The Rate of Demineralization in the Teeth Prepared by Bur and Er:YAG Laser

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Abstract
Introduction: The present in vitro study evaluated the recurrence rate of caries following cavity preparations with bur (conventional technique) and irradiation by Erbium:Yttrium–Aluminum–Garnet (Er:YAG) laser through micro hardness test.
Methods: A total of 72 human extracted molars were randomly divided into 3 groups and class 5 cavities were prepared on them with 3 different methods: G1) conventional bur, G2) Er:YAG laser irradiation alone and G3) laser irradiation + laser treatment. The specimens were immersed in the artificial caries solution with pH of 2.0 and 5.0 (12 days) and then immersed in re-mineralizing solution with pH of 7.0 (25 days). The specimens were longitudinally sectioned and their Vickers micro hardness was determined. Data were statistically analyzed by means of three-way analysis of variance (ANOVA) and Tukey multiple comparisons tests.
Results: The micro hardness of the samples was affected by substrate type (enamel and dentin) and low values were achieved in dentin (P<0.001). Moreover, no significant difference was observed between preparation methods by bar and laser irradiation alone (P≤0.499). Although laser irradiation + laser treatment decreased micro hardness of enamel compared to other methods. In dentin samples, different methods of preparation showed no significant effect on micro hardness (P≤0.874).
Conclusion: Due to the similar values of micro hardness following G1 and G2, it seems that Er:YAG laser alone is as much effective as the conventional bur to prevent recurrence caries. However, because of the high prices of laser instruments, bur preparations can be done commonly.
Keywords: Bur preparation; Er:YAG laser; Micro hardness.

Introduction
Dental caries is a common and almost inevitable disease with high prevalence in the population.¹ Therefore, identification of dental caries risk factors and use of preventive and therapeutic procedures are very critical.² From past to present, rotary devices were used to remove decays. In recent years, various laser devices have been used as alternatives for cavity preparation, treatment of tooth sensitivity, surface treatment of enamel and dentin for increased bond strength and other purposes. Erbium:Yttrium–Aluminum–Garnet (Er:YAG) laser is the most popular type of laser used for cavity preparation.³–⁹ Laser irradiation was employed to prevent cavities due to its strong interaction with the dental hard tissue.⁸ The mechanisms behind increased enamel resistance against acid after laser application include (1) decreased enamel permeability via melting and reforming enamel surface crystals, (2) decreased enamel solubility by forming compounds with more resistance against solubility like tetra calcium diphosphate monoxide, (3) decreased enamel solubility by changing enamel structure like reducing water and carbonate content and increasing hydroxyl ions, forming pyrophosphates and breaking up protein chains.⁷ It was also reported that small spaces are created inside the enamel by laser irradiation. Calcium, phosphor and fluoride ions are trapped inside them and deposit in enamel.¹⁰ Since 97 wt% enamel structure is made of carbonate hydroxyapatite and the rest from water, protein and fat, laser energy is absorbed quickly by water and hydroxyapatite, and its thermal variations

may change chemical and morphological structure of enamel.\textsuperscript{11,12}

The energy of Er:YAG laser with 2940 nm wavelength is largely absorbed by water which is equal to the amount of energy that is absorbed by hydroxyapatite.\textsuperscript{4,13} Therefore, this laser has been recognized to be more effective than other lasers for removing dental hard tissues with minimal thermal damage to dental structures.\textsuperscript{13,14}

Thus in some studies, the efficiency of Er:YAG laser in dentistry was evaluated regarding cavity preparation and surface treatment of enamel and dentin for better bonding to restorative materials.\textsuperscript{13} The benefits of cavity preparation by laser compared with bur preparation include less vibration and noise, sterilization of the cavity and also seals of the dentin surfaces.\textsuperscript{15} Also, laser application melts inorganic components of dentin and cause re-mineralization; thus laser increases resistance to caries.\textsuperscript{16}

Accordingly, by comparing the effects of tooth preparation by laser irradiation and conventional preparation by bur, the best means can be identified for cavity preparation and reduction of recurrent caries. Therefore, this experimental study was performed to compare the recurrent caries rate in cavities prepared with 3 methods of bur preparation (conventional method), Er:YAG laser application, Er:YAG laser application + laser treatment. The null hypothesis of this study was that tooth preparation with Er:YAG laser does not reduce recurrent caries rate of enamel and dentin.

