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THE EFFECT OF TWO TYPES OF ERBIUM LASER AND LIGHT CURE FLUORIDE VARNISH ON ENAMEL DEMINERALIZATION AND SURFACE MICROHARDNESS AROUND METAL ORTHODONTIC BRACKETS: AN IN-VITRO STUDY

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KEYWORDS

Demineralization, Er:YAG laser,
Er Cr: YSGG laser,
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White spot lesion.

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ABSTRACT

Introduction: Development of white spot lesions on enamel is a significant and common problem during the fixed orthodontic treatment. **Aim:** This study was aimed to assess the effect of two types of erbium lasers (Er: YAG) laser, (Er, Cr: YSGG) laser and light cure fluoride on the enamel demineralization and surface microhardness around metal orthodontic brackets. **Material and Methods:** In vitro study, 90 freshly extracted human premolars were randomly allocated to six groups (n =15) of control (neither fluoride nor laser was used), light cure fluoride varnish (Clinpro XT) on the enamel surfaces, Er: YAG laser (2.94 μm Er: YAG irradiation of the teeth), Er: YAG laser then varnish™ XT, Er Cr:YSGG laser (2.78 μm Er Cr: YSGG irradiation of the teeth), Er Cr: YSGG laser then varnish ™ XT. PH cycling for 14 days through a daily procedure of demineralization and remineralization for 6 h and 18 h, respectively. Microhardness values of enamel were evaluated with Vickers test. Two sample of each group was prepared for SEM (scanning electron microscopy) and the data from the remaining teeth were analyzed with F-test (ANOVA). **Results:** Microhardness mean values from high to low were as follow: varnish ™ XT, Er Cr: YSGG laser then varnish™ XT, Er:YAG laser then varnish ™ XT, control, Er Cr: YSGG laser, Er :YAG laser. **Conclusion:** Fluoride varnish alone showed the best preventive measure against enamel demineralization. There was no different between the two types of erbium laser on the effect of enamel surface microhardness.

INTRODUCTION

Demineralization or white spot lesion (WSL) development in the enamel in association with orthodontic treatment with fixed appliances remains a well-known clinical complication for dental specialists(1).

The WSL has also been defined as “subsurface enamel porosity from carious demineralization” that presents itself as a milky white opacity when located on smooth surfaces(2). The increased risk of developing white spot lesions during orthodontic treatment is due to an over accumulation of plaque around the brackets; this is partially attributable to patients’ inability or failure to maintain adequate oral hygiene(3).

Enamel crystal dissolution begins with subsurface demineralization, creating pores between the enamel rods. The resultant alteration of the refractive index in the affected area and a consequence of both surface roughness and loss of surface shine and alterations in internal reflection, all resulting in greater visual enamel opacity, as porous enamel scatters more light than sound enamel(4).

Fluoride plays an important role in preventing demineralization during orthodontic treatment. It joins the crystalline structure of hydroxy apatite, it creates fluoride hydroxy apatite that is less soluble and more resistant to acidic attack, it increases the remineralization process and has antibacterial effects(5).

The possibility of increasing the acid resistance of enamel after laser irradiation was first reported with a ruby laser. The most common lasers employed for caries prevention on enamel are Nd: YAG; CO2; Er:YAG; Er,Cr:YSGG ; and argon(6). So, this study was designed to evaluate the effect of two types of erbium lasers (Er: YAG) laser, (Er, Cr: YSGG) laser and light cure fluoride on the enamel demineralization and surface microhardness around metal orthodontic brackets.

MATERIALS AND METHODS

Tooth selection and sample preparation

A total of 90 human extracted premolar teeth with no caries, defects or hypoplastic enamel, were collected and stored in artificial saliva solution. Scaling was done in order to removing calculus or tissue remnant. All the teeth were mounted in epoxy resin blocks (Acrostone cold cure cross-linked, Egypt.) using specially designed cylindrical teflon mold for holding the epoxy resin and tooth inside to form 90 blocks to facilitate the use during laser irradiation and fluoride application. The acrylic bases are differentiated by color coding into blue, red, green, yellow, pink and transparent.

Bonding of Brackets

At the time of bonding, the enamel surface was polished at low speed with a mixture of non-fluorinated paste and a polishing brush then the teeth were rinsed for 30 seconds and dried for 10 seconds with moisture-free air spray.

The enamel was etched by 37% phosphoric acid (Spident, Korea), rinsed with water and dried with oil free air for 15, 30 and 20 seconds, respectively, in order to obtain the appearance of a frosty white surface. Orthodontic premolar metal brackets (Mini Diamond® Twin,Ormco) were bonded to premolars with the non-fluoride releasing composite resin (Grengloo, Ormco). Manufacturer’s instructions were followed precisely in every stage. Brackets was placed in the correct position. The composite resin was cured for 20 seconds with an LED light-curing unit (Dental led light curing unit, Foshan Jerry Medical Apparatus Co., China.).

