



Dentin pretreatment with Er:YAG laser and sodium ascorbate to improve the bond strength of glass fiber post

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Abstract

Root-filled teeth that received fiber posts most frequently fail at the adhesive interface between resin cement and dentin. The objective of this study is to evaluate the effect of Er:YAG laser and/or sodium ascorbate (SA) on bond strength, microhardness of dentin, and penetration depth of cement into dentinal tubules. Forty-eight bovine incisor roots were endodontically treated, post spaces were prepared and equally divided into four groups ($n = 12$): G1—distilled water (control); G2—10% SA (10 min); G3—Er:YAG laser (150 mJ/4 Hz/40 s), and G4—Er:YAG laser + 10% SA. Glass fiber posts were cemented and roots sectioned into slices. In the first slice, the push-out bond strength (MPa) and failures were analyzed by confocal laser scanning microscope (CLSM). The second slice was subjected to microhardness test (KHN) and CLSM to assess the cement penetration. ANOVA and Tukey test were used for bond strength and microhardness data and Kruskal-Wallis and Dunn tests for the cement penetration ($\alpha = .05$). The SA-treated samples had higher bond strength ($10.02 \pm 5.45a$), similar to Er:YAG laser ($9.91 \pm 4.62a$) and Er:YAG laser + SA ($8.09 \pm 4.07a$). The least values ($P < .05$) were found on control ($4.02 \pm 2.39b$). Significant differences were observed on root thirds ($P < .05$): cervical > middle > apical. There was a predominance of adhesive failures. The microhardness test revealed no differences between groups ($P > .05$). The experimental groups (G2, G3, and G4) had highest penetration into dentinal tubules when compared to the control (G1). Dentin pretreatments with Er:YAG laser or SA improved bond strength of cement-post-dentin interfaces; however, no synergistic effect of both treatments combined was observed.

Keywords Adhesion · Resin cement · Sodium ascorbate · Laser

Introduction

Root-filled teeth are weakened due to caries, fracture, pulp pathologies, or multiple repeated restorations [1, 2]. They require special considerations for the final restoration, mainly where there has been extensive loss of tooth structure [3, 4]. In these situations, an intracanal post can provide better retention of restoration to compromised roots [2, 5, 6].

Glass fiber posts (GFP) were introduced aiming to improve biomechanical properties as compared to prefabricated metallic posts [2, 6, 7]. Studies have supported GFP permits a regular stress distribution of the strength to the root canal, reduce the incidence of root fracture, and provide favorable or repairable fracture patterns [8–11]. GFP also has superior esthetics, easier removal, and shorter treatment visits in comparison to posts of different materials [7, 12].

However, there are some reports about failures, including debonding of the resin cement from intracanal dentin due to hybrid layer degradation. These failures might be attributed to irrigation with sodium hypochlorite (NaOCl) during endodontic treatment. NaOCl acts as a dentin deproteinizing agent, reduces microhardness of intracanal dentin and its by-products hampers the polymerization of adhesive systems [13, 14]. To overcome this problem, different techniques have been proposed to treat intracanal dentin [15–17], and antioxidants such as sodium ascorbate (SA) have been demonstrated to reverse the negative

Clinical significance: The pretreatment of post space with Er:YAG laser and/or sodium ascorbate antioxidant agent can transform the intraradicular dentin to a substrate more receptive to adhesive procedures.

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effects of oxidants and to preserve resin-dentin bond integrity.

SA is considered to be a stimulant for the synthesis of collagen [9] and recent studies still discuss their ability in tissue repair [18–23]. It has free-radical scavenging properties and stabilizes collagen fibers, improving the adhesion of restorative materials [9, 24]. For this reason, studies have shown the positive bond strength effect of SA on the compromised dental substrate after tooth bleaching [25–27].

Another alternative method to modify compromised intracanal dentin, removing residual smear layer from dentinal tubules, is laser treatment for this purpose Er:YAG laser seems to be the most appropriate [28–31].

Erbium:yttrium-aluminum-garnet (Er:YAG) laser has attracted attention because its emission wavelength (2.940 nm) is highly absorbed by water, and creates a porous and rough surface by removing the smear layer and changing the inorganic compounds in the tooth structure, without increasing temperature [29, 31, 32]. It also has antibacterial activity inside dentinal tubules [33]. To date, few studies tested the bond strength of GFP luted in dentin that were pretreated with Er:YAG laser [31–36], and no study assessed the combined effect of Er:YAG laser with SA antioxidant solution.

