An In Vitro Comparison of the Bond Strength of Composite to Superficial and Deep Dentin, Treated With Er:YAG Laser Irradiation or Acid-Etching

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Abstract
Introduction: The aim of this study was to compare the micro-shear bond strength of composite resin on superficial and deep dentin after conditioning with phosphoric acid and Erbium-Doped Yttrium Aluminum Garnet (Er:YAG) laser.

Methods: Thirty human molars were selected, roots were removed and crowns were bisected to provide a total of 60 half-crowns. Specimens were ground to expose superficial and deep dentin. Samples were assigned to six groups: (1) AS (acid etching of superficial dentin); (2) AD (acid etching of deep dentin); (3) LAS (Er:YAG laser irradiation on superficial dentin); (4) LD (Er:YAG laser irradiation on deep dentin); (5) LAD (Er:YAG laser irradiation on superficial dentin followed by acid etching); (6) LAD (Er:YAG laser irradiation on deep dentin followed by acid etching). The adhesive protocol was performed. Samples were thermocycled and micro-shear bond strength was tested to failure. The data were submitted to statistical analysis with one-way analysis of variance (ANOVA) and Tukey post hoc test.

Results: The AS group demonstrated the greatest amount of micro-shear bond strength. Statistical analysis showed a decrease in bond strength in laser-treated groups which was more significant for deep dentin.

Conclusion: Preparation of dentin with laser did not improve bonding to superficial and deep dentin.

Keywords: acid-etch; composite; laser; micro-shear bond strength; bond strength.

Introduction
The last decade has been marked by a growing demand for aesthetic restorations and increasing priority of tooth structure preservation which have led to the development and improvement of adhesive materials. Micro-leakage at the tooth-restoration interface has been significantly reduced since the introduction of acid etching by Buonocore in 1955.¹ The strength and durability of the bond between biomaterials and enamel/dentin are significant characteristics to be considered for mechanical, biological, and aesthetic purposes. Pulpal irritation and recurrent caries caused by micro-leakage can be eliminated by precise marginal adaptation of restorations.² Extension of resin tags into the pores of the etched enamel surface provides micromechanical bonding.³ Resin bonding to dentin might lead to less desirable results in comparison to enamel. This might be due to the high organic content, presence of odontoblastic processes in dentinal tubules, smear layer, and surface moisture.⁴,⁵ Dentin surface treated with acid-etch undergoes physical and chemical alterations that allow for micromechanical and chemical bonding of adhesive materials.⁶ The bonding process occurs most efficiently when resin monomers impregnated into partially demineralized dentin create a dentin-resin inter-diffusion zone. This process is highly dependent on the microstructure of the dentin, composed of fluid-filled dentin tubules surrounded by a peri-tubular zone and an inter-tubular matrix.⁷ This structure differs noticeably between superficial and deep dentin with regard to the number of tubules and the amount of peri-tubular and inter-tubular zones. Accordingly, each type of dentin presents different characteristics and requirements for resin...
bonding. 4, 7, 8
In search for more advantageous tooth surface conditioning techniques to replace acid-etching, recent investigations have focused on laser application. 9, 10 During laser treatment, light is converted into heat, which causes ablation and dentin etching. Lasers have been employed for tooth surface modification and improving restoration bonding. 11, 12 Erbium-Doped Yttrium Aluminum Garnet (Er:YAG) laser has the capacity to produce a micro-retainent pattern when applied on tooth surfaces. The effect of Er:YAG laser depends on parameters such as energy output, frequency, pulse mode, and irradiation time. The safety and efficacy of application of Er:YAG laser on dental tissues has been approved. The literature available on Er:YAG laser presents varying parameters and results, yet it lacks a clarified conclusion on the actual efficacy of this technique on improving resin adhesion, particularly on deep dentin. The present study aimed to compare the effectiveness of Er:YAG laser for etching superficial and deep dentin for bonding of a composite resin material with that of acid-etching.

