

Chlorhexidine's Effect on Sealing Ability of Composite Restorations Following Er:YAG Laser Cavity Preparation

Vinicius R. Geraldo-Martins, DDS, MS; Fabio R. P. Robles, DDS, MS;
Adriana B. Matos, DDS, MS, PhD



Abstract

Aim: The aim of this study was to evaluate the influence of chlorhexidine on microleakage of composite restorations in class V cavities prepared with a carbide bur (CB) and a erbium:yttrium-aluminum-garnet (Er:YAG) laser.

Methods and Materials: Cavity preparations were created on the buccal surfaces of 48 bovine incisors using different methods then equally dividing them into four groups. Groups I and II were prepared with a #56 CB in a high speed handpiece while Groups III and IV were prepared with a Er:YAG laser at 350m J/4Hz on enamel and at 80.24 J/cm² on dentin. The cavity preparations were rinsed with 2% chlorhexidine solution before the application of Clearfill SE Bond system and placement of a Z100 composite resin restoration. After 24 hours, the teeth were polished, thermocycled, and sealed with cosmetic varnish. The samples were then immersed in 2% methylene blue for four hours and sectioned in the buccolingual direction to determine the degree of microleakage.

Results: After statistical analysis (Kruskal-Wallis, $p < 0.05$), no significant differences were verified among the tested groups and between the evaluated cervical or incisal margins of the restoration.

Conclusion: The use of the chlorhexidine did not interfere with the adhesion process regardless of whether a CB or the Er:YAG laser were used to prepare class V cavities when restored with the tested self-etching adhesive system.

Keywords: Chlorhexidine, Er:YAG laser, self-etch primer, adhesion, microleakage

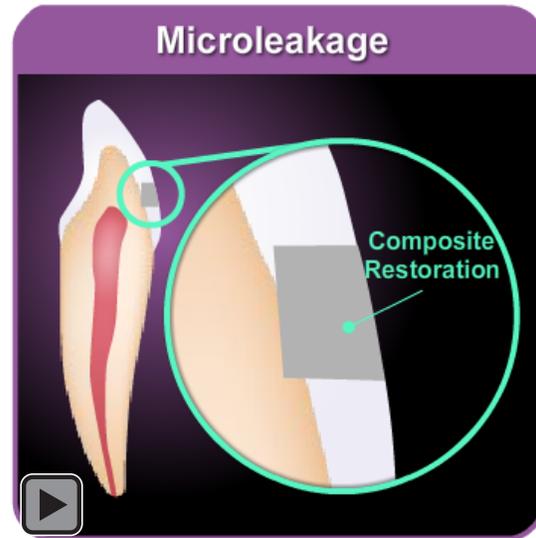
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Introduction

The first erbium:yttrium-aluminum-garnet (Er:YAG) laser for use in humans to prepare dental cavities and for soft tissue applications was approved in the United States by the Food and Drug Administration (FDA) in 1997. The emission wavelength of 2.94 μm coincides with the main absorption peak of water and hydroxyapatite resulting in a good absorption in all biological tissues. Its usage in dental hard tissues as well as in carious lesions results in effective ablation without thermal injury to the surrounding hard tissues such as carbonization or crack formation.¹

Clinical studies have demonstrated the Er:YAG laser system is more comfortable for patients than conventional methods of cavity preparation.² A cavity prepared with the Er:YAG laser does not produce a smooth surface. Scanning electron microscope (SEM) analysis of lased enamel revealed an irregular surface of grooves, flakes, shelves, and pits. In addition, lased dentin lacked a smear layer and the orifices of dentinal tubules were exposed.³ Due to these characteristics, most of these cavities are restored with composite resin in conjunction with a bonding system. As a result, an investigation to determine the adhesion between an Er:YAG laser-irradiated surface and composite resin is very important in order to ensure the best clinical result.