Methods

Seventy-two human molar teeth without caries or fractures were collected during 3 months and stored in normal saline. The teeth were extracted for various medical reasons and their use was allowed by the patients. The normal saline was exchanged weekly and in the last week the teeth were immersed in 1% thymol solution with pH = 7, for disinfection. Then, class 5 cavities (classic box) with dimensions of 3×5 mm and depth of 1.5 mm were created on the buccal surfaces of the teeth, in the way that the occlusal margins were placed in the enamel and cervical margins in the cement. The occlusal margin of the class 5 cavity was beveled. One-third of the teeth were prepared by diamond flat end fissure bur (G1).

Two-thirds of the teeth were randomly assigned for preparation with Er:YAG laser (2940D plus, Deka, Florence Italy). The characteristics of laser irradiation were: energy: 350 mJ; frequency: 10 Hz; power: 3.5 W; pulse duration: 470 µs, spot size: 1 mm, flounce of laser beam: 0.445, flow rate of water spray: according to the setting of instrument 50% water and 50% air. The laser was used in non-contact scanning mode, 4 mm above the surface. Irradiation was stopped when the preparation of enamel or dentin was achieved and a class 5 cavity was formed.

The samples prepared by laser were divided in 2 groups; without surface treatment by laser (G2) and with laser treatment (G3). Following parameters of laser treatment were used for surface conditioning: frequency of 10 Hz; energy of 50 mJ; power of 0.5 W and pulse duration of 230 µs. Laser was guided on the tissue surface by the transmitter arm (articulated arm) of the optical window and preparation was performed by sweep motion. Air and water spray were used during cavity preparation and surface treatment. The handpiece was placed perpendicular to the tooth surface and the samples were exposed to radiation once from every direction with horizontal and vertical slow motions, to create uniform radiations and to cover over the samples. The cavities were prepared by an expert dentist in laser instruments application.

In each group of 24 samples, half of the teeth were randomly immersed in the artificial cariogenic solution (CaCl\textsubscript{2}: 2.0 mmol/L; KH\textsubscript{2}PO\textsubscript{4}: 2.0 mmol/L), sodium acetate (0.075 mol/L) with pH = 5, and the other half were stored in the artificial cariogenic solution with pH = 2 both for 12 days. In a pilot study, we identified the proper time for demineralization of enamel and dentin. Then, the samples were placed in re-mineralization solution (CaCl\textsubscript{2}: 1.5 mmol/L; KH\textsubscript{2}PO\textsubscript{4}: 0.9 mmol/L; KCl: 150 mol/L) for 25 days. Samples were kept in the incubator at 37°C when they were immersed in the solution. Then, the teeth were mounted in the molds and were cut longitudinally (bucco-lingually) using a diamond blade mounted in Isomet low speed cutting saw machine. Then the samples were polished manually with silicon carbide 800, 1500, 2500 and 3000 grit.

Demineralization of the tooth structure as an indicator of recurrent caries was evaluated by measuring the micro hardness values. Micro hardness tests were performed by applying a constant force with Vickers device (Bareiss, Germany, V-test Serial No: vtp6060), power: 50 Nm; time of loading: 10 seconds and magnification: ×940. In micro hardness tests, enamel occlusal walls (in 3 points; the first point with 50 µm distance from the occlusal edge of cavity, and the second and third points with 50 µm and 100 µm distances from the first point in internal direction) and dentin axial walls (in 3 points; the first point with the 150 µm distance from the axial edge of cavity, and the second and third points with 150 µm and 300 µm interior distance from the first point) were measured. The Vickers device has the ability to accurately assess distances. Micro hardness was obtained through the formula of constant number × f (force) /d\textsuperscript{2}.