Methods
Specimen Preparation
This study was performed using 30 extracted intact human third molars. The teeth, all extracted within a duration of 6 months, were kept in 0.9% sodium chloride solution until the beginning of the experiment. The teeth were cleaned with non-fluoridated pumice and dental prophylactic cups. Roots were sectioned at 2 mm below the cementoenamel junction, and the remaining crowns were randomly divided into two groups and inserted into an epoxy resin block, with the upper portion of the crowns left unembedded. The teeth were inserted in a position parallel to the sides of the block. Each tooth should provide both superficial and deep dentin so a sectioning machine (Non Stop, Germany) was used to cut the crowns in half in a mesiodistal direction, providing a total of 60 half-crowns. The cutting process proceeded at low speed with refrigeration. In 30 of the half-crowns, superficial dentin was exposed within a 0.5-mm distance from the enamel at the central occlusal groove. In the remaining 30 specimens, deep dentin was exposed at a 0.5-mm distance from the highest pulp horn. Subsequently, the specimens were randomly allocated into six groups (each containing 10 specimens): group 1- AS (acid etching of superficial dentin); group 2- AD (acid etching of deep dentin); group 3- LS (Er:YAG laser irradiation on superficial dentin); group 4- LD (Er:YAG laser irradiation on deep dentin); group 5- LAS (Er:YAG laser irradiation on superficial dentin followed by acid etching); group 6- LAD (Er:YAG laser irradiation on deep dentin followed by acid etching).

Surface Treatment
Dentin surfaces treatment with acid etching in groups 1 and 2 included conditioning the surface by 37% phosphoric acid gel (3M–ESPE, St. Paul, MN, USA) for 15 seconds followed by thorough water rinsing and air drying. In group 2 to 6, surface treatment was performed with an Er:YAG laser machine (Dr Smile, Italy). According to the manufacturer’s instructions, laser irradiation was oriented perpendicular to the surface. Focal distance was 10 mm and laser beam spot size was 0.63 mm. With an energy of 80 mJ and a frequency of 2 Hz, laser scanning was performed for 20 seconds with 5 mL/min water irrigation. In groups 5 and 6, laser irradiation was followed by acid etching in a similar procedure as mentioned above.

Following surface treatment, a uniform layer of adhesive (single bond 2, 3M–ESPE, St. Paul, MN, USA) was applied on the dentin surfaces using disposable brush tips and following the manufacturer’s instructions. After gentle air-drying the specimens for 5 s, adhesive materials were polymerized for 20 seconds using a light-curing unit (XL 3000, 3M Dental Products, USA). The output of the light curing device was measured with a radiometer (Demetron/Kerr, Danbury, CT, USA) which showed 450 mW/cm².

From each group, 1 sample was randomly selected to be analyzed with scanning electron microscopy. The rest of the samples received composite resin materials (2250 3M – ESPE, St. Paul, MN, USA) on the prepared dentin surfaces. All prepared dentin surfaces in all groups were completely covered by composite. Composite resin material was manually applied and Light-cured by a visible-light-curing unit (XL 3000, 3M Dental Products, USA) for 60 seconds.

The samples were stored in a physiological solution at body temperature. After 7 days, all samples were subjected to 500 rounds of thermal cycling, each consisting of 20 seconds in water bath at 5°C and 55°C, with an exchange time of 10 s between the baths. Subsequently, specimens were sectioned vertically using the sectioning machine (Non Stop, Germany), to provide 36 dentin-composite rectangular slabs with a cross-section of 1*1 mm². 13

Bond Strength Testing
The micro-shear bond strength of the samples was tested on a micro-tensile tester machine (COMPACT GAUGE 200N, Bisco, Inc.). The samples in acrylic resin cylinders were protected on the platform, so as the axis of the composite slab perpendicular to the direction of the cross-head travel. The crosshead speed was 0.5 mm/s. The force at which the composite was dislodged from the dentine surface was recorded in N, and the shear bond strength was measured from the cross-sectional area of the composite. The averages and standard deviations were calculated and the data were submitted to statistical analysis by one-way analysis of variance (ANOVA) and Tukey post hoc test.

Results
The average bond strengths in superficial dentin were AS: 5.33 ± 2.15, LS: 2.95 ± 0.66, LAS: 4.97 ± 1.34 (P<0.001). The statistical analysis revealed that there was no statistically significant difference between the groups AS and LAS; however, the group LS showed significantly lower
An In Vitro Comparison in of the Bond Strength

bond strength (Figure 1). The average bond strengths in deep dentin were AD: 4.70 ± 1.21, LD: 2.74 ± 0.64, LAD: 1.92 ± 0.5 (P < 0.001). There was no statistically significant difference between the LD and LAD groups, however, the ED group showed a significantly higher bond strength (Figure 1).

