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Laboratory Research

Effect of Desensitizing Agents on the Microtensile Bond Strength of a Two-step Self-etch Adhesive to Dentin

HD Arısu • E Dalkıhç • MB Üçtaşlı

Clinical Relevance

Adhesive composite resin restorations may be performed after dentin hypersensitivity treatment procedures. However, the effect of desensitizers on the bond strength of adhesive restorations is controversial.

SUMMARY

The aim of this *in vitro* study was to evaluate the effect of cervical hypersensitivity treatments (neodymium yttrium aluminum garnet [Nd:YAG] laser and conventional techniques) on the microtensile bond strengths of adhesives to treated dentin. The buccal cervical enamel of 42 freshly extracted human mandibular third molars was ground flat to expose the cervical dentin. The dentin surfaces were polished with a series of silicon carbide papers, and the smear was removed with an ethylenediamine tetra-acetic acid solution. The teeth were randomly divided into six groups as follows: group 1, Vivasens; group 2, BisBlock; group 3, fluoride gel; group 4, Nd:YAG laser; group 5, Clearfil SE + Nd:YAG laser; and group 6, no treatment (control). The specimens were then restored with a two-step self-etch adhesive, with the exception of group 5. Five specimens from each group were restored with a nanohybrid composite resin. The adhesive interface of two specimens from each group was examined using scanning electron microscopy. The specimens were sectioned perpendicularly to the adhesive interface to produce beams (adhesive area 1 mm^2). The

Hacer Deniz Arısu, DDS, PhD, associate professor, Gazi University Faculty of Dentistry, Operative Dentistry and Endodontics, Ankara, Turkey

*Evrim Dalkıhç, DDS, PhD, assistant professor, Yüzüncüyıl University, Faculty of Dentistry, Department of Operative Dentistry and Endodontics, Van, Turkey

Mine Betül Üçtaşlı, DDS, PhD, professor, Gazi University, Faculty of Dentistry, Department of Operative Dentistry and Endodontics, Ankara, Turkey

*Corresponding author: Yüzüncüyıl University Faculty of Dentistry, Department of Operative Dentistry and Endodontics, KAMPUS, VAN, 65080, Turkey 0505 6878137; e-mail: eeliguzeloglu@yahoo.com

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beams were then attached to a microtensile tester and stressed to failure at 1 mm/min. The data were compared using one-way analysis of variance at a significance level of 0.05. The microtensile bond strengths of the control group were significantly higher than those found for group 1, group 2, group 3, and group 4 ($p < 0.05$). No significant difference was found between group 5 and the control group. Most of the premature failures were seen in group 2 (80%), and the fewest premature failures were seen in group 5 (13.3%). The SEM findings verified the microtensile test findings. In conclusion, desensitizing treatment procedures (with the exception of Clearfil SE + Nd:YAG laser) reduced the microtensile bond strength of a two-step self-etch adhesive to dentin.

INTRODUCTION

Dentin hypersensitivity is characterized by short, sharp, pain arising from exposed dentin in response to stimuli (thermal, tactile, osmotic, evaporative, chemical) that cannot be explained by any dental defect or pathology.[1] This clinical problem may arise as a result of loss of enamel and/or root surface denudation with exposure of underlying dentin.[2] Exposure of dentin is closely related to dentinal hypersensitivity.[3] Periodontal disease, periodontal treatment, and improper brushing habits can also result in gingival recession contributing to sensitive teeth.[4]

Dentin hypersensitivity can be treated with invasive (gingival surgery, pulpectomy, application of resins, laser) and noninvasive (topical agents and dentifrices that contain a desensitizing ingredient) procedures. Noninvasive treatment options are considered to be the simplest, most cost-effective, and most efficacious first line of treatment for most patients.[5] Topical agents containing fluoride, oxalate, potassium nitrate, and calcium phosphate occlude dentinal tubules and decrease the permeability of dentin.[6-11] In addition to these methods, lasers are playing an important role in treating dentinal hypersensitivity, including the helium neon (He-Ne) laser, gallium aluminum arsenium (Ga-AlAs) laser, neodymium yttrium aluminum garnet (Nd:YAG) laser, carbon dioxide (CO_2) laser, and erbium yttrium aluminum garnet laser.[12] The effectiveness of lasers for treating dentin hypersensitivity varies from 5% to 100%, depending on the type of laser and the treatment parameters.[13]

Adhesive composite resin restorations may be performed after dentin hypersensitivity treatment procedures. However, the effect of desensitizers on the bond strength of adhesive restorations is controversial. Pashley and others[14] reported that dentin surfaces were less favorable bonding substrates after using desensitizing agents.