This study evaluated the pretreatment of root canal dentin with Er:YAG laser on GFP bonded with self-adhesive resin cement, combined or not with 10% SA solution, analyzing bond strength, microhardness of treated dentin, and morphological characteristics of adhesive interface using CLSM. The null hypothesis is that there is no difference in the adhesion of resin-based cement when using SA and Er:YAG laser inside the root canal.

Materials and methods

Bovine incisors with completely formed roots and closed apices and without any excessive curvature were stored in 0.1% thymol solution at 4 °C, cleaned, and examined to discard those with cracks or structural anomalies. Radiographic exam was used to confirm the presence of a single root canal.

Sample preparation

Forty-eight teeth had their crowns removed (Isomet 1000; Buehler) and roots trimmed coronally to achieve a standardized length of 15 mm and their dimensions were checked with a digital caliper (Mitutoyo).

The working length was again established at 1 mm from the root apex and the canals were prepared using a size 40 K-file (Dentsply Sirona), introduced passively until it reached the apical foramen and then subtracting 1 mm. Root canal shaping was performed with K-file starting in size 40 until

80 (Dentsply Sirona). Irrigation was carried out with 2 mL of 1% NaOCl at each change of instrument and a final rinse with 0.5 mL of 17% EDTA for 3 min. Then, canals were irrigated with 1 mL of distilled water and dried paper points. The technique for obturating root canals was lateral condensation. After 7-day storage at 100% humidity at 37 °C, 10-mm post spaces were prepared using size 5 Gates drills (Dentsply Sirona) and a #3 low-speed drill (Exacto #3; Angelus), leaving a 4-mm apical seal.

The roots were divided into four groups according to intraradicular dentin treatment ($n = 12$): G1—irrigation with distilled water (control); G2—10% SA solution; G3—Er:YAG laser (Fidelli Er III, Fotona), and G4—Er:YAG laser with 10% SA solution; according to the settings summarized in Table 1.

The Er:YAG laser energy was delivered in pulsed mode (duration of 50 μ s), with Xpulse flexible fiber tip (1.0 mm thickness and 13.5 mm length) inserted into root canal in helicoidal movements (from cervical to apical and apical to cervical), using a R14 handpiece. The pulse repetition rate was 4 Hz, energy of 150 mJ during 40 s using a water spray of 4 mL/min. Then, canals were dried with absorbent paper points.

The 10% SA solution was prepared using 10 g of powder of sodium ascorbate (Sigma-Aldrich) and 100 mL of distilled water. The posts from all groups were cleaned using alcohol 96% for 30 s and silane-coupling agent (Prosil, FGM) was applied. Before luting a glass fiber post, the root canals were irrigated using 2 mL of distilled water and dried with absorbent paper points.

The self-adhesive resin cement Relyx U200 (3 M ESPE) was manipulated according to the manufacturer's instructions and was applied to the post surface, and this was seated to full depth in the prepared spaces with standardize finger pressure. After initial chemical polymerization, excess resin cement was removed with a microbrush, and it was light polymerized (650 mW/cm²; Raddi Plus, SDI) for 90 s as recommended by manufacture. Figure 1 illustrates laser irradiation and GFP cementation inside root canal.

The adhesive interface was sealed with zinc oxide cement without eugenol and the specimens were stored at 100% humidity at 37 °C for 24 h. After 24 h, the specimens were subjected 1800 thermal cycles (Ética Equipamentos Científicos) at a temperature ranging from 5 and 55 °C. The samples remained in each bath for 30 s, and the transfer time for another bath was 10 s.