A gap between the tooth surface and restorative material is one of the primary causes of failure of a restoration. A marginal gap can lead to formation of secondary caries when cariogenic bacteria seep under restorations. Although the long-term fate of residual bacteria is as yet unknown, results of investigations have shown the presence of bacteria in dentin after the removal of infected dentin as well as dye-stainable dentin.^{4,5} The use of disinfectant solutions to cleanse cavity preparations is an alternative to reduce or eliminate bacteria from cavity preparations.⁶ Some antibacterial solutions such as: chlorhexidine, sodium hypochlorite, and fluoride solutions have been evaluated, but studies have reported adhesion could be impaired by a series of previous dentin treatments. Results of these *in vitro* studies in permanent teeth are controversial with regard to whether the use of disinfectants in cavity preparations affect adhesion.^{7,8,9,10,11}



To avoid gap formation between restorative material and the dental surface, a good interaction between an adhesive system and the restorative material as well as between the bonding agent and dentin/enamel surface are necessary. The quality of dentin bonding is thought to be determined by the demineralization depth, diffusion extent, and monomer impregnation into the calcium-depleted zone of etched dentin. When dentin demineralization depth exceeds the dentin diffusion/impregnation depth, an area of hydroxyapatite-depleted collagen fibers is left exposed and inadequately infiltrated or hybridized.^{12,13} This zone of exposed collagen may be unstable and subjected to hydrolysis.^{14,15} Self-etching dentin bonding systems that do not require smear-layer removal by acid etching are being developed to avoid this situation. In theory these self-etching systems simultaneously decalcify the inorganic component of dentin and infiltrate collagen fibers at the same time through the action of acidic primers which minimize the potential for voids.¹³ The clinical procedure is less complicated and time consuming because there is no need for rinsing.

Self-etching primers are mainly aqueous mixtures of acidic monomers like phosphate ester or carboxylic acid and hydroxyethyl methacrylate. The latest commercially available self-etching primer incorporates resin monomers, a photoinitiator, and a tertiary amine accelerator into a single bottle which eliminates an additional

mixing step.^{16,17} The combination of etchant, primer, and adhesive resin into an all-in-one adhesive is advantageous because it shortens the application time and reduces errors that can occur at each bonding step.¹³

To date there have been no reports on the influence of chlorhexidine solution used as a cavity preparation disinfectant on the microleakage of cavities prepared with an Er:YAG laser and conditioned with a self-etching bonding system. Therefore, the aim of the present study was to evaluate the influence chlorhexidine has on the microleakage of Er:YAG laser prepared cavities.

Methods and Materials

Forty-eight bovine central incisors from freshly killed two to four year-old slaughterhouse animals were used in this study. After extraction, the teeth were cleaned with a water pumice slurry and the roots were removed below the cementoenamel junction then stored in a 0.1% thymol solution for up to one week at 4°C. Forty-eight class V cavities with occlusal margins in enamel and cervical margins located 1.0 mm above the cementoenamel junction were prepared on the buccal surfaces. Cavity dimensions were standardized utilizing a template to trace an outline with mesiodistal width and occlusogingival dimensions of 2 mm onto the buccal surface. The depth of the cavity was 1.5 mm as measured with a marked periodontal probe. The specimens were randomly assigned to four groups (n=12 per group).

For Groups I and II, cavities were prepared using a #56 carbide bur (CB) (Jet Equipment & Tools, Vancouver, Canada) in a high speed handpiece with air/water spray. New burs were used after every five preparations. Bevels were not placed on enamel margins. Each preparation was rinsed

for 20 seconds with a water spray and dried with compressed air for five seconds.

For Groups III and IV, cavities were prepared using a Kavo Key Laser 3, a pulsed Er:YAG laser, (Kavo Co., Biberach, Germany). An output energy of 350 mJ and a repetition rate of 4 Hz (112.34 J/cm²) was used for enamel and an output of 250 mJ and 4Hz (80.24 J/cm²) was used for dentin under a water spray coolant (5 ml/min). Each preparation was rinsed for 20 seconds with air/water spray and dried with compressed air for five seconds.

After preparation, Groups II and IV cavities were rinsed with a 2% chlorhexidine digluconate cavity cleanser (Cav-Clean, Herpo, Brazil). This solution was applied for 40 seconds according to the manufacturer's instructions and dried with absorbent paper (Table 1).

Following cavity treatment, Clearfil SE Bond self-etching bonding system (KURARAY Co. Ltd., Osaka, Japan) was applied to cavities in all four groups according to the manufacturer's instructions. Primer was applied for ten seconds and dried gently. The bonding agent was then applied and light-cured at 500 mW/cm² (Ultralux Eletrônica, Dabi-Atlante, Brazil) for ten seconds.