The aim of this *in vitro* study was to determine the effect of different dentin hypersensitivity treatment procedures on the microtensile bond strength of a two-step self-etch adhesive resin and a nanofill composite resin to dentin.

MATERIALS AND METHODS

Forty-two freshly extracted noncarious human mandibular third molar teeth were cleaned of tissue remnants and stored in 0.2% thymol solution. The teeth were cleaned using a slurry of pumice and water with a slowly rotating rubber cup. The experimental setup is schematically presented in Figure 1. Each of the buccal cervical enamel surfaces was ground with a series of silicon carbide disks (numbers 600, 800, 1000, 1200) under water coolant until the cervical dentin was exposed. Dentin surfaces were treated with 17% ethylenediamine tetra-acetic acid (EDTA) for 15 seconds to remove the smear layer. The exposed surfaces were treated with 2.5% NaOCL; for 5 seconds to stop the effect of EDTA and rinsed with distilled water. Teeth were randomly divided into six treatment groups (Table 1):

- •Group 1: Dentin surfaces were treated with Vivasens (Ivoclar Vivadent AG, Schaan, Liechtenstein) solution for 10 seconds and gently air dried for 10 seconds.
- •Group 2: Dentin surfaces were etched with 37% phosphoric acid for 10 seconds, rinsed with water, and gently air dried. Bis-Block (Bisco Inc, Schaumburg, IL, USA) solution was then applied for 30 seconds and rinsed with water.
- •Group 3: Dentin surfaces were treated with fluoride gel for 4 seconds (Dental Medical, Conegliano [TV], Italy).
- •Group 4: Dentin surfaces were treated with Nd:YAG laser (120 mJ, 20 Hz, 2.4 W) for approximately 10 seconds at 1 mm distance from the surface in a circular motion to scan the entire dentin.
- •Group 5: Dentin surfaces were treated with Clearfil SE primer and bond (Kuraray, Osaka, Japan) according to the manufacturer's instructions then Nd:YAG laser (120 mJ, 20 Hz, 2.4 W) was applied for 10 seconds at a 1 mm distance in a circular motion

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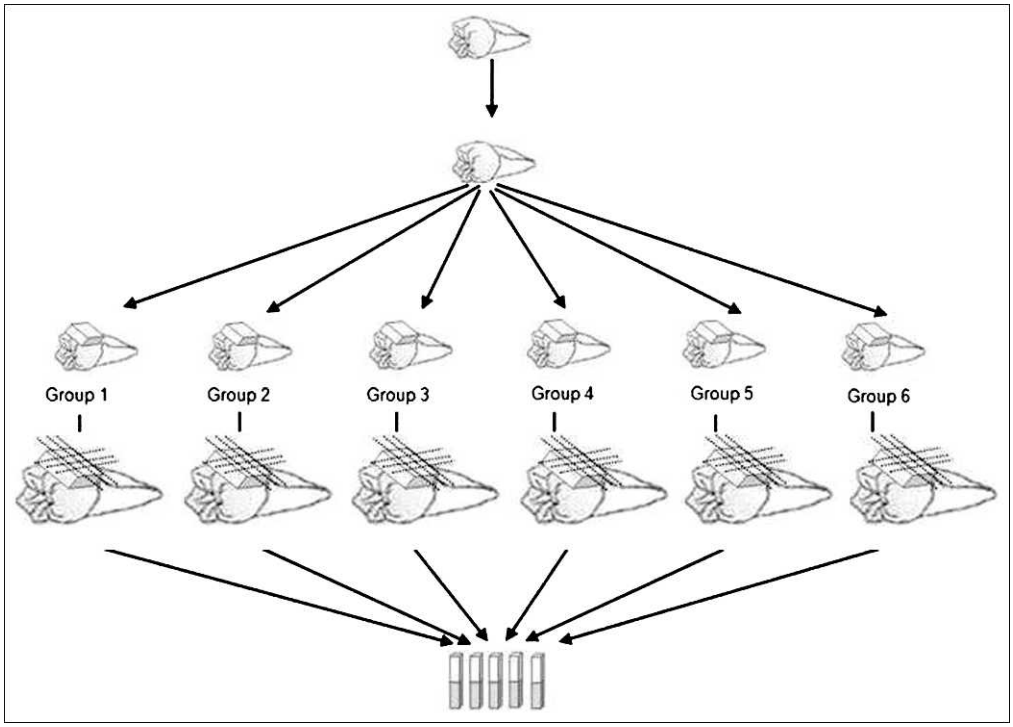


Figure 1. *Schematic representation of the study design.*

before being light cured for 10 seconds with a halogen light-curing unit (LCU; Hilux Ultra Plus, Benlioğlu, Ankara, Turkey). The irradiance of the halogen LCU was 600 mW/cm^2 with a wavelength of 450–520 nm. Light intensity was measured with a radiometer (Hilux Curing Light Meter, Benlioğlu, Ankara, Turkey).

