

Micro-shear bond strength of Er:YAG-laser-treated dentin

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Abstract This study tested if dentin adhesion is affected by Er:YAG laser. Ninety dentin disks were divided in groups ($n=10$): G1, control; G2, Er:YAG laser 150 mJ, 90° contact, 38.8 J/cm²; G3, Er:YAG laser 70 mJ, 90° contact, 18.1 J/cm²; G4, Er:YAG laser 150 mJ, 90° non-contact, 1.44 J/cm²; G5, Er:YAG laser 70 mJ, 90° non-contact, 0.67 J/cm²; G6, Er:YAG laser 150 mJ, 45° contact, 37.5 J/cm²; G7, Er:YAG laser 70 mJ, 45° contact, 17.5 J/cm²; G8, Er:YAG laser 150 mJ, 45° non-contact, 1.55 J/cm²; and G9, Er:YAG laser 70 mJ, 45° non-contact, 0.72 J/cm². Bonding procedures were carried out and the micro-shear-bond strength (MSBS) test was performed. The adhesive surfaces were analyzed under SEM. Two-way ANOVA and multiple comparison tests revealed that MSBS was significantly influenced by the laser irradiation ($p<0.05$). Mean values (MPa) of the MSBS test were: G1 (44.97±6.36), G2 (23.83±2.46), G3 (30.26±2.57), G4 (35.29±3.74), G5 (41.90±4.95),

G6 (27.48±2.11), G7 (34.61±2.91), G8 (37.16±1.96), and G9 (41.74±1.60). It was concluded that the Er:YAG laser can constitute an alternative tool for dentin treatment before bonding procedures.

Keywords Dentin · Er:YAG laser · Micro-shear bond strength · Morphology · Self-etching primers

Introduction

Roughness and free surface energy play an important role on the bonding procedure and can increase the adhesion of composite materials to dentin. The increase on adhesion strength depends on the adhesive ability to spread on dentin surface, which is referred to as wettability. To improve wettability and bonding, the free surface energy of dentin must be increased by the use of conditioners and primers [1, 2].

It is well known that acid-etching leads to physicochemical changes on dentin surface, beneficial to micro-mechanical and chemical attachment of resin to the dentin. The most effective approach appears to be the formation of a dentin–resin inter-diffusion zone or *hybrid layer* [1]. The use of self-etching primers has been extensively reported in the literature and results support that there is an increase in dentin bonding, even with a thick hybrid layer formation [3, 4]. Although these simplified systems are convenient to use, a perfect bond to dentin still remains as one of the most important and challenging features to be considered when choosing adhesive materials.

Some authors [1, 5] reported that the surface topography also interferes on bonding procedures; the degree of surface roughness seems to indicate the degree of mechanical anchorage in bonding process.