Then, the samples were randomly assigned to the following 6 experimental groups (15 teeth in each group):

Group 1 (control): The teeth received no additional enamel surface treatment.

Group 2 (varnish ™ XT): the enamel surfaces were cleaned, rinsed and dried around orthodontic bracket using dry air for 10 seconds. Thirty- seven % phosphoric acid etching was applied for 30 seconds then rinsed with water for 30 seconds and air dried for 10 seconds with a moisture free-air spray. The clicker dispenser was clicked once to dispense equal amounts of the two pastes and mixing with small spatula on paper pad for 10-15 seconds. The paste was applied on the enamel surface using micro brush applicator then light cure for 20 seconds.

Group 3 (Er:YAG laser): Er:YAG laser (Light Walker® AT, Fotona Inc., USA) was operated at a wave length of 2.94 μm with a cylindrical tip of a

1.3 mm diameter and 8mm long. The energy output was 80 mJ per pulse, and a pulse duration of 300μs and pulse frequency of 2 Hz were used. The laser beam was applied for 20 Sec in non-contact, focused mode. The hand piece (H14) was held perpendicular to enamel surface moving slowly horizontally and vertically for homogenous irradiation in a scanning style with 60% air and 80% water cooling system around metal orthodontic bracket.

Group 4 (Er:YAG laser then varnish™ XT): Enamel was treated with Er: YAG laser with the same parameters as used in Group 3 (Er: YAG laser) then light cure fluoride was applied as mentioned before in Group 2 (varnish ™ XT).

Group 5 (Er, Cr: YSGG laser): Er, Cr: YSGG laser was operated at a wave length of 2.78 μm with a MZ10 tip of a diameter of 1000mm and 6mm long in non-contact focused mode. The power of 0.75 w, and a pulse duration of 60μs and pulse frequency of 20 Hz were used. The laser beam was applied for 20 Sec. The gold hand piece was held perpendicular to enamel surface moving slowly horizontally and vertically for homogenous irradiation in a scanning style with 60% air and 80% water cooling system around metal orthodontic bracket.

Group 6 (Er, Cr: YSGG laser then varnish™ XT)

Enamel was treated with Er, Cr: YSGG with the same parameters as used in Group 5 (Er, Cr: YSGG laser) then light cure fluoride varnish was applied as mentioned in Group 2 (Varnish ™ XT).

PH cycling phase

The PH-cycling model used in this study was based on the one described by Yoshaskam Agnihotri(7). After treatment the specimens were placed in a demineralizing solution for six hours then rinsed with distilled water and placed in remineralizing solution for 18 hours for 14 days. The demineralizing solution contained (2.2 mM $CaCl_2$, 2.2 mM KH_2PO_4, 0.05M acetic acid having pH adjusted to 4.4 and 1 M KOH). The remineralizing solution contained (1.5 mM $CaCl_2$, 0.9 mM NaH_2PO_4, 0.15 M KCL and had a pH of 7.0. both of those solutions were Prepared by inorganic chemistry lab, Chemistry Department, Faculty of Science Suez Canal University.

Scanning electron microscope:

For the SEM analysis, the samples were coated with 40 nm to 60 nm of gold using a sputter coater (Spi-module sputter coater, USA).and then observed in the electron microscope (JSM-6510LV, JEOL, Japan) at the magnification of 5000X.

Enamel surface microhardness

For the purpose of testing the enamel microhardness of all the samples, Vickers hardness testing machine (Wilson® hardness – Buehler, Germany) was used.

In the Vickers test, the (100 gram) load was applied smoothly, without impact, forcing the indenter into the test specimen. The indenter was held in place for (10) seconds. The physical quality of the indenter and the accuracy of the applied load was controlled in order to get the correct results. After the load was removed, the indentation was focused with the magnifying eye piece and the two impression diagonals were measured, usually to the nearest 0.1-µm with a filar micrometer, and averaged.

Statistical analysis

All data was calculated, tabulated and statistically analyzed using suitable statistical tests. The IBM SPSS software program version 26.0 (Armonk, NY: IBM Corp) was used to examine the data. Range (minimum and maximum), mean, standard deviation, and median were used to characterize quantitative data. The significance of the acquired results was assessed at a 5% level. The tests that were utilized were Student t-test to compare between two groups. For normally distributed quantitative variables, the F-test (ANOVA) was used to compare more than two groups, whereas the Post Hoc test (Tukey) was used for pairwise comparisons.