- Group 6: No treatment was applied to dentin surfaces; this group served as the control.

After the desensitizing treatment, five teeth from each group were randomly selected and Clearfil SE primer and bond was applied to the dentin according to the manufacturer's instructions. All groups were restored with Filtek Supreme (3M ESPE, St Paul, MN, USA).

Five teeth from each group were embedded in acrylic blocks, horizontally to the long axis of the tooth, leaving the buccal surfaces facing up, and were placed on a low-speed (300 revolutions per minute) cutting device (Mecatome T201A, Pressi, Grenoble, France) to produce 1mm^2 adhesive surface area beams under water cooling. The beams were then attached with cyanoacrylate adhesive to a testing apparatus, and a tensile load was applied with a microtensile tester (Micro Tensile Tester T-61010 K, Bisco, Schaumburg, IL, USA) at a cross-head speed of 1.0 mm/min, until fracture.

Fracture surfaces were observed using a stereomicroscope (SZ-PT Olympus, Tokyo, Japan) at a magnification of 30× to determine the failure modes, which were classified as adhesive, cohesive, and mixed.

Results were expressed in megapascals (MPa) and the data were submitted to a one-way analysis of variance (ANOVA) and Tukey tests at a preset α of 0.05. Premature failures were recorded as zero values and were excluded in the statistical analyses.

The remaining two teeth from each group were sectioned perpendicularly to the bonding interface between the desensitizer and dentin. The sections were then coated with gold for scanning electron micrograph (SEM) examinations.

RESULTS

Mean microtensile bond strength values and significant differences between the groups are shown in Table 2. Between the groups, group 2 showed the lowest microtensile bond strength, and group 6 showed the highest bond strength values. When the one-way ANOVA and Tukey test ranked the mean microtensile bond strength values of groups, no significant difference was determined between group 1 and group 3 (p=0.823), group 1 and group 4 (p=0.984), group 1 and group 5 (p=0.095), group 2

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Table 1: *Materials and Groups That Were Examined in This Study*

Groups	Desensitizer	Adhesive	Composite
1	VivaSens (Ivoclar Vivadent)	Clearfil SE Bond (Kuraray)	Filtek Supreme (3M ESPE)
2	BisBlock (Bisco)	Clearfil SE Bond (Kuraray)	Filtek Supreme (3M ESPE)
3	Fluoride gel (Dental Medical)	Clearfil SE Bond (Kuraray)	Filtek Supreme (3M ESPE)
4	Nd:YAG laser (American Dental Technologies)	Clearfil SE Bond (Kuraray)	Filtek Supreme (3M ESPE)
5	Clearfil SE + Nd:YAG laser (American Dental Technologies)	-	Filtek Supreme (3M ESPE)
6	Control	Clearfil SE Bond (Kuraray)	Filtek Supreme (3M ESPE)

and group 3 (p=0.051), group 3 and group 4 (p=0.994), and group 5 and group 6 (p=0.728) (Table 3). There were significant differences between group 1 and group 2 (p=0.007), group 1 and group 6 (p=0.010), group 2 and group 4 (p=0.024), group 2 and group 5 (p=0.000), group 2 and group 6 (p=0.000), group 3 and group 5 (p=0.000), group 3 and group 6 (p=0.000), group 4 and group 5 (p=0.004), and group 4 and group 6 (p=0.000) (Table 3). Most of the premature failures were seen in group 2 (80%), and the fewest premature failures were seen in group 5 (13.3%). The distribution of failure modes is presented in Table 4. Bond failures were determined to be largely adhesive and mixed.

The microtensile bond strength results were supported with the SEM findings.