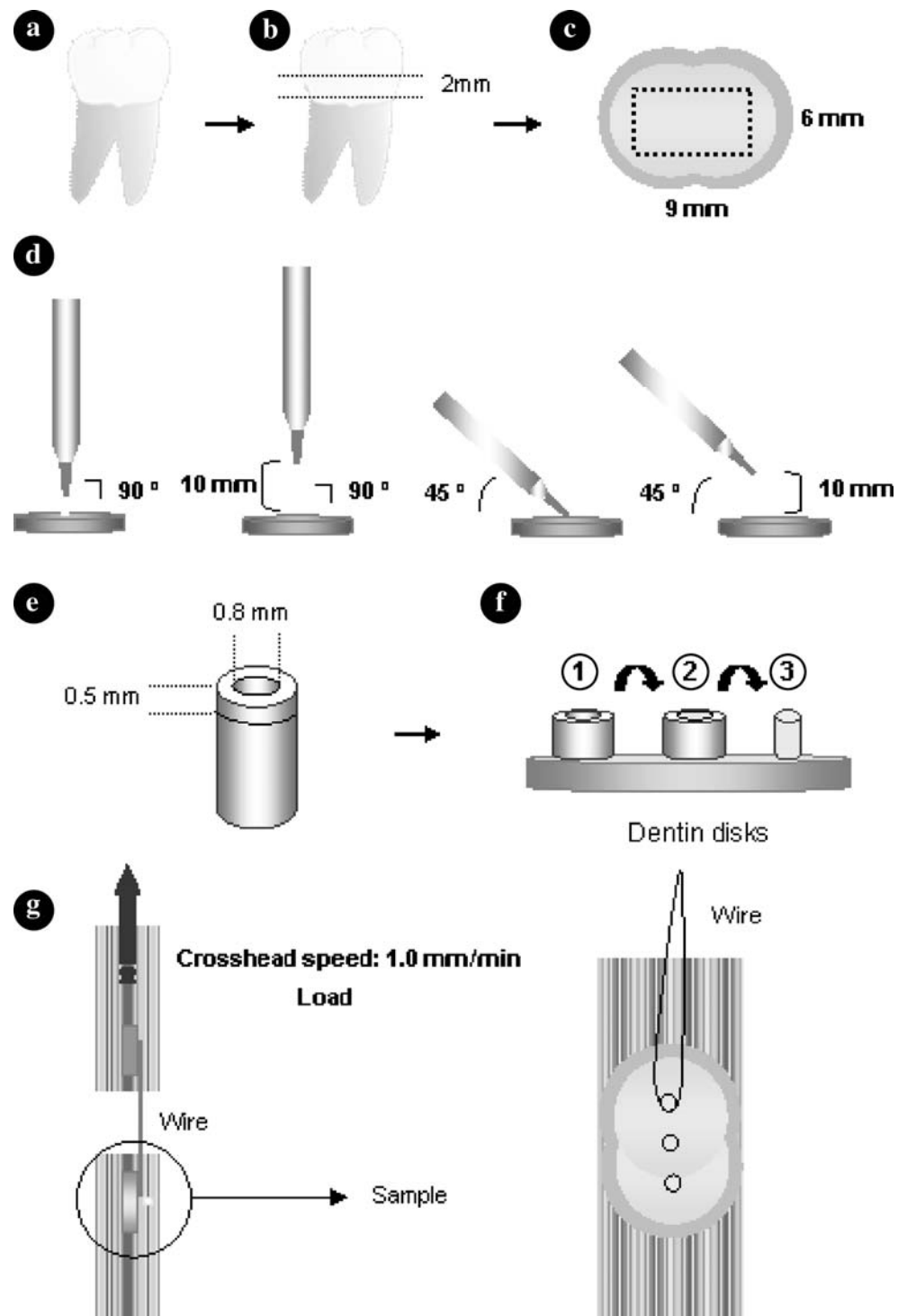
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Fig. 1 **a** Third molars were collected and cleaned. **b** Dentin disks of 2 mm were obtained from the crowns. **c** An area of 6×9 mm was delimited on the dentin surface of each sample. **d** Laser irradiation was performed with different parameters (contact/non-contact mode of irradiation, beam angle, and energy). **e** A tygon tube was cut to obtain matrix of 0.8 mm diameter. **f** Bonding procedures were carried out. The self-etching adhesive system was applied and the matrix was placed. After the light curing, the composite resin was placed and then light-cured. **g** The micro-shear bond test was carried out



Laser has been introduced in clinical dentistry for removal of dental hard tissues before the application of restorative materials [6]. One believes that the effects of laser irradiation on tooth structure are dependent on wavelength specificity, energy density, contact or non-contact mode of irradiation, and incidence angles. Er:YAG lasers emit a wavelength of 2.94 μm and can ablate dental hard tissues more effectively than other lasers [7, 8] due to

its high coefficient of absorption in both water and hydroxyapatite and because it only minimally increases temperature in the surrounding tissues, especially when constant cooling is achieved with continuous water spray [9].