After 24 h, at 100% humidity at 37 °C, the specimens were fixed in acrylic resin and mounted on a low-speed diamond Isomet 1000 (Buehler). Two slices were obtained from each third root (cervical, middle, and apical) with 1.5 mm thickness. The first slice of each third was used to the push-out test and in the second slice, it was

Table 1 Dentin pretreatments used in this study and instructions of use

Dentin treatment	Protocols/parameters
Distilled water (control)	0.5 mL of distilled water immersion and drying with absorbent paper points
10% sodium ascorbate	10 min immersion (20 mL), washing for 30 s with distilled water and drying with absorbent paper points
Er:YAG laser	Laser energy was delivered in pulsed mode, with a repetition rate of 4 Hz, energy of 150 mJ for 40 s. Water irrigation of 4 mL/min and drying with absorbent paper points

analyzed the microhardness and the penetration depth of the resin cement into dentin using CLSM.

Push-out bond strength test

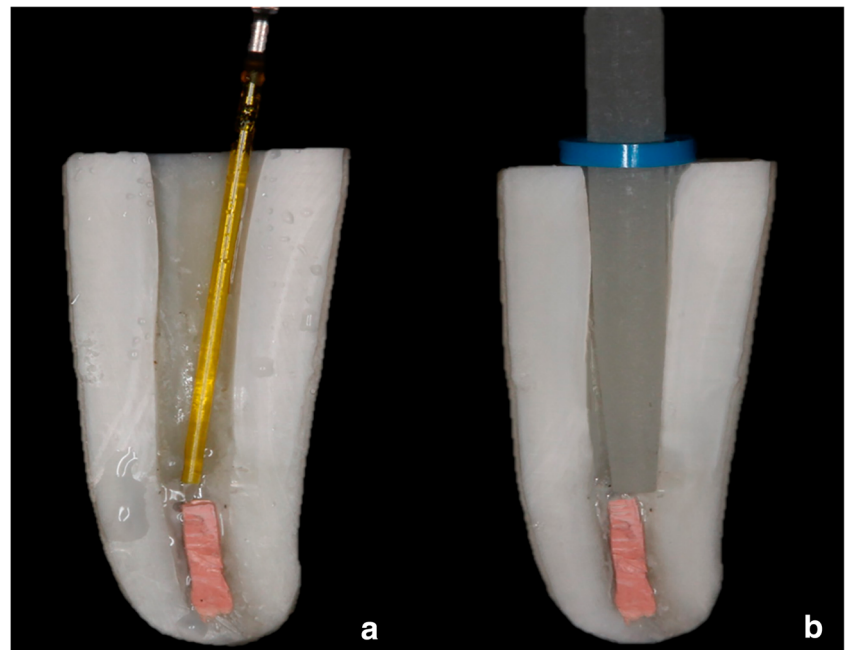
The push-out tests were performed using the universal testing machine (Instron Corporation), each *slice* was situated with its coronal side facing the metallic base. The posts were pushed out with cylindrical plungers with different diameter: cervical—1.9 mm, middle—1.7 mm, apical—1.5 mm. The plunger tip was positioned in such a way to touch only the post, avoiding any contact with the materials and dentin. The load was applied on the apical side of the root *slice* in an apical-coronal direction at a 1 mm/min crosshead speed, until the failure occurred. Bond failure was recorded by the extrusion of the post section from the root *slice*. In order to express the bond strength in MPa, the load at failure recorded in N was divided by the area of the bounded interface, which was calculated using the following equation: $a \frac{1}{4} 2\pi r h$ where p is the constant 3.14, r is the post radius, and h is the thickness of the *slice* in mm.

The failure type was evaluated by means of CLSM (LEXT 3D OLS 4000 laser microscope, Olympus, FAPESP grant 2011/12901-7) at $\times 50$ magnification. Failures observed after debonding were determined on a percentage basis and classified into four types: adhesive (between the relining material and dentin), cohesive in the relining material, cohesive in dentin and mixed (combination of adhesive and cohesive in the relining material).

Microhardness test

The specimens were ground wet with 600 and 1200-grit silicon carbide papers, polished with felt disks embedded in aluminum oxide paste at low speed, washed in running water, dried with gauze, and examined at $\times 40$ magnification to confirm smoothness. Dentin microhardness was measured with a Knoop indenter (Shimadzu HMV2) under 25-g load and 10-s dwell time. For each slice, three indentations were performed at each depth, starting from the root canal lumen towards the cement, located 30, 60, and 120 μm .