RESULTS

1. Assessment of surface microhardness between all the six groups

As presented in Table (1), Comparison between studied groups according to Vickers's microhardness. The highest microhardness value was exhibited by Group 2 followed by Group 6, Group 4 and Group 1, Group 5, Group 3. The least microhardness value was observed in Group 3. Level of significance of mean microhardness within groups and between groups was analyzed using one-way ANOVA test. There was a statistically significant difference between group 1 and 2 (p1=0.004*). There was a statistically insignificant difference between group 1 and 3 (p2=1.000). There was a statistically insignificant difference between group 1 and 4 (p3=0.967). There was a statistically significant difference between group 1 and 5 (p4=0.015*). There was a statistically significant difference between group 1 and 6 (p5=0.001*).

It was cleared that there was no a statistically significant difference between the two type of erbium laser and the combination between laser and fluoride increasing the enamel surface microhardness.

2. Scanning electron microscope:

SEM evaluation of enamel surface of Group 1 revealed circumferentially arranged enamel rods filled with inter rod material producing a typical keyhole appearance (**Fig. 1A**).

Evaluation of Group 2 showed streaks of particle deposition of less than 1 μm in size on the enamel surface. Areas of slight cracks were also visible near the bracket tooth interface (**Fig. 1B**).

Group 3 showed uneven melting of enamel rods and irregularities. Enamel has no smooth surface (**Fig. 1C**).

Evaluation of Group 4 demonstrates a relatively smoother and more homogeneous surface with cracks and granular particles (**Fig. 1D**).

The SEM evaluation of Group 5 revealed a smooth, homogenous enamel surface with well coalesced enamel rods. Porous structure of enamel was lost giving rise to a smooth surface (**Fig. 1E**).

Evaluation of Group 6 showed smoother and more homogeneous enamel surface with granular particles(**Fig.1F**).

Table (1) *Comparison between studied groups according to Vickers's microhardness.*

	Group 1	Group 2	Group 3	Group 4	Group 5	Group 6	p
Vickers's microhardness	266.61 ± 32.28	315.19± 35.74	221.69± 42.75	276.69± 17.58	224.07± 29.23	277.76± 30.35	<0.001*
P0	*P1=0.004*, P2=1.000, P3=0.967,P4=0.015*, P5=0.001*, P6=1.000, P7=0.043*, P8=0.001*, P9=0.001*, P10=0.001*, P11=1.000, P12=0.001*, P13=0.001*, P14=1.000, P15= 0.001**						