Cavities were restored with shade A4 Z100 hybrid resin composite (3M Dental Products, St. Paul, MN, USA) inserted in three increments. The first increment was placed against the gingival wall and light-cured for 40 seconds. Excess materials were removed with a high speed 170 bur. The specimens were placed in distilled water at 37°C for 24 hours after being finished and polished using the Sof-Lex disk system (3M Dental Products Division, St. Paul, MN, USA).

Table 1. Description of the four group procedures.

Groups	Cavity Preparation	Cavity Disinfection
I	No. 56 Bur	No
II	No. 56 Bur	Yes
III	Er:YAG Laser	No
IV	Er:YAG Laser	Yes

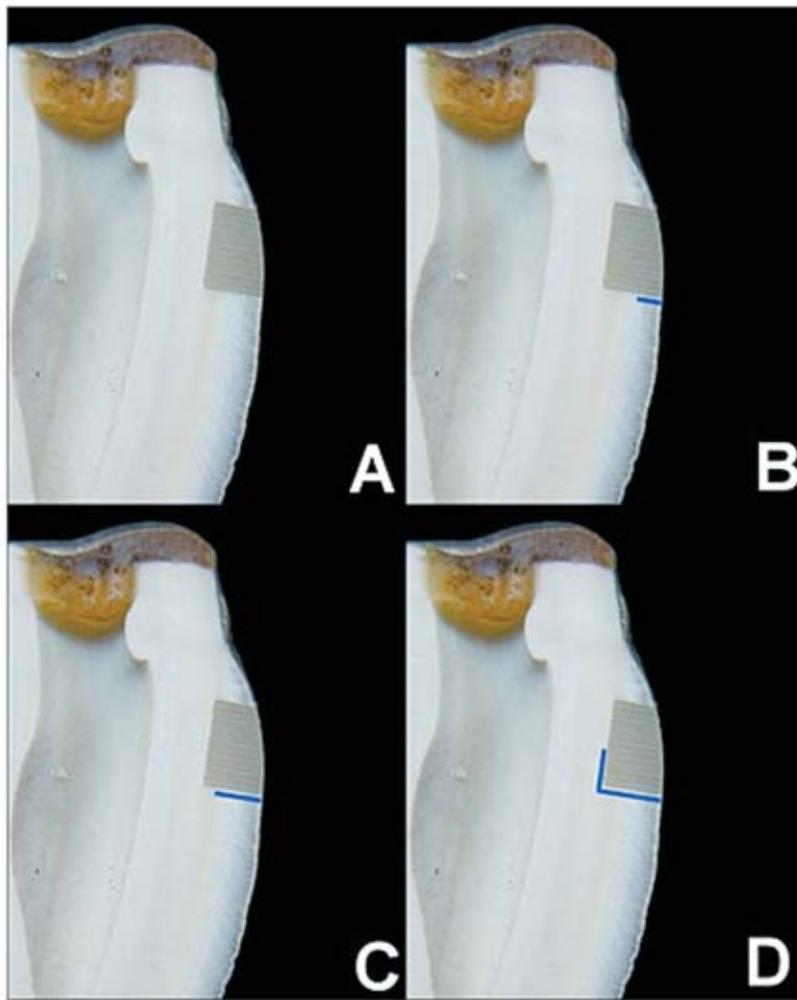


Figure 1. The graded criteria for leakage scores. Score/criterion: **(A)** 0= no dye penetration; **(B)** 1= dye penetration along the interface to the dentinoenamel junction; **(C)** 2= dye penetration into 1/2 of the dentin wall; **(D)** 3= dye penetration to over 1/2 of the dentin wall and along the axial wall.

The specimens were then thermocycled (500 cycles) between 5 and 55°C with a dwell time of one minute and three seconds of transfer time between baths. The tooth surface was then covered with two coats of fingernail polish leaving a 1-mm-wide varnish-free margin around the restoration margin. The teeth apices were sealed with epoxy resin (Araldite, Brascola, São Bernardo do Campo, Brazil) and coated with fingernail polish 1 mm short of the restoration margins in order to reduce the possibility of other leakage.