In group 1, clear resin tags were found and no distortion or separation was seen between the resin-dentin bonding area (Figures 2 and 3). When the adhesive interface was examined in group 2, rare and thin resin tags were seen (Figures 4 and 5). Additionally many separated areas were observed in the resin-dentin interface and most of the dentinal tubules were occluded (Figure 4). In group 3 and group 4, thin resin tags were seen (Figures 6 and 7); however, some regions of distortion and separations were observed (Figures 8 and 9). In group 5, long resin tags and a continuous hybrid layer were seen (Figures 10 and 11). In group 6, the dentinal tubules and hybrid layer were seen clearly. Many thick resin tags were also observed (Figures 12 and 13).

DISCUSSION

Dentinal hypersensitivity is a problem that irritates many dental patients. When a patient has dentinal hypersensitivity symptoms, he or she should be examined and informed of the multiple treatment options available to solve the problem. The initial treatment choice is to cover the tubules with topical agents to desensitize the nerves or interfere with the transmission of the pain signal at the synapse.[2]

In this current study, three topical agents (Vivasens, BisBlock, fluoride gel) were used for desensitizing. Studies have reported that these agents desensitize by occluding the dentinal tubules.[2,4] Fluorides decrease the permeability of dentin, possibly by precipitation of insoluble calcium fluo-

Table 2: *Mean Microtensile Bond Strength Values, Standard Deviations of Groups and Significant Differences Between Them*

Group	n′/n	Mean and Standard Deviations
1 (Vivasens + Clearfil SE)	10/30	8.17 ± 5.43 ^{A}
2 (BISBlock + Clearfil SE)	6/30	0.90 ± 2.22 ^{B}
3 (Fluoride gel + Clearfil SE)	18/30	6.28 ± 2.89 ^{ABD}
4 (Nd:YAG laser + Clearfil SE)	13/30	7.06 ± 3.22 ^{AD}
5 (Clearfil SE + Nd: YAG laser)	26/30	12.05 ± 2.62 ^{AC}
6 (Control) (no treatment + Clearfil SE)	13/30	13.91 ± 6.43 ^{C}

• *n′: tested specimens, n: total number of specimens*
• *Capital letters identify statistically similar groups.*

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Table 3: P *values of the Groups*

	1	2	3	4	5	6
1	-	0.007	0.823	0.984	0.095	0.010
2	0.007	-	0.051	0.024	0.000	0.000
3	0.823	0.051	-	0.994	0.000	0.000
4	0.984	0.024	0.994	-	0.004	0.000
5	0.095	0.000	0.000	0.004	-	0.728
6	0.010	0.000	0.000	0.000	0.728	-

ride within the tubules. Vivasens contains potassium fluoride, which comes into contact with the dentinal fluid. Precipitation of calcium ions and proteins in the dentinal fluid block the tubules. Another property of this agent is the low pH value (pH 2-3), which accelerates the penetration and transport of the active ingredients deep into the tubules.[15] Because of this acidic property, excellent bond strength of adhesives to these surfaces can be obtained. In the current study, although clear resin tags and a continuous hybrid layer without distortion or separation were seen in the Vivasens group (group1), the bond strength values were found to be lower than that of the control group (group 6). Fluoride gel contains 33% sodium fluoride, and it desensitizes by occluding the dentinal tubules. This tubule occluding effect can be clearly seen in the resin-dentin interface of this group. Thin and rare resin tags,

Table 4: *Failure Modes of Groups*

Group	Adhesive	Cohesive in Dentin	Cohesive in Composite	Mixed
1	6	-	2	2
2	6	-	-	-
3	17	-	-	1
4	12	-	-	1
5	25	-	-	1
6	13	-	-	-

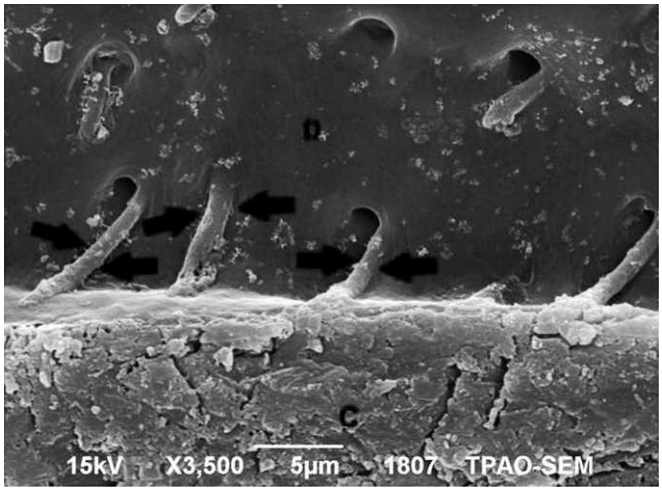


Figure 2. *SEM image (3500×) of group 1. Resin tags (arrows) are seen. D, dentin; C, composite.*

distortions in the hybrid layer, and lower microtensile bond strengths may be the result of tubule occlusion.