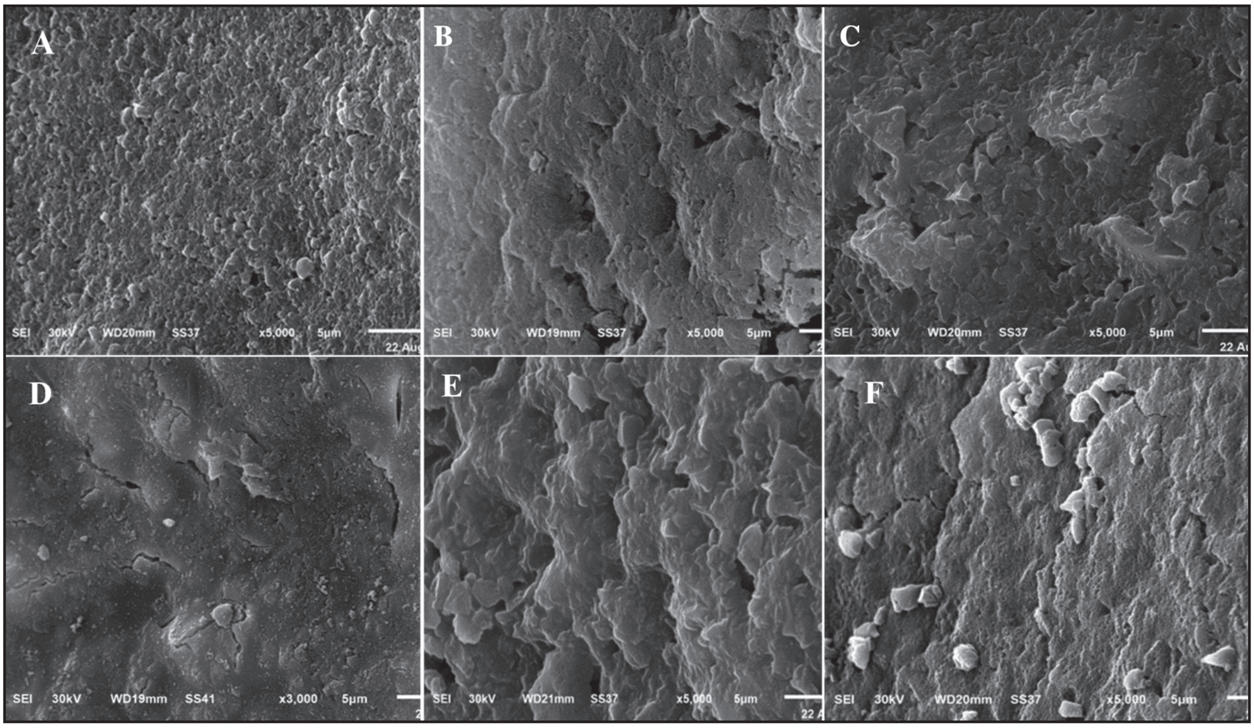

Fig. (1) **(A)** SEM showed circumferentially arranged enamel rods filled with inter rod material. **(B)** SEM showed streaks of particle deposition of less than 1 μm in size on the enamel surface. **(C)** SEM showed uneven melting of enamel rods and irregularities. **(D)** SEM showed a relatively smoother and more homogeneous surface with cracks and granular particles. **(E)** SEM revealed a smooth, homogenous enamel surface with well coalesced enamel rods. **(F)** SEM showed smoother and more homogeneous enamel surface with granular particles.

DISCUSSION

Orthodontic treatment has many advantages such as esthetic improvement, function and self-estimation enhancement. Orthodontic appliances make plaque removal and oral hygiene more difficult and so, increase the risk of white spot lesions. Clinically visible white spot lesions are observed in patients who received orthodontic treatment (8).

According to *Gorton and Featherstone* (9), approximately 50% of patients present clinically visible white spot lesions during treatment for approximately 2 years. The use of fluoride is one of the most studied, known and effective methods to prevent dental caries(10). Much of the success attributed to fluoride is due to its capacity of reversing the beginning and progression of caries(11).

The application of erbium laser on the enamel surface has been studied with great interest since the 1988s. Studies show that this type of laser causes structural and ultrastructural changes in enamel(12). There are several explanations about how reactivity occurs in the enamel treated with erbium laser. One explanation is that the lower permeability of enamel results from the fusion of microparticles on its surface(13). Another explanation is that the relation between decreased permeability with melting, fusion and recrystallization of the enamel particles creates a barrier on the tooth surface(14).

The results of this study showed that light curable fluoride varnish (clinpro XT) was effective to increase enamel microhardness. These result support previous studies that concluded that clinpro XT is more effectieve in preventing enamel demineralization(15, 16).

The result of the present study showed a significant difference between Er Cr: YSGG laser and synergism of Er Cr: YSGG laser with light curable fluoride when compared with control group.

These result of this study was in agreement of *de Freitas et al*(17) as it was studied the effect of different parameters of Er Cr: YSGG on enamel mineral loss in a simulated caries model. As it was concluded that low energy densities could produce a cariostatic potential. Another study be compatible with the present study as it was evaluate the effect of the Er, Cr:YSGG laser and fluoride application on the acid resistance of enamel adjacent to orthodontic brackets. It was concluded that Combination of Er, Cr:YSGG laser with fluoride and fluoride alone decreased enamel solubility significantly more than laser alone(18).

The result of the present study showed insignificant difference between Er: YAG laser and synergism of Er: YAG laser with light curable fluoride when compared with control group. The result of the present study was in agreement with *Ulkur et al*(19) whereas Er:YAG laser irradiation did not show a remarkable effect on a Vickers microhardness test and disagreement with Cecchini et al as laser was applied from 12mm distance while in the current study laser was applied similar to the study of *Correa-Afonso et al*(20), who indicated that Er:YAG laser was efficient in preventing demineralization at a 4 mm distance using water cooling.

Incontrast with the current study, *Liu et al* (21) was determined the mechanism of a combined fluoride-laser treatment using microcomputed tomography. The author was found that subablative low-energy Er:YAG laser irradiation following fluoride treatment may instantaneously transform enamel hydroxyapatite into fluoridated hydroxyapatite to reduce enamel solubility as a preventive treatment for enamel demineralization. This is due to different parameter used of Er:YAG laser radiation as frequency and pulse duration.

CONCLUSION

Fluoride varnish alone showed the best preventive measure against enamel demineralization evidenced by the highest surface enamel microhardness. There was no different between the two types of erbium laser on the effect of enamel surface microhardness. Combined treatment between laser and fluoride showed a higher surface enamel microhardness than the use of laser irradiation alone and lower than fluoride alone.

Clinical Recommendation

- Orthodontist should be advised to use fluoride varnish as preventive measure against enamel demineralization.
- Combining treatment between laser and fluoride can be use as preventive measure against enamel demineralization rather than laser alone.

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