Specimens were then immersed in a 2% methylene blue solution (Fórmula & Ação, São Paulo, Brazil) for four hours (neutral pH).^{18,19} To

assess the extent of microleakage specimens were sectioned in a buccolingual direction through the middle of the restoration with a diamond disk mounted in EXTEC® Labcut 1010 Low Speed Diamond Saw (Extec Corp., Enfield, CT, USA). The most stained half of the tooth was used to evaluate the microleakage. All selected specimens were inspected under a Reichert Model No. 338 44 0 optical microscope (Reichert, Vienna, Austria) at a 40x magnification.

Each specimen was assigned a numerical score after placement into one of four categories labeled 0 to 3 on the basis of the criteria shown on Figure 1.

Incisal and gingival scores for each group of restorations were compared using the Kruskal-Wallis non-parametric statistical test to identify any statistically significant differences among the four procedures. Significance was considered at 0.05 level of confidence.

SEM Observations

Two samples were observed for each type of cavity preparation. These samples were dehydrated in increasing concentrations of ethanol to a critical point of 100% then dried and gold sputter-coated using Polaron Sc500 Sputter Coater device (VG Microtech Inc., Sussex, England). The dentin surface was examined using Jeol SEM Model JSM-5600 (JEOL Ltd, Tokyo, Japan). Photomicrographs of a representative area of the surface were taken between 1000x and 5000x magnification.

Results

None of the procedures tested in this study completely eliminated microleakage. The data showing the extent of leakage scored for the incisal and gingival portions of the restorations are shown in Figures 2 and 3.

When the scores of microleakage at the gingival margins of the four groups were compared, there were no statistical differences found ($p > 0.05$).

The same leakage occurred at the incisal margins of all groups. Analysis of the average amount of microleakage obtained for each group revealed Group 4 samples presented smaller values at the occlusal and gingival margins followed by the samples from Groups 1, 3, and 2, respectively. Comparing the gingival and the incisal surface positions in each group, no significant differences were exhibited.

Discussion

In recent studies various laser systems have been evaluated in terms of their effectiveness in cavity preparation. The most effective system is the Er:YAG laser which emits radiation in the mid-infrared region and exhibits highly efficient absorption in water and hydroxyapatite. A Er:YAG laser in combination with a cooling water spray is effective and efficient in removing hard dental tissues and causes the least amount of thermal damage. Several researchers^{20,21,22} have investigated the temperature inside the pulp chamber during Er:YAG laser irradiation and found energy densities of 112.5 J/cm² (for enamel) irradiation and 80.38 J/cm² (for dentin) are safe parameters for cavity preparation.

The Er:YAG laser produces microexplosions during hard tissue ablation resulting in macroscopic and microscopic irregularities.

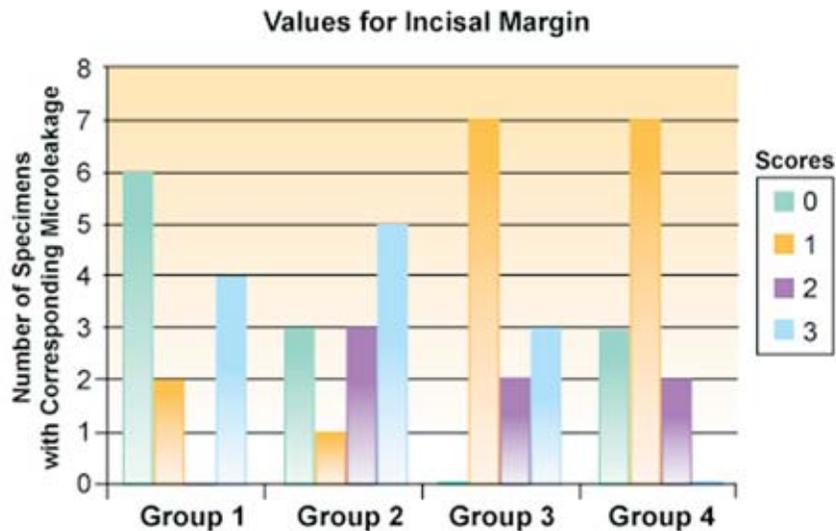


Figure 2. Distribution of specimens with the extent of microleakage on incisal margins. The heights of bars correspond to the number of specimens within each group that exhibited a specific degree of microleakage.