As the laser energy absorbed by dental tissue produces surface modifications, it has been suggested that laser could be used for the pretreatment of dentin and for enhancing the

Table 1 Parameters used for the irradiation of the dentin surface

Group	Energy (mJ)	Repetition rate (pulse/s)	Incidence angle	Laser beam delivery	Exposure time (s)	Energy Density (J/cm ²)
G1	Control—without laser irradiation					
G2	150	20	90°	Contact	52	38.8
G3	70	20	90°	Contact	52	18.1
G4	150	20	90°	Non-contact	52	1.44
G5	70	20	90°	Non-contact	52	0.67
G6	150	20	45°	Contact	52	37.5
G7	70	20	45°	Contact	52	17.5
G8	150	20	45°	Non-contact	52	1.55
G9	70	20	45°	Non-contact	52	0.72

bonding of adhesive restorative materials [10, 11]. Several characteristics of lased dentin have previously been considered as advantageous for resin bonding. They include the formation of a microscopically rough surface without demineralization and open dentinal tubules without smear layer production [10–16].

To obtain a higher bond strength of resin to dentin, this study aimed to evaluate the effect of different parameters of the Er:YAG laser irradiation on dentin micro-shear bond strength (MSBS). In addition, the treated dentin surfaces were evaluated morphologically under SEM. The null hypothesis tested was that there is no difference in the MSBS of the dentin irradiated with different parameters of the Er:YAG laser and the non-irradiated surface, when followed by the application of a self-etching primer.

Understanding how laser treatment contributes to the adhesion of resin to dentin may help the professional in selecting the best parameter of irradiation for a specific clinical application. This understanding is the key for clinical success, where excellence in esthetics should parallel oral health maintenance.

Materials and methods

Preparation of the specimens

Forty-five non-carious freshly extracted human third molars were cleaned with a rotary brush with pumice and stored in physiological serum at 4°C until its use. The teeth were fixed with cyanoacrylate adhesive (Zapit Base, Dental Ventures of America, Corona, USA) on a metal disk. To obtain samples of similar size, the teeth were sectioned at the cementum–enamel junction. Ninety dentin disks of approximately 2-mm thick were obtained from the lingual and buccal surfaces by slow-speed sectioning with a diamond saw (Isomet-Buehler Lake Buff, IL, USA). Each slice was ground with a 600-grit silicon carbide paper (Struers, Tokyo, Japan) under running water to provide uniform surfaces of superficial dentin. An area of 6×9 mm

was delimited on the surface of each specimen on which the laser irradiation was performed. Figure 1 illustrates the experimental design.

Laser device and irradiation

The dentin disks were randomly assigned in nine groups ($n=10$), as shown in Table 1.

All groups, except the control (G1), were irradiated with dentin disks of approximately 2-mm thick were obtained. The Er:YAG laser (Osada, Tokyo, Japan) works with a wavelength of 2.94 μm . Regarding the handpiece used (HPER-S), the energy delivered at the end of the tip was 30% lower than that depicted on the equipment display¹. Therefore, the groups irradiated with 150 and 70 mJ were, in fact, irradiated with 105 and 49 mJ, respectively. When using the handpiece at a non-contact mode, the laser beam was delivered at a work distance of 10 mm. The laser irradiation was performed on the dentin surface for 52 s, scanning the surface in both directions (vertical and horizontal) under water cooling (5.0 ml/mm). The irradiation was performed by hand, screening the dentin surface with a uniform motion. The handpiece was fixed on a metallic device for micrometric movements for which the angles of 1–180° were depicted.

Bonding procedures

The bonding and restorative procedures adhered strictly to the manufacturer's instructions. The bonding system used was the Clearfil SE Bond System (Kuraray Co, Osaka, Japan), which includes a self-etching primer and a bonding agent. All dentin disks (G1–G9) were treated with Clearfil SE Bond primer for 20 s and dried. The Clearfil SE Bond was applied, air-thinned and light-cured for 20 s.

