 seconds and rinsed and dried until a frosty, white appearance was obtained. Transbond XT (3M Unitek, Monrovia CA, USA) primer was applied as a thin, uniform coat. Stainless-steel premolar brackets (Dynalock; 3M Unitek) were bonded to teeth. Transbond XT composite (3M Unitek, Pucheim, Germany) was applied to the base of the bracket; the bracket was placed in the middle of the exposed surface and pressed firmly against the tooth surface. The excess adhesive was removed, and the adhesive was light cured for 20 seconds from the mesial and distal directions with a quartz tungsten halogen light-curing unit (Hilux; Benlioglu Dental, Ankara, Turkey).

Each sample obtained from the same individual was allocated to 1 of 4 groups. In this way, the sections of a patient were equally allocated to 4 groups (10 samples in each group). Groups were as follows:

- Group I (control): this group was considered as the control of the other groups. No enamel treatment was performed.
- Group II (F group): enamel was treated with 1.23% APF (Sultan, Topex NJ, USA) and left undisturbed for 4 minutes, then washed gently with distilled water.
- Groups III and IV were laser and fluoride combination groups. In Group III (LF group), fluoride was applied to the laser-irradiated surface, whereas laser irradiation was performed on the fluoridated enamel surface in Group IV (FL group).

Laser Irradiation

Laser irradiation was performed using an Er:YAG laser used in the study (Smart 2940D Plus; DEKA Laser, Firenze, Italy) with a wavelength of 2.94 μ m. The irradiations were performed with a contact handpiece (N327A2), and the parameters used were 100 mJ/pulse, 10 Hz, 1 W. The beam diameter at the focal area was 1.0 mm. The energy density was calculated as 12.73 J/cm². The surface was cooled with water spray at a rate of 5 mL/min during irradiation.

Demineralization Process

After enamel treatments, control and treatment samples were subjected to artificial demineralization via an acidic solution (pH 4.6) for 96 hours. The solution was composed of 1.5 mM CaCl₂, 0.9 mM KH₂PO₄, and 50 mM acetic acid (all 3 from Merck, Darmstadt, Germany). Samples were immersed in experimental tubes containing 2.5 mL acidic solution; each polypropylene tube contained 1 sample. Samples were stored in a stove at 37°C. After 96 hours, the teeth were removed from the solution, and the solutions were evaluated for Ca ion concentration with atomic absorption spectrometry.

Atomic Absorption Spectrometry

Calcium assessment was performed using atomic absorption spectroscopy (ATI Unicam 939; Unicam Atomic Absorption, Cambridge, UK). Measurements were done under acetylene flame (3.8 L/min) with the atomic emission method. In order to prevent the chemical interaction resulting from phosphate ion, 0.1% (w/v) La(NO₃)₂ was used. Calcium standards of 0.5, 1.0, 2.0, 3.0, 5.0, and 10.0 mg/L were prepared, to include 0.1% (w/v) La(NO₃)₂. Using emission values recorded in atomic absorption spectrometer, mg/L Ca⁺² concentration calibration equation traced (y= 8.5838x + 10.045, y=emisyon, x=mg/L Ca⁺²) (18-20).

Statistical Analysis

All statistical analyses were performed with the Sigma Stat software (Sigma Stat 3.5; IBM SPSS, Inc, Armonk NY, USA). The normality test of Shapiro-Wilks and the Levene variance homogeneity test were applied to the data. The data were normally distributed, and there was not homogeneity of variance among the groups. Thus, the statistical evaluation of Ca ion release values among test groups was performed using nonparametric tests. Descriptive statistics, including the median and the 25% and 75% guartiles were calculated for the 4 groups. Comparisons of means of Ca ion release values were made with the Kruskal-Wallis test. Post hoc multiple comparisons were done with the Tukey honestly significant difference test. The statistical significance level was set at p < 0.05.

RESULTS

The descriptive statistics and intergroup comparisons for calcium contents in the demineralization solution were given in Table 1. As for the calcium contents in demineralization solution (ppm), the LF group showed the lowest values (median, 112.7 ppm), whereas the highest values were recorded for the control group (median, 217.9 ppm). The differences between the control and F groups and between the control and LF groups were statistically significant. No difference was recorded between the control and FL groups. The difference between the LF and FL groups was also found to be statistically significant.

DISCUSSION

While many studies have shown the effects of laser treatment on enamel acid resistance,^{7–14} there are still contradictions regarding the synergetic effects of ER:YAG laser treatment and acidulated phosphate fluoride application. In this study, the possible interaction between these two treatments was investigated with different schedules of laser and fluoride application.