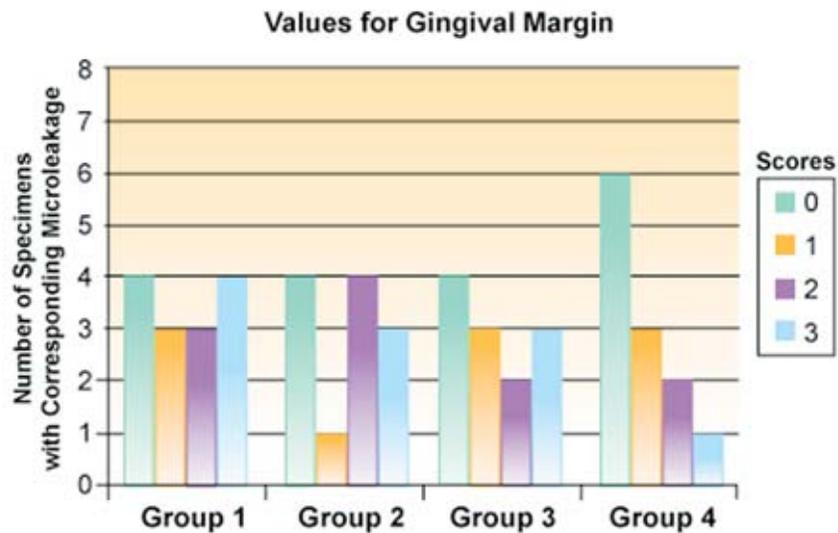


Figure 3. Distribution of specimens with the extent of microleakage on incisal margins. The heights of bars correspond to the number of specimens within each group that exhibited a specific degree of microleakage.

It initially causes vaporization of water and other hydrated organic tissue components. During vaporization, internal pressure builds up within tissue until the explosive destruction of inorganic substance occurs before its melting point is reached.²³

Using a Er:YAG laser for cavity preparation results in a surface with a significantly different morphology from the surface achieved using high speed diamond burs. Conventionally prepared enamel exhibits subsurface damage in the form of median-type cracks and microcracks.²⁴ Prepared dentin also exhibits clear grooved marks due to the rotary action of the bur which also creates a smear layer (Figure 4).

The surface configuration of irradiated dentin was smoother and more rounded with smaller pits and grooves with no smear layer and opened dentinal tubules (Figure 5).

At sub-ablative energy densities, the Er:YAG laser decreases the water content in dentin which can be partly restored later through water uptake. A relative decrease in organic tissue within dentin was detected in some studies which may indicate the Er:YAG laser selectively removes organic tissue.^{25,26} These surface alterations may affect hybridization and bonding effectiveness, even

when laser-irradiation is followed by acid-etching. This more acid-resistant surface could have reduced etching effectiveness especially when using the less acidic self-etching adhesive.

Adhesive systems have shown improved physical characteristics in the quest for materials and techniques that create a good seal between restorative materials and tooth structure, but microleakage at the tooth/material interface still occurs.

Self-etching primers combine dentin conditioning and a bonding agent in a single step. In such adhesive systems the acidic component of the primer dissolves the smear layer and incorporates it into the mixture as it demineralizes dentin and encapsulates collagen fibers and hydroxyapatite. In this way applying self-etching systems maintains the normal characteristics of dentinal tissue devoid of unprotected collagen with no increase in dentinal permeability.²⁷ However, incorporating smear layer particles into a hybrid layer is one potential disadvantage of these systems from the viewpoint of microbial content of the smear layer which could lead to pulpal infections.²⁸ This is why 2% chlorhexidine solution was applied before the bonding procedure in the present study.