¹ The energy delivered at the end of the tip was measured with a power meter (FieldMaster, Coherent, USA) before the laser irradiation.

Table 2 Mean values (MPa) and standard deviation (SD) of the micro-shear bond strength test for the specimens treated with the Er:YAG laser

Sample	G1	G2	G3	G4	G5	G6	G7	G8	G9
Mean (MPa)	44.97 A	23.83 E	30.26 D	35.29 C	41.90 B	27.48 D	34.61 C	37.16 C	41.74 B
SD	6.36	2.46	2.57	3.74	4.95	2.11	2.91	1.96	1.60

Different letters indicate significant statistical difference ($\alpha=0.05$)

Before light-curing the bonding resin, an iris was mounted on the dentin surface to restrict the bonding area. This iris was cut from microbore Tygon tubing (R-3603, Norton Performance Plastic, Cleveland, USA) with an internal diameter and a height of approximately 0.8 and 0.5 mm, respectively. A hybrid restorative resin composite, A3 shade (Clearfil AP-X, Kuraray), was placed into the iris and a clear cellophane sheet was placed over the resin and pressed gently. The resin was then light-cured for 40 s. In this manner, very small cylinders of resin of approximately 0.8 mm in diameter and 0.5 mm in height were bonded to the surface [16].

The specimens were stored at room temperature (23°C) for 1 h before removing the Tygon tubing. Then, they were stored in water at 37°C for 24 h.

Micro-shear bond test

After 24 h, the dentin disks were adhered with a cyanoacrylate adhesive (Zapit Base, Dental Ventures of America) that, in turn, was placed in a universal testing machine (Ez-test, 500 N, Shimazu, Kyoto, Japan) for micro-shear bond testing. A thin wire (diameter, 0.20 mm) was looped around the resin cylinder, making contact through half its circumference, and was gently held flush against the resin/dentin interface. A shear force was applied to each specimen at a crosshead speed of 1.0 mm/min until failure occurred.

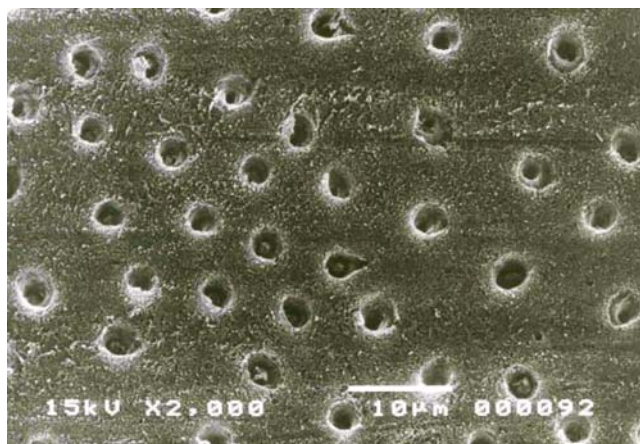


Fig. 2 Representative SEM picture of the morphological aspects of the non-irradiated dentin followed by the treatment with a self-etching primer

SEM evaluation

For the surface morphology analysis, 27 dentin disks ($n=3$) were treated in the same manner as in the micro-shear bond test. After bonding procedures, all specimens were analyzed under scanning electron microscopy (SEM; JXA840, Jeol, Tokyo, Japan), so that the surface morphological changes could be identified.

After laser irradiation and bonding procedures, the dentin samples were given an additional 60-s acetone rinse for the removal of any crystals or other residue from the primer. Finally, the surface was sputter-coated with gold and observed under SEM.

Statistical analysis

The data from the MSBT were statistically analyzed using two-way ANOVA and multiple comparison test. The analyses were performed using the Stat View J-4.5 for Windows. Statistical significance was defined as $p<0.05$.