Fig. 1 **a** Er:YAG laser tip inserted into root canal (11 mm) and **b** glass fiber post adapted into root canal



CLSM analysis

The slices used in the microhardness test were treated with 37% phosphoric acid for 15 s and washed with water for 1 min. The specimens were analyzed under CLSM at $\times 5$ and $\times 50$ magnifications. The depth of resin cement penetration into the dentinal tubules were assessed using a four-grade score table 4 circumferential points (3, 6, 9, and 12 o'clock). Two calibrated examiners (interexaminer agreement = 0.88, kappa test) did a blind evaluation of the resin cement penetration into dentin (Table 2, Fig. 2).

Statistical analysis

Data of the push-out bond strength, microhardness and analysis of cement penetration were evaluated using to Statistical Package for the Social Sciences software (SPSS 19). The assumptions of equality of variances and normal distribution of errors were checked for all data. For bond strength data, two-way ANOVA and Tukey test were performed. For microhardness, two-way ANOVA was used. Non-parametric analysis (Kruskal-Wallis and Dunn tests) was performed for cement penetration. All tests significances were set at 5%.

Results

Push-out test

The data used for this analysis correspond to the force required for the displacement of the filling material of the specimen. The two-way ANOVA (superficial treatment \times thirds) revealed that the bond strength was significantly affected by the dentin pretreatment and root thirds ($P < .05$). The mean push-out bond strength values and standard deviation are shown in Table 3.

The surface treated with SA had greater resistance to displacement ($P < .05$), statistically similar ($P > .05$) to the Er:YAG laser and laser + SA. The Er:YAG laser followed by SA was similar to treatments alone ($P > .05$). Distilled water had the lowest value of adhesive strength ($P < .05$) and differed statistically from the other groups. Significant

differences were observed on root thirds ($P < .05$): cervical $>$ middle $>$ apical.

The analysis of failures after the push-out test showed that the most common mode was the adhesive failure between the self-adhesive resin cement and dentin. No cohesive failures were identified in the dentin (Table 4).

Microhardness test (KHN)

The microhardness mean and standard deviations for each treatment are shown in Table 5. No significant differences between groups and depths were found ($P > .05$).

Analysis of the penetration depth of resin-based cement to dentin

The results of the resin-based cement penetration into dentinal tubules are displayed in Table 6. The scores for cement adaptation to the dentin walls were mostly 3 and 4 (more than 50% of the adaptation) irrespective of the laser irradiation or ascorbate solution used. The Kappa analysis showed a strong agreement between examiners ($K > .8$). The Kruskal-Wallis and Dunn tests reveal statistically significant differences between the control and experimental groups ($P > .05$). All the pretreatments tested improved the resin cement penetration into the dentinal tubules, mainly in the cervical region of the root.

Discussion

Most failures involving the reconstruction of post-and-core restorations are due to the bond strength failure in resin-based cement and dentin interface [12, 13, 36–38]. The luting process relies on the formation of resin tags in the dentin and depends on the treatment of the dentinal surface [5, 39]. The different pretreatments on the intraradicular dentin aim to improve bond strength of glass fiber post and luting cement [16, 35, 40].

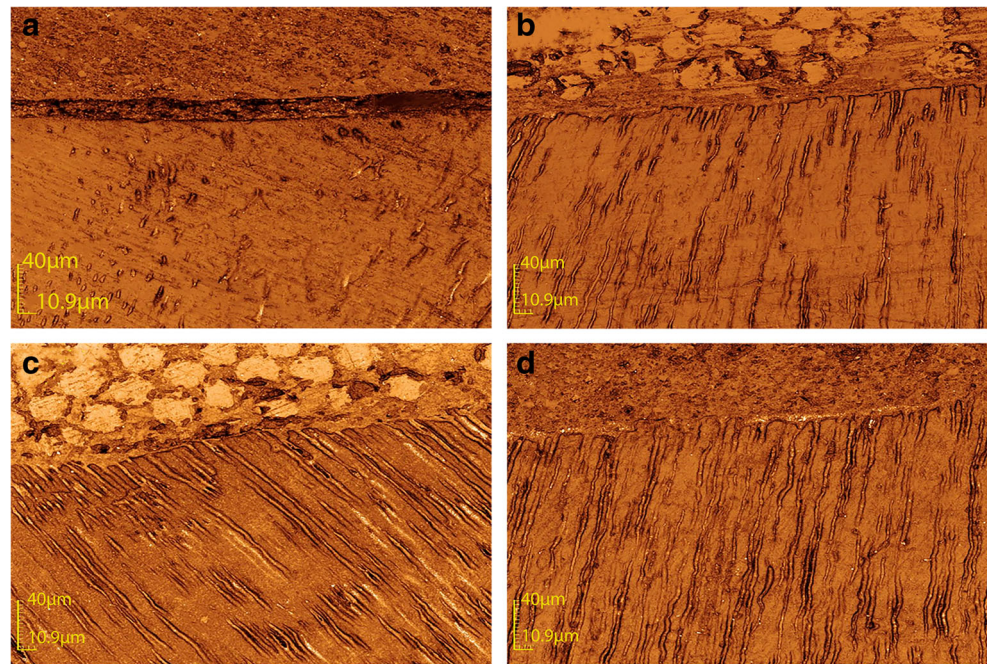
In this study, RelyX U200 self-adhesive resin cement (3 M ESPE) was used to lute the glass fiber post in the dentin, because it is a multistep bonding adhesive with low solubility [16] and reduces stress concentration in the interface [41].