Oxalate materials also work well for desensitization.[16] They react with calcium ions on dentin and in dentinal fluid to form insoluble calcium oxalate crystals.[17,18] Calcium oxalate crystals occlude open tubules in the cervical dentin.[19] In contrast with other oxalate desensitizers, with BisBlock's patented technique the total-etch procedure occurs before oxalate and adhesive placement.[20] This technique provides a durable effect because calcium is removed from the surface and oxalate crystals form deep within the dentinal tubules.[21] Yiu and others[22] reported that oxalate desensitizers did not negatively affect the bond strength of adhesives, such as

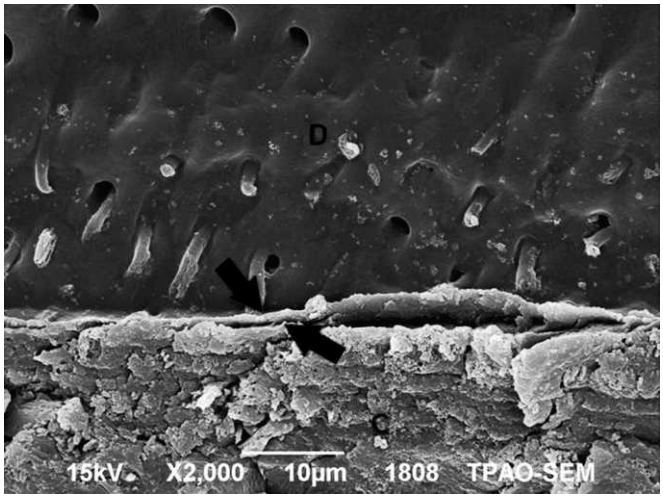


Figure 3. *SEM image (2000×) of group 1. D, dentin; C, composite.*

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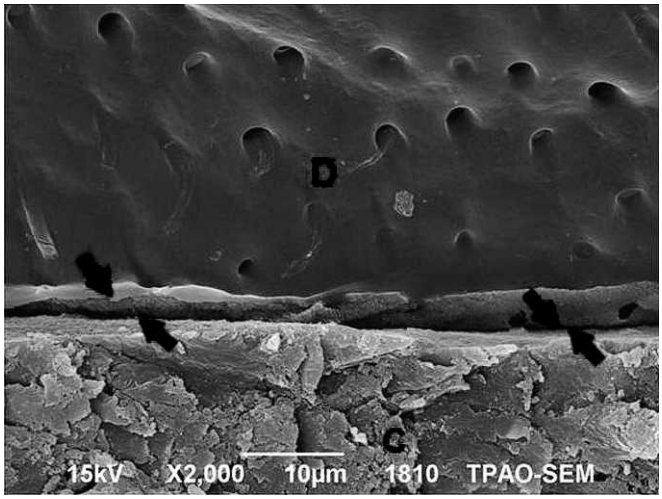


Figure 4. *SEM image (2000×) of group 2. There is no resin tag in resin-dentin interface. Distortions and separations (arrows) are seen in the hybrid layer. D, dentin; C, composite.*

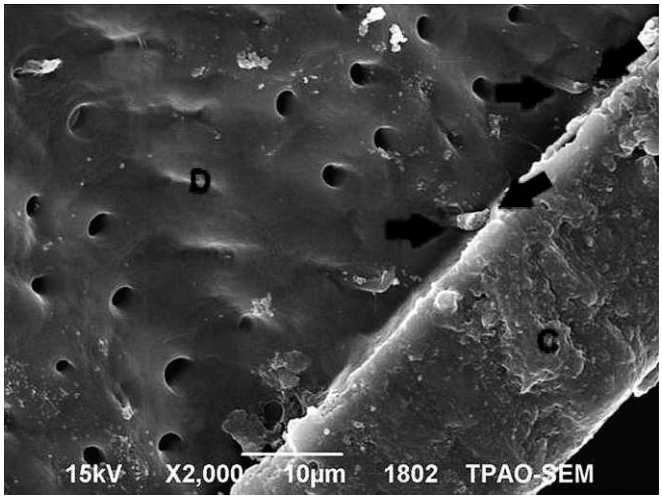


Figure 6. *SEM image (2000×) of group 3. Thin and rare resin tags (arrows) are seen. D, dentin; C, composite.*

Single Bond (3M ESPE) or One Step (Bisco Inc). However, Pashley and others[14] reported reduced bond strength because of crystal precipitation on the dentin surface. Pashley and others[14] and Tay and others[20] reported that when oxalates were used on acid-etched cavities that contained enamel margins, the enamel surfaces became covered with calcium oxalate crystals. A brief (10–15 second) acidic etch could dissolve apatite crystals beneath the acid-resistant oxalate crystals and leave etched enamel ready for resin infiltration after the oxalate crystals fall off.