Results

Micro-shear bond test

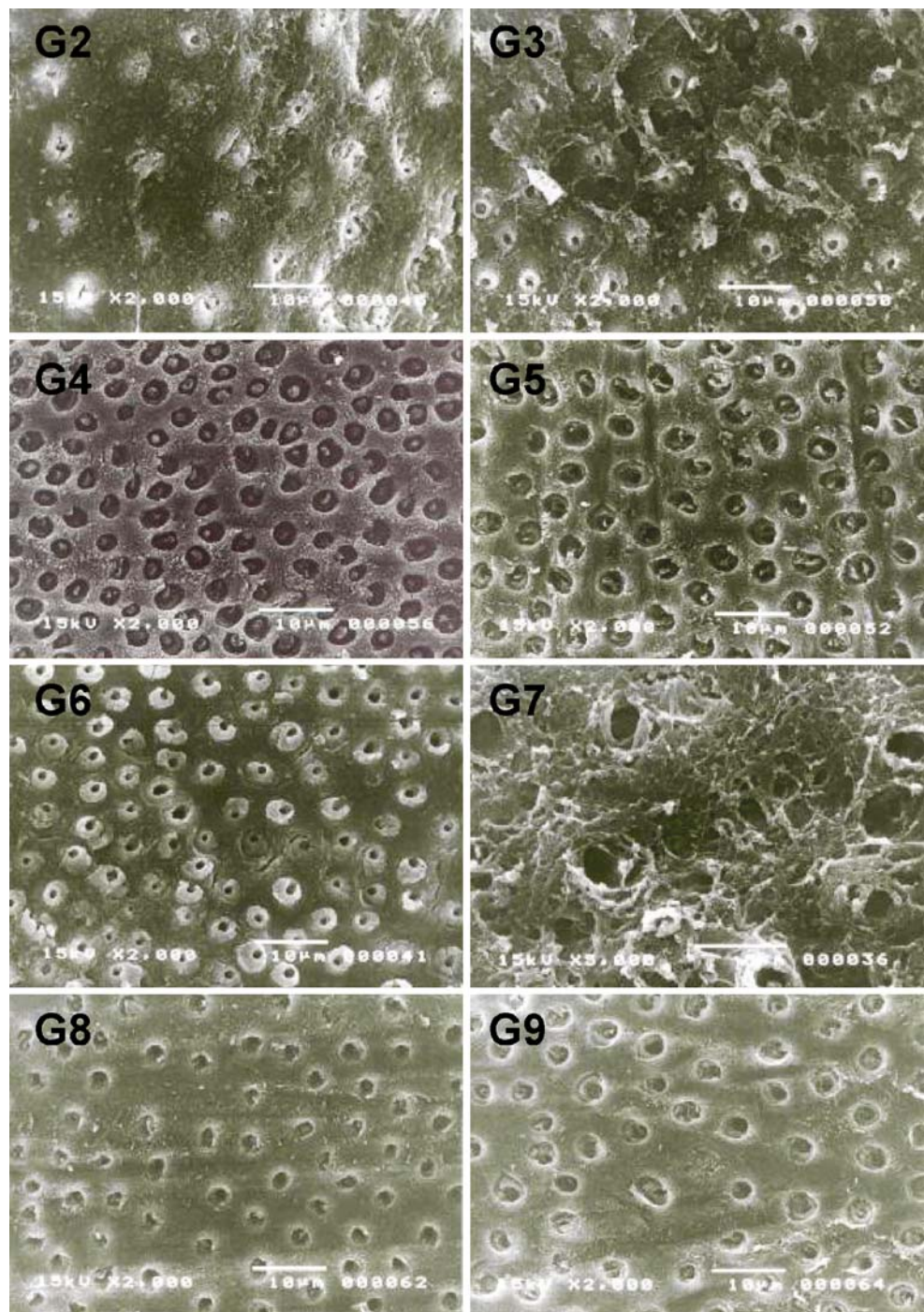
MSBS was significantly influenced by the treatment of the dentin surface ($p<0.05$), as shown in Table 2. Multiple comparison tests revealed that the use of higher energies and the handpiece in contact with the dentin surface (higher energy densities) yielded MSBS values that were lower than those achieved with the handpiece at a non-contact mode and 70 mJ (lower energy densities), regardless of whether the latter was used at 45 or 90° incidence angles.