No significant differences were found for Er:YAG laser-irradiation or phosphoric acid etching between superficial and deep dentin. The combination of phosphoric acid etching and Er:YAG laser irradiation improved the micro-shear bond strength of composite resin on superficial dentin as compared to Er:YAG laser (P < 0.05). The dentin tubules were shown to be exposed in SEM photographs of the superficial and deep dentin surfaces (Figure 2A-F).

Discussion

This study was conducted to compare Er:YAG laser irradiation and acid-etching for treatment of superficial and deep dentin surfaces prior to the application of adhesive materials. Chemical changes as well as surface roughening induced in dentin by laser irradiation might increase the bonding surface area, hence the adhesion.2, 14,16,17 The recent literature has not precisely verified the superiority of laser irradiation to acid-etching for treating dentinal surfaces prior to bonding.18-32 Previous reports have claimed that there are certain advantages in bonding to lased dentin because of an apparently enlarged surface area for adhesion based on the scaly and flaky surface appearance following Er:YAG irradiation.33-35 In a study, the shear bond strength in deep lased dentin was better than in superficial lased dentin.36 In addition, acid etching the previously laser-conditioned dentin surfaces has proved to be an effective technique. It was therefore concluded that the surface modification caused by Er:YAG laser does not improve the adhesion of bonding materials to dentin and cannot replace the conventional acid etching technique.37 Er:YAG laser diminishes the water content as well as the organic tissue of dentin at sub-ablative energy densities.37 Moreover, subsurface fissuring in dentin following treatment with Er:YAG laser can be detrimental to the adhesion.38 Laser-induced subsurface damage to dentin has been demonstrated by cracks created under the hybrid layer. Fe-SEM examination has demonstrated the presence of micro-cracks to be more abundant in laser-irradiated dentin surfaces than in fractured dentin surfaces.38 Dentin surface modifications made by laser irradiation, even if further altered by acid-etching, might not allow for proper adhesion of bonding resins, since laser irradiation provides an acid-resistant surface. On the other hand, as the hybridization is an imperative for adhesion of dentin bonding materials, removal of organic tissue by laser impedes this process. In addition, the previously mentioned subsurface damage might extend beyond the hybrid layer.

Figure 1. The Average Bond Strength Measured in the 6 Experimental Groups (small letters show significant difference between the groups – P<0.001)

Figure 2. Scanning electron microscopic photograph of acid etched superficial dentin (original magnification ×5000). (B) Scanning electron microscopic photograph of acid etched deep dentin (original magnification ×5000). (C) Scanning electron microscopic photograph of laser-treated superficial dentin (original magnification ×5000). (D) Scanning electron microscopic photograph of laser-treated deep dentin (original magnification ×5000). (E) Scanning electron microscopic photograph of laser-treated and acid-etched superficial dentin (original magnification ×5000). (F) Scanning electron microscopic photograph of laser-treated and acid-etched deep dentin (original magnification ×5000).
This condition weakens the substrate and causes cohesive dentin fracture.\textsuperscript{16,19,22,28} The potential role of subsurface damage and the limited hybridization in low tensile bone strength has been confirmed in laser-treated cavities; the superficial layer was removed by acid-etching or air abrasion.\textsuperscript{21,24} Laser irradiation seems not to be able to create an inter-diffusion zone similar to that created by acid etching.\textsuperscript{37,39,40}

It is worth mentioning that verification, i.e. recrystallization of dentin apatite and formation of more calcium phosphate, has not been evaluated thoroughly. Thanks to verification, acid-resistance, dental hardness and abrasion are increased. Dentinal permeability might also be reduced as a result of sealing the dentinal tubules up to a considerable depth.\textsuperscript{31-34} Such alterations in dentinal surfaces might impede the proper bonding of restorative materials and cause cohesive micro-fractures.\textsuperscript{19}

**Conclusion**

Within the limitations of this in vitro study, it may be concluded that laser treatment of dentinal surfaces negatively affects the bond strength when compared to acid etching. Contradictions still remain regarding the most effective way of accomplishing adhesion on Er:YAG laser-irradiated surfaces. This controversy might be partly due to the heterogeneity of methods for dentin conditioning with laser. Accordingly, it might be suggested that a standard energy output be defined for treatment of different dental tissues. Further research is required to precisely determine the effect of laser conditioning on adhesion of bonding materials to dentinal surfaces.

**Conflict of Interests**

The authors declare that there is no conflict of interest regarding the publication of this paper.

**References**

An In Vitro Comparison in of the Bond Strength