Contrary to previous studies the two step self-etch adhesive system presented the lowest bond strength values when it was used after oxalate desensitizing agent (BisBlock). This may be because the Clearfil SE primer did not remove crystal precipitation on the dentin surface. In addition, the highest premature failure was seen in this group. This highest premature failure percentage showed the slight adhesion between resin and dentin that was seen clearly in the SEM analysis. Separations and distortions and thin and rare resin tags in the resin-dentin interface may explain the lowest microtensile bond strength results. The manufacturer[21] claims that when BisBlock is applied on a decalcified (by acid etching and rinsing) dentin surface calcium oxalate crystals are formed only within the tubules, leaving the dentin surface unobstructed to readily accept the adhesive for ideally bonded restoration.

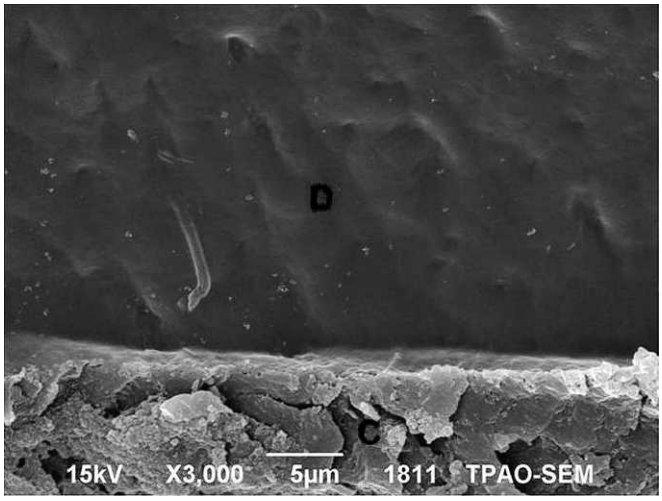


Figure 5. *SEM image (3000×) of group 2. D, dentin; C, composite.*

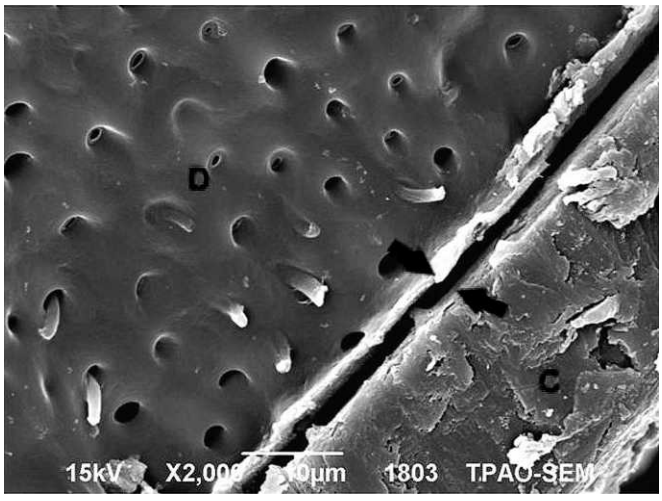


Figure 7. *SEM image (2500×) of group 4. Thin and rare resin tags (arrows) are seen. D, dentin; C, composite.*

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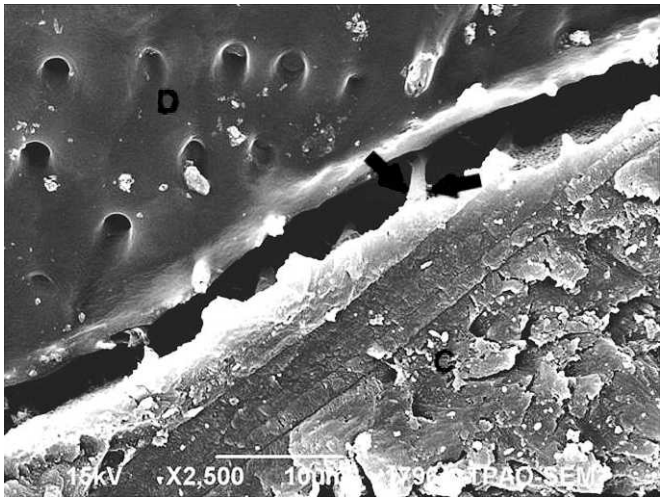


Figure 8. *SEM image (2000×) of group 3. Distortions and separations (arrows) are observed in the resin-dentin interface. D, dentin; C, composite.*

Findings of the current study, however, contradict the manufacturer reports and show that the two-step self-etch adhesive did not bond ideally to dentin surfaces. Because of this BisBlock cannot be recommended on dentin surfaces before the placement of direct restorations.