Table 2 Penetration criteria of self-adhesive resin cement to canal walls

Score	Criteria for adaptation	Criteria sealer penetration
1	< 25% adaptation	< 25% of penetration/short tags, few or absent
2	25 to 50% adaptation	25 to 50% of penetration/short but uniform tag formation
3	51 to 75% adaptation	51% to 75 of penetration/long but sparse tags
4	> 75% adaptation	> 75% of penetration/long tags with dense formation

Scores described by Moura-Neto et al. [31]

Fig. 2 Representative CLSM images to penetration depths. **a** Score 1. **b** Score 2. **c** Score 3. **d** Score 4



The intraradicular dentin was treated with laser and/or antioxidant solution and the results revealed that the irradiation with Er:YAG laser, irrigation with 10% SA or both treatments combined improved the bond strength of cement-post-dentin interfaces.

SA is a biocompatible and nontoxic antioxidant (salt of ascorbic acid—vitamin C) able to reduce a wide variety of oxidizing compounds, mainly free radicals [24, 26]. The higher bond strength achieved in roots treated with 10% SA solution might be explained by the reduction in the residual free radicals from the interior of the dentinal tubules [25] and dentin matrix [9, 27]. SA reverses the denaturing effect of acid etching, NaOCl or H₂O₂ on dentin collagen [13, 14] and was found to be a potent inhibitor of matrix metalloproteinases (MMPs) [37], offering protection against the long-term degradation of the adhesive material-dentin interface [42].

Table 3 Mean and standard deviation results in MPa, of the maximum force of displacement the fiber pin of the specimen, according to the superficial treatment

Experimental groups	Mean and standard deviation
Distilled water	4.02 ± 2.39b
Sodium ascorbate	10.02 ± 5.45a
Er:YAG laser	9.91 ± 4.62a
Er:YAG laser + sodium ascorbate	8.09 ± 4.07a

Different letters indicate significant statistical difference (Tukey test, $p < .05$)

Khoroushi et al. [14] also indicated that the treatment with SA solution improved the bond strength of fiber posts to dentin.

Recently, Moura et al. [37] reported that the adhesion of self-adhesive cement seems to be associated with the presence of smear layer on the dentin surface and resin cements are more likely to be adversely affected by residuals free radicals. Probably for this reason, the roots without dentin pretreatment (control) had inferior bond strength results.

Laser technology is another alternative to clean the dentin surface, improve the wettability, and increase bond strength of resin-based materials to dentin [28, 29, 34]. According to the results of our study, Er:YAG laser irradiation in intracanal dentin (150 mJ, 4 Hz, and 40 s) improved the push-out bond strength of GFP. However, the effects of laser light in improving bond strength depend on the laser system and appropriate parameters should be selected [31]. Five microexplosions caused by Er:YAG laser irradiation eject organic and inorganic tissue, thereby cleaning dentinal walls [34]. This laser system is also able to expose the collagen fibers, create a rough surface, and thus allow the penetration of the resin-based material to dentin. Some authors have suggested the use of high power lasers for dentin pretreatment before luting a fiber post [34, 35].