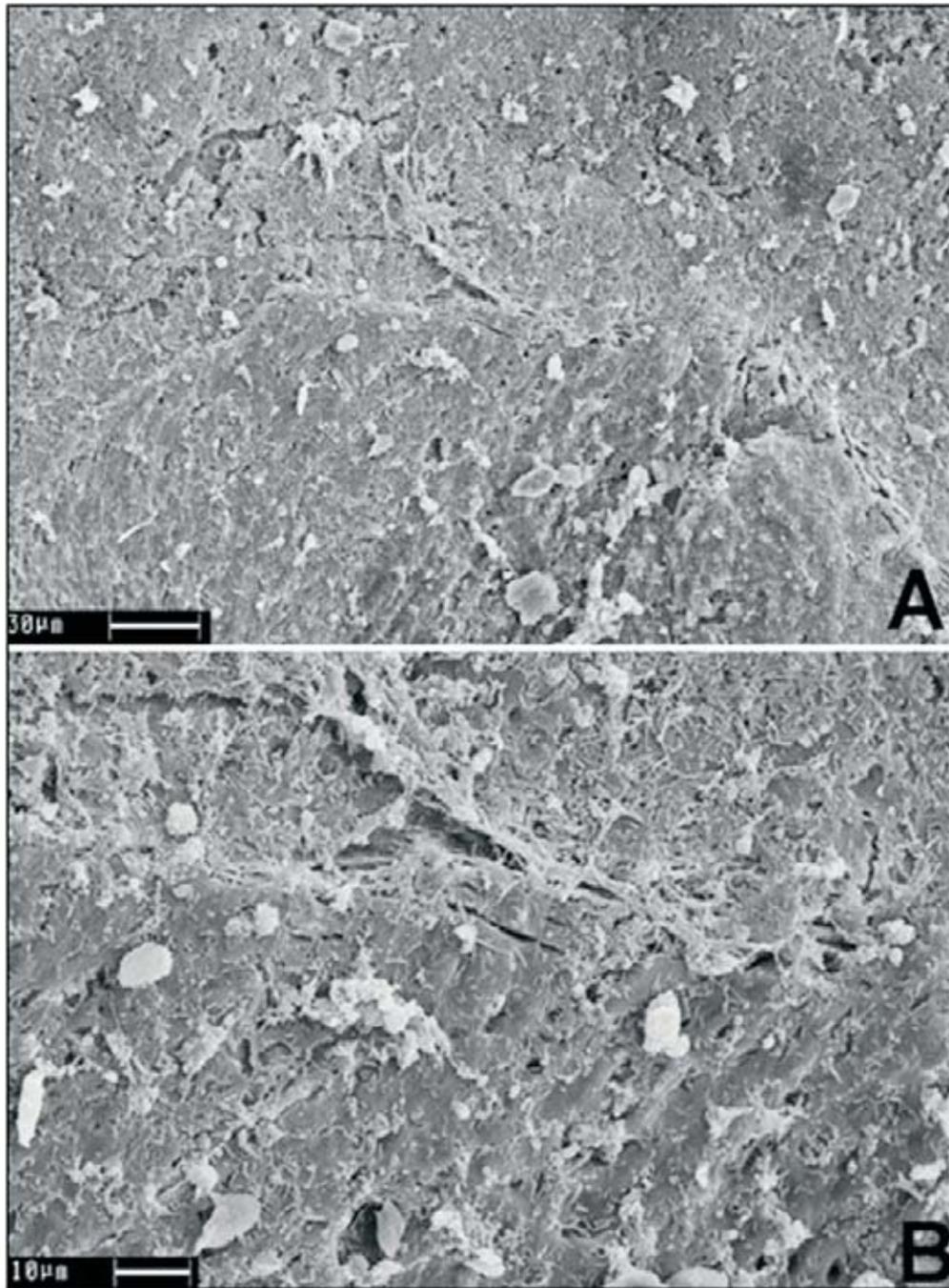


Figure 4. SEM image (A = 1000x and B = 5000x) of dentin roughened with a diamond bur. The presence of a smear layer and obliterated dentinal tubes are characteristics of this kind of preparation.