Lasers used for the treatment of dentin hypersensitivity are divided into two groups: low output power (low-level) lasers (He-Ne, GaAlAs) and middle output power lasers (Nd:YAG, CO_2).[13] The mechanism of the Nd:YAG laser's effect on dentin is caused by thermal energy absorption.[23] The hydroxyapatite crystals of dentin melt partly or completely, move, and increase in size once the activation energy is sufficient. Finally, the dentinal tubules become occluded. The sealing depth was reported to be 4μm.[24]

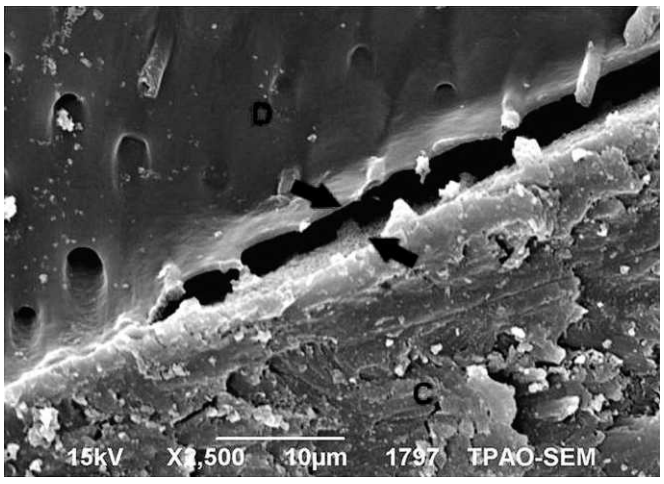


Figure 9. *SEM image (2500×) of group 4. Separations (arrows) are seen in the hybrid layer. D, dentin; C, composite.*

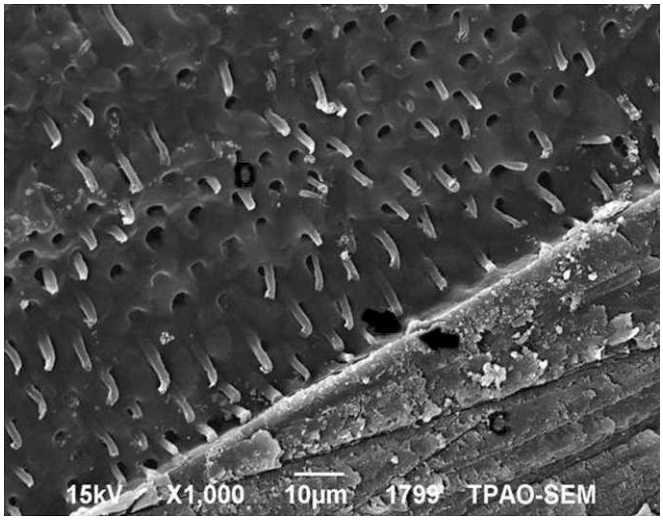


Figure 10. *SEM image (1000×) of group 5. Continuous hybrid layer (arrows) is seen clearly in the resin-dentin interface. D, dentin; C, composite.*

In the current study, a Nd:YAG laser was used in two groups. In group 4, the Nd:YAG laser was applied to the dentin surfaces for desensitizing. The bond strength values of the two-step self-etch adhesive were lower in group 4 than in the control group, which may be because of the mechanism of the Nd:YAG laser. The Nd:YAG laser treates hypersensitivity by narrowing the dentin tubules,[25,26] which may inhibit adhesion between resin and dentin. SEM observations of group 4 (thin resin tags and separations in the hybrid layer) supported the lower bond strength values. In group 5, the Nd:YAG laser was applied to the bonding surfaces before polymerization to increase the flow and diffusion of the resin monomer into the dentin tubules.[27] In group 5, the