Considering the normal distribution of data and assumption of the equal variances; three-way analysis of variance (ANOVA) was used to determine the effects of variables (type of substrate, cariogenic solution pH and cavity preparation methods in three different points of the cavity walls) on the micro hardness values of the samples. Also, Tukey test was used for multiple comparisons to compare the results between two groups after significant ANOVA results.

Results

The mean and standard deviation (SD) of micro hardness in enamel and dentin according to the type of cavity
preparation and demineralization solution pH is showed in Table 1. Three-way ANOVA showed that the substrate type (dentin, enamel) had a significant effect on the micro hardness ($P \leq 0.001$) and demineralization was higher for dentin, while that artificial cariogenic solution pH did not show a significant effect on the micro hardness values ($P \leq 0.877$).

Moreover, no significant difference was observed between preparation methods by bur and laser irradiation alone ($P \leq 0.499$). Although laser irradiation + laser treatment decreased micro hardness of enamel compared to other methods. This difference was statistically significant ($P \leq 0.001$). In dentin samples, different methods of preparation showed no significant effect on micro hardness ($P \leq 0.874$).

**Discussion**

In this study, the effects of tooth preparation with bur and Er:YAG laser on the enamel and dentin micro hardness were measured after applying artificial demineralization solution. To evaluate the effects of pH on the acid resistance of dental hard tissues, 2 pH values were used for demineralization solution ($pH = 2$ and $pH = 5$).

Seventy studies have been performed to evaluate the effects of preparation with different types of laser on the quality of dental hard tissues; enamel and dentin$^{17,18}$ Based on the results of the present study, cavity preparation methods (diamond bur, Er:YAG laser, Er:YAG laser + laser treatment) had significant effects on the recurrent caries rate or demineralization rate of the enamel (micro hardness values); Er:YAG laser + laser treatment (G3) increased the rate of demineralization in enamel. Artificial cariogenic solution pH showed no significant effects. Also micro hardness values of enamel samples were significantly higher than dentin samples.

The rate of demineralization in Er:YAG laser group (G2) was similar to bur group (G1). Several studies have shown increased enamel resistance against acid attacks in laser-prepared samples and expressed many theories.$^{14,19-21}$ The most acceptable theory states that an increase in temperature of the enamel surface over 100°C causes disintegration of adjacent carbonates. This continues to the melting point and increases acid resistance, thus decreases potential of enamel demineralization. Presence of carbonate in the network increases solubility of hydroxyapatite.$^{12,23}$ Most researchers believe that the loss of water and carbonate are important factors in caries prevention.$^{22,24}$ Increased acid resistance of enamel after laser irradiation is induced by photo thermal interactions, not photo mechanical.$^{23}$ It has been reported that thermal range between 100-650°C is essential for these effects and to increase the acid resistance of enamel.$^{25}$

Similar to our study, Hossain et al showed that Knoop hardness in cavities prepared by Er,Cr:YSGG laser had no significant difference compared to cavities prepared by bur.$^{17}$ In the present study there was no difference in Vickers micro hardness between the 2 groups of G1 and G2, which may be because the applied power could not cause the melting of enamel and dentin crystals. While using Er:YAG laser for caries prevention, observing the ablatable and sub-ablative conditions is very important for chemical changes to occur without morphological damage in enamel. In addition, laser energy should not cause thermal damage to the pulp and periodontal ligament.$^{26}$ Numerous studies have shown that the use of water/air cooling has negative effect on caries resistance of Er:YAG laser irradiation.$^{14,21}$