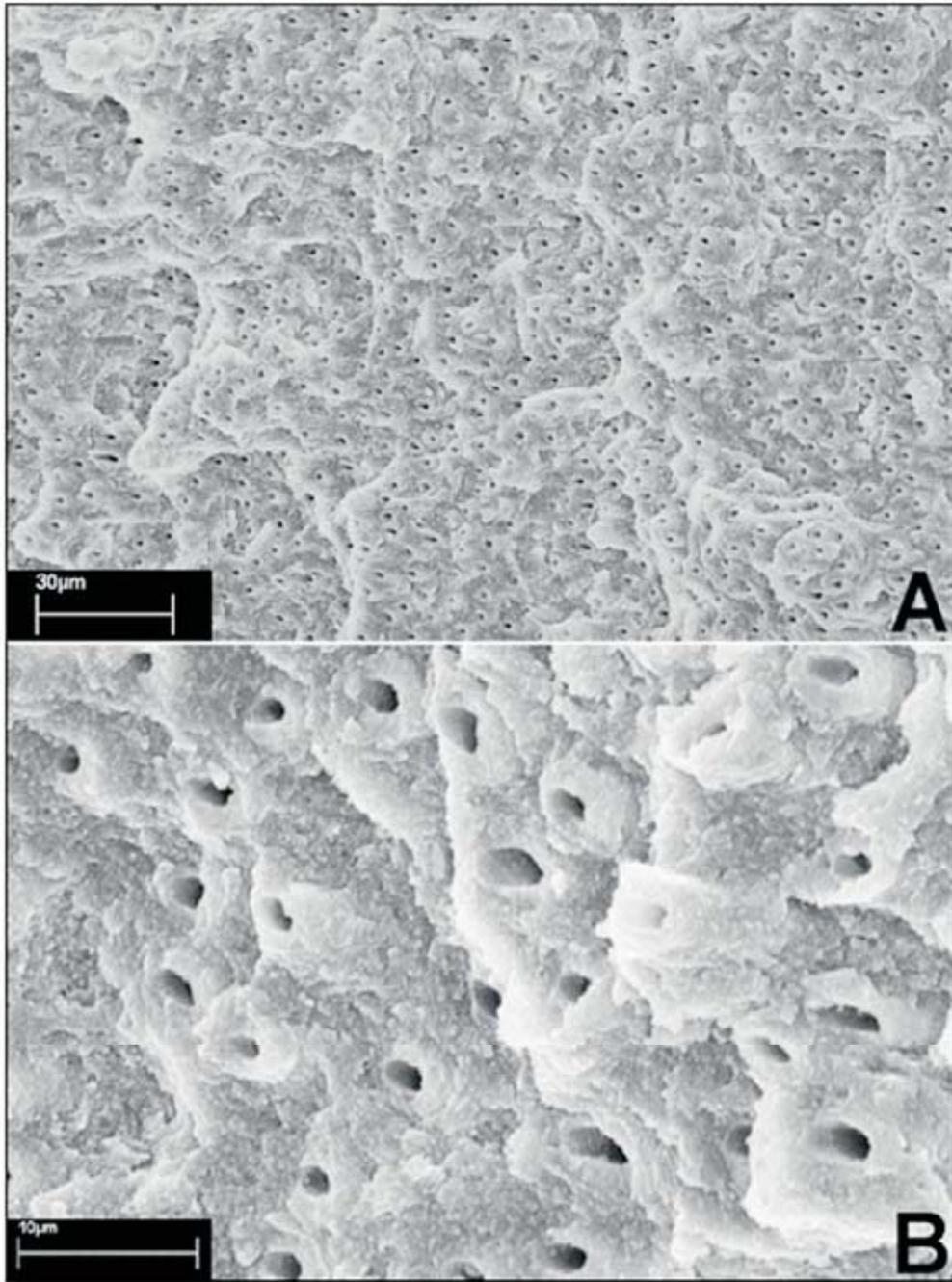


Figure 5. SEM image (A = 1000x and B = 5000x) of dentin prepared with a Er:YAG laser producing a surface with dentinal tubules clearly visible without a smear layer and a typical imbricate patterned topography.

Chlorhexidine is a broad spectrum antiseptic, and its use has been generalized over the past two decades for chemical control of bacterial plaque; the disinfection of therapeutic cavities has been adopted as a positive control for studies on bacterial growth or antibacterial activity. However, it is not known whether the application of a chlorhexidine solution in a cavity preparation influences the effectiveness of an adhesive and ultimately the marginal microleakage of resin composite restorations.

The results of the present study indicate using a cavity cleaning solution of 2% chlorhexidine prior to application of Clearfil SE Bond does not affect the sealing ability of this bonding system for both Er:YAG and bur generated cavity preparations. Until now no study has evaluated the effects of the chlorhexidine solution on the marginal sealing of resin restorations placed in cavities prepared using the Er:YAG laser.