For all the experimental groups, the MSBS values obtained between resin and the laser-treated dentin were statistically lower than the control group. However, MSBS values from groups G5 (41.90 MPa) and G9 (41.74 MPa) were similar from that of G1—control group (44.97 MPa).

SEM evaluation

Representative SEM pictures of the morphological aspects of the control group and experimental groups are shown in Figs. 2 and 3, respectively. After the application of the self-

Fig. 3 Representative SEM pictures of the morphological aspects of the irradiated dentin followed by the treatment with a self-etching primer. Micrographs A–H correspond to the results from groups G2–G9, respectively



etching primer, the samples from groups G2 (Fig. 3a) and G6 (Fig. 3e) exhibited a homogeneous surface with some occluded dentinal tubules. The collagen of the upper surface appeared to be packed and melted, with an appearance similar to that of glazing. In G3 (Fig. 3b) and G7 (Fig. 3f), it was possible to verify more exposed collagen. Tubules presented mineralized peritubular dentin matrix with cuff-like appearance.

Groups G4, G5, G8 and G9 showed opened tubules that appeared to be penetrated by adhesive resin. The smear

layer was removed and the collagen exposed. It was observed a reticular aspect of dentinal collagen fibers in both the intertubular zone and at the first part of the peritubular dentin.

Discussion

The present study investigated how the incidence angle of the laser beam can influence the irradiation pattern and the

adhesion to dentin by increasing the beam spot size and consequently, decreasing the energy density. Clinically, adhesion with self-etching primers can be compromised by laser parameters. Therefore, professionals should be aware of how laser works and which parameters select for different clinical applications.

A number of studies have been performed to evaluate the bonding ability of adhesives systems to dentin, including tensile and shear bond tests. However, there are still doubts about how laser-treated dentin can bond to adhesive systems. In the present study, bond strengths were assessed by means of a micro-shear bond test that can measure bonding to extremely small areas of substrate [17, 18].

Self-etching primers were used for the bonding procedures because it contains acidic adhesive monomers and water, so that it works as an etchant as well as a primer and thus offers simplification of the clinical procedure [19]. This adhesive system demineralizes and simultaneously infiltrates the dentin to form the hybrid layer [20] and was reported to create a dense and firm hybrid layer after polymerization of the bonding resin, enhancing adhesion to dentin [21].

Although some studies have reported that bond strength of Er:YAG laser-treated dentin was lower than that of non-irradiated dentin [13, 16, 22–24], the use of laser for cavity preparation or dentin conditioning before bonding procedures is expected to enlarge the adhesive area due to the typical flaky structure that dentin exhibits after the laser irradiation [8, 13, 15, 16, 25–27].

In the present study, the groups irradiated with the handpiece in contact with dentin surface (G2, 38.8 J/cm²; G3, 18.1 J/cm²; G6, 37.5 J/cm²; and G7, 17.5 J/cm²) presented a lower bonding quality, as revealed by the MSBS test. It is suggested that the irradiation with the handpiece in contact with the dentin surface and, consequently, with higher energies densities can led to changes on dentin that are not indicated for bonding procedures with the self-etching primers. After the laser irradiation and bonding procedures, the SEM evaluation revealed opened dentinal tubules lined by a slightly melted peritubular dentin, which may have contributed to the non-infiltration of the resin [16, 22, 27]. There was no open collagen matrix necessary for the interdiffusion of the adhesive. With dentinal tubules not fully impregnated by the bonding resin, it had possibly created a weak zone of bonding just below the interface, decreasing the MSBS values. The presence of peritubular dentin after acid etching can be due to the insufficient conditioning effect of the self-etching primer, which presents a weak acidity [28], or due to the possible increase in dentin acid resistance after the laser treatment [16, 29]. Corroborating with our study, Perdigão et al. (1996) [30] reported cuffs residual peritubular dentin matrix and suggested that the resin extension into the region of these

tubules probably compromises the resin retention or tubule sealing.