In our study secondary laser treatment reduced Vickers hardness in enamel samples compared to G1 and G2 groups. Our results were in accordance with the results of Apel et al$^{23}$ in 2005. These researchers evaluated the effectiveness of the erbium laser irradiation in sub-ablative manner and anti-caries effects on enamel. They stated that the sub-ablative Er:YAG laser irradiation in enamel caused microscopic changes and surface cracks. The fine cracks created after laser irradiation could act as starting points of acid attacks and deep demineralization, so counteract the positive effects of laser in caries prevention.$^{21}$

Chimello et al studied the effect of cavity preparation with Er:YAG laser and handpiece on enamel structure. They found no difference between groups with regard

<table>
<thead>
<tr>
<th>Preparation</th>
<th>pH</th>
<th>Substrate</th>
<th>Mean</th>
<th>SD</th>
<th>Number</th>
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<td>Bur (G1)</td>
<td>2</td>
<td>Enamel</td>
<td>303.0</td>
<td>43.92</td>
<td>12</td>
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<td></td>
<td></td>
<td>Dentin</td>
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<td>14.3</td>
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<td>5</td>
<td>Enamel</td>
<td>318.7</td>
<td>41.15</td>
<td>12</td>
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<tr>
<td></td>
<td></td>
<td>Dentin</td>
<td>56.76</td>
<td>10.5</td>
<td>12</td>
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<tr>
<td>Laser</td>
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<td></td>
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<td></td>
<td></td>
</tr>
<tr>
<td>Without laser treatment(G2)</td>
<td>2</td>
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<td>322.5</td>
<td>60.84</td>
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<tr>
<td></td>
<td></td>
<td>Dentin</td>
<td>50.19</td>
<td>13.05</td>
<td>12</td>
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<tr>
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<td></td>
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<td>55.79</td>
<td>16.80</td>
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<tr>
<td>With laser treatment(G3)</td>
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<td>50.39</td>
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<td></td>
<td>Dentin</td>
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<td></td>
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<td>Dentin</td>
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<td>10.23</td>
<td>12</td>
</tr>
</tbody>
</table>

Note: The same letters indicate no significant differences according to 3-way ANOVA ($P \geq 0.05$).
to demineralized area and presence or absence of cracks. They concluded that regarding alteration of enamel structure, preparation with Er:YAG laser was similar to high-speed handpiece. In a recent study Ahrari et al. evaluated demineralization resistance of the enamel after Er:YAG laser etching for bonding orthodontic brackets and concluded that the laser could not increase resistance of enamel against acid attack.

In the dentin samples, there was no statistically significant difference in Vickers micro hardness between G1, G2 and G3 groups. There are conflicting results regarding the demineralization rate and acid resistance after laser treatment of enamel and dentin. The sub-ablative laser irradiation causes fine cracks in the surface. In addition, the use of ablative laser with 400 mJ energy (without water) causes the lowest demineralization in enamel and dentin. Although at micro morphologic level, this treatment method induces thermal damages. In an in vitro study, the amounts of dissolved calcium and phosphor and their ratio were not different in non-lased and lased bovine dentin samples. This means that Er:YAG laser irradiation does not decrease nor increase dentin acid resistance. In another study, Hossain et al in 2004 showed that Knoop hardness and ca/p ratio in laser treated dentin samples were almost similar to the bur prepared samples.

As it is evident from the above contents, more studies should be done to find definite results. Therefore, it can be stated that regardless of the other benefits of tooth preparation with laser, this method compared with bur preparation does not have any advantages to increase the caries resistance.

Conclusion
Substrate type had a significant effect on micro hardness. The artificial cariogenic solution pH did not show significant effect on the micro hardness values. Due to similar values of micro hardness following G1 and G2, it seems that Er:YAG laser alone is as much effective as conventional bur to prevent recurrent caries. Though, laser irradiation + laser treatment decreased micro hardness of enamel compared to other methods. In dentin samples, different methods of preparation showed no significant effect on micro hardness.

Ethical Considerations
Not applicable.

Conflict of Interests
The authors declare that they have no conflict of interest.

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