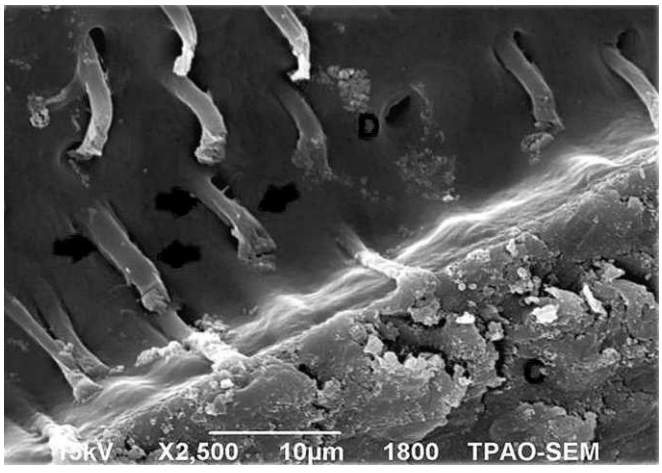


Figure 11. *SEM image (2500×) of group 5. Long resin tags (arrows) are seen. D, dentin; C, composite.*

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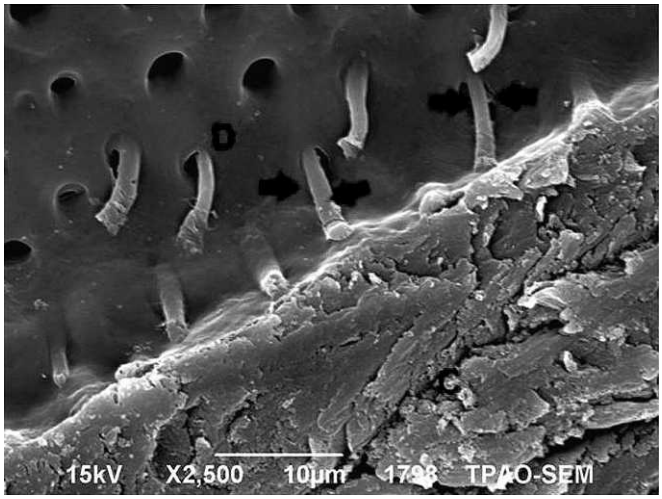


Figure 12. *SEM image (2500×) of group 6. Long resin tags (arrows) are seen. D, dentin; C, composite.*

bond strength values of the two-step self-etch adhesive were similar to those of the control group. In addition, many long resin tags were observed in the resin dentin interface, indicating adequate adhesion between resin and dentin.

This *in vitro* study was done with extracted teeth without simulating dentinal fluid pressure, so it is difficult to compare the results with clinical conditions. In clinical conditions when dentin is exposed to the oral cavity dentinal fluid flows from the pulp to exposed dentin because of the interstitial fluid pressure in the pulp. Some studies have reported that dentinal fluid flow effected the diffusion of adhesive resins into dentinal tubules.[28-30]

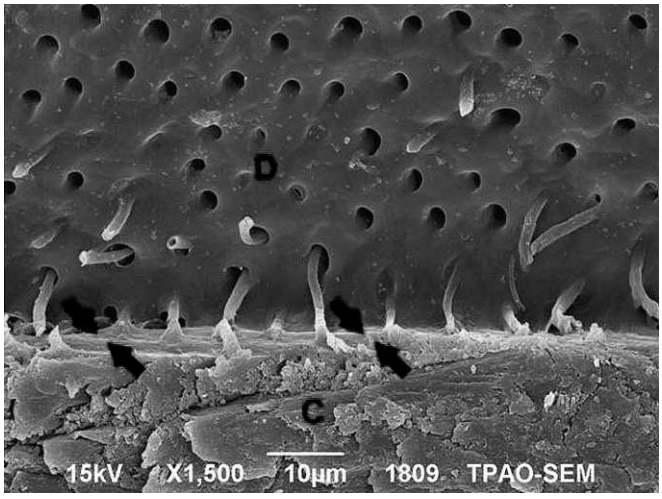


Figure 13. *SEM image (1500×) of group 6 (control). Continuous hybrid layer (arrows) is seen in the resin-dentin interface. D, dentin; C, composite.*

Within the limitations of this current study, desensitizing treatment procedures (except Clearfil SE + Nd:YAG laser) reduced the bond strength of a two-step self-etch adhesive to dentin. This may be due to the obliteration or narrowing of dentinal tubules. Hypersensitive dentin, which is treated with topical agents and laser, is a less favorable bonding substrate than normal dentin.

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