The diffusion of the monomer from the tubule lumina into the surrounding dentin could only occur if the mineralized peritubular dentin matrix was removed by the etching [4]. Once there are no widened dentin tubules, the obstruction of the infiltration of the self-etching primer into the dentin might occur and the primer could not fulfill its function to superficial layer of the Er:YAG laser-treated dentin [25]. This possibly justifies the lowest MSBS values obtained in the groups treated with the handpiece at a contact mode and higher energy densities.

Other interesting aspect of the groups irradiated with the handpiece in contact with the surface was the characteristic of the intertubular dentin. It appeared packed and melted, with an aspect similar of that of glazing. Opened collagen matrix was not visible, and the collagen fibrils were not found forming a porous network responsible for the increased porosity of dentin. Some authors [14, 16, 31] have reported that the presence of this fused layer, which lacks interfibrillar spaces, can probably restrict resin diffusion into the subsurface intertubular dentin, resulting in lower shear bond strengths. One important variable responsible for the formation of resin-infiltrated demineralized intertubular dentin by resin is the permeability of the demineralized dentin matrix (i.e., maintenance of collagen fibril separation) [4]. These characteristics were not found for the contact-irradiated groups.

When the restorative procedures were conducted in the specimens prepared with the handpiece at a non-contact mode of irradiation and very low energy densities (G5, 0.67 J/cm² and G9, 0.72 J/cm²), MSBS values were similar to those from the control group (non-irradiated dentin). Sealing of the tubules entrance was observed due to the penetration of monomers through the branching of dentinal tubules, which seems to be very important for the infiltration of the adhesive towards the deepest demineralized dentin zone [20].

Under SEM evaluation, it was observed that the dentin treated with the handpiece at a non-contact mode (G4, G5, G8, and G9) presented, after the self-etching primer application, dentinal tubules filled with resin suggesting the previous removal of the smear layer and exposed dentinal tubules by the laser irradiation. Regarding that these groups were irradiated with low energy densities (Table 1), it is suggested that the dentin was laser-conditioned and not submitted to a cavity preparation procedure, as revealed by the morphological analysis of the contact-irradiated groups. The collagen has a fibrillar and reticular aspect in both the intertubular and peritubular area. These aspects corroborate with the characteristics of diamond-prepared dentin etched with phosphoric acid [26, 27] and is possibly the one ideal for bonding procedures after laser irradiation.

Although dentin surface immediately after the laser irradiation was not considered in the current study, previous investigations have shown that the dentin irradiation with the handpiece at a contact or non-contact mode leads to different alterations on dentin morphology. For all experimental groups, the incidence angle did not greatly influence the MSBS results. The application of an acidic primer leads to the demineralization of both peritubular and intertubular dentin, resulting on the collagen matrix demineralization. The groups irradiated with 150 mJ also showed lower MSBS values when compared with the groups with the same parameters, but at 70 mJ. This can be attributed to physiochemical changes of the tissue due to the laser energy [16].

Regarding the energies densities evaluated, there was no evidence of charring or cracking of the dentin, emphasizing that minimal thermal damage of surrounding tissues can be expected when continuous water spray is used [9, 15].

Finally, one can conclude that different energy densities can adversely affect the bonding of composites to irradiated dentin. Both the contact/non-contact mode of irradiation and beam angle are directly related to the variation of the energy density. Despite the advances in adhesive dental materials and the achievement of greater bonding to dentin, professionals should be careful when using the Er:YAG laser for dentin irradiation and should not eliminate the etching step until further evidence-based studies report safe and effective parameters of erbium lasers for treating dentin surface.

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