Er:YAG laser is an important tool in dentistry and can be used for several procedures. Its wavelength is coincident with the main absorption band of water and is absorbed by enamel.^{21–23} Enamel consists of 85% mineral and 15% free water, protein, and lipid.²⁴ Absorbed laser energy in the enamel converts to heat, which boils water and forms highpressure steam. When this pressure exceeds the strength of the tooth, the ablation process begins and the water evaporates explosively with tooth particles. Because of this process, the Er:YAG laserablated surface becomes a flaky structure with an irregularly serrated and microfissured morphology.²⁵ According to Kim et al.,25 acid may reach the subsurface via these microfissures, which act as open channels. On the other hand, this surface morphology may be vulnerable to acid attack and mineral loss.26

In the literature, a wide dose range draws attention to increasing the acid resistance of enamel by laser application. Both ablative and subablative doses have been tested to try to decrease the acid solubility of enamel. The ablation threshold of the Er:YAG laser is also a controversial topic: it varies between 7 and 18.6 J/cm² in the literature.^{27–29} Fried *et al.*²⁷ preferred doses at the ablation threshold, suggesting that a minimum of 300°C of temperature increase would be required to create morphological changes like evaporation of water and loss of carbonate from enamel in order to increase acid resistance of the tissue. In the study of Bevilácqua *et*

	25%	Median	75%	p	Sig.
Group I (control)	176	217.9	340.8	0 < 0.001	А
Group II (fluoride)	123.6	131.5	171.9		В
Group III (laser+flouride)	93.8	112.7	128.9		В
Group IV (flouride+laser)	159.1	170	192.4		AC

Table 1. Descriptive statistics and statistical comparison of groups for Ca ion release

Sig.; significance

al.¹⁷ the lowest calcium levels were observed in the group with the highest energy density of an ablative dose (31.84 J/cm²). On the other hand, Yu et al.³⁰ and Cecchini et al.5 stated that higher energies at the enamel surface could induce more prominent chemical alterations, though this would not always mean an increase in acid resistance. Holcomb and Young³¹ reported that enamel proteins decomposed at about 350-400°C. It has been reported that decomposition of organic matrix might lead to a decrease in the organic blocking effect,³² which could enhance mineral loss and lesion progress.33 Liu et al.33 observed such a finding with a dose of 300 mJ and determined the ideal dose to be 100-200 mJ. Moreover, the temperature increase in pulpal tissue caused by laser irradiation with ablative doses should be considered. From this point of view, White and Goodish³⁴ determined the safe limits for pulpal health to be 1 W and 10 Hz. By considering the whole, we preferred a subablative dose, 100 mJ per pulse (12.73 J/cm²) in the present study, and obtained positive results in accordance with the results of Hsu et al.,³² Morioka et al.,³⁵ and Fried et al.,²⁷ who presented marked caries inhibition (40%) using subablative laser parameters.

Another aspect affecting the generated result is water cooling during irradiation. Water cooling has been reported to maximize the tendency for ablative processes via increasing the interaction between laser light and tissue. Laser application for caries prevention was found to be more effective without water cooling.^{29,33,35} Similarly, Hossain et al.³⁶ revealed that irradiation without cooling was more intense than it was with water cooling. This claim depends on the belief that the preventive effect emerges via chemical alterations in enamel caused by temperature increase. However, detrimental thermal damages such as cracking, carbonization and melting of the enamel, and inflammation or necrosis of pulpal tissue should be considered. Consequently, we preferred a subablative dose with water cooling.

The relationship between laser treatment and fluoride uptake has been investigated previously. Bevilácqua *et al.*¹⁷ and Huang *et al.*³⁷ reported that fluoride incorporation was increased with the laser treatment. On the other hand, Steiner-Oliveira *et al.*³⁸ observed no significant synergistic interaction between fluoride-laser combined therapies. Although different wavelengths were used, the main difference among these studies was the schedule of laser treatment. In light of this information, the effects of laser treatment performed prior to (LF group) and following fluoride application (FL group) were evaluated in the present study.

With the beforehand laser application in the LF group, calcium loss of enamel was significantly decreased compared to the other groups. This indicates the usefulness of the laser treatment in addition to acidulated phosphate fluoride gel in creating a synergetic effect. The laser-dependent crystallographic changes on the enamel surface might facilitate the integration of fluoride to the structure. Deposition of fluoride to the laser-created microspaces has been a possible explanation.^{26,39}

On the other hand, when laser treatment was performed after fluoride application, the preventive/ protective effect was found to be significantly decreased. This may depend on the fact that laser irradiation was performed under water cooling, which may cause the remnant fluoride to be sluiced. In this way, the effectiveness of fluoride later on might be reduced. Additionally, laser application might cause disintegration of the newly formed fluorapatite, resulting in a lower increase in the acid resistance of enamel. Hence, laser irradiation following fluoride application may be unsatisfactory.

Fluoride application without laser irradiation was also found to be effective in reducing Ca dissolution. The difference between the LF and F groups was not statistically significant. But the lowest Ca ion concentrations were obtained in the LF group. Thus, it may be that laser treatment followed by topical fluoride application is more effective than fluoride application alone.

CONCLUSION

Within the limitations of this study:

- Laser treatment followed by topical application of acidulated phosphate fluoride gel resulted in the lowest Ca ion dissolution in the demineralization solution. No statistically significant difference was found between the F and LF groups.
- No statistically significant differences were found between the control and FL groups regarding Ca dissolution.

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