Regarding bur preparations, Sung et al.²⁹ compared various disinfectant solutions and obtained similar results to the present study. These authors applied the antiseptic solutions after a 37% phosphoric acid etch (total etch technique) but the results were the same as the present study. Meiers and Kresin⁶ evaluated the effects of cavity disinfectants and anti-cariogenic agents on the microleakage of dental restorations. In this study a 2% chlorhexidine solution was applied prior to the use of a total etch adhesive system without affecting the dentin bonding agents' ability to prevent microleakage. These conclusions confirmed the results attained in this present study that chlorhexidine solutions do not adversely affect microleakage.

On the other hand, the results of the present study differed with previous studies. The results obtained by Owens et al.¹¹ showed the cavities disinfected by a chlorhexidine solution presented significantly less microleakage than those restored with no pre-treatment. This study used two kinds of chlorhexidine solution: one with abrasive

particles and the other without these particles. The solutions were applied after the acid etching, and the results obtained for the solution with abrasive particles indicated greater values for microleakage. According to the authors, these values could be explained by an abrasive slurry particle residue that remained in the preparations, thus, decreasing the wettability of the bonding adhesive at the dentin surface resulting in a poorer marginal seal.

Only one published study⁶ revealed an increase in microleakage using two bonding systems and chlorhexidine solution as a cavity antiseptic. Conclusions drawn from that study indicated perhaps the adverse effect of chlorhexidine was "material specific regarding the interactions with various dentin bonding systems and its ability to seal dentin."

Some studies showed a greater leakage in gingival walls.^{30,6} The authors' rationale for these findings is due to the thinner enamel margin at the gingival third than at the incisal third of the cavity preparation which allowed more leakage (frequency scores). The thickness of the incisal enamel prevented permeability resulting in a more resistant surface to dye penetration. In the present study no statistical differences were observed when the scores obtained for gingival and occlusal margins were compared which is in agreement with other studies.^{11,7}

Conclusion

The results of this *in vitro* study indicate a 2% chlorhexidine solution can be used as a cavity wash prior to the use of Clearfil SE Bond following cavity preparations with either a Er:YAG laser or diamond burs without affecting the dentin bonding agent's ability to prevent microleakage. However, more studies must be performed to elucidate the interaction between the self-etch bonding agents and chlorhexidine solutions to explain why this interaction does not affect the microleakage of resin composite restorations.

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About the Authors

Vinicius R. Geraldo-Martins, DDS, MS



Dr. Geraldo-Martins earned his dental degree from the Araraquara School of Dentistry in Sao Paulo, Brazil and a Masters of Science degree from the Energy and Nuclear Research Institute (IPEN). He is currently a PhD student in the Department of Restorative Dentistry at the University of Sao Paulo. He has worked as a Research Scholar in the Applied Molecular Photomedicine Laboratory at The Forsyth Institute in Boston, MA, USA. Dr. Geraldo-Martins is a member of the Brazilian Association of Lasers in Dentistry (ABLO). His research is focused on the use of high intensity lasers in restorative dentistry, dental materials, and caries prevention.

e-mail: vinigmartins@yahoo.com

Fabio R. P. Robles, DDS, MS



Dr. Robles earned his dental and Master of Science degrees from the University of Sao Paulo School of Dentistry in Sao Paulo, Brazil where he is currently a PhD student in the Department of Restorative Dentistry. He is a member of the Brazilian Society for Oral Research (SBPqO). Dr. Robles' research interests include use of lasers in restorative dentistry and caries prevention.

Adriana B. Matos, DDS, MS, PhD



Dr. Matos received her dental degree from the Federal University of Bahia, Brazil. She now serves as an Associate Professor in the Department of Restorative Dentistry of the School of Dentistry at the University of Sao Paulo in Sao Paulo, Brazil where she received her Master of Science and PhD degrees. Dr. Matos is a member of the Brazilian Association of Lasers in Dentistry (ABLO) and the Brazilian Society for Oral Research (SBPqO). Her research interests include the adhesion of etching systems and the use of lasers in restorative dentistry.

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