Besides, Er:YAG laser offers a better penetration of adhesive material to inaccessible areas of the dentin tubular network, proving a stable mechanical interlocking between the cement and root dentin. Akcay et al. [44] reported that Er:YAG laser properly activates the irrigant

Table 4 Percentage distribution (%) of the failure patterns observed after extrusion bond strength

Failure mode	Distilled water	Sodium ascorbate	Er:YAG laser	Laser + sodium ascorbate
Adhesive (dentin)	27 (75%)	31 (85%)	33 (92%)	32 (88%)
Adhesive (cement)	4 (12%)	3 (9%)	2 (5%)	2 (6%)
Mixed post/cement	3 (7%)	2 (6%)	1 (3%)	2(6%)
Cohesive cement	2 (6%)	–	–	–

The total of samples analyzed on each group was 36 ($n = 12 \times 3$ thirds)

solution, creating a surface pattern advantageous for sealer penetration.

In the present study, we observed differences in the bond strength of root thirds. This was an expected finding since the length of resin tags decrease from cervical to apical within the root canal, due to the morphological reasons and diameter of the dentinal tubules, which are larger cervical than apically [43]. In the cervical third is easier to irradiate the dentin surface than in the deeper canal areas. The decrease in bond strength values from the coronal to the apical regions is in agreement with findings of previous studies [34, 44]. Besides, it can be speculated that irradiation is not homogeneous through the entire root canal due to some limitations: hand-scanning process and incidence angle of the laser beam with the dentin surface.

Under CLSM, we could observe the resin tags on cement/dentin interface. There are two types of resin tags that can be formed in dentin: non-hybridized and hybridized. Long resin tags might not be necessary to increase bond strength. In fact, long or short hybridized tags (resin-based material that completely seal the dentinal tubules entrance) produce a good hybrid layer and this is the important point for adhesion and microleakage [45].

The bond failure location and mode provide information about the quality of the bond between the tooth and the adhesive. The analysis of failures after the push-out test revealed that the adhesive mode was predominant in all experimental groups, emphasizing the fragility of the

bonding interface. This finding is in accordance with other studies [14, 31, 37].

The microhardness of the dentin represents the quantity of calcified mineral matrix in a square millimeter of dentin [46]. The results of our study demonstrated that the microhardness of dentin adjacent to the root canal was not affected by the different surface pretreatments. On these concerns, the type of teeth might affect the results. Marcelino et al. [47] reported that 10% SA for 10 min reduces the intracanal dentin microhardness of human teeth and Oskoe et al. [46] found that 10% SA for 30 min did not alter the microhardness of bovine teeth. Regarding laser treatment, Chinelati et al. [48] ascertain that Er:YAG laser used for cavity preparation did not influence the dentin microhardness. However, to the best of our knowledge, no previous investigation assessed the microhardness around the root canal filling pretreated with Er:YAG laser.

In the present study, we could not confirm a synergistic effect with the improvement in bond strength on roots treated with Er:YAG laser combined to SA irrigation as compared to treatments alone. Nevertheless, both pretreatments have different mechanisms of action and distinct positive effects on dentin. It seems a good adhesive strategy to use laser light or an antioxidant agent before luting a GFP into root canal. Further studies in vitro and clinical trial should be conducted to evaluate the potential of other antioxidants combined with laser irradiation on tooth surfaces.

Table 5 Mean and standard deviation results in KHN of microhardness, according to the superficial dentin

Experimental groups	Mean and standard deviation
Distilled water	53.23 ± 8.60
Sodium ascorbate	54.19 ± 4.08
Er:YAG laser	61.65 ± 6.52
Ascorbate + laser	62.99 ± 3.40

ANOVA revealed no significant differences among groups ($P > .05$)

Table 6 Frequency distribution of scores of resin cement into root thirds of dentin ($n = 10$)

Groups	Scores				
	1	2	3	4	
Distilled water	3	3	2	2	b
Sodium ascorbate	0	1	5	4	a
Er:YAG laser	0	3	4	3	a
Sodium ascorbate + Er:YAG laser	0	2	4	4	a

Different letters denote statistically significant differences between groups

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Compliance with ethical standards

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

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